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Diagnostic yield of endobronchial ultrasound-guided transbronchial needle aspiration for isolated mediastinal lymphadenopathy in a tuberculosis-endemic region: first study from Northeast India

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Contributions: John Muchahary, Vijay Noel Nongpiur, Pranjal Kalita: conception and design of the study. John Muchahary, W. Valarie Lyngdoh, Pranjal Kalita, Evarisalin Marbaniang, Neeta Gogoi, Yepeto Lohe: data collection. John Muchahary, Vijay Noel Nongpiur, Vandana Raphael, Evarisalin Marbaniang: data analysis. John Muchahary, Vijay Noel Nongpiur, Pranjal Kalita, Neeta Gogoi: writing and proof-reading of the manuscript. All authors have read and approved the final version of the manuscript and agreed to be accountable for all aspects of the work.

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Ethics approval and consent to participate: the study protocol was reviewed and approved by the Institutional Ethics Committee of North Eastern Indira Gandhi Regional Institute of Health and Medical Sciences (NEIGRIHMS), Shillong, Meghalaya, India (Protocol No. P210/2024/210, dated 19th February 2024). The committee assessed the project as ethically unobjectionable. Written informed consent to participate was obtained from all study participants prior to enrolment in the study.

Informed consent: written informed consent was obtained from all participants for inclusion in the study. The manuscript does not contain any individual person's identifiable data in any form.

Patient consent for publication: obtained.

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Abstract

Endobronchial ultrasound-guided transbronchial needle aspiration (EBUS-TBNA) is a minimally invasive technique for evaluating mediastinal lymphadenopathy, especially in tuberculosis-endemic regions where differentiating infectious, inflammatory, and malignant causes remains challenging. This retrospective study aimed to assess the diagnostic yield of EBUS-TBNA in patients from the northeastern states of India, a region with a high burden of tuberculosis and limited diagnostic resources. A total of 74 patients with isolated mediastinal lymphadenopathy (IML) who underwent EBUS-TBNA between 2021 and 2023 at a tertiary care hospital in Meghalaya were included. The final diagnosis was based on histology, cartridge-based nucleic acid amplification test (CBNAAT), culture, and clinical-radiological follow-up for up to 1 year in inconclusive cases. EBUS-TBNA achieved an overall diagnostic yield of 78.6%. The most common diagnosis was malignancy (36.5%), followed by tuberculosis (23%), reactive lymphadenitis (13.5%), and sarcoidosis (5.4%). CBNAAT was positive in 47.1% of tuberculosis cases, with multidrug resistance detected in 11.7%. The sensitivity, specificity, positive predictive value, and negative predictive value of EBUS-TBNA were 90.6%, 100%, 100%, and 62.5%, respectively. To conclude, EBUS-TBNA is a safe, highly sensitive, and cost-effective diagnostic modality for IML in tuberculosis-endemic regions. The addition of CBNAAT enhances diagnostic precision and enables early detection of drug resistance. The study also highlights an emerging trend of higher malignancy rates in the Northeastern Indian population, underscoring the need for comprehensive evaluation in all cases of IML.

Key words: endobronchial ultrasound-guided transbronchial needle aspiration, isolated mediastinal lymphadenopathy, malignancy, tuberculosis, cartridge-based nucleic acid amplification test.

Introduction

Isolated mediastinal lymphadenopathy (IML) can occur due to metastatic tumors, lymphoma, tuberculosis (TB), sarcoidosis, and other granulomatous or inflammatory causes [1]. IML secondary to granulomatous disease is more common in patients of Asian and African origin (76%) [2]. Computed tomography (CT) scans of the thorax, magnetic resonance imaging and positron emission tomography are initial investigations for the evaluation of mediastinal lymphadenopathy but do not accurately distinguish between benign and malignant lesions and have a high false-positive rate of approximately 39% [3-6]. Mediastinoscopy is traditionally considered the gold standard for sampling mediastinal lymph nodes [7]. Owing to their high cost, invasive nature, requirement for general anesthesia and hospitalization, and potential for complications, these procedures are not feasible for use in all patients [8-10].

Endobronchial ultrasound-guided transbronchial needle aspiration (EBUS-TBNA) is a safer and minimally invasive technique that has become the procedure of choice for the evaluation of mediastinal lymphadenopathy. While EBUS-TBNA is widely documented for its role in the diagnosis and staging of lung cancer, research on its utility in developing countries, which are burdened by a high prevalence of TB, is limited. The prevalence of TB and malignancy is high in India; therefore, the diagnosis of mediastinal lymphadenopathy becomes necessary to distinguish between benign and malignant causes. The 2021 Global TB report states that in India, the incidence and prevalence of tuberculosis (TB) are 188 and 312 per lakh population respectively, making it a high burden country with TB. India accounts for a large global multidrug-resistant TB (MDR-TB) burden, with an estimated 27% of the global drug-resistant TB.

We present our two-year experience at our center in Northeast India to evaluate the diagnostic yield of EBUS-TBNA for isolated mediastinal lymphadenopathy and to assess its utility in detecting TB and MDR-TB.

Materials and Methods

This was a retrospective analysis conducted over a period of two years between 2021 and 2023 in the Department of Pulmonary Medicine at a tertiary hospital in Meghalaya, India. In this study, we analyzed the records of patients with isolated mediastinal lymphadenopathy on thoracic CT, without parenchymal lesions, pleural disease, or known extra-thoracic malignancy, who had undergone EBUS-TBNA. The patient's history, clinical examination, radiological findings and provisional diagnosis were recorded. Lymph nodes were deemed enlarged and suitable for transbronchial needle aspiration (TBNA) if the short-axis diameter on chest CT exceeded 1 cm. Nodal stations were classified according to the International Association for the Study of Lung Cancer (IASLC) lymph node map [11]. In patients with

multiple enlarged nodes, the most peripheral and technically accessible lymph node ≥1 cm was sampled first to safely and efficiently obtain representative tissue. Additional nodal stations were subsequently sampled based on sonographic characteristics, radiological suspicion, and procedural feasibility. All patients provided written informed consent before undergoing the procedure, and the study was approved by the institute's ethics review committee.

EBUS-TBNA was performed in the bronchoscopy suite on an outpatient basis. Nebulized lignocaine (4%) was administered, followed by the spraying of topical lignocaine (10%) in the posterior pharyngeal wall of the patients immediately before the procedure. It was performed under conscious sedation via the injection of midazolam and fentanyl. All patients underwent EBUS-TBNA via Fujifilm's EB-530US EBUS bronchoscope using a 22-gauge needle (EchoTip ProCore needle from Cook). The echo bronchoscope was introduced orally, and 2% lignocaine was administered to the vocal cords and airways using the spray-as-you-go technique. The lymph node stations were systematically examined under ultrasound guidance, and the targeted enlarged lymph node was punctured with a 22-gauge needle under real-time sonographic and endoscopic visualization. Three to five passes were taken from each node that was sampled. The aspirate obtained by the TBNA needle was smeared on slides for cytological examination. The samples were also sent in normal saline for cartridge-based nucleic acid amplification test (CBNAAT; GeneXpert) and acid–fast bacilli (AFB) culture. The tissue cores were collected in formalin and sent for histological examination. The presence of many lymphoid cells or malignant cells confirmed the adequacy of the TBNA sample. Rapid onsite evaluation (ROSE) could not be performed because of the unavailability of a pathologist during the procedure. Patients were monitored for 2 hours after the procedure in the recovery room for any complications before being discharged.

Mediastinal tuberculous lymphadenitis was diagnosed on the basis of fulfillment of one or more of the following criteria:

1) Necrotizing granulomatous inflammation on histology
2) AFB smear positive
3) Mycobacterium tuberculosis (MTB) detected via CBNAAT
4) Isolation of MTB via AFB culture

A diagnosis of sarcoidosis was established when the following three criteria were met:

1) The patient presented with clinical and radiological features typical of sarcoidosis.
2) The tissue samples revealed nonnecrotizing epithelioid granulomas, with no evidence of AFB on Ziehl–Neilsen staining, MTB not detected by CBNAAT and no mycobacterial growth in mycobacteria growth indicator tube (MGIT) culture.
3) The patient demonstrated clear clinical and radiological improvement following treatment with corticosteroids.

The presence of atypical or malignant cells was diagnostic of malignancy.
The data were collected and entered into Microsoft Excel. Statistical analysis of the data was performed using the statistical software IBM SPSS 27 version.

Results

A total of 74 patients were enrolled in the study, of whom 50 (67.6%) were male and 24 (32.4%) were female. The mean age was 50.23 years, and their clinical characteristics are summarized in Table 1. In total, 134 lymph nodes were sampled, with station 7 being the most frequently targeted lymph node. The mean number of nodes sampled per patient was 1.8. Overall, 270 passes were made, with an average of 3.64 passes per patient and 2.01 passes per node.

The EBUS features of lymph nodes with various etiologies are presented in Table 2. The mean long- and short-axis diameters of lymph nodes differed according to etiology, being largest in malignant nodes (35.7 mm and 23.6 mm) and smallest in reactive nodes (21.7 mm and 11.6 mm). Malignant nodes were predominantly round (18/27) and heterogeneous (19/27), with central necrosis in 4 nodes and calcification in 3. Tuberculous nodes were mostly oval (8/17) and heterogeneous (13/17), with central necrosis in 7 and calcification in 6. Lymph nodes in sarcoidosis were primarily oval (3/4) and homogeneous (3/4), with calcification observed in 1 node and no central necrosis. Reactive nodes were generally oval (7/10), homogeneous (10/10), and lacked necrosis or calcification.

The diagnosis of the 74 patients who underwent EBUS-TBNA for the evaluation of isolated mediastinal lymphadenopathy were malignancy in 27 patients (36.5%), tuberculosis in 17 (23.0%), sarcoidosis in 4 (5.4%), and reactive lymphadenitis in 10 (13.5%) as shown in Table 3. The remaining 16 patients (21.6%) had inconclusive results. Of the 27 patients diagnosed with malignancy, 11(40.7%) had adenocarcinoma, 7(25.9%) had squamous cell carcinoma, 4(14.8%) had small cell lung carcinoma, 4(14.8) had undifferentiated carcinoma, and 1(3.7%) had lymphoma.

Seventeen patients (23%) were diagnosed with TB. In all 17 patients, the diagnosis of mediastinal tuberculous lymphadenitis was supported by cytological evidence of granulomatous inflammation in EBUS-TBNA samples. The clinical and radiological features were strongly suggestive of MTB infection. Follow-up assessments demonstrated resolution of symptoms and radiological improvement in the mediastinal lymphadenopathy after the completion of antitubercular therapy. CBNAAT was positive in 8 of the 17 patients (47.1%), and TB culture was positive in 6 of the 17 patients (35.3%). Among these patients, 5 (29.4%) were positive by both CBNAAT and TB culture, 3 (17.6%) were CBNAAT positive but TB culture negative, 1 (5.9%) was CBNAAT negative but TB culture positive, and 8 (47.1%) were

negative by both tests. A total of 2 patients (11.7%) had MDR-TB. Of the 6 patients who were CBNAAT positive with rifampicin-sensitive TB, the TB culture was positive in 3 patients, whereas for 2 patients who were CBNAAT positive with rifampicin- resistant TB, the TB culture was positive in 2 patients as shown in Figure 1. AFB smears were positive in 4 patients.

EBUS-TBNA samples from 16 patients (21.6%) were inadequate for histopathological diagnosis due to excessive red blood cells and scant lymphoid cells. Therefore, these samples were rendered inadequate, leading to inconclusive results. Microbiological analysis for TB was negative in all these samples. Among the 16 patients with inconclusive EBUS-TBNA results, 10 underwent additional diagnostic evaluation combined with clinical–radiological follow-up. This included bronchoscopy with bronchoalveolar lavage, transbronchial lung biopsy, and endobronchial biopsy in four patients; repeat EBUS in five patients; and supraclavicular lymph node biopsy in one patient. Six patients were monitored with clinical–radiological follow-up alone. Among these 16 patients, 6 were subsequently diagnosed with a pathological cause of IML (sarcoidosis, n=2; lymphoma, n=1; TB, n=1; and reactive, n=2), and EBUS-TBNA was categorized as a false negative. The remaining patients were categorized as true negatives after completing a one-year follow-up period (n = 10; Figure 2). The procedure demonstrated high diagnostic accuracy, with a sensitivity of 90.6% (95% CI: 83.5–97.8%), specificity of 100% (95% CI: 69.2–100%), positive predictive value of 100% (95% CI: 94–100%), and negative predictive value of 62.5% (95% CI: 38.8–86.2%).

Discussion

This is the first study from the northeastern part of India in which EBUS-TBNA was used as an initial investigation for isolated mediastinal lymphadenopathy requiring pathological diagnosis. The diagnostic yield of EBUS-TBNA in our study was 78.6%. Adequacy was determined on the basis of the presence of cytological features such as atypical cells, granulomas, or reactive lymphoid tissue. Other studies evaluating suspected cases of mediastinal metastasis of lung cancer or a diagnosis of suspected malignancy reported diagnostic yields between 93.5% and 100% [12-14].

The diagnostic yield for suspected cases of malignancy in our study was 96.4%. However, studies have evaluated enlarged mediastinal lymph nodes irrespective of etiology, with a diagnostic yield of 88% by Dhamija et al., 78% by Gupta et al. and 69.2% by Srinivasan et al. [15-17]. Our findings were consistent with those of studies that have examined the learning curve and early experiences of centers initiating an EBUS-TBNA program, particularly those assessing mediastinal lymphadenopathy regardless of the underlying cause [17-19].

Recent studies have continued to support the diagnostic utility of EBUS-TBNA in the evaluation of both malignant and benign mediastinal lymphadenopathy, particularly in regions where

tuberculosis remains highly prevalent. In a recent study, Aljohaney et al. reported that EBUS-TBNA achieved a diagnostic yield of approximately 83.9% when histological and microbiological findings were considered together in patients evaluated for intrathoracic tuberculosis-like lymphadenopathy, further supporting its role as an effective and minimally invasive diagnostic modality in tuberculosis-endemic settings [20]. Likewise, Akyüz et al. demonstrated meaningful diagnostic sensitivity in a real-world cohort of patients with tuberculous lymphadenopathy, reinforcing the role of EBUS-TBNA as a minimally invasive first-line diagnostic approach [21]. In a TB-endemic setting such as India, a high index of clinical suspicion justifies the use of molecular diagnostic tools such as CBNAAT, which not only increases the diagnostic yield but also facilitates early detection of rifampicin resistance. In this study, 17 (23%) patients were diagnosed with TB. Among the 17 patients diagnosed with tuberculous lymphadenitis, microbiological confirmation via the CBNAAT was achieved in 8 patients (47.1%). Among these, rifampicin resistance was identified in 2 patients (11.7%). This facilitated timely diagnosis of MDR-TB and initiation of appropriate therapy, highlighting the utility of molecular diagnostics in optimizing TB management in high-burden settings. GeneXpert has demonstrated high sensitivity and specificity for diagnosing MTB and rifampicin resistance in respiratory samples, particularly in expectorated lower airway secretions. The integration of GeneXpert applied to EBUS-obtained samples, has further strengthened microbiological confirmation and enabled earlier detection of drug resistance in tuberculosis [22]. However, its diagnostic performance in nonrespiratory (extrapulmonary) samples remains poorly characterized, with relatively limited data available to date [23-26]. Overall, the sensitivity of GeneXpert for microbiologically proven TB in our study (47.05%) is consistent with reported sensitivities ranging from 38 to 77% in EBUS-TBNA samples [27-30]. Several studies evaluating the role of EBUS-TBNA in the diagnosis of mediastinal tuberculous lymphadenitis have reported culture positivity rates ranging between 25% and 62%. [29-33]. In the present study, 35.29% of the TB cultures were positive, similar to the findings of studies by Lucey o et al. [34]. The low culture positivity in mediastinal tuberculous lymphadenitis may be attributed to several factors, including the low number of AFB or nonviable TB bacilli in the lymph nodes, insufficient viable cellular material from necrotic areas, and technical difficulties in successfully culturing MTB [23]. The lower sensitivity of culture than GeneXpert is often linked to the paucibacillary nature of extrapulmonary samples, where bacilli are unevenly distributed and tend to form clumps [35].

Although EBUS-TBNA is a valuable diagnostic tool, it can be difficult at times to distinguish between sarcoidosis and TB, especially when TB patients present with noncaseating granulomas [36]. This was evident in our study, where two patients with positive cultures for

MTB also exhibited noncaseating granulomas on histopathology. Therefore, it is important to recognize that TB may also be associated with noncaseating granulomas [37].

Sarcoidosis is a common cause of mediastinal granulomatous lymphadenopathy in nondeveloping countries. Interestingly, a study by Kuo et al. revealed that TB can also be observed in patients with intrathoracic lymphadenopathy without lung involvement, even in countries where TB is common [38]. A definitive diagnosis relies on a combination of clinical and radiological findings, histological confirmation of noncaseating granulomas, and careful exclusion of other potential causes of granulomatous inflammation, including TB and lymphoma [39]. Trisolini et al. reported that the diagnostic accuracy of EBUS-TBNA in patients with sarcoidosis was 79% [40]. Other studies have demonstrated sensitivities of 80% to 91.4% [41-43]. The diagnostic accuracy in our study was 80%, which is consistent with the findings of published studies.

The etiological spectrum of mediastinal lymphadenopathy is influenced by regional epidemiology. In high-income countries, noninfectious conditions such as sarcoidosis, lymphoma, and intrathoracic malignancies predominate. However, in tuberculosis-endemic regions, infectious causes—particularly MTB—remain a leading cause and warrant early consideration in the diagnostic workup. However, the most common diagnosis in our study was malignancy, which was observed in 27 patients (36.5%). The use of both smoked and smokeless tobacco was notably more prevalent in this region than in other parts of the country, contributing to an increased risk for developing head and neck cancers [44]. Therefore, the high degree of malignancy observed in our study can be attributed to the widespread use of tobacco products such as cigarettes, betel nuts and smokeless tobacco (a mixture of crushed areca nuts, tobacco, slaked lime, catechu, sweeteners and flavoring agents), which are common in the seven states of Northeast India. Especially in states such as Assam and Meghalaya, chewing areca nuts—locally known as tamul or kwai—is a long-standing cultural tradition. It is typically prepared with betel leaves and a small amount of slaked lime and is deeply woven into daily life, symbolizing hospitality, respect, and social bonding [45,46]. Dietary habits such as the consumption of spicy food, hot beverages, fermented pork fat, smoked and dried salted meat and fish and the use of soda as a food additive have been linked to increased malignancy in this region [47]. A meta-analysis of regional data from South Asia and the Pacific revealed a strong epidemiological association between smokeless tobacco use and increased oral cancer incidence [48].

Of the sixteen patients with inconclusive EBUS-TBNA findings, ten were later classified as true negatives following a one-year period of clinical and radiological surveillance. The absence of any progressive or new disease in these patients suggests that the enlarged mediastinal nodes were pathologically inactive. Such findings can be explained by post-inflammatory or fibrotic

changes within lymph nodes that remain enlarged despite the resolution of the underlying process. In regions where TB and other granulomatous diseases are prevalent, healed or inactive lesions often persist radiologically even when microbiological activity has ceased. Additionally, transient immune-mediated nodal enlargement or overestimation of size on imaging could also account for the apparent lymphadenopathy. These explanations are consistent with earlier observations that benign or fibrotic lymph nodes may remain radiologically enlarged without harboring active pathology [14,29,31].

EBUS-TBNA demonstrated a favorable safety profile in our study. Only two patients experienced minor bleeding during the procedure, which resolved spontaneously without any need for medical intervention. No major or long-term complications were observed. These outcomes are in line with the literature reporting a low overall complication rate for EBUS-TBNA, supporting its excellent safety profile. Notably, this study was conducted in one of the largest referral centers in Northeast India, which, at the time, was the only facility offering EBUS-TBNA in the state. This allowed us to evaluate a broad and diverse group of patients from all the states of north east India, reflecting real-world clinical scenarios and referral practices. The cost of EBUS-TBNA was £10, making it affordable for all patients irrespective of their socioeconomic status. This is much lower than the costs of EBUS-TBNA estimated to be £1,050 and £1382 in the United States and National Health Service of the United Kingdom, respectively [1].

While our findings are encouraging, there are several limitations to consider. First, the retrospective and single-center nature of the study may introduce selection bias and limit the generalizability of the results. Second, ROSE data were not available during the study period because of the lack of an onsite cytopathologist. This may have affected real-time assessment of sample adequacy and potentially influenced the need for repeat procedures in some cases. Finally, the study excluded patients with anterior mediastinal lymphadenopathy, as these nodes are anatomically inaccessible via the EBUS approach. Despite these constraints, our study provides important insight into the performance of EBUS-TBNA in a real-world, high-burden setting and adds to the growing evidence supporting its role in the minimally invasive diagnosis of mediastinal pathology.

Conclusions

EBUS-TBNA is a safe and highly sensitive modality for evaluating IML. In patients with negative EBUS-TBNA findings, clinical surveillance may be a reasonable approach when carefully selected. The use of GeneXpert on TBNA samples plays a vital role in confirming mediastinal tuberculous lymphadenopathy and in detecting rifampicin resistance, thereby guiding timely and appropriate management. Furthermore, the study highlights an emerging epidemiological

trend in the Northeastern Indian population, where habitual and dietary factors may be contributing to a higher burden of malignancy, emphasizing the importance of comprehensive diagnostic evaluation in all cases of IML.

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Table 1. Clinical characteristics of patients undergoing EBUS-TBNA.

Characteristics of the study population n(%)	
Total patients	74
Male	50(67.6)
Female	24(32.4)
Average age (years)	50.23
Symptoms	
Cough	41(55.4)
Fever	23(31.08)
Breathlessness	11(14.8)
Weight loss	07(9.4)
Chest pain	03(4.05)

EBUS-TBNA, endobronchial ultrasound-guided transbronchial needle aspiration.

Table 2. Endobronchial ultrasound features of lymph nodes of various etiologies.

Axis Diameter(mm)				
	Tuberculosis	Sarcoidosis	Malignant	Reactive
Long Axis(mm)	33.14	31.38	35.72	21.70
Short Axis(mm)	14.36	18.42	23.64	11.62
Shape				
Round	04	01	18	03
Oval	08	03	09	07
Irregular	05	00	00	00
Echogenicity				
Heterogenous	13	01	19	00
Homogenous	04	03	08	10
Central necrosis sign	07	00	04	00
Calcification	06	01	03	00

Table 3. Diagnosis obtained through EBUS-TBNA.

Diagnosis	**n(%)**
Malignancy	27(36.5)
Tuberculosis	17(23)
Sarcoidosis	4(5.4)
Reactive	10(13.5)
Inconclusive	16(21.6)

EBUS-TBNA, endobronchial ultrasound-guided transbronchial needle aspiration.

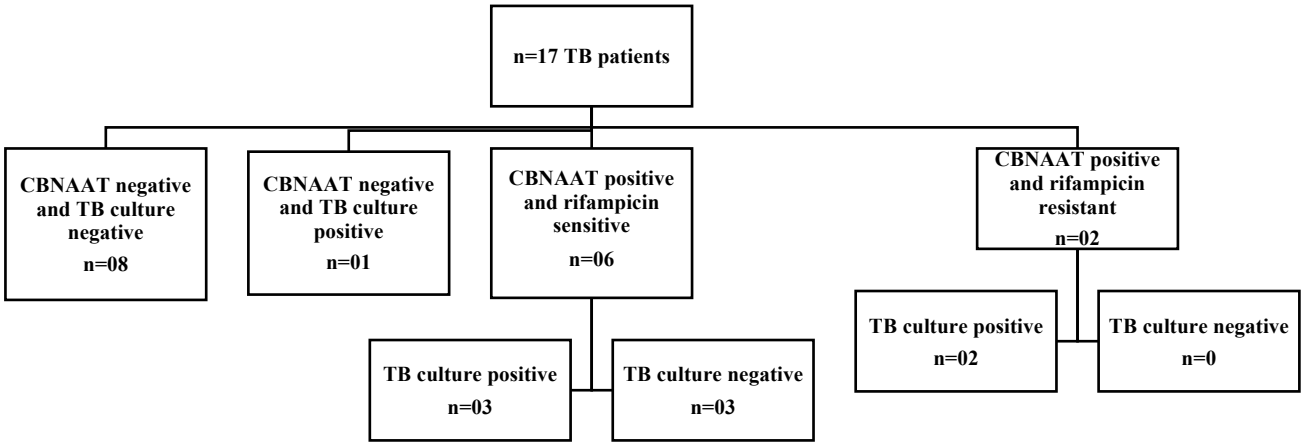

Figure 1. CBNAAT and tuberculosis (TB) culture in the study population diagnosed with TB. CBNAAT, cartridge-based nucleic acid amplification test.

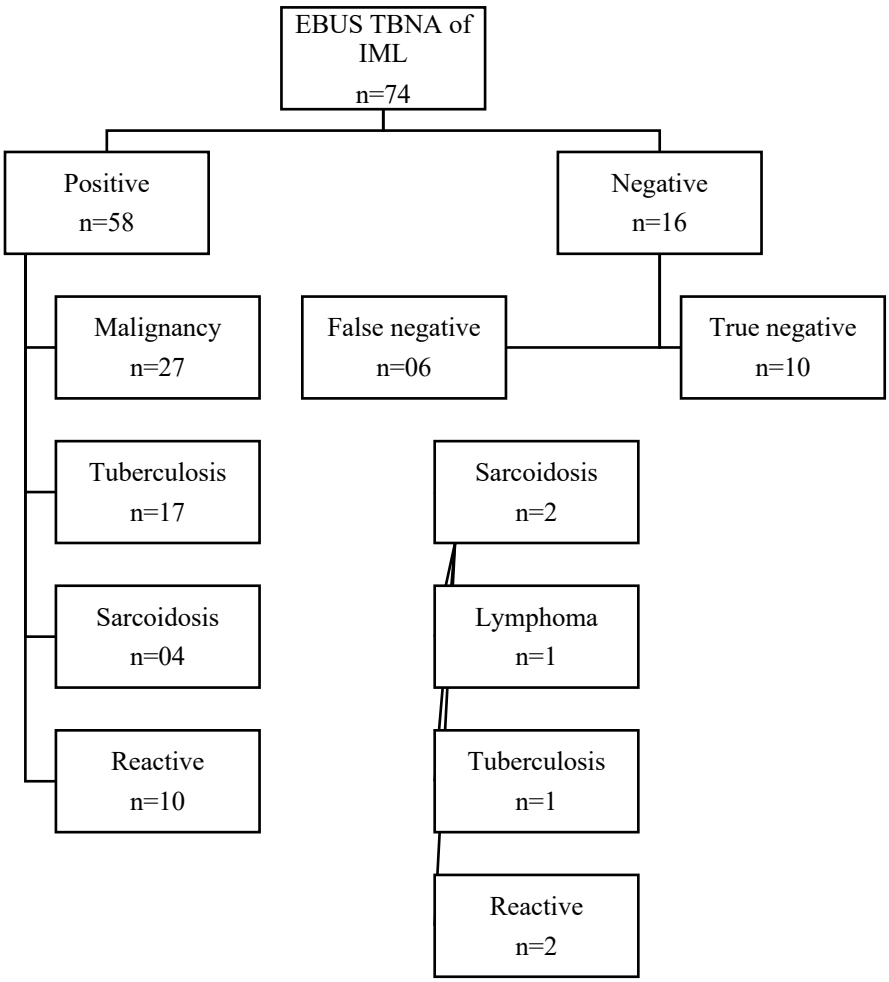

Figure 2. Diagram illustrating the diagnostic accuracy of endobronchial ultrasound-guided transbronchial needle aspiration (EBUS-TBNA) in patients with isolated mediastinal lymphadenopathy (IML).