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COMPARATIVE ACCURACY BETWEEN CARTRIDGE-BASED NUCLEIC ACID AMPLIFICATION TEST (CBNAAT) AND SPUTUM MICROSCOPY IN HUMAN IMMUNODEFICIENCY (HIV) PATENTS

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Abstract

Background: Among individuals with HIV, the most prevalent opportunistic illness is Tuberculosis (TB). TB bacilli is detected by a CBNNAT [cartridge-based nucleic acid amplification test]. The most significant obstacle to the worldwide control of tuberculosis bacteria is still the accuracy of tuberculosis diagnosis. For diagnosis of tuberculosis, Direct sputum smear microscopy is used. To diagnose tuberculosis, The Revised National Tuberculosis Control Program (RNTCP) uses the Cartridge-Based Nucleic Acid Amplification Test (CBNAAT). The current study aims to compare the accuracy of diagnosis between CBNNAT and Sputum microscopy for pulmonary tuberculosis (TB).

Material & Methods: This prospective study was conducted between July 1, 2022, and May 31, 2023, among patients of HIV with suspicion of pulmonary TB who visited the General Medicine department at the National Institute of Medical Sciences and Research, NIMS University, Jaipur.

Results: 110 study subjects were taken. Out of them, 55.4% were male, and the most prevalent age range was middle age (31years-40 years), presented with fever (60%), weight loss (30%), 22% of patients presented with cough followed by 18% with breathlessness, 15% with only fever, and 12% with cough. Sputum microscopy through ZN staining displayed 31.2% sensitivity and 89% specificity. CBNAAT showed high sensitivity (97.9%) and specificity (98%). Compared to sputum microscopy, it was substantially more accurate (0.95 vs. 0.72, mean diff 0.23 (0.096-0.37), $p<0.001$).

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Conclusions: According to the study's findings, TB diagnosis through culture is still the gold standard; however, CBNAAT has supplanted it as the diagnostic preferred approach because of its rapid turnaround time and excellent sensitivity.

Keywords: Cartridge nucleic acid amplification test (CBNAAT), Pulmonary Tuberculosis (PTb), Sputum Microscopy, Human Immunodeficiency virus (HIV).

1. BACKGROUND

The World Health Organization (WHO) claims that in the previous 20 years, TB has remained a significant worldwide health issue within developing nations; the highest TB prevalence is found in India, about 25% of all TB cases worldwide each year. Between 2015 and 2030, the WHO End TB Strategy objectives are to reduce the number of new infections by 80% and TB-related deaths by 90%. The proposal also seeks to guarantee that no family impacted by tuberculosis incurs unmanageable expenses due to the illness. The lifetime risk of acquiring tuberculosis is 20–30 times higher in patients with TB infection who also have HIV infection.[3][4]

Tuberculosis is a common infection that affects people with HIV. Although it can occur at any point in the depletion of CD4 T cells, it is more likely to happen in the early stages when the cells are still essentially normal. If tuberculosis is diagnosed accurately and treated promptly, it is curable in HIV/AIDS patients. Since the diagnosis and treatment are complicated and involve both HIV/AIDS and tuberculosis, special attention must be given to it.

One of the biggest challenges in controlling tuberculosis worldwide is accurately diagnosing the disease. Inadequate diagnosis leads to poor patient outcomes and raises the possibility of TB transmission. Patients on antiretroviral therapy (ART) may experience increased mortality rates and immune reconstitution inflammatory syndrome as a result of delayed TB diagnosis. Moreover, it is linked to the spread of tuberculosis in hospitals and the general public. In today's world, direct sputum smear microscopy is the most popular technique for TB diagnosis.

Most national TB control programs mainly rely on sputum microscopy for diagnosing tuberculosis. For HIV-positive patients, the range of sputum microscopy sensitivity is 43–51%. The sensitivity and specificity of sputum microscopy also differ widely. Currently, RNTCP uses CBNAAT for diagnosing tuberculosis, which is more accurate than sputum microscopy. The diagnosis accuracy of sputum AFB microscopy and CBNAAT for pulmonary tuberculosis has been examined in this study.

2. MATERIAL & METHODS

Study population & area:

This prospective comparative study was conducted between July 1, 2022, and May 31, 2023, among patients of HIV with suspicion of pulmonary TB who had visited the ART centre,

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General Medicine department at the National Institute of Medical Sciences and Research, NIMS University, Jaipur.

Inclusion Criteria:

All age groups and features suspicious of Pulmonary TB in PLHIV.

Exclusion Criteria:

1. Those receiving anti-tubercular therapy for TB or HIV-infected individuals.
2. unwilling patients.

Methodology:

After obtaining written consent, each study's patients were asked for personal details, chief complaints, and previous history in predesigned performa. Patients received three wide-mouthed containers and underwent the cartridge-based nucleic acid amplification test (CBNAAT) and sputum smear microscopy. The instructions are to produce phlegm and spit it in the container without adding saliva. It is necessary to gather one early morning sample and two spot sputum samples. One of the spot samples will be used for CBNAAT testing, while the other spot and early morning samples will be used for sputum microscopy. In terms of quantitative outcomes, these were compared for sensitivity and specificity.

3. RESULTS

This study was conducted among 110 study subjects. Out of them, 61 (55.4%) were male and 49(44.6%) were female, and the most prevalent age range was middle age (31-40 years). [Table no:-1]

Table 1. Age and Gender distribution of the study participants

VARIABLES	SUB GROUP	FREQUENCY (n=110)	PERCENTAGE
Gender	Male	61	55.4%
	Female	49	44.6%
Age	Less than 18 years	19	17.2%
	18-30 years	53	48.2%
	31-40 years	20	18.2%
	40 and above	18	16.4%

A majority of the subjects we studied had a fever (60%), followed by weight loss (30%), cough (22%), and breathlessness (18%). Some patients only had a fever (15%) or cough (12%). We performed cartridge-based nucleic acid amplification testing (CBNAAT) and sputum smear microscopy for every patient. CBNAAT positivity was found in 48 (43.6%) more than sputum smear microscopy 16 (14.5%). Of all the cases, 33 (30%) were positive for CBNAAT and negative for sputum AFB. On the other hand, only 1 case (1.2%) was found to be CBNAAT negative and sputum positive, demonstrating the excellent sensitivity and specificity of CBNAAT and the false negativity of sputum smear microscopy. Most patients with a CD4 count higher than 200 had negative sputum AFB and CBNAAT results,

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concerning CD4 count no, of cases negative for CBNNAT and positive for sputum, were less [Table 2]

Table 2. Comparison of Sputum microscopy with CBNAAT & CD4 count

CORRELATION	FREQUENCY	CD4=<200/>200 cells/µl
Both CBNAAT & AFB Positive	15(13.6%)	3/12
CBNAAT Positive & AFB Negative	33(30%)	16/17
CBNAAT Negative & AFB Positive	1(0.9%)	0/1
Both CBNAAT & AFB Negative	61(55.5%)	28/33
Total	110(100%)	47/63

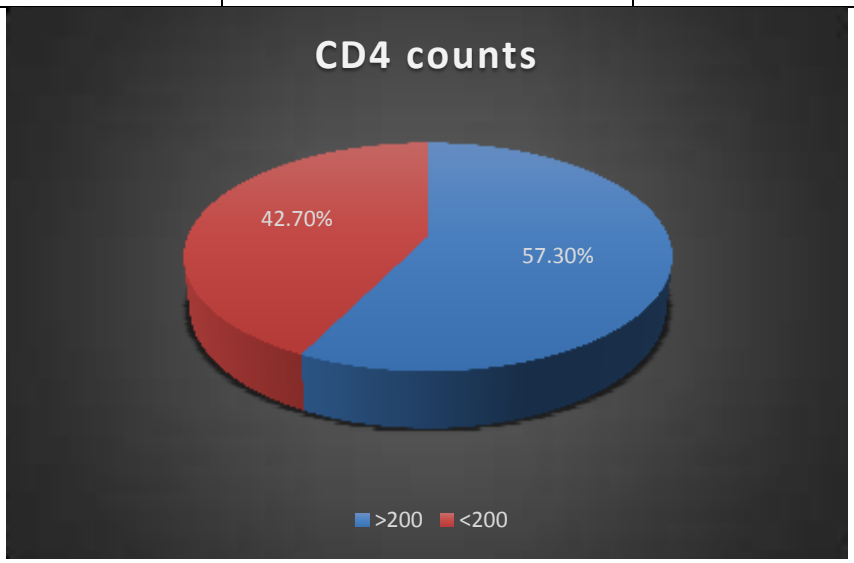

Figure 1: Distribution of CD4 count among study subjects

The sensitivity and specificity of sputum microscopy with ZN staining were 31.2% and 89%, respectively, whereas CBNAAT results showed a high sensitivity & specificity of 97.9% and 98%, respectively. CBNAAT demonstrated significantly higher accuracy than sputum microscopy with a mean difference of 0.23 (95% CI: 0.096-0.37, P<0.001).

4. DISCUSSION

Tuberculosis is a significant issue globally. The incidence of tuberculosis varies greatly within HIV-positive individuals globally, and it is rising quickly as a result of the pathogen's intensification. Early detection of Mycobacterium tuberculosis in HIV patients is crucial for prompt diagnosis and management. In this study, a total of 110 study subjects were involved. The results of sputum microscopy and CBNAAT were compared and statistically evaluated, considering CBNAAT the gold standard.

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When CBNAAT is regarded as the gold standard, Z-N staining revealed 0.9% positive cases in our investigation. In contrast, in studies conducted by Agarwal et al., [9] 2.2% of cases had positive ZN stain results, 7.3% by Jain et al.,[10] and 13.38% by Mittal et al.,[11] After adding sample reagent for homogenization, Z-N staining was performed, but Mycobacterium tuberculosis was not found. Compared to earlier studies, ours detected Mycobacterium tuberculosis in fewer cases using Z-N staining.

Our study found that the sensitivity of CBNAAT is 86%, which is higher than that of other comparable research carried out around the globe. For instance, Mittal et al. [11] found a sensitivity of 34.5% in their study conducted in Gorakhpur, India in 2017. Similarly, Basavaraj et al. [12] reported a sensitivity of 22.31% in their study conducted in Raichur, India. On the other hand, Bajrami et al. [13] found a sensitivity of 82.3% in their study conducted in Kosovo. Furthermore, according to our research, South India has less sensitivity than North India and other nations, as demonstrated in the survey conducted in Raichur [14].

In this study, out of 110 cases, only one case showed a correlation of 1.2% between the positive sputum and the negative CBNAAT, indicating that CBNAAT has almost 100% sensitivity. CBNAAT positivity was found in 48 cases (43.6%), more than the microscopy of sputum smears (14.5%). Fifteen cases showed CBNAAT and sputum smear-positive (13.6%). Sputum smear microscopy negative and CBNAAT positive were found in 33 cases (30%), sputum smear microscopy's high rate of false negative results and CBNAAT's excellent sensitivity and specificity. Previous studies have reported CBNAAT positivity in 41.9% of cases that were positive in sputum AFB, whereas 58.1% of sputum AFB results were negative (false negative). [15]16]

5. CONCLUSIONS

According to this study, even if the culture approach is still the most reliable for TB diagnosis, CBNAAT has become the preferred method because it produces results quickly and with high sensitivity. It is considered better than traditional microscopic methods such as Z-N staining. CBNAAT is a straightforward and precise technique that can correctly identify TB in HIV patients and also determine Rifampicin susceptibility, which aids in starting appropriate treatment for them.

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