

# Evaluation of Recurrent Lymphadenitis after Six Months Anti-Tuberculous Therapy

MMH CHOWDHURY<sup>a</sup>, MAJ CHOWDHURY<sup>b</sup>, SMM KAMAL<sup>c</sup>, MKANAM<sup>d</sup>,  
MT ISLAM KHAN<sup>e</sup>, FT ZOHURA<sup>f</sup>

## Abstract:

**Background:** Recurrence or persistence of tuberculous lymphadenitis even after treatment occurs due to paradoxical immune response, non-tubercular mycobacterial (NTM) infection, drug resistance tuberculosis or because of misdiagnosis. In these situations sometimes the patients are asked to continue category-1 more than six months on the basis repeat fine needle aspiration cytology (FNAC) findings. This study evaluated the cause of persistence or appearance of new lymph node after anti-TB category-1.

**Methods:** This observational study was done from January, 2013 to March, 2014 after getting ethical clearance from Bangabandhu Sheikh Mujib Medical University (BSMMU). Patients who had persistent or new lymphadenitis after completing anti-TB category-1 for six months were selected. After analysis of treatment records, patients were selected for either FNAC or biopsy with local surgical clearance. Cyto/histopathological analysis, AFB stain (LED fluorescent), AFB culture (MGIT and L-J media) and GeneXpert test was done. Follow-up of the patients were done after two weeks and six months of FNA/surgery.

**Results:** Thirty-two patients (M/F=11/21) with mean age 25(±12) years were investigated. Altogether 26 cases (81.25%)

were confirmed as tuberculous lymphadenitis and six (18.75%) were non-tuberculous (03 lymphomas, 01 sarcoidosis, 01 reactive lymphadenitis and 01 pyogenic abscess). Among the TB patients, granuloma was present in 23 cases (88.46%) (FNA-07, biopsy-16). Twenty-three (88.46%) cases were GeneXpert positive, comprising 21 rifampicin sensitive and 02 rifampicin resistant. One rifampicin resistant case was confirmed by culture and the remaining 25 were culture negative. One case was AFB stain positive and no NTM was found in AFB culture. After surgery, rapid healing was observed within two weeks among tuberculous lymphadenitis patients.

**Conclusions:** Persistent granuloma after anti-TB category-1 does not indicate active tuberculosis if it is culture negative. It may occur due to immunological reaction against dead bacilli in deep seated sinus. Local surgical clearance results rapid recovery and other diagnosis should be excluded without further continuation of anti-TB category-1.

**Key word:** TB Lymphadenitis, Recurrences, Anti-TB category-1

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- Dr. Mohammad Mainul Hasan Chowdhury, Lt Col, Classified Medicine Specialist, Combined Military Hospital, Ramu Cantonment, Bangladesh Army.
- Dr. M A Jalil Chowdhury, Professor of Medicine (Retired), Bangabandhu Sheikh Mujib Medical University, Shahbag, Dhaka, Bangladesh.
- Dr. S M Mostofa Kamal, Professor of Microbiology (Retired), Department of Pathology and Microbiology & Ex Co-ordinator of National Tuberculosis Reference Laboratory (NTRL), National Institute of Diseases of the Chest and Hospital (NIDCH), Dhaka, Bangladesh.
- Dr. M Khairul Anam, Associate Professor, Department of Respiratory Medicine, Shaheed Suhrawardy Medical College, Dhaka, Bangladesh.
- Dr. M Touhidul Islam Khan, Registrar, Department of Medicine, Kurmitola General Hospital, Dhaka, Bangladesh.
- Dr. Fatima Tuj Zohura, Medical Officer, Department of Haematology, BSMMU, Dhaka.

Address for Correspondence: Professor M A Jalil Chowdhury, Professor of Medicine (Retired), Bangabandhu Sheikh Mujib Medical University, Shahbag, Dhaka, Bangladesh Road No. 4, House No. 44, PC Culture Housing Society, Adabor, Dhaka-1207, Bangladesh. Email: [majchow@yahoo.com](mailto:majchow@yahoo.com)

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## Introduction

Tuberculous lymphadenitis is the commonest cause--of extra-pulmonary tuberculosis and in USA 30-40% of the extra-pulmonary tuberculosis cases present with lymphadenitis.<sup>1</sup> In a Danish report of more than 480 patients with TB, lymphadenitis was observed in 13.5 percent cases.<sup>2</sup> After starting anti-TB treatment, some of the TB-lymphadenitis patients develop new lymph nodes in the same or another site. Sometimes lymph nodes may persist or enlarge at the end of full course of anti-TB drugs (ATDs). This may be due to treatment failure, relapse of tuberculosis, paradoxical immune reaction, pyogenic infection, atypical mycobacterial infection or due to other causes. Paradoxical reaction during anti-TB treatment was first described in 1955,<sup>3</sup> defined as the clinical or radiological worsening of pre-existing tuberculous lesions or development of new lesions in a patient who initially improved with anti-TB therapy in the absence of disease relapse. Immune restitution, temporally related to recovery of the immune

system is believed to be a possible mechanism responsible for paradoxical immune reaction. Immune restitution leads to an acute symptomatic or paradoxical deterioration of a pre-existing infection.<sup>3</sup> It occurs in about 15.4 % of the patients with tuberculosis.<sup>4</sup> Though found commonly among HIV-positive patients, paradoxical immune reaction can occur in HIV-negative patients and frequently in those with extra-pulmonary and disseminated tuberculosis.<sup>5</sup> Cheng et al showed that age, gender and underlying co-morbidity posed no additional risk to the development of paradoxical immune reaction.<sup>4</sup> Paradoxical immune reaction may develop within 87 days (range 23 to 157 days) after initiation of anti-TB treatment in HIV negative TB patient.<sup>6</sup> Commonly affected sites include central nervous system, respiratory system, cervical and mediastinal lymph nodes.<sup>4</sup> The rapid and significant reduction of mycobacterial load with anti-tubercular treatment causes release of large amounts of tuberculo-protein and other cell wall products which initiate a cascade of cellular and cytokine inflammatory response, leading to local tissue damage by immune restitution syndrome, also known as paradoxical immune reaction.<sup>7</sup> The clinical severity of paradoxical immune reaction depends on the appropriateness of exaggerated immune recovery that results in excessive tissue damage.<sup>7</sup>

In Bangladesh, the overall prevalence of multi-drug resistant tuberculosis is very low (1.5%).<sup>8</sup> So, the chance of treatment failure or development of resistant strains in TB-lymphadenitis patient which is paucibacillary is unlikely, as because compliance is assured properly under TB control program. Diagnostic result of fine needle aspiration (FNA) for culture obtained after six weeks in TB-lymphadenitis patient were present in nearly half of the aspirate; sensitivity was 46%, specificity 100%, positive predictive value (PPV) 100% and negative predictive value (NPV) 94%.<sup>9</sup> But AFB culture from fine needle aspirate is not possible in most of the cases as it is time consuming. Fine needle aspiration cytology (FNAC), commonly used for diagnosis of TB-lymphadenitis on the basis of granuloma, can give 62% correct diagnosis with high false negative rate (38%).<sup>10</sup> But, 18% of the patients with granulomatous lymphadenitis may miss correct diagnosis by FNAC.<sup>11</sup> Usually atypical mycobacteria are isolated frequently among HIV positive TB-lymphadenitis patient in African countries. One Indian

study has isolated 01% atypical mycobacteria from HIV seronegative TB-lymphadenitis patient.<sup>12</sup> So, in a low HIV prevalent country like Bangladesh atypical mycobacteria is unlikely to be a common cause of persistent lymphadenitis among these groups of patients who received ATDs. Ormerod P has noticed that deep seated sinus tract or sterile abscess may develop because of inadequate drainage in a TB lymph node which was initially smaller in size and begins to enlarge after starting ATDs.<sup>13</sup> He has also observed that surgery with adequate local clearance will give rapid recovery within two weeks in these group of patients as because they have developed sterile abscess after starting ATDs. Though, lymph nodes may enlarge in TB-lymphadenitis patients due to many causes, they were sometimes advised to continue ATDs more than six months or start category-2 on the basis of findings of granuloma in FNAC. The objective of this study was to evaluate TB-lymphadenitis patients, whose lymph nodes persist or develop new lymph nodes even after successful category-1 anti-TB treatment. Evaluation was done by clinical, pathological and microbiological assessment of lymph nodes. Follow-up after surgery was also done for six months to see whether they had active clinical, pathological and microbiological evidences of tuberculosis.

### Methods

This observational study was done from January, 2013 to March, 2014 after getting ethical clearance from Bangabandhu Sheikh Mujib Medical University (BSMMU). Patients who had persistent or new lymphadenitis after completing anti-TB category-1 at least for six months were selected. Previous treatment records were analyzed after detailed history taking to confirm the diagnostic basis of TB-lymphadenitis. Improvement of fever, appetite, weight gain and compliance to anti-TB drugs were also justified to assess response. Any secondary local infection, previous episodes of major infections and family history of similar presentation were searched to exclude the possibility of immunodeficiency, which can cause recurrent TB-lymphadenitis. A thorough physical examination was done to find out the cold abscess and to exclude other causes of lymphadenitis like any focus of infection, or malignancy. BMI was calculated to evaluate nutritional status. Elevation of total white cell count and inflammatory markers like ESR and CRP were used to

assess prognosis of TB-lymphadenitis patient but were non-specified as their elevation can be contributed by both secondary infection and paradoxical immune reaction. Serum creatinine and blood sugar were done in all patients to exclude other co-morbid pathology like renal impairment or diabetes mellitus. Mantoux test (MT) was done to assess cell mediated immunity as immune system recovery may lead to paradoxical response. Chest radiograph was done to see mediastinal lymph node or any focus of infection in chest. All accessible lymph nodes were assessed clinically to evaluate the nature of lymph node. FNAC were done from easily accessible lymph nodes and at least one milliliter (ml) aspirate were taken for each of the sample. Those who agreed for surgery, adequate local surgical clearance was done to remove deep seated sinus. Three samples from each of fine needle aspirate and biopsy were taken. Fresh sample without any blood contamination were sent for analysis immediately after aspiration or biopsy. One sample was sent for Gram stain and bacteriological culture to Microbiology department of National Institute of Chest Diseases and Hospital (NIDCH). One fresh sample of FNAC or one formalin preserved tissue sample of biopsy material was sent to Pathology department of NIDCH. Another fresh sample was sent for AFB stain, AFB culture in MGIT 960 broth, Lowenstein Jensen Media and GeneXpert test to National Tuberculosis Reference Laboratory (NTRL). After collection, pus or tissue sample were sent to NTRL within two hours. If immediate transportation or processing could not be done samples were stored at 4°C for overnight and sent for analysis in the next morning. GeneXpert test was

done by Cepheid Xpert MTB/RIF analyzer and AFB stain was done by Auramin re-agent. AFB culture was done in both Liquid (MGIT broth 960) and solid media (Lowenstein Jensen media). Reading from culture was taken at one week interval for eight weeks. Specimens of FNA showing acute supportive inflammatory changes were considered as pyogenic abscess and if co-existed with residual caseating granuloma were defined as TB-lymphadenitis or cold abscess. Follow up of all patients were done at two weeks and six months after surgical clearance with or without ATDs as per their previous treatment schedule. Clinical improvement of sign/symptoms, healing of lesions, and further non-recurrence of lymph nodes was observed during follow-up. Excision biopsy and FNAC reports were carefully analyzed. All drug resistant cases were referred to NIDCH for further management. Informed written consent was taken from each of the participants.

### Results

At first 53 participants with TB-lymphadenitis were recruited in the study as per inclusion criteria. Twenty-one patients did not provide adequate FNA sample and denied surgical management and were excluded from the study. The remaining 32 participants were finally analyzed. Among them 11 (34%) were men and 21 (66%) were women with mean age 25 years (SD, ±12). Most of the participants belonged to the age group between 20 to 35 years (n=23, 72%) while mean age was 25(±12) years.

Table-I shows that most of the lymph nodes (68.8%) in the studied participants appeared or persisted within 6-

**Table-I**

*Development of Lymph Node by Treatment Duration and Site*

Site of Lymph node	Treatment duration (n=32)		
	Six to nine months (n=22) (68.8%)	Nine to twelve months (n=4) (12.5%)	More than twelve months (n=6) (18.8%)
New lymph node appeared (n=19) (59.38%) Cervical (n=12) Axillary (n=7)	12	1	6
Enlargement of previous lymph node (n=13) (40.62%) Cervical (n=10) Axillary (n=03)	10	3	0

9 months of treatment for which they were advised to continue the ATDs. Four patients (12.5%) had recurrences after nine to twelve months and six (18.8%) even after more than 12 months of starting ATDs. Thirteen participants (40.62%) (Table-II) developed enlargement of previous lymph node and 19 (59.38%) developed new lymph node either in the same or other site.

Out of 32 participants, 23 (71.87%) agreed for biopsy and 09 (28.13%) did not agree for biopsy but opted for

FNAC. Amongst the 23 biopsy group, 16 (69.57%) revealed granuloma with or without caseation suggestive of tuberculosis. Among these 16 biopsy patients with granuloma, all were negative by AFB culture, but 14 were positive by GeneXpert. Of the remaining granuloma negative 07 (30.43%) biopsy patients, 06 revealed alternate diagnoses (03 lymphoma, 01 sarcoidosis, 01 abscess and 01 reactive hyperplasia); 01 was GeneXpert positive and all 07 were AFB culture negative (Figure-1).

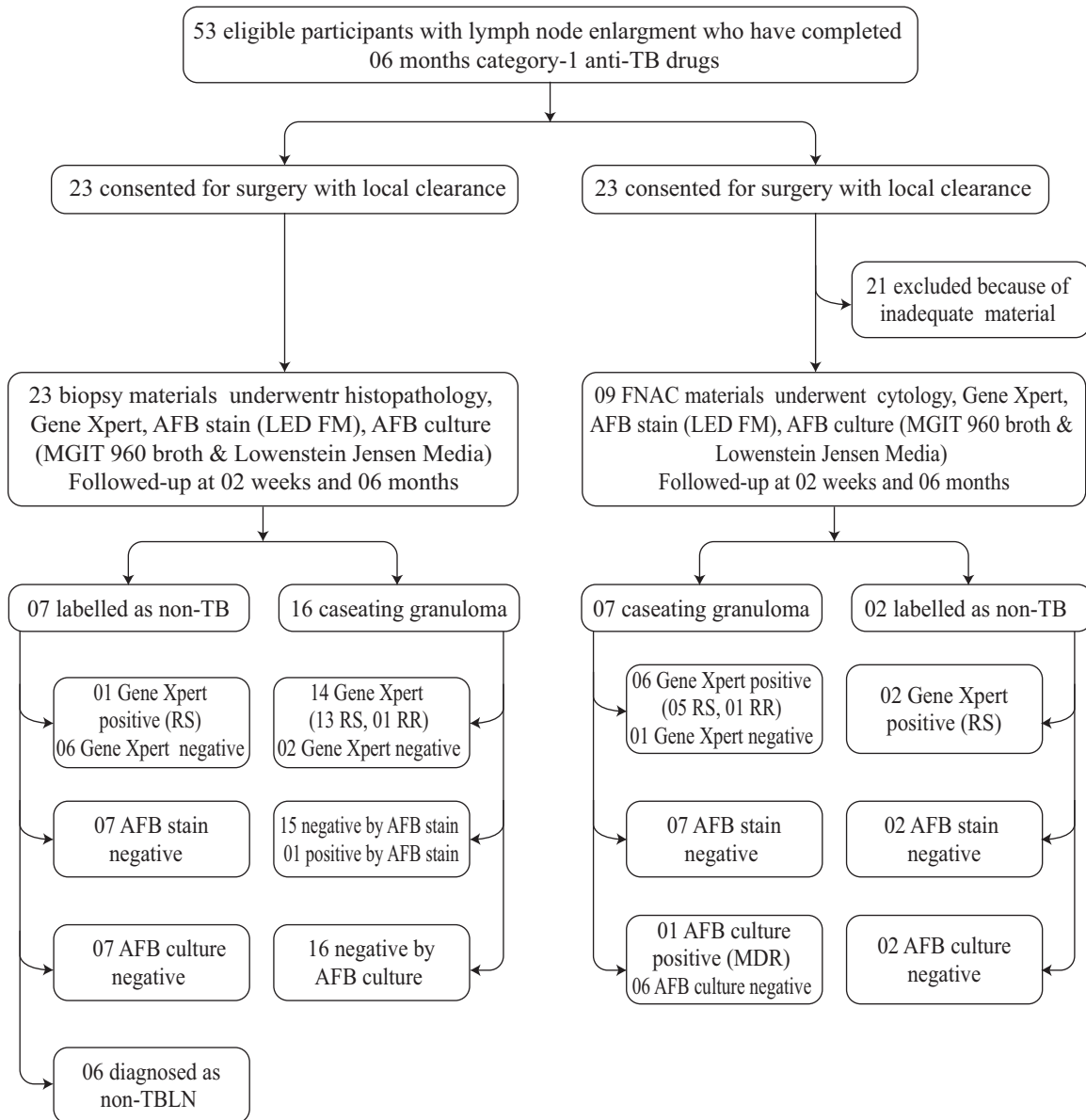


Fig.-1: Evaluation of the Enrolled Participants

**Table-II**

<i>Treatment Outcome of Recurrent TB Lymphadenitis Patient after Two Weeks and Six Months of FNAC and Surgery</i>					
FNAC	Two weeks (n=25) (Drop out, n=1)		Surgery	Improved*	Not improved**
	Improved	Not improved**			
Anti-TB (n=6)	2	4	Anti-TB (n=13)	12	1
No Anti-TB (n=2)	0	2	No Anti-TB (n=4)	3	1
Total (n=8)	2 (25%)	6 (75%)	Total (n=17)	15 (88%)	2 (12%)
Six months (n=25) (Drop out, n=1)					
FNAC	Surgery		Surgery	Improved	Not improved
	Improved	Not improved			
Anti-TB (n=6)	5	1	Anti-TB (n=13)	12	1
No Anti-TB (n=2)	2	0	No Anti-TB (n=4)	3	1
Total (n=8)	7 (87.5%)	1 (12.5%)	Total (n=17)	15 (88%)	2 (12%)

\* Rapid healing of lesion was observed within two weeks after surgery.

\*\* Persistence or recurrences of lymph node.

TBLN-tuberculous lymphadenitis, solid AFB culture-Lowenstein Jensen Media, liquid AFB culture- MGIT broth 960, LED FM- LED fluorescence microscopy, RS-rifampicin sensitive, RR- rifampicin resistance, FNA- fine needle aspiration, FNAC- fine needle aspiration cytology, MDR- multi drug resistance.

Among FNAC group (n=9), seven patients (77.78%) revealed granuloma with or without caseation suggestive of tuberculosis. Out of these 07 granuloma positive patients, 06 were negative and 01 was positive by AFB culture. Six patients were positive by GeneXpert. Remaining 02 patients of the FNAC group did not show granuloma but were GeneXpert positive.

Rapid healing among TB-lymphadenitis patients were observed within two weeks after surgery (n=15 out of 17, 88%) (non tubercular lymph node has been excluded from follow-up). No recurrences of lymphadenitis among this group at the end of six months (Table-II). None of the patients were receiving steroid.

### Discussion:

According to WHO global tuberculosis report, 2019, Bangladesh is one of the eight high TB burden countries that accounts for two-third of the new TB cases in the world.<sup>8</sup> About 18.7% of the TB patients are extra-pulmonary TB (EPTB) and among them 40.4% are tuberculous lymphadenitis.<sup>1</sup>In this study we have

examined patients who had persistent lymph node either enlargement of previous lymph node (40.56%) or appearance of new lymph node (59.44%) in the same or another site after completion of category-1 anti-TB treatment. Most of the participants were young and women which was consistent with previous findings.<sup>14</sup>Patients with lymphadenitis who were put on ATDs on the basis of granuloma with or without caseation necrosis on the basis of FNAC may present with persistently enlarged lymph node or new lymph node at the same or other site after completion of category-1. In this study it was revealed that six cases (18.75%) were wrongly diagnosed as TB lymphadenitis and all of them received category-1 anti-TB drugs on the basis of FNAC. Among them three lymphomas, one sarcoidosis, one reactive lymph node and one pyogenic abscess were confirmed and this findings was comparable with previous study.<sup>11</sup>Two participants labeled as non- tubercular on repeat FNAC was ultimately found GeneXpert positive. As per previous report, 18% of the patient with granulomatous lymphadenitis may miss correct diagnosis by FNAC.<sup>11</sup> So, if suspected or patient does not respond to anti-TB after FNAC diagnosis, bacteriological evaluation should be done along with histopathology as early as possible. We have also seen that GeneXpert was positive in 88.46% cases and two cases were found to be rifampicin resistant and one became culture positive for M. tuberculosis

and resistant to three first line anti-TB drugs. It has been reported that GeneXpert has high diagnostic accuracy in detection of tuberculous lymphadenitis with sensitivity 96.7% and specificity 88.9%.<sup>15</sup> But, GeneXpert cannot differentiate between live and dead bacilli.<sup>16</sup> In this study, all rifampicin sensitive GeneXpert cases were found culture negative and this is because of the fact that GeneXpert test cannot differentiate between live and dead bacilli. Though GeneXpert test is more sensitive in diagnosis of tuberculous lymphadenitis, it is not a good tool for monitoring the response of treatment. This study revealed only one AFB stain positive case which was rifampicin sensitive in GeneXpert test but negative in AFB culture. As gland-TB is usually pauci-bacillary, AFB stain alone cannot be utilized for diagnosis of tuberculous lymphadenitis patient and if AFB stain found positive it will give suspicion of excessive load of MTB organism.<sup>17</sup> Moreover conventional AFB stain cannot differentiate between live and dead bacilli. Possibly because of high organism load this patient had developed more reaction due to extensively released tuberculo protein which invite extensive immunological reaction and causes delayed recovery.<sup>18</sup> Only one participant was AFB culture positive and was resistant to three first line anti-TB drugs. No atypical mycobacteria were isolated during AFB culture. Atypical mycobacteria have frequently been isolated in HIV positive tuberculous lymphadenitis patient in African countries. Some of the Indian studies have noticed very limited number of cases of atypical mycobacteria (01%) from non HIV patient.<sup>12</sup>

Out of 32 participants 23 (16 biopsy group and 7 FNA group) revealed granuloma with or without caseation. But all except one had negative AFB culture. That one showed MDRTB. So it is obvious that mere presence of granuloma with or without caseation on repeat biopsy or FNA on enlarged lymph node during or at completion of standard anti-tuberculous chemotherapy does not mean active tuberculosis and does not justify continuation of anti-TB drugs beyond standard duration. Moreover repeat FNAC or biopsy only to monitor anti-TB response should not be done in these groups of patients as because it creates confusion about activity of tuberculosis. If there is possibility of drug resistance, though it is rare, AFB culture and sensitivity should be done on appropriate sample.

Our findings of rapid improvement by healing of lesion after surgery possibly explain presence of inactive tubercle bacilli as because some of the author has noticed that rapid healing after local surgical clearance indicate inactivity of tuberculous lymph node with formation of sterile abscess or deep seated sinus tract.<sup>13</sup> Deep seated sinus tract or smaller lymph node contains tuberculoprotein released by tubercle bacilli after starting ATDs. Inadequate drainage of pus from this deep sinus or lymph nodes leads to excessive immunological reaction to tuberculoprotein and resulting in enlargement of previous lymph node or appearance of new lymph node after starting ATDs. So, adequate drainage of locally collected sterile pus without further continuation of anti-TB will be the treatment of choice in this patients who develop persistent lymph node either new or enlargement of previous one after completion of category-1 ATDs. Patients' immunological or nutritional status also play vital role in pathogenesis of tuberculosis and treatment response to anti-TB drug by CD-4 T-lymphocyte and micronutrient.<sup>19</sup> So, immunological and nutritional status assessment in future will provide more precise information for evaluation and management of these groups of patients.

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