JPSIM Journal of Pakistan Society of Internal Medicine

Original Article

Optimizing Tuberculosis Diagnosis: The Role of GeneXpert in Smear-Negative Cases

Haris Saeed[1], Muhammad Kashif Munir[2], Sana Rehman[2]

[1]CMH Lahore Medical College and Institute of Dentistry, Lahore, [2]Health Research Institute (HRI)-National Institutes of Health (NIH)

Abstract

Objective: To observe the efficiency of GeneXpert in confirmation of smear negative pulmonary TB cases.

Methods: This descriptive study was conducted at HRI-NIH TB Research Centre King Edward Medical University/ Mayo Hospital Lahore during 23rd December 2020 to 26th May 2021. Those smear negative patients who already started their anti-tubercular treatment and coming for follow-up were not included in this study. After taking informed consent patients suspected of having pulmonary tuberculosis on clinical and radiological grounds but negative sputum smear microscopy were asked to provide a spot sputum specimen in a sterile container and GeneXpert MTB/RIF assay was performed.

Results: A total of 116 smear negative patients consisted of 64 (55.2%) females and 52 (44.8%) males with male to female ratio of 1:1.23. Mean age of patients was 39.7±15.46 with minimum age of patients as 18 years and maximum of 72 years. Presenting complaints were also recorded; low grade fever and weight loss were present in 100% patients while fatigue was also present in 90.5% patients and anorexia in 83.6% cases. A total of 21 (18.0%) smear negative patients came back as positive for MTB on GeneXpert; of these 13/21 (61.9%) were reported as showing very low quantity and remaining 8/21 (38.1%) showed low quantity while none of the patients showed Rifampicin resistance on GeneXpert.

Conclusion: Diagnostic delay of smear negative pulmonary TB could be improved by using GeneXpert as 18% smear negative TB cases were confirmed by using this technique in this study which ultimately can prevent considerable morbidity and mortality.

Keywords: Tuberculosis, GeneXpert, Early diagnosis, AFB smear.

How to cite this:

Saeed H, Munir MK, Rehman S. Optimizing Tuberculosis Diagnosis: The Role of GeneXpert in Smear-Negative Cases. J Pak Soc Intern Med. 2025;6(2): 123-127

Corresponding Author: Dr. Haris Saeed **Email:** harissaeed15@gmail.com

Received: 15.10.2024 **Revised:** 05.01.2025 **Accepted:** 15.05.2025

DOI: https://doi.org/10.70302/jpsim.v6i2.2524

Introduction

Mycobacterium tuberculosis complex (MTBC) is a group of infectious agents which are responsible for causing tuberculosis (TB) which is one of the 10 topmost causes of deaths due to a single agent.[1] The disease of TB is considered to be very ancient and has been found in the skeletal of old mummies. One of the major challenges of TB is that it spreads through air born droplets followed by attack on immune-compromised and vulnerable groups to cause pulmonary and or extra-pulmonary TB.

Prevalence of pulmonary TB is as high as >70% which is characterized by infection of TB bacilli in the lungs though it can cause infection in each and every part of body to cause extra-pulmonary TB.[2] World Health Organization (WHO) has currently reported a high global incidence of TB as 118-146 cases/100,000 populations and presented around 10 million new TB cases during 2018. Pakistan remained amongst top ten countries having maximum load of patients with an incidence of 265 patients per 100,000 populations with a burden of 562000 new TB cases in this period.[1]

Timely diagnosis and prompt treatment of TB cases is the key to success in terms of cure and further prevention from TB.[3] Observation of acid-fast bacilli

(AFB) smear microscopy is the basic tool of diagnosis and patients diagnosed with AFB are immediately put on anti-tubercular treatment (ATT) and further investigations like monitoring of drug resistant TB are processed side by side.[4]

Smear microscopy for AFB has a very good specificity but lacks sensitivity[5] which creates various problems for the patient and physicians at times. An older study in Netherlands revealed that around 42% smear negative pulmonary TB patients are being enrolled for treatment, although smear negative patients are less infectious as compared to smear positive pulmonary TB patients[6] but reports showed that smear negative TB patients are responsible to spread around 20% of total TB cases.[7]

Diagnosis of smear negative TB is challenging while a good number of TB infections are noted to be smear negative and empirical therapy is started on the basis of clinical presentation, general characteristics of patients and respiratory symptoms.[8] Culture of MTB is the only gold standard for definite diagnosis of pulmonary TB but delay the treatment considerably due to slow growth and other related factors.[5] GeneXpert MTB/RIF assay has proven to show a good sensitivity and specificity in diagnosis of pulmonary TB. Therefore, present study was undertaken to observe the efficiency of GeneXpert in conformation of smear negative pulmonary TB cases.

Methods

This descriptive study was undertaken at HRI-NIH TB Research Centre, King Edward Medical University/ Mayo Hospital Lahore during 23rd December 2020 to 26th May 2021. A sample size of 116 smear negative patients were calculated by taking confidence level of 95%, precision as 9% and expected prevalence of smear negative TB patients as 42%[6]. New smear negative pulmonary TB patients enrolled for treatment in outpatient's department of either gender, aged eighteen and above were included in this study. Smear negative patients already started their anti-tubercular treatment and coming for follow-up were not included in this study.

After taking an informed consent patient were asked to provide a spot sputum specimen in a sterile container. A semi-structured Performa was designed to collect data of each patient consisting of demographic characteristics, presenting complaints and history of treatment and contact. Sputum sample was used to prepare a new AFB smear and processed for GeneXpert MTB/RIF Assay as per prescribed protocol.[5] Smears were stained using auramine staining technique and observed under fluorescent microscope. Data analysis was done by using statistical package for social sciences (SPSS) software.

Results

A total of 116 smear negative patients consisted of 64 (55.2%) females and 52 (44.8%) males with male to female ratio of 1:1.23. Mean age of patients was 39.7±15.46 with minimum age of patients as 18 years and maximum of 72 years. Most of the patients (49.2%) were married and belonged to younger age groups. About half of the patients (50.8%) were illiterate and 90.5% patients' belonged to poor or lower middle socio-economic status. History of TB contact was established among 37.1% patients and previous history of anti TB treatment was also present among 19.8% patients as described in table 1.

Presenting complaints and further investigations of smear negative patients were also recorded where low grade fever and weight loss were present among 100% patients while fatigue was also present among 90.5% patients and anorexia among 83.6% cases. Chest x-ray of patients showed mild infiltration, moderate infiltration and calcification in 40.5%, 11.2% and 16.4% cases respectively as shown in table 2.

Table 2: *Presenting complaints and X-ray findings of Patients. (N = 116)*

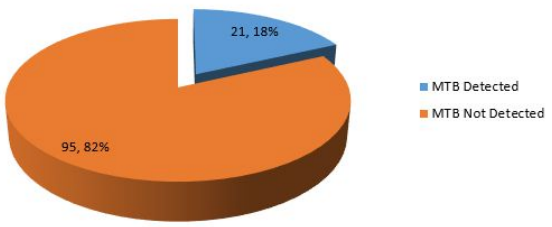
Complaints & Investigations		N	%
Low Grade Fever		116	100
Fatigue		105	90.5
Cough		85	73.3
Weight Loss		116	100
Chest Pain		42	36.2
Expectoration		36	31
Anorexia		97	83.6
Breathlessness		26	22.4
Chest x-ray Presentation	Mild infiltration	47	40.5
	Moderate Infiltration	13	11.2
	Cavitation	-	-
	Calcification	19	16.4
	Undefined	37	31.9

Table 1: *Demographic Characteristics and History of Patients.*

Demographic Factors	Characteristics	Mean±SD	N	%
Gender	Male		64	55.2
	Female		52	44.8
Marital Status	Married		57	49.2
	Unmarried		42	36.2
	Others*		17	14.6
Age in years	Mean	39.7±15.46		
	Range	18-72		
Age Classes	<25		38	32.8
	26-35		29	25
	36-45		21	18.1
	46-55		13	11.2
	56-65		9	7.7
	66+		6	4.5
	Range	14-Apr		
Education	Illiterate		59	50.8
	Primary		11	9.5
	Middle		9	7.8
	Matriculation		24	20.7
	Intermediate & Above		13	11.2
Socio Economic Status	Poor class		51	43.9
	Lower middle class		54	46.6
	Upper middle class		8	6.9
	Higher class		3	2.6
History of Smoking	Smokers		47	40.5
	Non smokers		69	59.5
Previous History of Anti TB Treatment			23	19.8
History of TB Contact	Present		43	37.1
	Not Established		73	62.9

A total of 21 (18.0%) smear negative patients remained positive for MTB on GeneXpert as depicted in figure I. Of these 13/21 (61.9%) were reported to be in very low quantity and remaining 8/21 (38.1%) were in low quantity among provided samples, none of the patient showed Rifampicin resistance on GeneXpert.

Figure I: Smear Negative Findings on GeneXpert



Discussion

The eventual target of TB control program is to reduce the MTB transmission by early detection and treatment of TB patients to prevent further spread. There are various tools and measures working together in this regard including contact & risk groups

screening, provision of free diagnostic and treatment facilities and follow up of defaulter TB patients to increase the vigilance of program.[3,9] Though, it is pertinent that improved detection of TB reduces the delay in treatment of patients. Although AFB smear has low sensitivity but its specificity, simplicity, cheapness and promptness in diagnosis could not be denied therefore still used as basic diagnostic tool for TB in developing countries.[10,11]

Benefits of GeneXpert MTB assay technique include higher sensitivity, specificity, delivery of results in two hours, non-infectious procedure and availability of rifampicin susceptibility within the same sample.[12] Presently 18.0% smear negative patients remained positive for MTB on GeneXpert while none of the patient showed Rifampicin resistance in this study. Results are though in accordance with the study which reported 14.2% AFB smears negative cases which were found to have MTB by GeneXpert technique.[13] Major sign and symptoms among patients remained fever (100%), weight loss (100%), cough (73%) and anorexia (83.6%) are comparable with another study focussed on diagnosis of smear negative pulmonary patients and reported these symptoms 91%, 97%, 100% and 80% respectively.[7]

Role of chest x-rays could not be denied which showed the pictures suggestive to have TB among 68.1% cases in this study and are not concomitant with former study which reported even higher rate 93% among smear negative pulmonary TB patients[7]. An older study from same settings has also reported 87.3% cases presenting changes in their chest x-rays.[14] Family history of TB contact, previous history of anti TB treatment, poor socio-economic status and low literacy rate are the related factors of occurrence of disease and are shown to be in agreement with previous studies.[4,9]

Smear positive TB patients although more infectious but are put on anti TB treatment at earliest as compared to smear negative patients as they require further investigations, observations and empirical treatment before starting the full course of TB therapy.[7] There is rigorous need to improve the diagnosis as early as possible for the sake of early treatment which may prevent the patient to become smear positive and decrease misery. Reports have shown that many patients are misdiagnosed as smear negative TB on the basis of sign & symptoms, radiography and clinical features while patients actually suffer from pneumonia, lung cancer and interstitial lung disease.[15,16]

In conclusion, diagnostic delay of smear negative pulmonary TB could be improved by using GeneXpert as is the case in present study where around 18% smear negative TB cases were confirmed by this technique which ultimately can prevent considerable morbidity and mortality.

Ethical Approval: The IRB/EC approved this study via letter no. 928/RC/KEMU Dated December 9, 2020.

Conflict of Interest: *None*

Funding Source: *None*

Authors' Contribution

HS: Conception

MKM: Design of the work

SR: Data acquisition, analysis, or interpretation

MKM, SR: Draft the work

HS: Review critically for important intellectual content

All authors approve the version to be published

All authors agree to be accountable for all aspects of the work

References

1. WHO Global tuberculosis report 2019. [Updated October 2019, Cited January 2020], Geneva, Licence: CCBY-NC-SA3-0IGO.available from website: [https://apps.who.int/iris/bitstream/-handle/10665/329368/9789241565714-eng.pdf?ua=1]
2. Ali A, Munir MA, Sarwar MI, Khaliq S, Munir MK, Rehman S. Tuberculous Pleural Effusions: Efficacy Utilization and Comparison of Various Diagnostic Techniques. Pak J Med Health Sci. 2018;12(4): 1461-4.
3. Munir MK, Rehman S, Iqbal R. Meeting the challenge, making a difference: multidrug resistant tuberculosis in Pakistan. Pak J Med Res. 2018;57(1): 1-2.
4. Munir MK, Rehman S, Iqbal R, Saeed MS, Aasim M. Comparison of gene Xpert MTB/RIF assays with conventional standard proportion method for determination of drug susceptibility in multidrug resistant TB suspects. Ann KEMU. 2018 ;24(1):570-6.
5. Munir MK, Rehman S, Aasim M, Iqbal R, Saeed S. Comparison of Ziehl Neelsen microscopy with GeneXpert for detection of Mycobacterium tuberculosis. IOSR JDMS. 2015;14(11):56-60.
6. Tostmann A, Kik SV, Kalisvaart NA, Sebek MM, Verver S, Boeree MJ et al. Tuberculosis transmission by patients with smear-negative pulmonary tuberculosis in a large cohort in the Netherlands. Clin Infect Dis. 2008;47(9):1135-42.

7. Linguissi LS, Vouvoungui CJ, Poulain P, Essassa GB, Kwedi S, Ntoumi F. Diagnosis of smear-negative pulmonary tuberculosis based on clinical signs in the Republic of Congo. BMC Res Notes. 2015;8(1):804.
8. Shital P, Anil J, Sanjay M, Mukund P. Tuberculosis with diabetes mellitus: clinical-radiological overlap and delayed sputum conversion needs cautious evaluation-prospective cohort study in Tertiary Care Hospital, India. J Pulm Respir Med. 2014;4(2):1-5.
9. Rehman S, Kashif Munir M, Iqbal R, Ahmed Salam A, Saeed S, Masud F et al. Active Case Detection Among Household Contacts of Multi Drug Resistant Tuberculosis Patients in a Tertiary Care Setting. Pak J Med Res. 2014;53(3).
10. Golub JE, Mohan CI, Comstock GW, Chaisson RE. Active case finding of tuberculosis: historical perspective and future prospects. Int J Tuberc Lung Dis. 2005;9(11):1183-203.
11. Rehman S, Iqbal R, Munir MK, Salam AA, Saeed S. Incremental yield of submitting three sputum specimens for the diagnosis of pulmonary tuberculosis. Pak J Med Res. 2013;52(2):35-8.
12. Shinnick TM, Starks AM, Alexander HL, Castro KG. Evaluation of the Cepheid Xpert MTB/RIF assay. Expert Review Mol Diag. 2015;15(1):9-22.
13. Marlowe EM, Novak-Weekley SM, Cumpio J, Sharp SE, Momeny MA, Babst A et al. Kawamura M, Pandori M. Evaluation of the Cepheid Xpert MTB/RIF assay for direct detection of Mycobacterium tuberculosis complex in respiratory specimens. J Clin Microbiol. 2011;49(4):1621-3.
14. Shabbir I, Nazir A, Iqbal R, Khan SU, Munir K. Clinico-pathological profile and treatment outcome in smear negative pulmonary tuberculosis patients at a teaching hospital. Pak J Med Res. 2010;49(2):28-31.
15. Laushkina Z, Filimonov P. Misdiagnosed pulmonary TB: Influencing factors and diagnostic chances in TB hospital. Eur Resp J. 2012;40(1):2702.
16. Akhter N, Rizvi NA. Interstitial Lung Diseases misdiagnosed as tuberculosis. Pak J Med Sci. 2018;34(2):338.