Accuracy and Adequacy of Computed Tomography–Guided Lung Biopsies: Experience From a Community Hospital

Barbara D. Florentine, MD; Richard J. Helton, DO; Michael M. Mitchell, DO; Kara E. Schmidt, DO; and David B. Kozlov, MD

From the Department of Pathology at St Lucie Medical Center in Florida (Dr Florentine); the Internal Medicine Residency Program in the Department of Medicine at the University of Florida in Gainesville (Dr Helton); the Pediatric Residency Program in the Department of Pediatrics at the University of South Carolina in Columbia (Dr Mitchell); the Internal Medicine Residency Program at the Palm Beach Consortium for Graduate Medical Education at West Palm Hospital in Florida (Dr Schmidt); and the Department of Radiology at St Lucie Medical Center in Florida (Dr Kozlov).

Financial Disclosures: None reported.

Support: None reported.

Address correspondence to Barbara D. Florentine, MD, Medical Director, Department of Pathology, St Lucie Medical Center, 1800 SE Tiffany Ave, Port St Lucie, FL 34952-7521.

E-mail: barbara.florentine@hcahealthcare.com

Submitted February 15, 2015; final revision received May 11, 2015; accepted June 5, 2015.

Context: Small tissue biopsies obtained through minimally invasive methods have become the primary diagnostic tools for the pathologic characterization and testing of lung masses. In view of recent advances in targeted therapy for non–small cell lung carcinoma, and lung adenocarcinoma in particular, pathologists are now expected to thoroughly characterize lung lesions microscopically while making certain that enough tissue remains for potential molecular analysis if indicated.

Objective: To report our experience with computed tomography (CT)–guided lung needle biopsies with particular concentration on diagnostic yield, diagnostic accuracy, and adequacy of tissue for molecular testing if indicated.

Methods: A retrospective observational study analyzed 224 biopsies in 222 patients undergoing CT-guided lung needle biopsies. Accuracy of diagnosis and adequacy of tissue for molecular testing, if applicable, was evaluated. A standardized protocol for specimen evaluation, triage, and processing was used. This protocol included intraprocedural real-time microscopic specimen evaluation and triage by a pathologist and use of a histologic protocol specifically designed to conserve tissue for ancillary testing. The initial biopsy was considered successful if the specimen was malignant, had specific benign features, or had nonspecific benign features with follow-up supporting benign lesion. Initial biopsy failure cases were those with inadequate tissue or a nonspecific result with highly suspicious imaging or clinical findings.

Results: Of the 224 biopsies, 8 cases with benign but nonspecific findings lacked follow-up and were excluded from the study. The biopsy was diagnostically successful in 189 of 216 (88%) cases. Of these 189 cases, 154 (81%) were malignant, and 35 (19%) were benign. There were 28 diagnostic failures. Subsequent tissue sampling of 13 of 28 diagnostic failures found 9 (69%) to be malignant. Molecular studies were requested on 25 cases: 24 had sufficient material for some of the requested tests, and 20 had enough tissue for all requested testing.

Conclusion: A standardized protocol and team approach for CT-guided lung needle biopsy optimizes the ability to achieve a high accurate diagnostic yield with adequate tissue for molecular testing.

J Am Osteopath Assoc. 2015;115(10):592-603.
doi:10.7556/jaoa.2015.120

Lung cancer is one of the most prevalent cancers in the United States today.[1] Of the different modalities used to diagnose lung cancer, computed tomography (CT)–guided percutaneous lung needle biopsy is a well-established procedure often used to obtain a diagnosis. Surgical pulmonary resections offer abundant tissue for testing; however many patients present with advanced disease and are not surgical candidates. Therefore, minimally invasive small tissue biopsies, such as CT-guided biopsies or endobronchial cytologic or surgical biopsies, are the primary diagnostic tools for the pathologic characterization and testing of malignant lesions in the lung and are also used to triage patients with a nonmalignant diagnosis for conservative management or excision.

Using limited tissue afforded by these minimally invasive biopsy methods, pathologists are now expected to thoroughly characterize lung lesions microscopically and, in the case of a malignant lesion, make certain that enough tissue remains for potential molecular analysis, particularly in primary lung adenocarcinoma. This represents a major paradigm shift from the past, when, in the case of primary lung carcinoma, pathologists only had to discriminate small cell carcinoma from non–small cell lung carcinoma (NSCLC). Today, the most common subtypes of NSCLC are subclassified into adenocarcinoma, squamous cell carcinoma, or carcinoma with an adenocarcinoma component, which are of utmost importance, because molecular tests (eg, epidermal growth factor receptor [EGFR], anaplastic lymphoma kinase [ALK]) performed on these specimens commonly guide treatment.[2,3] Immunohistochemical and mucin stains can aid in subclassifying NSCLC when the histomorphology is not definitive; however, injudicious use of such panels may exhaust tissue that might be needed for molecular studies.[3,4] The risks of the sampling procedure must be tempered with the need for obtaining adequate tissue. At the same time, the triage and processing of tissue requires more care than in the past when fewer tests were performed on the biopsy specimen. Harvesting adequate material for all anticipated studies, careful sample preparation, and judicious use of immunostains to subcategorize tumors requires forethought and necessitates a team approach with good communication between the procuring interventional radiologist, pathologist, laboratory staff, and other clinicians.[4]

Many CT-guided biopsies for the diagnosis of lung lesions occur in the community hospital setting. However, limited reports have evaluated the success of this procedure outside large tertiary medical centers. Our aim is to report our overall experience with the use of a standardized team-based protocol for procuring, processing, and diagnosing CT-guided lung biopsies with particular concentration on NSCLC. In this retrospective observational study of CT-guided lung biopsies at a single community hospital, we evaluated diagnostic yield, diagnostic accuracy, and adequacy of tissue samples for requested molecular testing.

Methods

The present study was conducted in a general 229-acute bed community hospital located in southeast Florida. The radiology department has 1 full-time interventional radiologist and 2 full-time pathologists. Many of the patients were outpatient referrals from community physicians, and the remainder were inpatients usually admitted for other reasons. This study was exempt from institutional review board approval.

All consecutive CT-guided percutaneous lung biopsies performed in our hospital from January 2012 through May 2014 were evaluated. We retrospectively reviewed the patients' electronic health records to determine demographic information, clinical history, clinical course, biopsy diagnosis, molecular testing results (if requested), imaging result and follow-up. Final diagnosis of each lung lesion was confirmed by either tissue pathology result alone, or tissue pathology result with supporting clinical or radiographic results.

Diagnostic Categories

Results from each initial CT-guided biopsy procedure were categorized into 4 diagnostic groups: malignant, benign specific, benign nonspecific, and nondiagnostic. Benign specific cases included a specific etiology (ie, benign neoplasms, lipoid pneumonia, granulomatous inflammation). All biopsies categorized as malignant and benign specific were considered true-positives. Biopsies categorized as benign nonspecific included cases in which biopsy specimens showed nonspecific tissue changes, such as chronic inflammation, fibrosis, or suppurative pneumonia, and the clinical presentation or radiologic images at the time of the procedure were noncontributory. Suppurative pneumonia was included in the benign nonspecific category because the differential diagnosis for this finding may include postobstructive pneumonia secondary to a nearby but unsampled neoplasm. A case was considered nondiagnostic if the biopsy specimen was entirely necrotic, entirely normal, had insufficient tissue for diagnosis, or the radiologic or clinical features at the time of the procedure were either highly suspicious for a malignant lesion or for a benign specific process, but the sample yielded only benign nonspecific results.

Diagnostic Accuracy

Diagnostic accuracy was determined by further dividing the initial biopsies into 2 groups: diagnostic success and diagnostic failure. The diagnostic success group included all malignant and true benign cases. True benign cases were defined as all cases in the benign specific category and benign nonspecific cases with radiologic or clinical follow-up supportive of a nonspecific benign process in keeping with the biopsy findings. The diagnostic failure group included all nondiagnostic cases and cases categorized as benign nonspecific but found on subsequent tissue sampling to either have a benign specific pathologic diagnosis or were missed cancers.

Adequacy of Tissue for Molecular Testing

Molecular studies were performed at the request of the treating physician at a regional or national commercial reference laboratory of the clinician's choice. The cases in which molecular testing was requested were divided into 3 groups: sufficient tissue for all requested studies, sufficient tissue for some studies, and insufficient tissue for any studies.

Biopsy Procedures

The majority of the procedures (75%) were performed by a single interventional radiologist. Most patients underwent a core needle biopsy (CNB); in a few cases, fine needle aspiration (FNA) biopsy was used. A pathologist attended all but 1 procedure to review the history and imaging findings with the radiologist, confirm adequacy of tissue and proper triage of the samples, and render a preliminary diagnosis.

Core Needle Biopsy

Semiautomatic coaxial core needles were used, with a 20-gauge needle used most often. Select CNB specimens were lightly and carefully touched or rolled onto a glass slide prior to immersing the biopsy specimen into formalin. The touch prep (TP) cytology slide was then stained with a Romanowksy stain variant for onsite microscopic evaluation of the cells. If the pathologist reported that the diagnosis would be challenging and would require an expanded panel of immunostains or if a primary lung adenocarcinoma was likely, additional cores were obtained to assure adequate tissue for all required ancillary studies. If an infectious agent was suspected, a separate fresh CNB specimen was sent in a small amount of saline to the microbiology laboratory. All CNB samples were then placed in buffered formalin and submitted for paraffin embedding and tissue sections.

The histology laboratory processed the CNB samples using a special tissue protocol specifically designed to conserve tissue for ancillary testing. Ten sections were initially cut in every case, with 1 section placed on each of

10 slides. Hematoxylin-eosin stain was used on slides 1 and 10, and the intervening sections remained unstained for immunostaining and any other ancillary studies.

Fine-Needle Aspiration

A portion of each FNA biopsy sample was expressed onto glass slides. One or 2 were stained with a Romanowksy stain variant, and the remainder fixed in 95% alcohol for later Papanicolaou staining. The FNA material not used for smears was rinsed in 95% alcohol and processed into a cell block, which was embedded in paraffin and cut into tissue sections, using the same method as for the CNBs. A pathologist performed an onsite preliminary diagnosis and material triage. If the pathologist reported the diagnosis would require an expanded panel of immunostains or if a primary lung adenocarcinoma was likely, additional passes for cell block were obtained to assure adequate tissue for all required ancillary studies. When the lesion was suspected of being infective, additional aspirates were procured and rinsed into a small amount of normal saline before sending for culture.

Postprocedure Processing of NSCLC Cases

After each NSCLC case was processed, the pathologist attempted to definitively subclassify each case into squamous, adenocarcinoma, carcinoma with an adenocarcinoma component, or other. First, the cytomorphology from the TP/FNA cytology slide and histopathology from the paraffin block slide were studied morphologically using routine stains. In cases where the morphology was indeterminate, immunostains and occasionally a mucin stain were used. The immunostain used to confirm primary adenocarcinoma was a positive thyroid transcription factor-1 (TTF-1) with or without a positive napsin-A. A positive p63 stain or CK5/6 stain, or p40 stain with negative TTF-1 was used to confirm squamous cell carcinoma. If a definitive subtype could not be given, the pathologist attempted to favor 1 subtype over another. Favored subtype cases as defined in the present study are cases in which the pathologist favored 1 subtype over the other but felt that there was not enough evidence to render a definitive NSCLC subtype. When the morphology or immunostains were nondiscriminatory or there was insufficient tissue to subclassify the NSCLC further, NSCLC, not otherwise specified, was rendered.

Statistical Analysis

Data were summarized using descriptive statistics. The calculations used the formulas below.

(1) Sensitivity of the overall procedure (S_{op}, benign and malignant) was calculated as:

$$S_{op}=DS_{fb}/PM+TBS+TBNS$$

where DS_{fb} is the number of all diagnostic success cases from first biopsy; PM is the number of all positive malignant cases; TBS is the number of all true benign specific cases; and TBNS is the number of all true benign nonspecific cases.

(2) Diagnostic accuracy of the overall procedure (DA_{op}) was determined by:

$$DA_{op}=TBS+TBNS+PM/TBS+TBNS+PM+TBF+TMF$$

where TBF is the number of true benign failure cases and TMF is the number of all true malignant failure cases.

(3) Sensitivity for the diagnosis of malignant lesion (S_{dm}) was calculated using the following equation:

$$S_{dm} = PM_{fb}/ATM$$

where PM_{fb} is the number of all cases diagnosed as malignant on the first biopsy and ATM is the number of all true malignant cases.

(4) Diagnostic accuracy for malignant lesion (D_{am}) was calculated using the following equation:

$$D_{am}= TMN+TMP/(TMN+TMP+FMN+FMP)$$

where TMN is true malignant negative; TMP is true malignant positive; FMN is false malignant negative; FMP is false malignant positive. (In this study all malignant tissue biopsy results were assumed to be true-positives.)

(5) Negative predictive value for malignant lesion was calculated using the following equation:

(NPV_m)= TMN/TMN+FMN

Results

In total, 224 cases in 222 patients who underwent CT-guided percutaneous needle lung biopsies were evaluated for inclusion. Two patients had 2 CT-guided biopsies at different times. Eight patients in the benign nonspecific category lacked follow-up and were excluded. In total, 216 cases were included the study. Of these patients, 112 (52%) were women and 104 (48%) were men. The mean age was 70 years. Follow-up varied from 8 to 29 months. Lesion size was measured radiographically and ranged from 0.6 cm to 14.6 cm, with a mean of 3.5 cm. Of 216 patients, 208 (96%) initially underwent a CNB, and 8 (4%) underwent an FNA.

Initial CT-Guided Biopsy Results

The breakdown of the 216 initial biopsy cases by diagnostic category and sex is detailed in *Table 1*. A pathologic diagnosis of malignant lesion was made in 154 of 216 (71%) cases; of these, 16 (10%) were metastatic from other sites. Of all primary lung carcinomas, 119 were NSCLCs, and 13 were SCLCs. A definitive subtyping of NSCLC was made in 102 of 119 (86%) cases and a favored subtype in 10 of 119 (8%). In 7 of 119 (6%) cases, we were unable to subtype the NSCLC further, so it was classified as NSCLC, not otherwise specified. Twenty benign specific and 19 benign nonspecific cases were diagnosed, leaving 23 nondiagnostic cases.

Diagnostic Success and Failure

The 216 initial biopsies were further divided into a diagnostic success group and a failure group. The biopsy was diagnostically successful in 189 of 216 (88%) cases. Of these cases, 154 (81%) were malignant, 20 (11%) were benign specific, and 15 (8%) were true benign nonspecific cases. *Table 2* details the supportive radiologic and clinical follow-up of the 15 true benign nonspecific diagnostic successful cases. *Table 3* details the 28 diagnostic failure cases. Subsequent biopsy tissue sampling was performed in 13 of the 28 cases. Of these 9 (69%) were found to be malignant. The 4 other cases were specific benign diseases in which diagnostic material was not adequately represented at the primary procedure. Follow-up CT scan of a patient 1 year later showed nodule growth, although repeated tissue sampling was not done. No additional analysis could be performed in the remaining 14 nondiagnostic cases because these patients were either lost to follow-up or no follow-up procedure was done. Thus, the initial CT biopsy specimen was inadequate in 28 of 216 (13%) cases because of sampling error.

In the present study, the diagnostic accuracy, negative predictive value, and sensitivity of CT-guided lung biopsies for malignant lesion was 95%, 80%, and 94%, respectively (*Table 4*). The sensitivity and diagnostic accuracy of the overall procedure (benign and malignant) was 95% and 94%, respectively.

Molecular Studies

In 25 cases, molecular studies were requested and CNB paraffin-embedded tissue was used in all cases. The 25 cases comprised 22 primary lung NSCLCs and 3 metastatic tumors. The variety of tests requested is shown in *Table 5*. The 3 metastatic tumors had sufficient material for all requested molecular studies. In 21 of the 22 (95%) primary lung cancer cases, enough tissue was available to do some of the requested studies, and in 17 of 22 (77%), enough tissue was available to successfully perform all of the requested molecular testing. One case (5%) had insufficient material for any of the requested studies. In 17 of the 22 (77%) cases, ALK and EGFR testing were requested and successfully performed.

Discussion

The present study evaluated diagnostic yield, diagnostic accuracy, and adequacy of tissue samples for requested molecular testing of a team-based protocol for procuring, processing, and diagnosing CT-guided lung biopsies, with particular concentration on NSCLC.

Diagnostic Yield

The overall accuracy (benign and malignant) of 94%, and diagnostic sensitivity, accuracy, and negative predictive value for malignant lesion of 94%, 96%, and 80%, respectively, are within the range reported in the literature.[5-8] In a study of 226 CT-guided lung CNBs, Quint et al[5] reported a diagnostic sensitivity, accuracy, and negative predictive value in the diagnosis of malignancy of 91%, 92%, and 68%, respectively. Loh et al[7] reported a diagnostic sensitivity, accuracy, and negative predictive value of CT-guided thoracic biopsies for malignant lesion of 96%, 97%, and 88%, respectively, in 384 patients who underwent 399 CT-guided thoracic biopsies in Singapore. In the same study,[7] the authors concluded that their high rate of success was partly due to onsite evaluation of FNA specimens, as well as the use of combined FNA and CNBs in some of their patients. Quint et al[5] did not evaluate their specimens onsite, and we believe our higher diagnostic sensitivity, accuracy, and negative predictive value for malignant lesions are at least partly attributable to having a pathologist onsite to evaluate the specimens in real time. Previous studies have reported that immediate onsite evaluation by a trained cytology professional increases the diagnostic ability of image-guided needle biopsies.[7-10]

The results of the present study are in keeping with those of previous studies,[5-8,11] indicating that CT-guided biopsies have a relatively high false-negative rate for the diagnosis of malignant lesion. In the present study, 9 of 13 (69%) unsuccessful cases that had subsequent biopsy sampling represented missed cancers. Gelbman et al[11] reported 170 patients who underwent a CT-guided FNA

Table 1.
Initial CT-Guided Lung Biopsy Cases by Diagnostic Category and Sex

	Patients, No.		
Diagnostic Category	**Female**	**Male**	**Total**
Benign			
Benign nonspecific[a]	6	13	19
Benign specific			
Granulomatous inflammation	10	3	13
Lipoid pneumonia	1	0	1
Miscellaneous[b]	6	0	6
Malignant			
Metastatic malignant tumor[c]	9	7	16
Miscellaneous[d]	1	1	2
Neuroendocrine carcinoma			
Small cell lung cancer	9	4	13
Well differentiated neuroendocrine carcinoma, carcinoid, and atypical carcinoid[e]	2	2	4
NSCLC			
Adenocarcinoid	1	0	1
Adenocarcinoma	31	28	59
Adenosquamous	1	0	1
NSCLC, favor adenocarcinoma	4	1	5
NSCLC, favor adenosquamous cell carcinoma	1	1	2
NSCLC, favor squamous cell carcinoma	2	1	3
NSCLC, not otherwise specified	3	4	7
Squamous	15	26	41
Nondiagnostic[f]	11	12	23
Total	**113**	**103**	**216**

[a] Out of 27 benign nonspecific cases, 8 were excluded from the study because of lack of follow-up, which left a total of 19 evaluable cases.
[b] Miscellaneous benign specific includes 5 patients with hamartoma/chondroma and 1 patient with neurofibroma.
[c] Metastatic malignant tumor includes the numbers of patients with the following cancers: breast, 3; gastrointestinal, 2; melanoma, 3; hepatocellular, 1; prostate, 1; female genital tract, 1; renal cell, 2; sarcoma, 2; and uncertain primary origin, 1.
[d] Miscellaneous cancers include 1 patient with malignant tumor not otherwise specified and 1 patient with lymphoma.
[e] Well differentiated neuroendocrine carcinomas are categorized separately from the NSCLC group.
[f] Nondiagnostic cases include 8 patients with normal lung, 7 patients with radiological features highly suspicious for malignant tumor or for a benign specific process, 5 patients with necrotic tissue, and 3 patients with insufficient tissue for diagnosis.

Abbreviations: CT, computed tomography; NSCLC, nonsmall cell lung cancer.

Table 2.
Follow-up Findings Supporting the True Benign Nature of 15 CT-Guided Lung Biopsy Cases in the Benign Nonspecific Group[a]

Patient No.	Pertinent CT Findings at the Time of the Procedure	Comment/Follow-up
9	Mass in the anterior left lower lobe (16 mm) worrisome for malignant neoplasm	CT scan 6 mo later revealed nodule no longer visible
33	Large consolidative mass-like density extending from the right hilum to the right anterior and lateral chest walls involving the right middle and upper lobes, measuring approximately 14.6×9.0 cm with diffuse mediastinal, hilar, and subcarinal adenopathy, large areas of air bronchograms, thickening of the interlobular septa, and crazy paving appearance; these findings, sometimes seen with severe pneumonia, are worrisome for malignancy with lymphangitic carcinomatosis	CT scan 3 wk later showed decrease in size of the right lateral lung mass and clearing of the diffuse infiltrates that were present in the right upper and right middle lobe previously; patient was discharged from hospital with a diagnosis of pneumonia
39	Right upper lobe nodular density (12 mm), indeterminate for malignancy	CT scan 1 mo later showed the previous right lung nodule regressing in size and much less apparent
44	Patchy bilateral ill-defined interstitial opacities thought to represent infiltrates superimposed on chronic interstitial changes	Patient treated for bilateral pneumonia with improvement
51	Middle lobe thick walled cavitary mass (6.1×4.2×4.0 cm) with adjacent fluffy airspace disease and micronodules felt to represent infectious or neoplastic process	Discharged from hospital 3 wk after with diagnosis of ventilator-dependent respiratory failure and empyema
64	Large soft tissue attenuating spiculated mass in the posterior right upper lobe measuring approximately 5.3 cm in transverse dimension with air bronchograms; increased in size compared with previous examination and worrisome for malignancy	Conservative follow-up only; CT scan 2 y later showed lesional regression with a 1.0×1.5 cm slightly lobular noncalcified intraparenchymal pulmonary nodule in the right lateral perihilar region
93	Wedge-shaped soft tissue attenuating mass involving the medial aspect of the left upper lobe/lingual	CT scan 5 mo later revealed lesional regression
97	Multifocal bilateral airspace opacities, including a small cavitary lesion in the left upper lobe with a small right pleural effusion	Patient's symptoms improved after being treated for pneumonia
105	Masslike area consolidation in the left mid lung field worrisome for malignancy	CT scan 1 mo later showed marked reduction in size of the mass
151	Left upper lobe 7-mm cavitary lesion	CT scan 1 mo later showed the cavitary lesion previously seen being less cavitary on the current study and slightly smaller or stable in size; bronchoscopy 5 mo later negative for malignancy
159	Left lower lung nodule	CT scan 1 mo later showed partial involution of the lesion in the posterior aspect of the left lower lobe when compared with the previous study
160	Multiple bilateral cavitary lung nodules with the largest up to 4 cm	CT scan 2 wk later showed improving peripheral cavitary nodules bilaterally
162	Patchy ill-defined bilateral infiltrates worrisome for pneumonia/fluid overload	Patient discharged with diagnosis of pneumonia; CT scan 1 y later showed bilateral lower lobe patchy opacity
185	1.3-cm pulmonary nodule within the left lingula with additional scattered smaller subcentimeter pulmonary nodules	CT scan 3 y later revealed stable findings
222	Right lower lobe spiculated nodule, highly suspicious for malignancy[b]	CT scan 1 y later showed no findings other than a wedge shaped subsegmental atelectasis at the right base; patient received conservative treatment

[a] The benign nonspecific group included cases in which the initial CT-guided biopsy results showed nonspecific tissue changes, such as chronic inflammation, fibrosis, or acute pneumonia, and the clinical presentation or radiological images at the time of the procedure were non-contributory.
[b] Although the imaging for case 222 was reported highly suspicious for malignancy, follow-up confirmed that this represented an overcall by radiology, and the case was therefore classified with the benign nonspecific group.

Abbreviation: CT, computed tomography.

Table 3.
Initial CT-Guided Diagnosis, Follow-up, and Final Diagnosis of Lung Biopsy Specimens in the Diagnostic Failure Group

Patient No.	Initial Diagnosis	Follow-up Procedure	Final Pathologic Diagnosis
1, 4, 6, 7, 8,150, 176, 179, 180, 182, 183, 187, 193, 225	Nondiagnostic	Patient lost to follow-up or no follow-up procedure done	NA
17	Nondiagnostic	Lung wedge resection	Granulomas
25	Benign nonspecific	Repeat CT-guided lung biopsy	Squamous cell carcinoma
28	Benign nonspecific	Lymph node biopsy	Adenocarcinoma of primary lung origin
31	Nondiagnostic	Repeat CT-guided lung biopsy	Pneumonia-abscess
52	Nondiagnostic	Lung lobectomy	Granulomatous
70	Nondiagnostic	Brain biopsy	Metastatic malignant melanoma
111	Nondiagnostic	Lung wedge resection	Adenocarcinoma
122	Nondiagnostic	Lung wedge resection	Squamous cell carcinoma
135	Nondiagnostic	Lobectomy	NSCLC, not otherwise specified
152	Benign nonspecific	Lung wedge resection	Granulomatous
175	Nondiagnostic	Lung wedge resection	NSCLC, favor squamous cell
181a	Nondiagnostic	Mediastinal lymph node	Lymphoma
213	Benign nonspecific	Repeat CT scan 1-year later showed nodule growth from 0.6 cm to 1.4 cm	NA
221	Nondiagnostic	Bronchial biopsy	Small cell carcinoma

[a] Although the initial CT biopsy was suspicious for lymphoma, this case was classified in the nondiagnostic category because the tissue was insufficient for definitive diagnosis.

Abbreviations: NA, not applicable; CT, computed tomography; NSCLC, nonsmall cell lung cancer.

lung biopsy yielding an initial benign result, with adequate follow-up, and found that 18 proved to be false-negatives. Tsukada et al[6] reported that out of 91 true malignant tumors, 21 were diagnosed as benign (false-negative) on initial CT-guided needle biopsy. Thus, a benign nonspecific result cannot be relied on for the exclusion of malignant lesions.[3,5,11] Therefore, it is especially important to have close clinical and imaging follow-up in patients with noncorrelating clinical or imaging and biopsy results with a low threshold for a repeated biopsy.

A limited number of prior studies discuss the ability to subclassify NSCLCs into adenocarcinoma, squamous cell carcinoma, or a mixed carcinoma with an adenocarcinoma component using small biopsies, partly because of the recent clinical need for this distinction. Similar to other reports,[12-14] the current study found that ancillary immunostains were helpful in cases where the cytologic and histomorphologic findings were ambiguous on routine hematoxylin-eosin staining because of poor tumor differentiation or scanty tissue. Several useful immunohistochemical algorithms using various combinations of TTF-1, napsin A, p63, p40, and CK5/6 have been published, which allow for accurate subclassification of most ambiguous NSCLC cases while conserving tissue for potential molecular marker testing.[3,15,16] Use of special stains as a component of NSCLC subtyping is also recommended by the International Association for the Study of Lung Cancer, the American Thoracic Society, and the European Respiratory Society.[17]

Table 4.
Diagnostic Accuracy of Computed Tomography–Guided Lung Biopsy Specimens for Malignant Lesions

Diagnostic Accuracy	Patient Cases No. (%)
Diagnostic accuracy for malignant lesion[a]	189 (95)
False-negative rate for malignant lesion	9 (6)
Negative predictive value for malignant lesion[b]	35 (80)
Sensitivity for malignant lesion[c]	154 (94)

[a] Diagnostic accuracy for malignant lesions was calculated as the number of true-negative malignant cases plus the number of true-positive malignant cases divided by the sum of the number of true-negative malignant cases plus the number of true-positive malignant cases plus the number of false-negative cases and the number of false-positive cases.
[b] Negative predictive value for malignancy was calculated as the number of true malignant cases divided by sum of the number of true-negative malignant cases and the number of false-negative malignant cases.
[c] Sensitivity for diagnosis of malignancy is calculated as the number of cases diagnosed as malignant on first biopsy divided by the number of true malignant lesions.

Sigel et al[12] compared small tissue biopsy and cytologic specimens in the subtyping of 101 NSCLC cases. These authors reported tissue biopsy categories of definitive vs favored vs unclassified at 71%, 23%, and 6%, respectively.[12] However, when both cytology and biopsy paired specimens were combined, the rate of definitive diagnoses by at least 1 method was increased to 84%, and the unclassified rate decreased to 4%.[12] The authors concluded that optimal results are attained when the 2 modalities are considered jointly.[12] Similar to Sigel et al,[12] we successfully subclassified 86% of the NSCLC cases in the present study and favored a subtype in an additional 8% of cases. In addition, 6% of our NSCLC cases were unclassifiable (NSCLC, not otherwise specified). We believe the use of immunostains along with the combined use of cytology on the TP slides and histomorphology substantially increased our ability to subtype our NSCLC CNB cases, even though we did not specifically assess these parameters in this study.

Adequacy of Tissue for Molecular Studies

Although it can be challenging to acquire adequate amounts of tissue for NSCLC molecular testing, the present study demonstrated that sufficient material can be provided in most cases. Twenty-one of 22 NSCLC cases (95%) contained enough tissue to do some of the requested studies, and 17 of 22 (77%) cases contained enough tissue for us to successfully perform all of the requested molecular testing. The most commonly ordered tests were ALK and EGFR. Both of these tests were successfully performed in 17 of 22 (77%) cases; in 21 of 22 (95%) cases either test was successfully performed. Several studies[18-20] have addressed the adequacy of small lung biopsy specimens for molecular testing. Although our results fall within the range reported by others, it is difficult to compare results and draw conclusions because of the great variability in the way individual institutions procure and process tissue, what test menus and test platforms they use, and whether they reflex test all cases.[18-20] Solomon et al[18] reported that 16 of 18 (89%) patients had adequate CNB specimens for EGFR and KRAS mutational analysis. Hsiao et al[19] reported on 132 successful (80%) results for EGFR testing on 164 CNB specimens. Schneider et al[20] reported a success rate of 67% for EGFR, KRAS, and ALK testing on 52 CNB specimens with an even lower success rate (46%) for FNA. They concluded that when paraffin-embedded tissue is used for molecular testing, CNB specimens are more likely than FNA specimens to provide adequate tissue.[20] It cannot be confirmed, but part of our success with the molecular studies might have been due the use of CNB paraffin-embedded tissue rather than FNA cell block. Our use of a standard protocol in the present study also likely contributed to preserving adequate material for molecular studies.

Table 5.
Adequacy of Computed Tomography–Guided Needle Lung Biopsy Specimens for Molecular Studies

Patient No.	Molecular Test(s) Requested by Physician	Material Sufficient for All Requested Studies	Comment
23	ALK, EGFR, KRAS	Yes	…
34	ALK, EGFR	Yes	…
47 (metastatic)	HER-2,	Yes	…
48	ALK, EGFR, ERCC1, KRAS	Yes	…
49	ALK, EGFR, KRAS	Yes	…
54 (metastatic)	BRAF	Yes	…
57	ALK, EGFR, ROS1	Yes	…
59	ALK, EGFR	Yes	…
63	ALK, EGFR	Yes	…
68	ALK, EGFR, ERCC1, KRAS	Yes	…
69	ALK, EGFR, KRAS, ROS1	Yes	…
79	ALK, cMET, EGFR, ERCC1, KRAS, PBK, RRM1, Ts	No	Sufficient material only for ALK
82	ALK, EGFR	Yes	…
131 (metastatic)	BRAF	Yes	…
134	ALK, EGFR, KRAS	No	Sufficient material only for ALK
138	ALK, EGFR, ERCC1, KRAS	No	Sufficient material for EGFR, KRAS, ALK
139	ALK, EGFR, ERCC1, KRAS	Yes	…
143	ALK, EGFR, KRAS, ROS1	Yes	…
155	ALK, cMET, EGFR, FGFR1, KRAS, RET, ROS1,	Yes	…
157	ALK, EGFR, ERCC1, KRAS,	No	Sufficient material for EGFR, ERCC1, KRAS
163	ALK, EGFR, ROS1	No	Sufficient material for EGFR, ROS1
166	ALK, cMET, EGFR, ERCC1, KRAS, PBK, ROS1, RRM1	No	Insufficient material for all studies
186	ALK, EGFR, KRAS	Yes	…
202	ALK, EGFR, ROS1	Yes	…
226	ALK, EGFR, KRAS	Yes	…

Abbreviations: ALK, anaplastic lymphoma kinase; BRAF, B-Raf proto-oncogene, serine/threonine kinase; c-MET, mesenchymal-epidermal transition; EGFR, epidermal growth factor receptor; ERCC1, excision repair cross-complementation group 1; FGFR1, fibroblast growth factor receptor 1; HER-2, human epidermal growth factor receptor 2; KRAS, Kirsten rat sarcoma viral oncogene homolog; PBK, PDZ binding kinase; RET, rearranged during transfection; ROS1, ROS proto-oncogene 1, receptor tyrosine kinase; RRM1, ribonucleotide reductase M1; Ts, thymidylate synthase.

Limitations

The present study had several limitations. A major limitation is the lack of availability of outcome data from the patients without follow-up in the nondiagnostic and benign nonspecific groups that could have notably affected the results. Second, the follow-up time of benign nonspecific lesions may not have been long enough to ensure that none represented a false-negative result. Third, it is not clear whether some of the failures in molecular testing could have been caused by variations in individual reference laboratory methods or inadequate tissue. Finally, the data collected are from 1 center, which makes generalization more difficult.

Conclusions

The results of the present study show that CT-guided percutaneous lung needle biopsy in a community hospital setting can achieve a high accurate diagnostic yield with adequate tissue for molecular and other ancillary testing. We attribute our success to having a team composed of radiologists, pathologists, clinicians, and laboratory specialists who are dedicated to optimizing the procurement, processing, and diagnosis of cases. We approach each case as if ancillary studies might be required. The radiologist is always prepared to obtain additional tissue for such studies when the onsite pathologic evaluation shows that it might be indicated. Staff at the histology laboratory are careful to conserve tissue using proactive cutting procedures, and the pathologist is careful not to waste tissue by doing unnecessary immunostains. The literature emphasizes that this type of team approach is optimal for obtaining sufficient material,[4,21] and our results support this finding.

Acknowledgements

We thank Cody Anderson, DO, and Brittany Nobilette, DO, for their helpful contribution to the present study by gathering and tabulating data. We also thank Andrea R. Gwosdow, PhD, for her help preparing this manuscript for publication.

Author Contributions

Drs Florentine and Helton provided substantial contributions to conception and design, acquisition of data, or analysis and interpretation of data; Dr Schmidt contributed to acquisition of data and analysis; Drs Florentine, Helton, and Mitchell drafted the article or revised it critically for important intellectual content; Dr Kozlov gave final approval of the version of the article to be published; and Dr Florentine agrees to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

References

1. What are the key statistics about lung cancer? American Cancer Society website. http://www.cancer.org/cancer/lungcancer-non-smallcell/detailedguide/non-small-cell-lung-cancer-key-statistics. Accessed August 13, 2015.
2. Lindeman NI, Cagle PT, Beasley MB, et al. Molecular testing guideline for selection of lung cancer patients for EGFR and ALK tyrosine kinase inhibitors: guideline from the College of American Pathologists, International Association for the Study of Lung Cancer, and Association for Molecular Pathology [published online April 3, 2013]. *Arch Pathol Lab Med*. 2013;137(6):828-860. doi:10.5858/arpa.2012-0720-OA.
3. Mukhopadhyay S. Utility of small biopsies for diagnosis of lung nodules: doing more with less. *Mod Pathol*. 2012;25(suppl1):S43-S57. doi:10.1038/modpathol.2011.153.
4. Aisner DL, Marshall CB. Molecular pathology of non-small cell lung cancer: a practical guide. *Am J Clin Pathol*. 2012;138(2):332-346. doi:10.1309/AJCPFR12WJKCEEZZ.
5. Quint LE, Kretschmer M, Chang A, Nan B. CT-guided thoracic core biopsies: value of a negative result. *Cancer Imaging*. 2006;6:163-167.
6. Tsukada H, Satou T, Iwashima A, Souma T. Diagnostic accuracy of CT-guided automated needle biopsy of lung nodules. *AJR Am J Roentgenol*. 2000;175(1):239-243.
7. Loh SE, Wu DD, Venkatesh SK, et al. CT-guided thoracic biopsy: evaluating diagnostic yield and complications. *Ann Acad Med Singapore*. 2013;42(6):285-290.
8. Anderson JM, Murchison J, Patel D. CT-guided lung biopsy: factors influencing diagnostic yield and complication rate. *Clin Radiol*. 2003;58(10):791-797.
9. Schmidt RL, Witt BL, Lopez-Calderon LE, Layfield LJ. The influence of rapid onsite evaluation on the adequacy rate of fine-needle aspiration cytology: a systematic review and meta-analysis. *Am J Clin Pathol*. 2013;139(3):300-308. doi:10.1309/AJCPEGZMJKC42VUP.
10. Tsou MH, Tsai SF, Chan KY, et al. CT-guided needle biopsy: value of on-site cytopathologic evaluation of core specimen touch preparations [published online November 22, 2008]. *J Vasc Interv Radiol*. 2009;20(1):71-76. doi:10.1016/j.jvir.2008.10.011.

11. Gelbman BD, Cham MD, Kim W, et al. Radiographic and clinical characterization of false negative results from CT-guided needle biopsies of lung nodules. *J Thorac Oncol.* 2012;7(5):815-20. doi:10.1097/JTO.0b013e31824abd9c.

12. Sigel CS, Moreira AL, Travis WD, et al. Subtyping of non-small cell lung carcinoma: a comparison of small biopsy and cytology specimens. *J Thorac Oncol.* 2011;6(11):1849-1856. doi:10.1097/JTO.0b013e318227142d.

13. Mukhopadhyay S, Katzenstein AL. Subclassification of non-small cell lung carcinomas lacking morphologic differentiation on biopsy specimens: utility of an immunohistochemical panel containing TTF-1, napsin A, p63, and CK5/6. *Am J Surg Pathol.* 2011;35(1):15-25. doi:10.1097/PAS.0b013e3182036d05.

14. Khayyata S, Yun S, Pasha T, et al. Value of P63 and CK5/6 in distinguishing squamous cell carcinoma from adenocarcinoma in lung fine-needle aspiration specimens. *Diagn Cytopathol.* 2009;37(3):178-183. doi:10.1002/dc.20975.

15. Rekhtman N, Ang DC, Sima CS, Travis WD, Moreira AL. Immunohistochemical algorithm for differentiation of lung adenocarcinoma and squamous cell carcinoma based on large series of whole-tissue sections with validation in small specimens [published online May 27, 2011]. *Mod Pathol.* 2011;24(10):1348-1359. doi:10.1038/modpathol.2011.92.

16. Bishop JA, Teruya-Feldstein J, Westra WH, Pelosi G, Travis WD, Rekhtman N. p40 (ΔNp63) is superior to p63 for the diagnosis of pulmonary squamous cell carcinoma [published online November 4, 2011]. *Mod Pathol.* 2012;25(3):405-415. doi:10.1038/modpathol.2011.173.

17. Travis WD, Brambilla E, Noguchi M, et al. International Association for the Study of Lung Cancer/American Thoracic Society/European Respiratory Society: international multidisciplinary classification of lung adenocarcinoma: executive summary. *Proc Am Thorac Soc.* 2011;8(5):381-385. doi:10.1513/pats.201107-042ST.

18. Solomon SB, Zakowski MF, Pao W, et al. Core needle l ung biopsy specimens: adequacy for EGFR and KRAS mutational analysis. *Am J Roentgenol.* 2010;194(1):266–269. doi:10.2214/AJR.09.2858.

19. Hsiao SH, Chung CL, Lee CM, et al. Suitability of computed tomography–guided biopsy specimens for subtyping and genotyping of non–small-cell lung cancer [published online July 25, 2013]. *Clin Lung Cancer.* 2013;14(6):719-725. doi:10.1016/j.cllc.2013.06.002.

20. Schneider F, Smith MA, Lane MC, Pantanowitz L, Dacic S, Ohori NP. Adequacy of core needle biopsy specimens and fine-needle aspirates for molecular testing of lung adenocarcinomas. *Am J Clin Pathol.* 2015;143(2):193-200. doi:10.1309/AJCPMY8UI7WSFSYY.

21. Moreira AL, Thornton RH. Personalized medicine for non-small-cell lung cancer: implications of recent advances in tissue acquisition for molecular and histologic testing [published online March 14, 2012]. *Clin Lung Cancer.* 2012;13(5):334-339. doi:10.1016/j.cllc.2012.01.004.