

## Comparison of TCM GeneXpert MTB/RIF Ultra Examination Results with AFB Microscopic Examination in Pulmonary TB Patients with MTB Detected

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**Abstract:** Tuberculosis (TB) is a disease in Indonesia with the second largest number of cases in the world after India. One of the TB-free strategies is early diagnosis with bacteriological laboratory examination for rapid treatment, thereby reducing the incidence rate. WHO recommends the GeneXpert molecular rapid test for confirming the diagnosis of TB. The TB diagnosis is confirmed by Acid-Fast Bacilli (AFB) microscopic examination for laboratories with difficulty accessing rapid molecular tests. The study aims to compare the rapid molecular test GeneXpert MTB/RIF Ultra examination with Ziehl-Neelsen stained AFB microscopic examination in diagnosing lung patients who have been detected with *Mycobacterium tuberculosis* very low, low, medium, and High (MTB detected)—the type of observational analytical study, with a cross-sectional design to understand the differences. The sampling technique is a total sampling of 30 samples, and data analysis using the Wilcoxon statistical test. Of the 30 samples tested for *Mycobacterium tuberculosis* (MTB), the Molecular Rapid Test identified all samples as positive (100%). In contrast, AFB microscopy detected only 23 positive cases (76.67%) and failed to detect MTB in 7 samples (23.33%). Based on bacterial load classification, three samples (10%) were categorized as very low, with AFB results showing two negatives (6.67%) and one scanty (3.33%). Among the nine low-load samples (30%), five were AFB-negative (16.67%) and four were positive (1+) (13.33%). In the seven medium-load samples (23.33%), microscopy detected five samples as 1+ (16.67%), two as 2+ (6.67%), and one as 3+ (3.33%). Of the 11 high-load samples (36.67%), AFB identified three as 2+ (10%) and eight as 3+ (23.33%). The conclusion of the results of the Molecular Rapid Test examination with AFB microscopic there shows a significant difference ( $P < 0.025$ )  $< \alpha$  (0.05). It is recommended that further research be conducted to compare three bacteriological examinations, namely rapid molecular test, AFB microscopic, and culture, as a gold standard examination.

**Keywords:** Acid-Fast Bacilli (AFB) microscopy; GeneXpert MTB/RIF Ultra; *Mycobacterium tuberculosis* (MTB) detected; rapid molecular test.

## INTRODUCTION

Tuberculosis (TB) is one of the oldest known diseases affecting humans and remains a significant public health challenge globally<sup>1</sup>. This disease is caused by *Mycobacterium tuberculosis* (MTB) and is the leading cause of death from a single

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infectious agent. According to the Global TB Report 2023, there were an estimated 10.6 million new TB cases worldwide in 2022<sup>2,3</sup>.

TB poses a high risk of transmission. A single active case of TB can infect several people before the patient receives treatment, creating a persistent reservoir of infection<sup>4</sup>. Therefore, early and accurate diagnosis is crucial for effective TB management and improving patient clinical outcomes. However, TB diagnosis still faces various challenges, especially in developing countries. Traditional diagnostic methods, such as Ziehl-Neelsen (ZN) smear microscopy, have low sensitivity and risk producing false-negative results<sup>5,6</sup>. This test also requires multiple patient visits and often fails to provide accurate results due to technical limitations. Although mycobacterial culture is considered the gold standard, it is a slow process, taking two to six weeks to obtain results. Culture also requires adequate laboratory infrastructure and technical expertise<sup>7</sup>.

As a solution to these limitations, the WHO recommended using the GeneXpert MTB/RIF test to diagnose TB and detect resistance to Rifampicin (RIF) in December 2010<sup>8</sup>. GeneXpert is a DNA-PCR-based Molecular Rapid Test that can automatically detect MTB DNA and mutations associated with RIF resistance in approximately two hours<sup>9,10</sup>. This test offers high sensitivity and specificity, especially for pulmonary TB, and is more efficient for use in resource-limited healthcare settings.

In Indonesia, TB remains a major health problem. The WHO Global TB Report 2018 noted that in 2017, there were 842,000 new TB cases in Indonesia (319 per 100,000 population), which increased to 969,000 cases (354 per 100,000 population) in 2021. With this figure, Indonesia ranks second with the highest TB burden globally after India<sup>11</sup>.

As a national effort, the Indonesian Government set a target in Presidential Regulation of the Republic of Indonesia Number 67 of 2021, namely to reduce the incidence of TB to 65 per 100,000 population and the mortality rate to 6 per 100,000 population, as well as achieving 90% detection and treatment coverage. One important strategy to achieve this target is improving early diagnosis, including through molecular rapid tests such as GeneXpert, which the WHO recommends<sup>11</sup>.

However, not all health care facilities in Indonesia have direct access to the Molecular Rapid Test. Therefore, despite its lower sensitivity, AFB microscopy with Ziehl-Neelsen (ZN) staining is still commonly used as the primary diagnostic method. In addition, pulmonary TB treatment response in Indonesia is routinely monitored using AFB microscopy with ZN staining<sup>11</sup>.

Temindung Community Health Center in Samarinda City is one of the health facilities that has access to the Molecular Rapid Test. This facility serves the Sungai Pinang Dalam and Mugirejo sub-districts. Based on SITB application data from January–July 2024, there were 66 MTB Detected and 309 MTB Not Detected results from Molecular Rapid Test examinations. Although GeneXpert is used, AFB microscopic examination is still performed in parallel for comparison and emergency situations.

Several studies have shown that GeneXpert has higher sensitivity than the ZN staining method. Murtafi'ah et al (2020) showed that GeneXpert detected 33% of positive cases from 30 samples, while ZN only detected 26%<sup>12</sup>. Relasiskawati (2020) also showed that GeneXpert detected 22 positive case patients from 182 samples, while ZN only detected 18 patients<sup>13</sup>. A similar thing was reported by Nuryaningsih E et al (2023), who found that GeneXpert detected 16.7% of positive cases, while ZN only detected 12.5%<sup>14</sup>.

However, most of these studies have not specifically examined the relationship between GeneXpert quantification levels (very low, low, medium, or high) and AFB microscopic results (negative, scanty, 1+, 2+, or 3+). Therefore, this study aims to compare the GeneXpert MTB/RIF Ultra examination results with ZN staining in pulmonary TB patients detected with MTB, to identify the agreement between the two and the potential degradation of microscopic results based on the GeneXpert detection level. The results of this study are expected to be the basis for more efficient diagnostic decision-making, especially in primary care.

## MATERIALS AND METHODS

This research is an analytical observational study with a cross-sectional approach (cross-sectional), which aims to compare the results of the Molecular Rapid Test GeneXpert method with the results of the AFB microscopic examination using Ziehl-Neelsen (ZN) staining in pulmonary tuberculosis patients with the results *Mycobacterium tuberculosis* (MTB) was detected. This research was conducted from February to April 2025 and took place in the Temindung Community Health Center Laboratory and the Samarinda City Health Laboratory.

The population in this study was all patients with GeneXpert test results indicating MTB detection during the study period. A total of 30 sputum specimens were used, collected using a total sampling technique. The independent variable in this study was the Molecular Rapid Test GeneXpert test results, while the dependent variable was the AFB microscopic examination results. Both variables were assessed ordinally based on the MTB detection level (low, medium, high) and AFB result categories (negative, scanty, 1+, 2+, and 3+).

The research instruments included laboratory equipment such as slides, flat and pointed sticks, Bunsen burners, tweezers, a light microscope, and a GeneXpert machine. The materials used included morning sputum specimens, ZN staining reagent, immersion oil, and special GeneXpert cartridges and reagents. Primary data were obtained directly from Molecular Rapid Test laboratory examinations and AFB microscopy results, while secondary data came from relevant literature and supporting documents.

This research has received permission from the research ethics commission of the Poltekkes Kemenkes Banjarmasin with certificate number: 1183/KEPK-PKB/2024. Molecular Rapid Test. The procedure followed standard methods based on WHO guidelines and the Indonesian Ministry of Health. The specimen used was morning sputum from patients who met clinical criteria. This specimen was processed by mixing the volume of specific reagent twice into a sputum pot, then shaking thoroughly and letting it sit for 10 minutes at room temperature. Afterward, the mixture was shaken and incubated for 5 minutes. The homogenization process was repeated if lumps remained until the mixture was completely homogeneous.

Next, 2 mL of the mixture is slowly injected into the GeneXpert cartridge using a special pipette. The cartridge is then inserted into the GeneXpert Molecular Rapid Test device, which is connected to the software. After the cartridge barcode is scanned and the patient data is entered according to the system format (including the NIK, laboratory registration number, and patient name), the examination is initiated by pressing the "Start" button. Test". The examination lasts for  $\pm 80$  minutes, and the system will automatically interpret the results based on the fluorescence signal.

GeneXpert results are categorized as MTB detected with gradation very low, low, medium, or high based on the cycle threshold (Ct) value, namely: very low (Ct >28), low (Ct 22–28), medium (Ct 16–22), and high (Ct <16). In addition, this tool also detects possible resistance to Rifampicin by identifying mutations in the gene *rpoB*. Inspection results are declared valid when all internal control indicators, such as Probe Check and SPC (Sample Processing Control), show "PASS" status. For inspection, microscopic AFB, the same sputum specimen was prepared into a smear on a glass slide. The smear was made with a flat stick to form an oval layer measuring approximately 2 x 3 cm, then flattened using a pointed stick. Once dry, the slide was fixed by passing it over a Bunsen flame for 1–2 seconds, 2–3 times. Staining was performed using the Ziehl-Neelsen method, namely by adding carbol fuchsin and heating it until it gives off steam (not boiling), then leaving it for 5 minutes and rinsing with running water.

The next step is decolorization using acid alcohol for 10–20 seconds until the red color fades. Then, rinse again and add methylene blue as a counterstain for 1 minute. After the final rinse, the slide is dried on a drying rack. Readings are performed using a light microscope with a 10x objective lens to find the field of view, followed by a 100x lens using immersion oil. Readings are performed horizontally on at least 100 fields of view from left to right.

Microscopic results are assessed based on the classification of the International Union Against Tuberculosis and Lung Disease (IUTLD), which divides the results into several categories: negative, scanty (1–9 bacilli per 100 fields of view), 1+ (10–99 bacilli per 100 fields of view), 2+ (1–10 bacilli per field of view on a minimum of 50 fields), and 3+ (more than 10 bacilli per field of view at a minimum of 20 fields).

The results of the Molecular Rapid Test and AFB examinations were each coded on an ordinal scale to facilitate analysis. To determine whether there were significant differences between the results of the two examination methods, a Wilcoxon statistical test was performed because the data were ordinal, paired, and not normally distributed. This analysis aimed to evaluate the level of agreement and possible degradation of results between the molecular and microscopic methods in detecting MTB in pulmonary TB patients.

## RESULTS AND DISCUSSION

This study used a total of 30 samples of MTB pulmonary TB patients and detected Rifampicin-sensitive (Rif Sen). The study was conducted for two months, from February 22 to April 27, 2025.

Table 1 shows that of the 30 respondents, the majority were male (19 people) (63.33%), while 11 were female (36.67%). Based on age group, the majority were in the 15–54 years age range (20 people) (66.67%), followed by nine people aged ≥55 years (30.00%), and only one person aged <15 years (3.33%). The type of sputum specimen obtained was mostly phlegm (26 specimens) (86.67%), while only four specimens (13.33%) contained mucus. In terms of specimen volume, 20 specimens (66.67%) had a volume of ≥3 mL, and the remaining 10 specimens (33.33%) had a volume of <3 mL.

Table 2 shows that out of 30 tested samples, the Molecular Rapid Test identified all 30 as positive, yielding a 100% positivity rate. In contrast, the AFB Microscopic Test detected only 23 positive samples, corresponding to a 76.67% positivity rate, and missed seven samples (23.33%) found positive by the molecular method.

Table 1. Characteristics of Respondents and Sputum Specimens

Variables	Category	Frequency (n)	Percentage (%)
Gender	Male	19	63.33
	Female	11	36.67
Age	< 15 years	1	3.33
	15–54 years	20	66.67
	≥ 55 years	9	30.00
Specimen Type	Phlegm	26	86.67
	Mucus	4	13.33
Specimen Volume	≥ 3 mL	20	66.67
	< 3 mL	10	33.33

Table 2. Comparison of Molecular Rapid Test and AFB Microscopic Results

Diagnostic Method	Positive Samples (%)	Negative Samples (%)	Total Samples (%)
Molecular Rapid Test	30 (100.00%)	0 (0.00%)	30 (100.00%)
AFB Microscopic	23 (76.67%)	7 (23.33%)	30 (100.00%)

Table 3. Univariate Results of AFB Microscopic

AFB Microscopic Results	Amount	Percentage
Negative	7	23.33
Scanty	1	3.33
Positive (1+)	9	30.00
Positive (2+)	5	16.67
Positive (3+)	8	26.67
Total	30	100,00

Table 4. Univariate Molecular Rapid Test Results (MTB Detected)

Molecular Rapid Test Results	Amount	Percentage
Very Low	3	10.00
Low	9	30.00
Medium	8	27.00
High	10	33.00
Total	30	100,00

Based on the results of the AFB microscopic examination in Table 3, seven samples (23.33%) showed negative results, while 1 sample (3.33%) showed scanty results. The positive results were divided into Positive 1+ in 9 samples (30.00%), Positive 2+ in 5 samples (16.67%), and Positive 3+ in 8 samples (26.67%). This indicates that the majority of samples had positive AFB results with varying degrees.

The results of the Molecular Rapid Test examination in Table 4 show that of the total 30 samples, 10 samples (33.00%) had a high bacterial load (High), eight samples (27.00%) were at the Medium level, nine samples (30.00%) were at the Low level, and

the remaining three samples (10.00%) were at the Very Low level. This indicates that most samples had MTB detection with a moderate to high load.

Table 5. Cross-tabulation of Specimen Quality Characteristics with Molecular Rapid Test Results

Characteristics	Category	Very Low n (%)	Low n (%)	Medium n (%)	High n (%)	Total n (%)
Gender	Male	2 (6.67%)	8 (26.67%)	3 (10%)	6 (20%)	19 (63.33%)
	Female	1 (3.33%)	1 (3.33%)	5 (17%)	4 (13%)	11 (36.67%)
Age (years)	<15	0 (0%)	1 (3%)	0 (0%)	0 (0%)	1 (3.33%)
	15–54	1 (3.33%)	6 (20%)	6 (20%)	7 (23.33%)	20 (66.67%)
	≥55	2 (6.67%)	2 (6.67%)	2 (6.67%)	3 (10%)	9 (30%)

Table 6. Cross-tabulation of Specimen Quality Characteristics with Molecular Rapid Test Results

Specimen Quality	Category	Very Low n (%)	Low n (%)	Medium n (%)	High n (%)	Total n (%)
Volume	≥ 3 mL	1 (3.33%)	6 (20%)	6 (20%)	7 (23.33%)	20 (66.67%)
	< 3 mL	2 (6.67%)	3 (10%)	2 (6.67%)	3 (10%)	10 (33.33%)
Type	Phlegm	1 (3.33%)	3 (10%)	5 (16.67%)	8 (27%)	17 (56.67%)
	Mucus	0 (0%)	2 (6.67%)	3 (10%)	2 (6.67%)	7 (23.33%)
	Drooling	2 (6.67%)	4 (13.33%)	0 (0%)	0 (0%)	6 (20%)

Table 5 shows the relationship between respondent characteristics and the Molecular Rapid Test MTB examination results. Of the total of 30 respondents, the majority were male (63.33%), with the most results in the category Low (26.67%) and Medium (10%). Meanwhile, women (36.67%) showed more results in the category Medium (17%) and High (13%). Based on age group, most respondents were aged 15–54 years (66.67%) and also dominated the results of the category High (20%). In the age group >54 years (30%), the distribution of results was quite even across all categories, while the age group <15 years only contributed one respondent with similar results, Low (3%).

Table 6 illustrates the relationship between specimen quality and MTB Molecular Rapid Test results. Detected Specimens with a volume of ≥3 mL (66.67%) showed a more optimal distribution of results, especially in the category Low (20%) and High (23.33%), compared to specimens with a volume of <3 mL (33.33%), which were more numerous

in the category Low (10%). Based on the type of specimen, the majority was sputum (56.67%), with dominant results in the category Medium (16.67%) and High (27%), indicating good examination quality. Mucus (23.33%) and liquid (20%) specimens tended to produce MTB detection in the lower category. This finding suggests that the success of MTB detection through Molecular Rapid Test is influenced by individual characteristics, especially age and gender, and the quality of the volume and type of specimen used.

Table 7. Comparison of Molecular Rapid Test Results and AFB Microscopic Results

Results AFB Microscopic	Result Molecular Rapid Test MTB Detected								Total	%
	Very Low		Low		Medium		High			
	n	%	n	%	n	%	n	%		
Negative	2	6.67%	5	16.67%	0	0%	0	0%	7	23.33%
Scanty	1	3.33%	0	0%	0	0%	0	0%	1	3.33%
Positive (1+)	0	0%	4	13.33%	5	16.67%	0	0%	9	30%
Positive (2+)	0	0%	0	0%	2	6.67%	3	10%	5	16.67%
Positive (3+)	0	0%	0	0%	1	3.33%	7	23.33%	8	26.67%
Total	3	10%	9	30%	8	27%	10	33%	30	100%

Table 7 shows the research data of 30 Molecular Rapid Test examination samples. GeneXpert MTB/RIF Ultra Rif Sen's positive result was found in the examination of 3 samples very low with microscopic examination results of 2 negative samples (6.67%) and one scanty sample (3.33%). Results of 9 samples were low, with microscopic examination results of 5 negative samples (16.67%) and four positive samples (1+) (13.33%). The results of 7 samples medium with microscopic examination, five samples were positive (1+) (16.67%), two samples were positive (2+) (6.67%), and 1 sample (3+) (3.33%). Results of 10 samples were high with microscopic examination, three samples were positive (2+) (10%), and seven samples were positive (3+) (23.33%).

Table 8. Wilcoxon Signed Ranks Test Results

Test Statistic	AFB Result – Molecular Rapid Test Result
Test Type	Wilcoxon Signed Ranks Test
Number of Samples (N)	30
Negative Ranks	7
Positive Ranks	0
Ties	23
Z-Value	-2.236
Asymp. Sig. (2-tailed)	0.025
Effect Size (r)	0.41 (moderate effect)
Interpretation	There is a statistically significant difference between AFB and Molecular Rapid Test results

A Wilcoxon signed-ranks test was conducted to compare the AFB and the Molecular Rapid Test results. The test showed a statistically significant difference ( $Z = -2.236$ ,  $p = 0.025$ ), with an effect size of 0.41, indicating a moderate effect. Of 30 samples,

23 had tied results, while 7 showed negative ranks, meaning the Molecular Rapid Test detected more positive cases than AFB. No positive ranks were found. These results suggest that the Molecular Rapid Test is more sensitive than AFB in detecting tuberculosis.

Based on the Molecular Rapid Test examination results presented in Table 1, of the 30 samples detected with MTB, the majority came from male patients, namely 19 (63.33%), while only 11 were female (36.67%). This finding is in line with research by Susanti D. (2013), which states that more men are found to be positive for TB than women. One of the causes is the more common smoking habit in men, which can increase the risk of pulmonary TB infection<sup>15</sup>. Research by Nisa W (2022) also showed that positive Molecular Rapid Test results were most often found in men aged 15–54<sup>16</sup>. This is thought to be related to high levels of activity and work as a productive age group, making them more susceptible to exposure to TB bacteria.

Gender and age factors have been recognized as individual risk factors in TB transmission, as stated in the Presidential Regulation of the Republic of Indonesia—Indonesian Law Number 67 of 2016 concerning Tuberculosis Control. TB prevalence surveys show that men suffer from TB more often than women, which can also be linked to smoking behavior. Smoking increases the risk of developing pulmonary TB by 2.2 times. Young adults, especially those of productive age, are the group most vulnerable to TB exposure<sup>17</sup>.

Table 1 shows that the quality of specimens examined with the Molecular Rapid Test mostly had a volume of  $\geq 3$  mL, amounting to 20 samples (66.67%). Based on specimen type, sputum specimens dominated, amounting to 17 samples (56.67%), followed by mucus, seven samples (23%), and saliva, six samples (20%). However, according to Rafika (2022), there was no significant difference between variations in sputum volume and Molecular Rapid Test results in pulmonary TB patients. Even a sputum volume as small as 0.5 mL can still detect the presence of TB. *Mycobacterium tuberculosis*<sup>18</sup>. In addition, Listyowati (2024) stated that the type of specimen, such as phlegm, mucus, or saliva, does not affect the results of Molecular Rapid Test examinations<sup>19</sup>. The ideal specimen for Molecular Rapid Test examination is phlegm with a 1–4 mL volume, greenish yellow (mucopurulent), thick, and not containing food residue or solid particles<sup>20</sup>.

However, specimens with a volume of  $< 3$  mL and in the form of saliva—5 samples (16.67%) and six samples (20%) respectively—did not meet the standards for good specimens for AFB microscopy. These negative results may be due to the number of *Mycobacterium tuberculosis* being too low in the specimen and the limited sensitivity of microscopic examination, which depends on the officer's expertise. However, microscopic examination can still be carried out on saliva specimens if the specimen has been taken repeatedly and it is noted that the specimen does not meet the requirements<sup>21</sup>. Research Aminah et al. (2017) also confirmed that specimen quality affects the results of AFB examinations, where specimens containing pus (purulent) and mucus are more likely to show positive results than saliva<sup>22</sup>.

Regarding the Molecular Rapid Test and AFB microscopic results (Table 6), two samples were found in the "very low" category and five samples in the "low" category for the Molecular Rapid Test. Still, all showed negative results for AFB microscopically. This discrepancy is likely due to the different working principles between the two methods. The



Molecular Rapid Test GeneXpert MTB/RIF Ultra test detects the presence of DNA *Mycobacterium tuberculosis* and resistance to Rifampicin using the Real-Time PCR method with high sensitivity (up to 12 cfu/mL) and an examination time of less than 80 minutes<sup>20</sup>. This examination can detect specific *rpoB* and IS1081/IS6110 genes, so that "trace" results in cases with very low bacterial counts can still be recognized, especially in patients with HIV, pediatric TB, extrapulmonary TB, and patients with a history of treatment of more than 5 years<sup>20</sup>.

In contrast, based on Ziehl-Neelsen staining, the AFB microscopic method relies on Carbol Fuchsin staining and a heating process to penetrate the lipid-rich bacterial cell wall. Although resistant to decolorization, this method has the disadvantage of requiring a high bacterial count (at least 10,000–100,000 bacteria/mL) for positive results and is highly dependent on the preparation's quality and the examiner's expertise<sup>21</sup>. Therefore, this method tends to have low sensitivity. In addition, careful handling of reagents and equipment, such as immersion oil, must be carried out to prevent cross-contamination between preparations<sup>21</sup>.

Study Murtafi'ah N.M. (2020) showed that the GeneXpert examination gave higher positive results than ZN staining<sup>12</sup>. This was supported by research by Nuryaningsih E. (2023), who found that the GeneXpert method detected more TB than ZN staining, which tended to give negative results<sup>14</sup>. Da Silva et al. (2024) also stated that the proportion of positive results on GeneXpert was significantly higher compared to the microscopic method ( $p < 0.05$ )<sup>23</sup>.

India also faces a large TB burden, accounting for about a quarter of global cases<sup>24</sup>. There, traditional methods such as ZN staining are still used because they are fast and specific, but have low sensitivity<sup>25</sup>. In contrast, molecular techniques such as GeneXpert have revolutionized TB diagnosis because they are rapid, sensitive, and specific. Their widespread availability at the district level through NTEP has expanded access to free TB diagnosis across India.[26]. Other studies have shown that GeneXpert has a much higher sensitivity than ZN, as reported by Chinedum et al. (65.7% vs 38.6%) and Bajrami et al. (29.3% vs 14.6%)<sup>26,27</sup>.

Other studies have also shown very high specificity of ZN (99.21%) but low sensitivity (84.85%), as reported by Dzodanu, Chen, and colleagues<sup>28,29,30</sup>. In contrast, GeneXpert has shown sensitivity approaching 100% and high specificity (98.81%) in several studies, with an overall accuracy of up to 99.06%<sup>31,32,33</sup>.

Of the 30 samples tested for *Mycobacterium tuberculosis* (MTB), all were positive by the Molecular Rapid Test, while 7 yielded negative results by AFB microscopy. This result confirms the conclusion that differences in the principles and sensitivity of the examination methods cause differences in results. The statistical test results showed a  $p$  value of  $0.025 < \alpha = 0.05$ , which means there is a significant difference between the results of the Molecular Rapid Test GeneXpert MTB/RIF Ultra examination and AFB microscopy in patients with MTB lung detection.

This study has several limitations. First, the sample size was limited to only 30 MTB-detected patients, making it incapable of representing a broad population. Second, quantitative analysis of microscopic specimen quality (e.g., viscosity or visual classification of sputum) was not performed. Furthermore, culture confirmation was not performed, the gold standard for validating findings from Molecular Rapid tests and microscopic examinations.

The strength of this study is the use of the GeneXpert MTB/RIF Ultra method, which has high sensitivity and rapid detection times and can provide information on rifampin resistance, which is crucial in TB control programs. These findings support the use of the Molecular Rapid Test as a primary method for rapid TB screening, particularly in facilities with limited microscopy or in high-risk populations.

Expanding the scope of research with a larger sample size and a wider variety of specimens is necessary. Adding culture methods as a comparison is highly recommended to confirm Molecular Rapid Test and AFB microscopy results. Furthermore, routine training for laboratory personnel on sputum specimen collection and handling needs to be improved to improve diagnostic quality. The government is also advised to expand access to GeneXpert to areas with high TB rates to ensure early detection and prompt and appropriate TB treatment.

## CONCLUSION

Based on the research results, it can be concluded that there is a significant difference between the results of the GeneXpert MTB/RIF Ultra Molecular Rapid Test and the microscopic examination of Ziehl-Neelsen stained AFB in MTB detected lung patients ( $p = 0.025 < \alpha = 0.05$ ), with Molecular Rapid Test showing higher sensitivity in detecting *Mycobacterium tuberculosis*, especially in the low bacterial concentration categories (very low and low). Therefore, it is recommended that the Molecular Rapid Test GeneXpert MTB/RIF Ultra examination be used as the primary method in TB diagnosis, especially for early detection and in high-risk patients. In contrast, microscopic examination remains used as a complement. Further research is recommended to compare three diagnostic methods—Molecular Rapid Test, AFB microscopy, and culture—to better understand the most effective method as the standard for TB diagnosis.

## CONFLICT OF INTEREST

In this study there is no conflict of interest

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