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Clinical Stem 1

Nonsmoking female with new right upper lobe lung mass and three PET positive mediastinal lymph nodes[1]

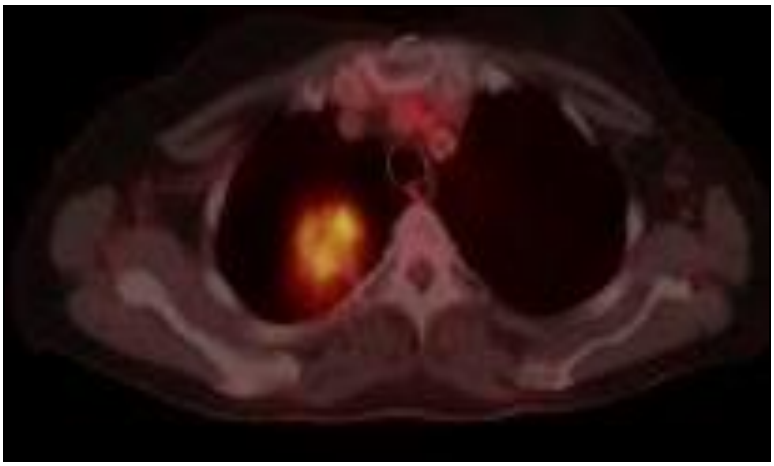
Learning objectives: the webcast participant will be able to:

1. Describe mediastinal staging and EBUS-guided nodal-sampling strategies.
2. Describe the rationale for downstream molecular analysis of small specimens.
3. Describe ways to assure specimen adequacy for diagnosis and molecular testing.
4. Describe indications for repeat biopsy in case of disease progression.
5. Describe how molecular analysis of adequate specimens helps determine prognosis and potential response or resistance to therapy.

[1] Disclaimer: This is a fictitious clinical case scenario based on a conglomerate of real patient data, modified to avoid any possibility for patient identification and to help meet educational objectives. Any resemblance to real persons, living or deceased is purely coincidental.

Case Description

A 65 year old white, nonsmoking female required preoperative medical clearance before knee surgery. Past medical history was positive for asthma since childhood. She complained of occasional dyspnea and wheezing. Physical examination was normal except for signs of left medial meniscus injury. She had minor memory loss but was independent in activities of daily living. She lived alone and had no children. Family history was unremarkable. A chest radiograph showed a 4 x 5 cm mass in the right upper lobe. Computed tomography confirmed the mass and also showed a 1.2 cm right lower paratracheal lymph node, a 1 cm left lower paratracheal node, and a 1.7 cm subcarinal node. Whole body PET CT showed increased uptake in the primary tumor and in mediastinal lymph nodes stations 4R, 7 and 4L. Brain MRI was normal.



Clinical Stem

Question 1: If this patient has primary lung cancer, which of the following most accurately predicts survival?

A. Productive cough
B. Exertional dyspnea
C. Performance status

Answer: C

Estimated survival is an important factor for decision making in all disease processes. If this patient has primary lung cancer, she would likely be clinically staged IIIB because of the contralateral mediastinal lymph node at station 4L. In such cases of advanced cancer, prognostic considerations are important because treatment goals may change from prolonging life at any cost, to palliating symptoms, preserving quality of life, and maintaining dignity.

Estimating survival based on objective data is warranted because subjective assessments of predicted survival are often incorrect and overly optimistic. Patients usually want their doctors to be realistic yet hopeful prognosticators. While exact statistics are not always shared, physicians who address prognosis can conduct meaningful and honest discussions with patients and their families.

Performance status, along with heart rate, blood pressure, temperature, respiratory rate and pain level, is an important vital sign in clinical oncology. Because performance status is the strongest prognostic indicator of survival in patients with cancer, it is frequently used as an entry criterion and adjustment factor in clinical trials. One commonly used measure of performance is the Karnofsky Performance Status score that uses a 0-100 range in ten point increments to measure functional impairment. Lower scores correlate with worsened survival for most serious illnesses.

Results from the analysis of 100 variables from several studies showed that dyspnea, dysphagia, weight loss, xerostomia, anorexia, and cognitive impairment were strongly and independently associated with cancer patient survival. These signs and symptoms were outranked, however, by the assessment of performance status.

References:

1. Lamont EB, Christakis NA. Survival estimates in advanced cancer. Available at www.UpToDate.com.
2. Hollingsworth HM. Wheezing and stridor. *Clin Chest Med* 1987;8:231–240.

Question 2: Which of the following four procedures would you perform next?

A. Mediastinoscopy
B. EBUS-guided TBNA
C. Conventional TBNA
D. Esophageal ultrasound guided FNA

Answer: B

Any of the four procedures could be performed for diagnosis, so decisions might depend on equipment availability, available expertise, or institutional bias. For lymph node station 4L, the yield is usually higher using EBUS-TBNA than using conventional TBNA. Data from a meta-analysis of EBUS-TBNA for patients with confirmed or suspected NSCLC showed that a subgroup of patients selected on the basis of CT or PET positive results had a pooled sensitivity of 94%; higher than a subgroup of patients who had not been selected according to CT or PET results.

This patient has an enlarged, PET positive lymph node. The yield of EBUS-TBNA is expected to be greater than 90%. A negative EBUS-TBNA in patients with highly suspected mediastinal nodal metastases, however, should be followed by mediastinoscopy. Mediastinoscopy allows systematic

exploration and biopsy under visual guidance of nodal stations 1, 2, 3, 4 and 7 but has associated morbidity in lesser experienced hands.

Diagnosis can probably be obtained bronchoscopically: stations 4R, 4L, and 7 can be accessed using conventional TBNA, especially using rapid on-site cytology examination, also known as ROSE. Diagnosis might also be possible using EUS-FNA to sample nodal stations 4L and 7.

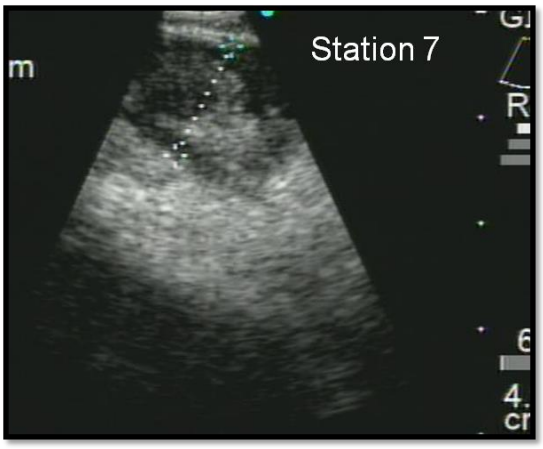
Up to 28% of patients known or suspected lung cancer and a high clinical suspicion of nodal disease might have mediastinal nodal metastases confirmed by mediastinoscopy after negative EBUS-TBNA.

References:

1. Herth FJ, Lunn W, Eberhardt R, et al. Transbronchial versus transesophageal ultrasound-guided aspiration of enlarged mediastinal lymph nodes. Am J Respir Crit Care Med. 2005 May 15;171:1164-7
2. Gu P, Zhao Y, Jiang L, et al. Endobronchial ultrasound-guided transbronchial needle aspiration for staging of lung cancer: A systematic review and meta-analysis. Eur J Cancer 2009; 45, 1389-1396
3. Defranchi SA, Edell ES, Daniels C E et al. Mediastinoscopy in Patients with Lung Cancer and Negative Endobronchial Ultrasound Guided Needle Aspiration. Ann Thorac Surg 2010; 90: 1753-8.

The Case Continues

White light bronchoscopy, EBUS-TBNA and ROSE were performed while the patient was under general anesthesia. No airway abnormalities were seen. EBUS examination of the mediastinum was normal except for enlarged nodes at levels 4L, 7, and 4R.



Question 3: Which lymph node station should be sampled first?

A. Station 4R
B. Station 4L
C. Station 7

Answer: B

Diagnostic yield from either conventional TBNA or EBUS-TBNA is highest from subcarinal station 7 nodes. While this might support initial sampling of the subcarinal node in most instances, the first station that should be sampled in this patient is station 4L because the patient has a right upper lobe mass. A positive 4L, in this case a contra-lateral mediastinal node, would confirm N3 disease. The patient might also have positive nodes at levels 7 and 4R consistent with N2 disease. Should the contralateral node be negative, than stations 7 and 4R could be sampled. If either of these N2 nodes is positive, the patient's tumor would be staged III A using the revised IASLC classification.

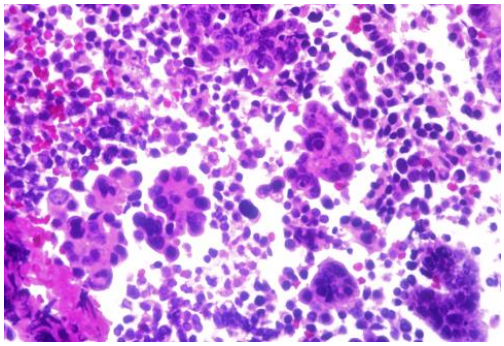
If N3 nodes are positive for malignancy on rapid on-site cytological evaluation, and if the procedure is performed for diagnosis and mediastinal staging only, the procedure could be stopped and the tumor would be staged III B.

References:

1. Rusch VW, Asamura H, Watanabe H, et al; Members of IASLC Staging Committee. The IASLC lung cancer staging project: a proposal for a new international lymph node map in the forthcoming seventh edition of the TNM classification for lung cancer. J Thorac Oncol. 2009; 4: 568–577

The Case Continues

A first aspirate was done from the level 4L node. Rapid on-site examination was positive for malignancy, favoring adenocarcinoma. Using a 22 gauge needle, the second aspirate showed core tissue. Part of the core was placed in formalin for histopathology and part was placed in stabilizing solution to be later used for DNA and RNA extraction.



Question 4: At this point you choose to:

A. End the procedure.
B. Perform at least two more aspirates from the level 4L node, and then end the procedure.
C. Perform aspirations from level 7 and level 4R nodes, and then end the procedure.

Answer: B

All choices are reasonable options. Bronchoscopists should communicate with the Pathology department in their institutions to determine a level of confidence for diagnosing lung carcinoma on rapid on-site evaluation. In general, there is a good correlation between ROSE and the final cytologic diagnosis, so ending the procedure may be appropriate. In this case, after specimen adequacy was established and the diagnosis of malignancy was made by N3 node involvement, the tumor was classified stage III B; therefore, the procedure could end after the first two aspirates. In part, this is because evidence suggests that optimal results are obtained after three aspirations per lymph node station or after two aspirations per station when at least one tissue-core specimen is obtained by the first or second aspiration.

Obtaining at least two additional aspirates from station 4L, however, is highly desirable in this patient because adequate specimen is needed for molecular analysis. Sampling station 7 and 4R is also appropriate as more tissue is needed for downstream molecular analysis. Additional samples would not be necessary for staging.

ROSE helps to confirm the presence of malignant cells and the adequate cellularity of the sample before submitting it to the molecular pathology laboratory. Some practitioners do not use ROSE because it can increase the duration and cost of the procedure. Sending the specimen directly for molecular analysis without first confirming the presence of malignancy, however, may not be cost effective because biopsy specimens may not contain sufficient carcinoma cells suitable for molecular testing.

If this patient's diagnosis is confirmed adenocarcinoma as suspected using ROSE, biomarker-directed therapy might be considered as first or second line treatment for locally advanced disease depending on biomarker analysis. Adequate specimens are therefore needed. For instance, specimens are considered adequate for EGFR analysis if they contain more than 40% malignant cells which could be obtained by four "good" fine needle aspirations or 2-3 core needle tissues.

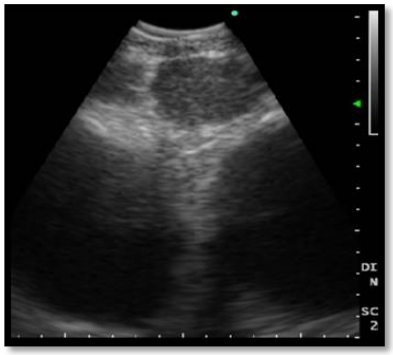
References:

1. Lee HS, Lee GK, Lee HS, et al. Real-time endobronchial ultrasound-guided transbronchial needle aspiration in mediastinal staging of non-small cell lung cancer: how many aspirations per target lymph node station? Chest. 2008;134:368-74.
2. http://labmed.ucsf.edu/uploads/369/166_24JonesMolecularPathologyOfLungCancer.pdf (accessed 8/23/11).

3. Coghlin CL, Smith LJ, Bakar S, et al. Quantitative analysis of tumor in bronchial biopsy specimens. J Thorac Oncol. 2010;5:448-52.

The Case Continues

After rapidly obtaining two more aspirates from level 4L, you chose to sample the level 7 lymph node. Using ROSE, the cytopathologists says the first aspirate at this level is also positive for malignancy and is most likely adenocarcinoma. You obtain three additional samples to send to the laboratory for molecular analysis.



Question 5: You choose to submit these samples for which of the following tests?

A. EGFR (mutation analysis)
B. KRAS (mutation analysis)
C. ALK gene rearrangement
D. All of the above

Answer: D

Results from all three tests can alter patient management. Activating EGFR mutations at exons 18-21 will predict response to Tyrosine Kinase Inhibitors, also known as TKIs. Relevant to this patient, EGFR mutation analysis is feasible in needle biopsy/aspiration paraffin-fixed specimens such as those obtained by EBUS-TBNA. EGFR mutation has been reported in 10-37% of EBUS-TBNA specimens. In one study using a PCR technique, the overall specimen insufficiency rate for EBUS was only 4%, a rate that is lower than that obtained using CT guided biopsy.

KRAS mutation occurs in approximately 15-30% of NSCLC, mostly in lung adenocarcinoma, rarely in squamous cell carcinoma and is mutually exclusive with EGFR mutations. Its presence confers resistance to treatment with TKIs. Clinically relevant mutations found in 3.5-7% of EBUS-TBNA specimens can be detected by RT-PCR or DNA sequencing. The optimal methodology for the detection of KRAS mutation for needle biopsied samples is uncertain at this time; in fact, needle specimens may be inadequate compared with resected specimens which show the mutation at higher frequencies.

Approximately 2-7% of NSCLC harbor ALK fusions and in the vast majority of cases, ALK rearrangements are non-overlapping with other oncogenic mutations such as EGFR and KRAS mutations found in NSCLC. The anaplastic lymphoma kinase (ALK) is a receptor tyrosine kinase that is aberrant in a variety of malignancies. The EML4-ALK fusion oncogene results from a small inversion within chromosome 2p. This leads to expression of a chimeric tyrosine kinase, in which the N-terminal half of echinoderm microtubule-associated protein-like 4, also known as EML4, is fused to the intracellular kinase domain of ALK. Clinically, the presence of ALK fusions is associated with EGFR TKI resistance. ALK gene rearrangement predicts response to inhibitors of the chimeric tyrosine kinase synthesized by this oncogene and could therefore assist in the management of this patient.

Epidermal Growth Factor Receptor, also known as EGFR, is a growth promoting protein lying within the cytoplasmic membrane. Its external domain binds growth factors and is the target of monoclonal antibody drugs while the internal domain, including the tyrosine kinase domain, is the target of small molecule drugs, known as tyrosine kinase inhibitors Fine needle aspirates, unstained slides and Formalin Fixed Paraffin Embedded tissues, when core tissue is obtained, can be sent for EGFR mutation analysis or increased gene copy number.

An established test for ALK gene rearrangement is fluorescence in situ hybridization, also known as FISH. Some molecular laboratories require at least 2 mL of special media for fine needle aspirates or 10% neutral buffered FFPE tissue; ALK gene rearrangement can also be detected by immunohistochemistry, PCR and DNA sequencing[2], and is feasible in EBUS-TBNA specimens. As of June, 2013, FISH is the only FDA-approved test in the United States.

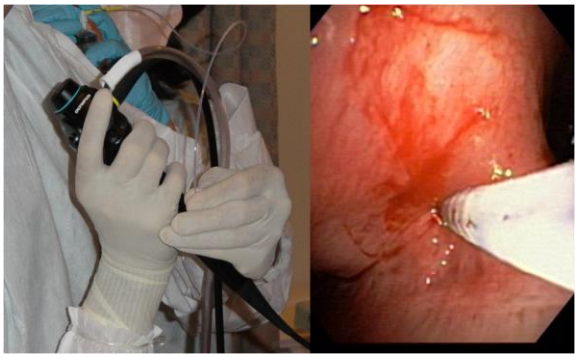
References:

1. Rosell R, Moran T, Queralt C, et al; Spanish Lung Cancer Group. Screening for epidermal growth factor receptor mutations in lung cancer. N Engl J Med. 2009;361:958-67.
2. Rosell R, Gervais R, Vergnenegre A, et al. Erlotinib versus chemotherapy (CT) in advanced non-small cell lung cancer (NSCLC) patients (p) with epidermal growth factor receptor (EGFR) mutations: Interim results of the European Erlotinib Versus Chemotherapy (EURTAC) phase III randomized trial. J Clin Oncol 29: 2011 (suppl; abstr 7503).
3. http://www.mycancergenome.org/molecular-pathology (accessed 8/23/11).
4. Nakajima T, Yasufuku K, Nakagawara A, et al. Multi-gene mutation analysis of metastatic lymph nodes in non-small cell lung cancer diagnosed by EBUS-TBNA. Chest 2011 Apr 28. [Epub ahead of print].
5. Sakairi Y, Nakajima T, Yasufuku K, et al. EML4-ALK fusion gene assessment using metastatic lymph node samples obtained by endobronchial ultrasound-guided transbronchial needle aspiration. Clin Cancer Res. 2010;16:4938-45.

[2] As of this writing, the only FDA approved test in the United States is FISH.

The Case Continues

After obtaining all the specimens and sending them to the appropriate laboratories, including for all three molecular analysis tests, the procedure was ended. The patient was transferred to the recovery area and discharged home. On follow-up visit, the final results of the procedure were discussed. The patient had primary lung adenocarcinoma, stage IIIB. EGFR mutation analysis showed deletion of exon 19; KRAS mutation was negative. ALK rearrangement was negative by FISH.



Question 6: In case of personalized therapy, this patient will most likely benefit from which of the following?

A. ALK-directed therapy
B. EGFR-TKI
C. Folate analogue metabolic inhibitors

Answer: B

For patients with advanced NSCLC whose tumors harbor ALK rearrangement, there is evidence of a benefit from ALK TKI treatment. Our patient, however, tested negative for ALK rearrangement.

EGFR TKIs administered to Asian patients who were EGFR mutation positive improved progression free survival when compared with those who received chemotherapy with carboplatin/paclitaxel. The response rate in mutation positive patients was 73.7% versus 30.7 % in the chemotherapy group. Progression free survival was 10.8 months versus 5.4 months, favoring EGFR TKIs.

Folate analogue metabolic inhibitors have been studied in NSCLC patients with low levels of thymidylate synthase, also known as TS, expression. This enzyme involved in DNA biosynthesis is responsible for maintaining intracellular levels of thymidine, important for DNA synthesis and repair. Immunohistochemistry and RT-PCR on FFPE tissue can be analyzed for TS expression, which is higher in patients with squamous cell carcinoma compared to adenocarcinoma and is correlated with resistance to Folate analogue metabolic inhibitors.

In Caucasians, a 58% objective response rate was reported using EGFR TKIs compared with 15% response after chemotherapy. Progression free survival improved from 5.2 months to 9.4 months. Because our patient tested positive for EGFR mutation, initiation of EGFR TKI is appropriate.

References:

1. Kwak EL, Bang YJ, Camidge DR, et al. Anaplastic lymphoma kinase inhibition in non-small-cell lung cancer. N Engl J Med 2010; 363:1693-1703.
2. Mok TS, Wu YL, Thongprasert S, et al. Gefitinib or carboplatin-paclitaxel in pulmonary adenocarcinoma. N Engl J Med. 2009;361: 947-57.
3. Maemondo M, Inoue A, Kobayashi K, et al; North-East Japan Study Group. Gefitinib or chemotherapy for non-small-cell lung cancer with mutated EGFR. N Engl J Med. 2010;362:2380-8.
4. Rosell, R. Gervais R, Vergnenegre A, et al. Erlotinib versus chemotherapy (CT) in advanced non-small cell lung cancer (NSCLC) patients (p) with epidermal growth factor receptor (EGFR) mutations: Interim results of the European Erlotinib Versus Chemotherapy (EURTAC) phase III randomized trial. J Clin Oncol 29: 2011 (suppl; abstr 7503).
5. Scagliotti GV, Parikh P, von Pawel J, et al. Phase III study comparing cisplatin plus gemcitabine with cisplatin plus pemetrexed in chemotherapy-naive patients with advanced-stage non-small-cell lung cancer. J Clin Oncol 2008; 26:3543-3551.

The Case Continues

The patient elected to have biomarker-directed therapy rather than chemoradiation. She was started on EGFR TKI targeted therapy. She tolerated treatment well except for mild diarrhea and an acne type rash over her neck and face which disappeared after several weeks of treatment. Her follow up PET CT scans were stable until 12 months later when she showed increased lymphadenopathy size and activity in stations 4R, 4L and 11L. She reported intermittent hemoptysis, but remained active and independent in her activities of daily living. She had strong social support and wanted additional therapy. This is discussed at your multidisciplinary lung cancer conference.

Question 7: What do you recommend next?

A. Chemotherapy with carboplatin and taxol
B. Confirm progression of disease before recommending additional therapy
C. Consult palliative care
D. Combined chemoradiation

Answer: B

Initiating chemotherapy or palliative care only without confirming progression of disease is probably undesirable. It is also noteworthy that some patients may have a granulomatous disorder that can mimic cancer. Patients may also have tumor markers, such as over expression of ERCC1, which predicts resistance to platinum based chemotherapy. Consulting palliative care medicine is appropriate, particularly because progressive disease is a concern, but may not be the next immediate step in this patient's management because her quality of life and activities of daily living have not deteriorated.

In this patient with lung adenocarcinoma and history of EGFR mutation, repeat biopsy is done to confirm disease progression and to evaluate for secondary mutations which might offer prognostic value and help guide referral towards clinical trials.

Patients with a diagnosis of cancer and evidence of mediastinal and or hilar lymphadenopathy on PET CT should undergo a tissue sampling procedure to avoid inaccurate upstaging and inappropriate therapeutic management.

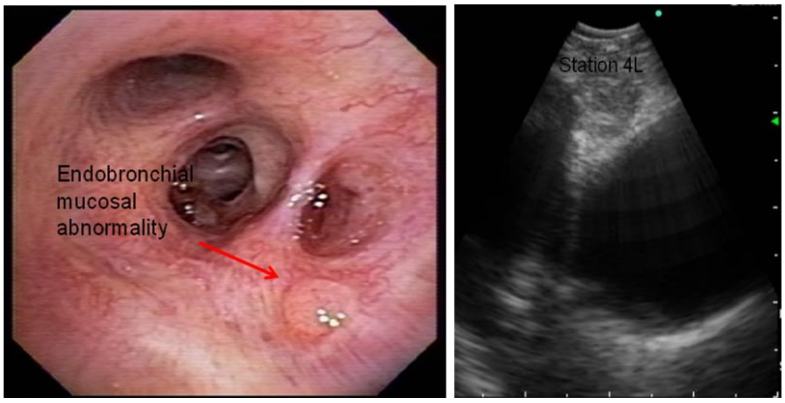
References:

1. Sequist LV, et al. Genotypic and histological evolution of lung cancers acquiring resistance to EGFR inhibitors. Sci Transl Med 2011;3:75ra26.
2. Kennedy M, Jimenez C, Mhatre A, et al. Clinical implications of granulomatous inflammation detected by endobronchial ultrasound transbronchial needle aspiration in patients with suspected cancer recurrence in the mediastinum. J Cardiothorac Surg. 2008; 3:8.
3. Olaussen KA, Dunant A, Fouret P, et al. DNA repair by ERCC1 in non-small-cell lung cancer and cisplatin-based adjuvant chemotherapy. N Engl J Med. 2006;355:983-991.

The Case Continues

The patient is discussed in the multidisciplinary chest conference. A decision is made to proceed with bronchoscopy in order to evaluate for possible airway involvement with tumor given the patient's new onset of hemoptysis, and to perform EBUS-TBNA in an attempt to confirm progression of disease within the mediastinum given the suspected progression of disease on PET-CT. Bronchoscopy revealed

mucosal abnormalities within the right bronchus intermedius and EBUS imaging confirmed the presence of mediastinal and hilar lymphadenopathy.



Question 8: What would you do next?

A. Endobronchial biopsy, lavage and brushings of the abnormality in the bronchus intermedius.
B. Sequential EBUS-TBNA from stations 11L, 4L and 4R
C. Sequential EBUS-TBNA from stations 4L, 4R, and 11L, stopping should any of these nodal stations be positive.
D. Endobronchial sampling by biopsy, lavage, and brushing in addition to sequential EBUS-TBNA from stations 11L, 4L, and 4R.

Answer: D

For central lesions such as those visible during flexible bronchoscopy, the diagnostic yield of endobronchial biopsy is 74% as compared to 46% for peripheral lesions. To achieve 90% probability of positive biopsy for malignancy, 5 samples are usually required for visible tumor. For molecular markers analysis, endobronchial and trans-thoracic biopsies provide adequate tissue for DNA sequencing in 89% of samples.

On the other hand, for cytological diagnosis, the diagnostic yield of bronchial brushings in central lesions is 59% compared to about 46% for peripheral lesions. The utility for molecular markers analysis has not been clearly determined.

Bronchioloalveolar lavage has a diagnostic yield of 48% for central lesions and 43% for peripheral lesions. There is emerging data on the use of lavage for molecular analysis but testing a major limitation relates to tumor cellularity.

EBUS-TBNA from abnormal mediastinal/hilar nodes in patients with known or highly suspected lung cancer has a diagnostic yield for malignancy of 94% with usually at least 3 specimens per nodal

station being obtained for diagnosis if ROSE is not utilized; two aspirations per nodal station are considered acceptable when at least one core obtained by first two specimens. For molecular markers, adequacy of EBUS-TBNA specimens varies in the published literature between 72-99%.

In this patient combined sampling techniques are warranted. Using EBUS-guided TBNA, left sided nodes should be sampled first since the patient's primary tumor was on the right. A reasonable approach, therefore, would be to start with the newly PET positive 11L node followed by sampling nodes at level 4L and potentially 4R.

Published literature supports a practice of combined techniques to increase the yield for central lesions for which the diagnostic yield is 88% versus 69% for peripheral lesions. Brushing and washing increases diagnostic yield by up to 17%.

Because the utility of bronchoscopic specimens for molecular markers analysis is based on number of intact malignant cells provided for analysis, and because specimens with malignant cells in suspension can be prepared in cell block for immunohistochemistry or submitted for molecular testing, a combination of endobronchial techniques and EBUS-TBNA techniques seems to be the most advantageous in terms of specimen acquisition for molecular analysis.

References:

1. British Thoracic Society guidelines on diagnostic flexible bronchoscopy Thorax 2001;56:(suppl I) i1–i21.
2. Arcilla M, et al. Rebiopsy of Lung Cancer Patients with Acquired Resistance to EGFR Inhibitors and Enhanced Detection of the T790M Mutation Using a Locked Nucleic Acid-Based Assay Clin Cancer Res 2011;17:1169-1180.
3. Van der Drift M, et al. The Additional Value of the Ras-Association Domain Family 1A Gene Methylation and Kirsten Rat Sarcoma 2 Viral Oncogene Homolog Mutation Analyses in Washings in Nondiagnostic Bronchoscopy Chest 2012;141;169-175.
4. Tanner NT, et al. Utilizing Endobronchial Ultrasound With Fine-Needle Aspiration to Obtain Tissue for Molecular Analysis J Bronchol Intervent Pulmonol Volume 18, Number 4, October 2011.

The Case Continues

Five endobronchial biopsies were performed from the airway lesion. Small tissue fragments were fixed in formalin 10% and sent to pathology to prepare formalin fixed paraffin embedded tissues, also known as FFPE. Five EBUS-guided TBNA aspirates were obtained performed from nodal station 11L. Three slides were air dried and stained with Romanowsky solution for rapid on site evaluation which showed the presence of malignant cells consistent with NSCLC. Three slides were fixed in alcohol and sent to the pathology laboratory for Papanicolau staining. Aspirates were also placed in Cytolyt solution for cell block preparation.

Question 9: What would you do next if the patient wants to be considered for clinical trials using targeted therapy?

A. Discuss the adequacy of the specimen with your pathologist on-site prior to requesting molecular tests.
B. Given the scant amount of tissue, order molecular testing without confirming non squamous non small cell carcinoma by immunohistochemistry.
C. Wait for final results. After malignancy is confirmed, refer the patient for chemotherapy without ordering molecular testing.
D. Wait for final results. After malignancy is confirmed, initiate discussions about quality of life and refer the patient to a palliative care specialist.

Answer: A

It is important to assure that samples contain adequate cellularity and representative tissue for analysis. Care should be taken to avoid wasting tissue on unnecessary IHC studies, and to conserve as much tissue as possible for molecular analysis when indicated. Because the literature suggests that specimens should contain at least 50-70% tumor cells for mutation analysis, sending specimens directly for molecular testing without a pathologist's review for quality and quantity is not recommended.

In this patient, specimens were available for analyzing markers predictive of response and or resistance to certain chemotherapy agents and molecular targets. Evidence suggests that high expression of thymidylate synthase, for example, could predict resistance to pemetrexed. Initiating chemotherapy without performing molecular testing may not be cost-effective.

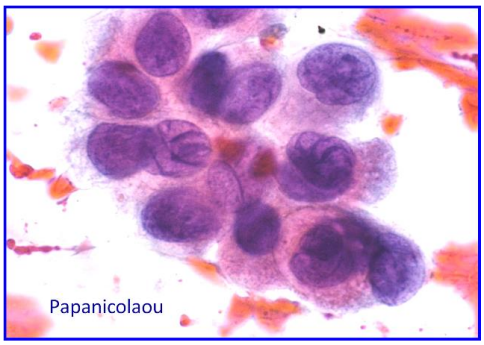
Operators collecting small specimens should carefully use appropriate smear techniques, attempt to obtain tissue cores, and communicate on-site with their pathologists to assure sample adequacy and appropriate processing.

References:

1. Pirker R, et al. Consensus for EGFR mutation testing in non-small cell lung cancer: results from a European workshop. Thorac Oncol. 2010 Oct;5(10):1706-13
2. Takezawa K. Identification of thymidylate synthase as a potential therapeutic target for lung cancer et al. British Journal of Cancer (2010)

The Case Continues:

Papanicolau stains were positive for malignancy, most likely adenocarcinoma. Both endobronchial biopsy and EBUS-TBNA specimens were analyzed by the cytopathologist and considered adequate for molecular analysis. IHC confirmed adenocarcinoma. TTF1 was positive and P63 was negative. Given the patient's history of primary lung malignancy and no clinical suspicion for extrathoracic malignancy metastatic to the lung, specimens were not processed for further Immunohistochemistry.



Courtesy M. Ferretti, with permission

Question 10: Which molecular markers would you test for now?

A. ERCC1, RRM 1 and TS
B. T790M mutation
C. MET amplification
D. All of the above

Answer: D

While data regarding clinical use is still emerging, all markers listed above could impact management. Testing for these markers is relevant if clinical trials are accessible to the patient.

ERCC1, RRM1 and TS could predict response /and or resistance to certain chemotherapy agents. ERCC1 is an important component of nucleoside excision repair. Because platinum-based chemotherapy works by creating platinum-DNA adducts, increased levels of ERCC1 indicates resistance to platinum based therapy while low levels indicate sensitivity. Regulatory subunit of ribonucleotide reductase, also known as RRM1, is the target of gemcitabine. In some studies, high levels are associated with gemcitabine resistance and poor outcome. High levels of TS indicate pemetrexed resistance, but levels are usually higher in squamous cell compared to adenocarcinoma, which may be why squamous cell carcinoma does not respond to pemetrexed.

In patients treated with TK inhibitors for EGFR positive lung cancer, adaptive resistance commonly develops during therapy. Mechanisms of resistance include T790M mutation and MET amplification. If tested positive, this patient could be enrolled in clinical trials directed towards these molecular markers.

References:

1. Olaussen et al: DNA Repair by ERCC1 in Non–Small-Cell Lung Cancer and Cisplatin-Based Adjuvant Chemotherapy. N Engl J Med. 2006;355:983-991.
2. Rosell R et al: Ribonucleotide reductase mRNA expression and survival in gemcitabine/cisplatin-treated advanced non-small-cell lung cancer patients. Clin Cancer Res 10:1318-1325, 2004.
3. Kobayashi S et al. EGFR mutation and resistance of non-small-cell lung cancer to gefitinib. N Engl J Med. 2005;352(8):786.
4. Cipriani NA et al. MET as a target for treatment of chest tumors. Lung Cancer. 2009;63(2):169.

Case conclusion:

T790M mutation was positive and MET was not amplified. ERCC1 and TS levels were low. RRM1 was also low. The patient wanted to receive standard chemotherapy. She refused enrollment in a clinical trial. Given her tumor's low expression of ERCC1 and TS, she is more than likely to respond to a combination of platinum and Folate analogue metabolic inhibitors such as pemetrexed.

The patient was referred to a palliative care specialist in consultation only. By integrating palliative care consultation, the multidisciplinary team felt the patient's overall quality of life might be improved. Studies show a benefit of integrating palliative care with oncologic care. Both chemotherapy and performance status have been shown to positively impact survival in patients with advanced NSCLC.

Literature shows that the early introduction of palliative care prolongs survival among patients with advanced III B and IV non-small-cell lung cancer stage.

References:

1. Olaussen et al. DNA Repair by ERCC1 in Non–Small-Cell Lung Cancer and Cisplatin-Based Adjuvant Chemotherapy. N Engl J Med. 2006;355:983-991
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Credits

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