Is endobronchial ultrasound-guided transbronchial needle aspiration an effective diagnostic procedure in restaging of non-small cell lung cancer patients?

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ABSTRACT

Background and Objectives: Selecting the diagnostic procedure for mediastinal restaging after chemotherapy and/or radiotherapy in Stage IIIA-N2 non-small cell lung cancer (NSCLC) patients remains a problem. The aim of the study was to determine the efficacy of endobronchial ultrasound-guided transbronchial needle aspiration (EBUS-TBNA) for the evaluation of mediastinal lymph nodes in the restaging of NSCLC patients. Materials and Methods: The present multicentric study retrospectively analyzed the results of Stage IIIA-N2 NSCLC patients who had undergone EBUS for mediastinal restaging after preoperative chemotherapy or radiotherapy or both. Results: In 44 patients with 73 N2 nodes, malignant cells were identified in EBUS-TBNA from 23 patients (57.5%) and 25 lymph nodes (34.2%). Twenty-one patients (42.5%) and 48 lymph nodes (65.8%) were negative for nodal metastasis. All of these patients with negative results subsequently underwent mediastinoscopy or surgery (n = 9 and n = 12, respectively). Metastasis was detected in 5 (23.8%) of 21 patients and 6 (12.5%) of 48 lymph nodes. The diagnostic sensitivity, specificity, positive predictive value, negative predicted value and accuracy of EBUS-TBNA based on number of patients were 82.1%, 100%, 100%, 76.2%, and 88.6%, respectively. Conclusions: EBUS-TBNA should be done before invasive procedures in restaging of the mediastinum in patients previously treated with neoadjuvant therapy because of high diagnostic accuracy rate. However, negative results should be confirmed with invasive procedures such as mediastinoscopy and thoracoscopy.

Key words: Lymphatic diseases, neoplasm staging, neoplasms, ultrasonography

INTRODUCTION

Malignant lymph nodes are common targets for the evaluation of non-small cell lung cancer (NSCLC)
therapeutics. The absence of mediastinal metastases at the time of resection is a major prognostic factor for survival in Stage IIIA NSCLC patients.\(^1\) Today, noninvasive techniques such as computed tomography (CT) or positron emission tomography (PET)/CT are not accurate enough to make the final decision about mediastinal restaging based on their results. CT has low accuracy in primary staging, so it also has low accuracy in restaging of the mediastinum, as expected.\(^2\) Although PET/CT has better results than CT, the occurrence of false negative (FN) and also false positive (FP) results remains a problem.\(^2\)–\(^5\)

In recent years, repeat mediastinoscopy, an invasive procedure, has been used for restaging of the mediastinum in experienced hands even though it was known as difficult to perform because of scar tissue development. Its sensitivity was reported to range widely from 29% to 87.5%\(^6\)–\(^8\) and in some cases, re-mediastinoscopy did not adequately sampled subcarinal lymph node.\(^9\) Transcervical extended mediastinal lymphadenectomy (TEMLA) had better results than re-mediastinoscopy, but it is also another surgical procedure, such as anterior mediastinotomy and videothoracoscopy.\(^9\)–\(^10\)

Nowadays, minimal invasive procedures such as endobronchial ultrasound-guided transbronchial needle aspiration (EBUS-TBNA) and transesophageal ultrasound-guided fine needle aspiration (EUS-FNA) are used for the sampling of mediastinal lymph nodes. Both techniques have had successful results for the initial staging of lung cancer.\(^11\)–\(^12\) There have been an increasing number of reports that analyze EBUS-TBNA accuracy in the restaging of the mediastinum in NSCLC patients.\(^13\)–\(^16\) Therefore, we aimed to analyze the success rates of EBUS-TBNA results for restaging mediastinal lymph nodes after neoadjuvant therapy for NSCLC Stage IIIA-N2 patients in a multicenter study.

**MATERIALS AND METHODS**

The present study was approved by the Ethics Review Board of Erciyes University (No. 2015-286).

**Patients**

The present multicentric study retrospectively analyzed the results of Stage IIIA-N2 NSCLC patients in four centers who had undergone EBUS for mediastinal restaging after neoadjuvant therapy between January 2011 and December 2013. The inclusion criteria were as follows:

1. NSCLC patients with ipsilateral or subcarinal lymph node metastases (Stage IIIA-N2) which had been previously diagnosed pathologically
2. Patients who received at least three cycles of a platinum-based chemotherapy regimen or radiotherapy or both
3. Patients who had stable disease or partial response after preoperative therapy defined by radiologists
4. Negative results for lymph node metastasis by EBUS-TBNA verified by cervical mediastinoscopy or thoracotomy.

Eighty patients underwent EBUS-TBNA in the study period. We excluded 36 patients who had a negative result by EBUS-TBNA and which were not verified by mediastinoscopy or thoracotomy. Although 16 of these 36 patients had clinical follow-up, we excluded them because we had no pathological data. Ten patients did not accept the surgical procedure, and we had no follow-up data on the remaining ten patients due to the retrospective design of the present study. Eventually, we analyzed the data of 44 patients who had positive results after EBUS-TBNA or had pathological confirmation surgically (thoracotomy or mediastinoscopy) after negative EBUS-TBNA results. If the first staging was done by EBUS-TBNA, mediastinoscopy was performed to verify the negative EBUS-TBNA results. If the first staging was done by mediastinoscopy, thoracotomy was performed to verify negative EBUS-TBNA results.

**Endobronchial ultrasound-guided transbronchial needle aspiration procedure**

Endobronchial ultrasonography was conducted using a fiberoptic ultrasound bronchoscope (Convex Probe EBUS; BF-UC 160F-OL8; Olympus Medical Systems, Tokyo, Japan). The location, shape, and structure of the lesions were examined with ultrasound. The locations of the stations were named and numbered using the lymph node map proposed by Mountain.\(^17\) After the bronchoscope was guided to the target area, during real-time imaging, a 22-gauge aspirating needle with a syringe connected proximally (model NA-201SX-4022, Olympus, manufactured for this purpose) was pushed out from the distal tip of the bronchoscope, and samples consisting of cells or tissue fragments were obtained as previously described.\(^18\) The aspirate was smeared onto glass slides, air-dried or fixed immediately.
with 95% alcohol, and stained with hematoxylin and cosin (H and E). Histological cores were fixed with 10% neutral buffered formalin and stained with H and E. Immunohistochemical staining was also performed when considered necessary. A rapid onsite cytopathological examination was not performed. Cytopathological specimens were categorized as (i) positive (adequate sample with the presence of malignant cells) or (ii) negative (sample consisting of mature lymphocytes and no malignant cells). Samples from all patients contained lymphocytes, and there were no inadequate samples by EBUS-TBNA.

**Mediastinoscopy and thoracotomy**

In the present study, when the EBUS-TBNA cytopathologic results were positive, they were assumed to be true positives (TPs), and additional diagnostic procedures were not performed. However, if the cytopathologic results were negative, cervical mediastinoscopy was performed. For patients whose initial staging was done by cervical mediastinoscopy, restaging by EBUS-TBNA was confirmed during thoracotomy.

**Statistical analysis**

Descriptive statistics are presented in frequency, percentage, median, and minimum and maximum values. The diagnostic sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and accuracy of EBUS-TBNA were calculated as follows: Sensitivity (TP/[TP + FN]), Specificity (True negative(TN)/[TN + FP]), PPV (TP/[TP + FP]), Negative predictive value (TN/[TN + FN]), Diagnostic accuracy ([TP + TN]/total patients).

The data were entered into a database and analyzed with the SPSS statistical software package (SPSS 18.0 Chicago, Illinois, USA). The FN results were compared between each two groups by the two proportions test. \( P < 0.05 \) was considered to be statistically significant.

**RESULTS**

EBUS-TBNA was performed in 44 IIIA-N2 NSCLC patients after neoadjuvant therapy. There were 39 male and 5 female patients, with a mean age of 58.3 ± 8.6 years. Pathology showed squamous cell carcinoma in 22 patients, adenocarcinoma in 10 patients, and mixed NSCLC and small cell lung cancer (SCLC) in 1 patient. NSCLC was not subtyped pathologically in 11 patients. The characteristics of the patients are shown in Table 1.

Seventy-three N2 nodes were sampled in 44 patients. Samples from all patients contained lymphocytes. The most frequent sampled lymph node was in the right lower paratracheal station (47.9%). The median short-axis size of the lymph nodes seen at EBUS was 10 mm (range, 4.6–35 mm). The median number of lymph nodes sampled was 1 (range, 1–3). There were no serious complications related to EBUS-TBNA. The characteristics of lymph nodes are shown in Table 2.

In 44 patients with 73 N2 nodes, malignant cells were identified in biopsies from 23 patients (57.5%) and 25 lymph nodes (34.2%). Twenty-one patients (42.5%) and 51 lymph nodes (69.9%) were negative for nodal metastasis. All of these patients with negative results subsequently underwent mediastinoscopy or surgery (\( n = 9 \) and \( n = 12 \), respectively). Metastasis was detected in 5 (23.8%) of 21 patients. There were six FN results for EBUS-TBNA per lymph node basis in these five patients. The location and numbers of FN lymph nodes were as follows: station 4R (\( n = 3 \)) and station 7 (\( n = 3 \)). We had 16 (76.2%) true negative (TN) results for patient basis and

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### Table 1. Characteristics of patients

| Characteristics                  | n (%) |
|----------------------------------|-------|
| **Gender**                       |       |
| Male                             | 39 (88.6) |
| Female                           | 5 (11.4) |
| **Mean age (years)**             |       |
| Right                            | 58.3±8.6 |
| Left                             | 58.3±8.6 |
| **Tumor location**               |       |
| Right                            | 34 (77.3) |
| Left                             | 10 (22.7) |
| **Histology**                    |       |
| NSCLC (not subtyped)             | 11 (25.0) |
| Squamous                         | 22 (50.0) |
| Adenocarcinoma                   | 10 (22.7) |
| Mixed NSCLC and SCLC             | 1 (2.3) |

Data are presented as n (%) or average±SD. NSCLC: Non-small cell lung cancer, SCLC: Small cell lung cancer, SD: Standard deviation

### Table 2. Characteristics of 73 lymph nodes sampled by endobronchial ultrasound-guided transbronchial needle aspiration

| Examined site | n (%) |
|---------------|-------|
| 4 right       | 35 (47.9) |
| 4 left        | 9 (12.3) |
| 7             | 29 (39.7) |
| **Node size** (mm) |       |
|               | 10 (4.6–35) |
| **Number of lymph nodes per patient** | 1 (1–3) |

*Data represent median (minimum-maximum)
42 (87.5%) TN results per lymph node basis. The location and numbers of TN results for lymph nodes were as follows: station 4R \( (n = 17) \), 4L \( (n = 3) \), and 7 \( (n = 22) \). Figure 1 shows the outcomes for patients with Stage IIIA-N2 NSCLC who underwent EBUS-TBNA. Seven patients had negative results after mediastinoscopy, and all of them had also negative results after thoracotomy.

The diagnostic performance results of EBUS-TBNA for the diagnosis of mediastinal metastases in patients with NSCLC are shown in Table 3. The diagnostic sensitivity, specificity, PPV, NPV, and accuracy rates of EBUS-TBNA as per number of patients were 82.1%, 100%, 100%, 76.2%, and 88.6%, respectively. The diagnostic sensitivity, specificity, PPV, NPV, and accuracy rates of EBUS-TBNA as per number of nodal stations were 80.6%, 100%, 100%, 91.4%, and 90.7%, respectively. The FN rates for 4R and subcarinal stations were 8.6% (3/35) and 10.3% (3/29), respectively, and the difference was not statistically significant \((P > 0.05)\). There were no FN results for 4L \((0\%, 0/9)\). The FN rate difference for 4L with 4R and subcarinal stations was also statistically not significant \((P > 0.05)\). The diagnostic accuracy rates of 4R, 4L, and subcarinal stations were 91.4%, 100%, and 89.6%, respectively. In addition, 33 lymph nodes were \(>10 \text{ mm} \) and 40 lymph nodes were \(\leq 10 \text{ mm} \). The sensitivity, NPV, and diagnostic accuracy were 88.2%, 88.9%, 93.9% and 71.4%, 86.7%, and 90.0% in the lymph nodes \(>10 \text{ mm} \) and \(\leq 10 \text{ mm} \), respectively.

Eventually, the prevalence of mediastinal lymph node metastases in the present study per patient basis was 63.6% (25/44) and per lymph node basis was 42.5% (31/73). Twenty-three patients (57.5%) had positive results for metastasis after EBUS-TBNA, so they were not required to undergo invasive surgical procedures.

**DISCUSSION**

Three major reports evaluated the mediastinum after neoadjuvant therapy in NSCLC patients with EBUS-TBNA, and they had a sensitivity of 50%–76% and diagnostic accuracy of 76%–89%.\(^{14-16}\) In the present report, the sensitivity of EBUS-TBNA was 82.1%, and the diagnostic accuracy was 88.6% in patients with Stage IIIA-N2 NSCLC; the sensitivity rate is better than the previous reports, and the success rate is similar with them. In addition, the sensitivity and the diagnostic accuracy were 80.6% and 90.7% per node station basis in our report. In fact, EBUS-TBNA seems to be a good diagnostic minimally invasive procedure in patients with Stage IIIA-N2 NSCLC.

CT has low accuracy in restaging of the mediastinum with a diagnostic accuracy of 58%–60%.\(^{12,13}\) In the previous reports, PET/CT had high sensitivity rates of 73%–92%.\(^{12,4,8}\) However, because of FP results,
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its specificity changed between 62% and 89%.\cite{2,4,5}
In tuberculosis endemic countries, such as ours, this problem increases due to the lymph node involvement of tuberculosis and the specificity of PET/CT can be decreased up to 35%.\cite{19} From another perspective, Collaud et al. showed that PET/CT revealed new fluorodeoxyglucose-positive lesions in 6 of 31 (20%) Stage III NSCLC patients at the restaging time, but all these lesions proved to be benign with invasive procedures.\cite{20} However, in the previous reports, EBUS-TBNA had a specificity of 86%–100%, and in the present study, it also had a specificity of 100%.\cite{14,16,17}

Thus, all of these results show us that histopathologic diagnosis is required because reliance on imaging techniques alone is not sufficient to confirm metastasis.

EUS-FNA had a sensitivity of 44%–75%, NPV of 42%–67%, and diagnostic accuracy of 60%–83% in patients with NSCLC after neoadjuvant treatment.\cite{21,22}

The sensitivity and diagnostic accuracy results of EUS-FNA were almost the same as EBUS-TBNA results. However, there are no reports about EBUS combined with EUS (combined ultrasound) in restaging. In a recent report, Zielinski et al. evaluated the mediastinum in NSCLC after neoadjuvant treatment by EBUS in 32 patients, EUS in 6 patients, and CUS in 50 patients.\cite{13}
They found that the sensitivity and NPV were 64.3% and 82.1%, respectively. They also compared the restaging results with TEMLA, and the sensitivity and NPV were significantly superior in favor of TEMLA (the sensitivity was 96.6% and NPV was 98.5% in the TEMLA group). However, there was no morbidity in the EBUS, EUS, or CUS groups, but the morbidity rate was 6.4% in the TEMLA group.

In addition, they compared all EBUS, EUS, and CUS results with TEMLA; however, they did not compare just the CUS results with TEMLA. Thus, good results alone with EBUS and EUS show us that a study comparing CUS results with surgical procedures is needed.

Another invasive procedure, namely, re-mediastinoscopy had very different sensitivity rates in the previous reports.\cite{6,23}\n
Pauwels et al. evaluated re-mediastinoscopy success in a small number of patients \((n = 15)\), and they found that it had a sensitivity of 87.5% and diagnostic accuracy of 93.7% which are the highest reported rates in the literature.\cite{8} Three studies with large patient goups \((n = 96, n = 104 \text{ and } n = 165)\) found that it had a sensitivity of 61%–88% and diagnostic accuracy of 87%–93% in the restaging of the mediastinum.\cite{23,24}\n
However, de Leyn et al. found that re-mediastinoscopy had a sensitivity of 29% and diagnostic accuracy of 60% in 30 patients.\cite{8} Because of adhesions and fibrosis, they did not adequately sample in 67% of cases, so they explained these low rates by this technical difficulty. Although mediastinoscopy is a successful procedure for initial staging, adhesion and fibrosis may be an explanation for the low success rates of re-mediastinoscopy.\cite{9}

Another point to remember is that mediastinoscopy did not reach N1 nodes such as 10R, 10L, 11Rs, and 11L. However, it is easy to sample these nodes by EBUS-TBNA.\cite{25}

Initial staging of the mediastinum with EBUS-TBNA in patients with NSCLC also had higher rates than restaging with the same procedure.\cite{13}\n
These low rates for the second procedure are due to the FN results. Herth et al. explained this difference in different ways.\cite{13}

First, the metastatic lymph nodes undergo necrosis and fibrosis after chemotherapy. Second, malignant cells may be focal within the node, and finally, necrosis within the aspirate makes pathologic interpretation more difficult. In addition, Szlubowski et al. explained these FN results with the fact that the posterior and inferior parts of stations 4L and 7 are more difficult to visualize and to conduct a biopsy on.\cite{14}\n
Moreover, they found FN results only in small nodes. However, in our report, there are no significant differences among the 4R, 4L, and subcarinal stations with NPVs. In addition, we found that lymph nodes >10 mm or ≤10 mm did not affect the NPV rates (88.9% vs. 86.7%, respectively). Therefore, our findings are similar to the first report that explained FN results with the lymph node necrosis, fibrosis, and the focal localization of malignant cells in the node.

In the present study, 57.5% of patients had positive results for metastasis after EBUS-TBNA so that they were not required to undergo invasive surgical procedures. However, 57.1% of patients with EBUS-TBNA negative results were restaged by thoracotomy because these patients’ initial staging was done by mediastinoscopy. After thoracotomy, we found that one-fourth of these patients had mediastinal metastasis. Herth et al. also evaluated the mediastinum by thoracotomy after restaging with EBUS-TBNA.\cite{13}

They found that 28 of 35 (80%) EBUS-TBNA negative patients had positive results. These data show us that the negative results obtained with EBUS-TBNA should be confirmed with invasive methods before surgery in restaging of the mediastinum. The first staging should
be done by EBUS-TBNA to reserve mediastinoscopy for the second staging because re-mediastinoscopy sensitivity rates varied widely in the previous reports.

The present study had several limitations. It was retrospective in design and involved a relatively small number of patients. In addition, NSCLC was not subtyped in some of the patients.

CONCLUSIONS

EBUS-TBNA should be conducted before invasive procedures in restaging of the mediastinum in patients previously treated with neoadjuvant therapy because of its high diagnostic accuracy rate. However, negative results should be confirmed with invasive procedures such as mediastinoscopy and thoracoscopy.

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Conflicts of interest
There are no conflicts of interest.

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