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## RESEARCH ARTICLE

# ROLE OF CBNAAT (CARTRIDGE BASED NUCLEIC ACID AMPLIFICATION TEST) IN THE DIAGNOSIS OF TUBERCULAR PLEURAL EFFUSION

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**ABSTRACT**

**Introduction:** Tuberculosis (TB) has existed for millennia and remains a major global health problem. Among extra-pulmonary tuberculosis, lymph node tuberculosis is the most common type constitutes about 35%cases followed by pleural effusion (20%), bone and joint (10%), genitourinary TB (9%), TBM (5%), other (13%). Cartridge based nucleic acid amplification test (CBNAAT), specific for Mycobacterium tuberculosis has been recently introduced for detection of tubercular pleural effusion. It has an added advantage of detecting rifampicin resistance. **Material and methods:** With an aim to determine the role of pleural fluid CBNAAT in tubercular pleural effusion, the study was conducted in department of Internal medicine, R.N.T. Medical College Udaipur, Rajasthan. Patients with symptoms suggestive of pleural effusion were enrolled. Pleural fluid was drawn using standard procedure protocol and sent for CBNAAT test and routine as well as bacteriological examination. **Results:** 200 patients were included, with male to female ratio of 3.5:1. The most common affected age group was below 45 years and mostly from rural area (84%). 114 (57%) of 200 patients were underweight. Mostly were farmers and most were smoker. Most common morbidity was DM followed by COPD, HTN and HIV. In our study, most common symptom was chest pain. 52 patients had pulmonary involvement and 28 were sputum AFB positive. Pleural fluid CBNAAT was positive in 37 patients out of which 31 were sensitive to rifampicin. **Conclusion:** While the cytology of Pleural Fluid gives a very good estimation of the positivity status of a patient than CBNAAT as the higher cell count is suggestive of TB but NAAT helps in diagnosing both the positivity status as well as rifampicin resistant state of the patient. To conclude, CBNAAT has the potential to significantly improve and escalate the diagnosis of pleural fluid specimens at both hospitals as well as point-of-care settings in regions not only with high TB burden but also with overlapping HIV. Also, detection of Rifampicin resistance aids in prompt initiation of appropriate therapy and thus improving the overall quality of TB care.



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## INTRODUCTION

Tuberculosis (TB) has existed for millennia and remains a major global health problem. In 2015, there were an estimated 10.4 million new (incident) TB cases worldwide, of which 5.9 million (56%) were among men, 3.5million (34%) among women and 1.0 million (10%) among children (WHO 2016). Most of the estimated number of cases in 2015occurred in Asia (61%)and the WHO African Region (26%); smaller proportions of cases occurred in the Eastern Mediterranean Region (7%), the European Region (3%) and the Region of the Americas (3%). The six countries that stood out as having the largest number of incident cases in 2015 were (in descending order) India, Indonesia, China, Nigeria, Pakistan and South Africa (combined, 60% of the global total). The incidence rate in India was 217 (112-355) per lakhpopulation in 2015, according to WHO global report 2016. The best estimate is that there were 1.4 million TB deaths in 2015, and an additional 0.4 million deaths resulting from TB disease among HIV positive people. Globally, the absolute number of TB deaths (excluding TB deaths among HIV-positive people) and the TB incidence rate has fallen since 2000. The number of TB deaths fell from 1.8 million in 2000to 1.4 million in 2015. However, the global rate of decline in the TB incidence rate was only 1.5% from 2014 to 2015 and the CFR in 2015was 17% (WHO, 2016). Pulmonary TB is the most important clinical manifestation, as it is the most common presentation and practically the only form of the disease that is infectious. While pulmonary tuberculosis is the most common presentation, extra-pulmonary tuberculosis (EPTB) is also an important clinical problem (Fanning, 1999; Iscman, 2000). Extra pulmonary TB represented 15% of the 6.1 million incident cases that were notified, ranging from 8% in the WHO Western Pacific Region to 23% in the Eastern Mediterranean Region (WHO 2016). The percentage of patients with EPTB in tertiary carecenters in India was between 30% and 53%, while the percentage estimated by the national control program in

India for HIV-negative adults are between 15% and 20%. Among extra-pulmonary tuberculosis, lymph node tuberculosis is the most common type constitutes about 35% cases followed by pleural effusion (20%), bone and joint (10%), genitourinary TB (9%), TBM (5%) abdominal tuberculosis (3%), other (10%) (Sharma and Mohan, 2004). The diagnosis of EPTB is challenging because of paucibacillary in nature, lack of specific sign and symptoms and often negative acid-fastbacilli smear of biological specimens. Indirect methods like tuberculin skin test and interferon gamma release assay are adjunctive diagnostic tools but it may be negative in presence of disease. In developing countries like India where tuberculosis is highly endemic, tuberculin skin test and gamma interferon result alone are not enough evidence todiagnose EPTB (Kumar, 2001; Sharma *et al*., 2002; Singh *et al*., 1969; Kumar and Muralidhar, 1999; Rooney *et al*., 1970; Biehl, 1958). Histo-cytological examination has its limitations as it cannot differentiate between TB and other related diseases like sarcoidosis or non-tubercularmycobacterial. Serological assays including antigen and antibody detectionwhich were used very frequently in past had a reputation of creating morediagnostic confusion than solving the problem and lead to wide spread misleading results; these tests were very rightly banned by the government of India. Other tests that are also being employed for the diagnosis of EPTB include tuberculin test and polymerase chain reaction (PCR) assays; however, thes specificities and sensitivities of these tests are variable. Although the culture remains the gold standard for the diagnosis of the tuberculosis but it can take up to 8-10 weeks using a solid media.

Rapid identification is essential for initial treatment initiation, improved patient outcome as well as for more effective public health intervention and it relies mainly on Nucleic acid amplification techniques (NAAT) (Pai and Ling, 2008). Cartridge based nucleic acid amplification test (CBNAAT), specific for Mycobacterium tuberculosis has been recently introduced for detection of TB. It has an added advantage of detecting rifampicin resistance as it targets the rpoB gene of mycobacteria, which is the critical gene associated with rifampicin resistance. The Gene Xpert MTB/RIF (Cartridge Based Nucleic Acid Amplification Test CBNAAT) is an automated real time polymerasechain reaction (PCR) assay designed for the rapid and simultaneous detection of Mycobacterium tuberculosis and Rifampicin resistance within 2 hours (Helb *et al*., 2010; Boehme *et al*., 2010; Van Rie *et al*., 2010). Based on systematic review, (Claudia *et al*., 2014) WHO recommends Xpert over conventional tests for diagnosis of EPTB which permits rapid TB diagnosis through detection of the DNA of mycobacterium TB and simultaneous identification of a majority of the mutations that confirm Rifampicin resistance which is highly predictive of MDR TB. The original WHO policy guidance on Xpert MTB/RIF issued in2010 recommends its use as the initial diagnostic test in individuals suspected of having MDR-TB or HIV-associated TB (strong recommendations). A policy update in 2013 expanded its recommended uses, including for the diagnosis of TB in children, on selected specimens for the diagnosis of EPTB, and for all individuals suspected of having pulmonary TB (conditional recommendations).

## MATERIAL AND METHODS

This is a hospital based prospective study conducted from Jan 2017 to Jan, 2018, on 200 patients. Patients who were attended department of General Medicine R.N.T. Medical College, Udaipur with signs and symptoms, and radiological examination suggestive of tubercular pleural effusion included in this study. Their detailed clinical history, demographic profile, socioeconomic status and anthropometric data, contact number and consent was taken and recorded. Previous history of tuberculosis, history of contact with pulmonary tuberculosis, past history of medical illness and h/o co-morbid illnesses were also taken. General physical examination as well as complete systemic examination was done carefully with more emphasis on involved system. A fresh digital chest radiograph was advised to study population with suspected pleuraleffusion, hydro-pneumothorax or pyo-pneumothorax. Sputum samples from study population, who had cough for any duration, were sent for AFB examination by light microscopy under RNTCP. Afterclinico-radiographic suspicion, pleural effusion was aspirated. After pleural aspiration, a check X-ray was done to see the parenchymal involvement which was curtained by pleural effusion. Pleural fluid was sent for sugar, protein, cell count, cell type, ADA, gram stain and culture. 2 mlof pleural fluid or pus was sent for Gene X-pert. Based on MTB result, the study population were divided into 'MTB detected' and 'MTB not detected' groups. MTB detected group was further divided into two sub groups i.e. 'Rif' Resistant and 'Rif' sensitive. All the collected information was filled in predesignedproforma in excel sheet for final analysis.

**Inclusion criteria**

- Patients of both sex (>18 years) admitted in RNT Medical College and attached hospital.
- 2. Medical history compatible with tubercular pleural effusion.
- 3. Pleural effusion by clinical examination**,** chest x ray, USG.

**Exclusion criteria**

- Age ≤18 years.
- Patient's refusal for pleural fluid aspiration.
- Transudative pleural effusion.
- Contraindication to thoracentesis.
- Patients on anti-tubercular therapy.

## RESULTS

Males predominantly affected by tubercular pleural effusion were included in this study. Male to female ratio was 3.5:1. Overall, 168 (84%) patients were from rural areas and 32 (16%) patients were from urban areas. 113 (56.5%) patients in this study were below 45 years of age and 87(43.5%) patients were above the age of 45 years. 114 (57%) patients were underweight, 75 (37.5%) had normal BMI and 11 (5.5%) were overweight. In this study, 126 (63%) patients were literate and 74 (37%) patients were illiterate. Out of 200 patients, 66 (33%) were farmer, 32 (16%) were skilled worker, 24 (12%) were semi-skilled worker, 14 (7%) were professional, 22 (11%) were student and 42 (21%) were housewife (Table 1). In our study, 126 (73%) patients were from the upper-class while 74 (37%) were from lower class. 135 patients had current substance abuse. Among these, Smoker was most prevalent followed by tobacco chewer (Table 2). Most common morbidity was DM followed by COPD, HTN and HIV (Table 3). In our study, most common symptom is chest pain, followed by cough, anorexia and fever (Table 4). Most

common physical finding was anaemia followed by clubbing and lympha denopathy. Out of 200 patients diagnosed as tubercular pleural effusion had pulmonary lesions in chest radiograph in 52 (26%) patients. Out of 52 Patients with pulmonary lesions in their chest radiograph, most common finding was infiltrations in lung followed by consolidation, cavitation (Table 5). Interestingly 5 patients had pyo-pneumothorax. In this study of 200 patients only 28 (14%) cases had positive AFB sputum and 172(86%) cases had negative AFB status. In this study of 200 patients, maximum 66(33%) cases pleural fluid protein level was in range of 4.5-5.5 mg/dl mg/dl. Mean pleural fluid protein level was 3.6 mg/dl and S.D. for this was 1.01 (Table 6). In this study of 200 patients complete pleural fluid analysis and pleural fluid cell counts were done. In majority of patients, cell counts were lymphocytic pleocytosis (Table 7). In this study ADA level in pleural fluid was analysed and observed that majority of the patients had pleural fluid ADA level in range of 40-100 U/Litre (Table 8). CBNAAT was able to detect MTB in 37 (18.5%) different extra pulmonary TB samples out of 200 samples subjected to this test. CBNAAT was more successful in detection of MTB in patients with tubercular pleural effusion who had pulmonary involvement in their chest radiograph and sputum AFB positive patients. 'R' resistant was also detected in those patients (Table 9 & 10).

**Table 1. Distribution of Study Population according to their Occupation (n=200)**

| Occupation | Frequency | % |
|---|---|---|
| Farmer | 66 | 33.0 |
| Housewife | 42 | 21.00 |
| Student | 22 | 11.0 |
| Semi-skilled | 24 | 12.0 |
| Skilled | 32 | 16.0 |
| Professional | 14 | 7.0 |
| Total | 200 | 100 |

**Table 2. Distribution of study population according to their current substance abuse (n=135)**

| Substance abuse | Frequency | % |
|---|---|---|
| Smoker | 22 | 16.3 |
| Alcoholic | 7 | 5.2 |
| Tobacco chewer | 19 | 14.1 |
| Smoker + Alcoholic | 30 | 22.2 |
| Smoker + Tobacco chewer | 24 | 17.8 |
| Smoker + Alcoholic + Tobacco chewer | 33 | 24.4 |
| Total | 135 | 100 |

**Table 3. Co-morbidities in study population (n=43)**

| Comorbid Illness | Frequency | % |
|---|---|---|
| COPD | 13 | 30.2 |
| COPD + HTN | 3 | 7.0 |
| DM | 12 | 28.0 |
| DM + HTN | 5 | 11.6 |
| HTN | 6 | 14.0 |
| HIV | 4 | 9.2 |
| TOTAL | 43 | 100 |

**Table 4. Various symptoms in the study population**

| Symptoms | Frequency | % |
|---|---|---|
| Cough | 141 | 70.5 |
| Expectoration | 60 | 30.0 |
| Dyspnoea | 76 | 38.0 |
| Chest pain | 143 | 71.5 |
| Haemoptysis | 16 | 8.0 |
| Fever | 116 | 58.0 |
| Anorexia | 125 | 62.5 |
| Weight loss | 114 | 57.0 |
| Night sweats | 88 | 44.0 |

**Table 5. Distribution of Tubercular Pleural Effusion with Pulmonary Involvement according to radiological presentation (N=52)**

| Radiographic Presentation | Frequency | % |
|---|---|---|
| Infiltration | 22 | 42.3 |
| Infiltration + Cavity | 9 | 17.3 |
| Infiltration + Consolidation | 16 | 30.8 |
| Infiltration + Pyo-pneumothorax | 3 | 5.8 |
| Infiltration + Consolidation + Pyo-pneumothorax | 2 | 3.8 |
| Total | 52 | 100 |

**Table 6. Pleural fluid protein in pleural fluid analysis of study population**

| Serial no. | Pleural fluid Protein level | No. of patients |
|---|---|---|
| 1. | < 2.5 mg/dl | 2 (1%) |
| 2. | 2.51-3.5 mg/dl | 38 (19%) |
| 3. | 3.51-4.5 mg/dl | 60 (30%) |
| 4. | 4.51-5.5 mg/dl | 66 (33%) |
| 5. | >5.5 mg/dl | 34(17%) |
| | Total | 200 |
| | Mean pleural fluid Protein level | 3.6 mg/dl |
| | S.D. | 1.01 |

**Table 7. Pleural Fluid cell count instudy population**

| Serial no. | Pleural Fluid cell count Cells/microliter | No. of patients |
|---|---|---|
| 1. | <1500 cells/microliter | 100(51%) |
| 2. | 1501-3000 cells/microliter | 59 (42%) |
| 3. | 3001-4500 cells/microliter | 26 (4%) |
| 4. | >4500 cells /microliter | 15 (3%) |
| | Total | 200 |

**Table 8. ADA Level in Pleural Fluid analysis of study population**

| Serial no. | ADA level in Pleural fluid (U/Litre) | No. of patients |
|---|---|---|
| 1. | 40-100 U/Litre | 163 (9%) |
| 2. | 101-150 U/Litre | 27(37%) |
| 3. | 151-200 U/Litre | 5(43%) |
| 4. | >200 U/Litre | 5(11%) |
| | Total | 200 |

**Table 9. Distribution of patients with tubercular pleural effusion according to their sputum microscopy and pleural fluid CBNAAT result**

| Sputum Microscopy result (N=200) | Pleural fluid CBNAAT result | | |
|---|---|---|---|
| | MTB Detected | MTB not detected | Total |
| Positive (28) | 22 | 6 | 28 |
| Negative (172) | 15 | 157 | 172 |
| Total | 37 (18.5%) | 163 (81.5%) | 200 |

**Table 10. Distribution of tubercular pleural effusion with or without pulmonary involvement according to CBNAAT result**

| CBNAAT | No Pulmonary involvement | Pulmonary involvement |
|---|---|---|
| Negative | 132 | 31 |
| Positive | 16 | 21 |
| Total | 148 | 52 |

## DISCUSSION

In this study, 156 (78%) patients were male and 44 (22%) patients were female and the male to female ratio was 3.55: 1. Xinyu Zhang *et al*. (2011) conducted a study composed of 5,684 TB patients, which represented approximately 99% of mycobacterial culture-confirmed non-paediatric TB cases (≥ 15 years of age) diagnosed in Denmark during January 1, 1992–December 31, 2007.Of these 5,684 patients, 3,332 (58.7%) were males and 2,341 (41.3%)were females. In

mentioned literature male were more frequently affected than man, there is clear evidence that socioeconomic and cultural factors leading to barriers in accessing health care may cause under notification in women, particularly in developing countries, biological mechanisms may account for a significant part of this difference between male and female susceptibility to TB investigations should be conducted to clearly understand the role of sexual hormones, sex-related genetic background and genetic regulations, and metabolism, among other factors, in susceptibility differences between men and women. Other confounding factors, such as smoking, alcohol and druguse, exposure to out door pollution, migration of males to high prevalence areas could be the other reasons of male predominance. In low income countries, women often have a reduced access to economic resources and fewer educational opportunities as compared to male. As a result, many women are unable to locate and reach appropriate health services. The decision regarding a woman's treatment is also made by the husband or senior members of the family. Furthermore, the stigma attached to a positive diagnosis leads many women to forego seeking necessary medical attention.

In the low-income countries like India, women tend to self-medicate or seek out traditional healers instead of accessing public health facility because they are afraid of being recognized as a TB patient by members of the community. The higher rates of TB among male is due to a higher prevalence of infection among men. In our study, 113 (56.5%) patients in this study were below 45 years of age and87 (43.5%) patients were above the age of 45 years. In a study by Xinyu Zhang *et al*[16], out of 1449 EPTB patients, 1011(69.8%) patients were below 44 years of age and 438 (30.2%) patients were above 45 years of age. In this study patients of productive age groups were frequently involved by this clinical entity. In the mentioned literature and by WHO stated that the mycobacterium tuberculosis infection is more prevalent in productive age groups. Poor and vulnerable groups are at greater risk of infection with Mycobacterium tuberculosis compared with the general population because of overcrowded and substandard living or working conditions, poor nutrition, interaction with other diseases (such as HIVand AIDS), and migration from, or to, higher-risk communities ornations.

In this study, 168 (84%) patients were from rural areas whereas 32(16%) patients were from urban areas. In a study by Shrivastava *et al*. (2015) from central India, 62.30%patients were belonged to urban areas and only 37.69% belong to ruralareas which emphasize the fact that awareness, diagnosis and reporting of EPTB is still lacking in rural areas of Madhya Pradesh. RNTCP programme is working to combat tuberculosis in this part of Rajasthan since 2000. This programme not only provide diagnostic and treatment facilities but also sensitize health personals and citizens from the grass root levels by community meetings and patients providing meeting. RNTCP also use mass media for IEC to aware about tuberculosis. Various NGO are also working in collaboration with RNTCP for various activities. Impact of RNTCP's various activities in rural areas could be reason for a greater number of rural populations in our study. In addition to that the patients attending our institution are mostly from rural areas as, rural population is higher in Udaipur Zone which is the Southern part of Rajasthan. These might be reasons for higher rural population in our study. In this present study, 114 (57%) patients had BMI < 18.5 kg /m2and 75 (37.5%) had BMI between 18.5 to 24.9 kg /m2. 11 (5.5%) patients with tubercular pleural effusion were overweight and had BMI between 25-29.9 kg/ m2. Underweight patients (BMI < 18.5 Kg/M2) are of high risk of developing EPTB. And to a lesser extent overweight and obese patient (BMI > 25 Kg/M2) also develop EPTB. Many studies also show that having a BMI < 18 (Kg/M2) is considered as a risk factor for developing TB, contrarily, having a BMI > 25 (Kg/M2) has a protective effect against TB.79,80 It is well known that nutritional status influences the functioning of the cell-mediated immune system. Though the exact pathways are not fully understood, there is no doubt that several nutritional factors also influence the capacity of the cell-mediated immune system to fight TB bacilli. In our study out of 200 patients, 74 (37%) patients were illiterate and126 (63%) patients were literate. Illiteracy is the main constraint for any health programme. People with low health literacy may have access to health information but they often fail to use the information properly.

A common reason for misunderstanding health instruction may be the patient's low health literacy skills. Patient with limited health literacy are often considered noncompliant. Illiterate patient makes medication and treatment errors because they cannot understand or follow instruction properly. In our study, out of 200 patients 66 (33%) patients were farmer,32(16%) patients were skilled worker, 24 (12%) were semi-skilled worker, 14 (7%) were professional, 22 (11%) were student of different standard and42 (21%) females were housewives. In the study by Bag *et al*. (2015) from Bhubaneswar, India, 2596 (18.51%) had extra pulmonary involvement of tuberculosis including 2492 with exclusive extra pulmonary disease and Eighty percent (80%) were with rural back ground mostly labour classes and slum dwellers. Manjusha Sajith *et al*. (2015) analysed socioeconomic character in both pulmonary and extra-pulmonary tuberculosis and find that, out of 112 TB cases there were 49 extra-pulmonary tuberculosis cases and among these19 were employed, 18 were unemployed, one was retired and 10 were student.

The association between poverty and health is well documented. The founders of social medicine have established the powerful relationship of poverty and ill health that was attributed to abysmal housing, overcrowding, insanitation and poor working conditions. Exactly how poverty may lead to tuberculosis remains unclear. Poor SES with its attendant poor education is associated with poor knowledge of TB, risks of infection and dissemination, and with inadequate and/ordelayed availability of health care. Poverty also results in poor nutrition and low body weight, which are likely to render the immune system more vulnerable to the invading organisms 85. In our study, out of 200 study subjects,135 had different substance abuse habit and among these 22 (16.3%) patients had smoking habit,19 (14.1%) patients were tobacco chewer and 7 (5.2%) were alcoholic.30 (22.2%) patients were both smoker and alcoholic, 24 (17.8%) were both Smoker and tobacco chewer and 33 (24.4%) patients were smoker, alcoholic and tobacco chewer. Chandrashekhar T Sreeramareddy *et al*. (2008) from Nepal, compare pulmonary and extra-pulmonary cases. In their study, current smokers and current alcoholic was 34 (14.8%) and 24 (10.4%) respectively. They also conclude that the smoking and alcoholic habit were more frequently seen in pulmonary tuberculosis than extra-pulmonary tuberculosis. In the study by Carlos Pérez-Guzmán *et al*. (2014) history of smoking or drug addiction was found in 21% and 12% of patients with PTB, respectively, while none of the patients with extra-pulmonarytuberculosishad these

habits. Bag *et al*. (2015), from Bhubaneswar Odisha, invariably observed alcohol consumption and smoking habit by the patients with extra-pulmonarytuberculosis. Smoking habit in the patients were more in this study in comparison to other published study. The reason behind a greater number of the smoker in extra-pulmonary tuberculosis cases in our study may be due to illiteracy as 37% patients were illiterate. Secondly, 84 % of the patients were from the rural areas and maybe the y were unaware about health hazards of tobacco use. Lack of infiltration of health information regarding health hazard to the grass root levels and poor execution of the tobacco law may be the other possibilities. Only 43 (21.5%) patients in our study had co-morbidities. 17patients had diabetic mellitus, 16 patients had obstructive air way disease, 14 had hypertension and 4 patients were HIV positive. So most common co-morbid illness observed with this study was diabetes mellitus followed by COPD and HTN.

The most common systemic disorders accompanying EPTB in the study by Aysel Sunnetcioglu *et al* 72 were DM (n = 9; 0.04 %) and chronicrenal failure (n = 8; 0.04 %), while chronic renal failure (n = 9; 0.04) and chronic obstructive pulmonary disease (n = 6; 0.03 %) were those detected frequently in PTB group. In InêsSanches *et al*. (2015) study, HIV, DM and cancer were frequentco-morbidities associated with extra-pulmonary tuberculosis and seen in15.8% (20), 6.3% (8) and 4.8% (6) patients respectively. Soham Gupta *et al*. (2011), from South India conducted a study and in this study, it was observed that 31.8% of the pulmonary TB patients had DM as a co-morbid factor, which was significantly higher than HIV (8.85%). Conversely, in the case of extra-pulmonary TB the situation was reversed, with 32.43% HIV and 5.4% DM. Extra-pulmonary TB is more prominent among patients with HIV-co infection. In literatures, DM is a predominant co-morbidity in extra pulmonary as well as pulmonary except in countries where prevalence of HIV infection is high. DM is a well-known risk factor for Tuberculosis. Depressed cellular immunity, dysfunction of alveolar macrophages, low levels of interferon gamma, pulmonary microangiopathy, and micronutrient deficiency have been implicated in the occurrence of tuberculosis in Diabetic patients. Only 4 (2%) patients with tubercular pleural effusion were HIV positive.

HIV is a major risk factor for tuberculosis. The risk of developing TB is estimated to be between 20-37 times greater in people living with HIV than among those without HIV infection. All the facilities for Diagnosing Tuberculosis and HIV are made available at free of cost at district level hospital stoperipheral health institution level under this scheme. ICTC and ART centres are also working efficiently too in the districts. So, extra-pulmonary tuberculosis co-infected with HIV is diagnosing in the district level hospitals and directly referred to nearby DOTS centre or selectively referring to this hospital. Second reason might be the CBNAAT machine which is deployed in almost all districtsin Rajasthan. So, the diagnosis of extra-pulmonary tuberculosis is now become easy at district level. In this study, 52 (26%) patients out of 200tubercular pleural effusion patients had pulmonary involvement. most common finding is infiltration followed by consolidation, cavity and pyo-pneumothorax. The co-existence of parenchymal disease in association with pleural effusion has been observed on chest radiograph in up to 50% ofpatients,98 and occurs on the same side in almost all cases.78 Observed parenchymal changes are in the upper lobes in three quarters of cases, suggesting reactivation as the cause of TB. In the remaining patients, parenchymal disease is in the lower lobe suggesting primary TB infection. In this study, we examined 200 pleural fluid samples and out of it CBNAAT was able to detect MTB in 37 (18.5%) samples. Patil Shital *et al*. (2014) all 100 pleural fluid samples subjected for DNAPCR and observed positive in 74 (74%) of the cases. Reechaipichitkul *et al*. (2000) mentioned a sensitivity of 50% and specificity of 61% and had PCR positive in 100% of culture positive TB effusion and only in 30-60% of culture negative pleural fluid. Bahador *et al*. (2005) reported a PCR positive in 66 (84%) of 78 patients studied. Chakravarthy *et al*. (2005) found 40 PCR positive cases out of 53patients studied (75.47%), with a sensitivity of 75.5% and specificity of93.8% PPV 97.6% and NPV 53.6%.In our study, 200 pleural fluid samples were tested for CBNAAT and MTB was detected in 37 samples and out of it 6 were 'Rif' Resistance. CBNAAT was more successful in detection of MTB in patients with tubercular pleural effusion who had pulmonary involvement in their chest radiograph and sputum AFB positive patients. 'R' resistant was also detected in those patients. In the study by Soma Chakraborty *et al*. (2005) out of the 240 extrapulmonary samples 13 (5.41%) were positive for AFB by ZN staining and 23 samples (9.2%) were stain and culture positive for Acid fast bacilli as well as detected as positive for Mycobacterium tuberculosis on Gene Xpert. 2 (8.69%) out of 23GeneXpert positive extra-pulmonary samples were found to be Rifampicin resistant, 2 (8.69%) out of 23 Gene Xpert positive extra-pulmonary samples were found to be Rifampicinresistant. Avashia *et al*. examined 300 various extra-pulmonary samples from suspected extra pulmonary tuberculosis and out of it, MTB was detected in 111 samples and 105 samples were 'Rif' sensitive and 6samples were 'Rif' resistant (5.40%).

**Conclusion**

While the cytology of Pleural Fluid gives a very good estimation of the positivity status of a patient than CB-NAAT as the higher cell count is suggestive of TB but NAAT helps in diagnosing both the positivity status as well as rifampicin resistant state of the patient. To conclude, CBNAA Thas the potential to significantly improve and escalate the diagnosis of pleural fluid specimens at both hospitals as well as point-of-care settings in regions not only with high TB burden but also with overlapping HIV. Also, detection of Rifampicin resistance aids in prompt initiation of appropriate therapy and thus improving the overall quality of TB care. Physician and surgeon of other departments from tertiary care centres should have the knowledge about this diagnostic facility and WHO recommendations for tubercular pleural effusion and should be send the pleural fluid sample using WHO guideline for the CBNAAT testing. Treating doctor should also have knowledge that CBNAAT machines, are installed in almost every district TB clinic of Rajasthan and available at free of cost.

## RERERENCES

Avashia, S., Bansal, D. and Ahuja, K. Comparison of conventional methods with gene Xpertmtb/rif assay for rapid detection of mycobacterium tuberculosis and rifampicin resistance in extra pulmonary samples.

Bag, S., Deep, N. and Padhy, S. 2015. Resurgence of extra pulmonary tuberculosis. SAARC j Tuber Lung Dis HIV/AIDS, xii (1).

Bahador, A., Etemadi, H., Kazemi, B., Ghorbanzadeh, R., Nakhjavan, FA., *et al*., 2005. Performance Assessment of IS1081-PCR for Direct Detection of Tuberculous Pleural Effusion: Compared to rpoB-PCR. *Research Journal of Agriculture and Biological Sciences,* 1: 142-145.

Biehl, JP. 1958. Miliary tuberculosis. A review of sixty-eight adult patients admitted to a municipal general hospital. *Am Rev TubercPulm Dis.,* 77: 605-22.

Boehme, CC., Nabeta, P., Hillmann, D., Nicol, MP., Shenai, S., Krapp, F., Allen, J., Tahirli, R., Blakemore, R., Rustomjee, R., *et al*., 2010. Rapid molecular detection of tuberculosis and rifampicin resistance. *N Eng J Med.,* 363: 1005-1015.

Carlos Pérez-Guzmán, MSc, Mario H Vargas, 2014. MSc, María del Rosario Arellano-Macías. Clinical and epidemiological features of extra-pulmonary tuberculosis in a high incidence region. *Salud Pública Méx.,* Vol. 56(2):189-196.

Chandrashekhar, T., Sreeramareddy, Kishore V Panduru, Sharat, C Verma *et al*., 2008. Comparison of pulmonary and extrapulmonary tuberculosis in Nepal- a hospital-based retrospective study. *BMC Infectious Diseases,* 8: 8 doi: 10.1186/1471-2334-8-8.

Claudia, M., Denkinger, Samuel G. Schumacher, Catharina C. Boehme, *et al*., 2014. Xpert MTB/RIF assay for the diagnosis of extrapulmonary tuberculosis: a systematic review and metaanalysis. ERJ Express. Published on April 2, as doi; 10.1183/09031936.00007814.

Fanning, A. 1999. Tuberculosis: 6. Extrapulmonary disease. CMAJ., 160: 1597-603.

Helb D, Jones M, Story E, Boehme C, Wallace E, Ho K, Kop J, Owens MR, Rodgers R, Banada P, *et al*. Rapid detection of Mycobacterium tuberculosis and rifampicin resistance by use of on-demand, near patient technology. J Clin Microbiol 2010; 48: 229-237.

InêsSanches A, Aurora Carvalho, Raquel Duarte *et al*., 2015. Who are the patients with extra-pulmonary tuberculosis? *Rev Port Pneumol.,* 21(2):90---93.

Iscman, MD. 2000. Tuberculosis in relation to human immunodeficiency virus and acquired immunodeficiency syndrome. In: Iseman MD, editor. A clinician's guide to tuberculosis. Philadelphia: Lippincott Williams and Wilkins; p. 199-252.

Kumar, A. 2001. Lymph node tuberculosis. In: Sharma SK, Mohan A, editors. Tuberculosis. New Delhi: Jaypee Brothers Medical Publishers; p. 273-84.

Manjusha Sajith, Ansu Thomas, Jaydeep J Kothia *et al*., 2015. Socio-Demographic characteristics of tuberculosis patients in a tertiary care hospital. International Journal of Medical and Health Research. Volume 1; Issue 3; Page No. 25-28.

Kumar, B. and Muralidhar, S. 1999. Cutaneous tuberculosis: a twenty-year prospective study. *Int J Tuberc Lung Dis.,* 3: 494-500.

Pai, M. and Ling, DI. 2008. Rapid diagnosis of extra pulmonary tuberculosis using nucleic acid amplification tests: what is the evidence? *Future Microbiol.,* 3:1-4.

Patil Shital, Halkanche Gajanan and Ayachit Rujuta, 2014. Role of nucleic acid amplification test (NAAT) in tuberculous pleural effusion: Where it fits in Routine diagnosis workup. *J Cell Sci Ther.,* 5; 4.

Reechaipichitkul, W., Lulitanond, V., Sungkeeree, S. and Patjanasoontorn, B. 2000. Rapid diagnosis of tuberculous pleural effusion using polymerase chain reaction. *Southeast Asian J Trop Med Public Health.,* 31:509-514.

Rooney, JJ., Crocco, JA. and Lyons, HA. 1970. Tuberculous pericarditis. *Ann Intern Med.,* 72: 73-8.

Sharma, S.K. and Mohan, A. 2004. Extra-pulmonary tuberculosis. *Review Article Indian J Med Res.,* 120, 316-353.

Sharma, SK., Mitra, DK., Balamurugan, A., Pandey, RM. and Mehra, NK. 2002. Cytokine polarization in miliary and pleural tuberculosis. *J Clin Immunol.,* 22: 345-52.

Shrivastava, AK., Brahmachari, S., Pathak, P., *et al., 2015.* Clinico-Epidemiological Profile of Extra-pulmonary Tuberculosis in Central India. International Journal of Medical Research and Review. Vol 3, No 02.

Singh, MM., Bhargava, AN. and Jain, KP. 1969. Tuberculosis peritonitis: an evaluation of pathogenic mechanisms, diagnostic procedures and therapeutic measures. *N Engl J Med.,* 289: 1091-4.

Soham Gupta, Vishnu Prasad Shenoy, Indira Bairy, *et al*., 2011. Diabetes mellitus and HIV as co-morbidities in tuberculosis patients of rural south India. *Journal of Infection and Public Health*, Volume 4, Issue 3, Pages 140–144.

Soumitesh Chakravorty, Manas Kamal Sen and Jaya Sivaswami Tyagi, 2005. Diagnosis of Extrapulmonary Tuberculosis by Smear, Culture, and PCR Using Universal Sample Processing Technology. J Clin Microbiol 43: 4357–4362.

Van Rie, A., Page-Shipp, L., Scott, L., Sanne, I. and Stevens, W. 2010. Xpert MTB/RIF for point of care diagnosis of TB in high-HIV burden, resource-limited countries: hype or hope? *Expert Rev Mol Diagn.,* 10: 937-946.

WHO Global Tuberculosis Report 2016

Xinyu Zhang, Aase B. Andersen, Troels Lillebaek, 2011. Effect of Sex, Age, and Race on the Clinical Presentation of Tuberculosis: A 15- Year Population-Based Study. Am. *J. Trop. Med. Hyg.,* 85(2), pp. 285–290.

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