Intraoperative Sentinel Node Mapping with Technitium-99 in Lung Cancer

Results of CALGB 140203 Multicenter Phase II Trial

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Introduction: Sentinel node mapping with radioactive technetium in non-small cell lung cancer has been shown to be feasible in several single institution reports. The Cancer and Leukemia Group B designed a phase II trial to test a standardized method of this technique in a multi-institutional setting. If validated, the technique could provide a more accurate and sensitive way to identify lymph node metastases.

Methods: Patients with clinical stage I non-small cell lung cancer amenable to resection were candidates for this trial. Intraoperatively, tumors were injected with technetium sulfur colloid (0.25 mCi). The tumor and lymph nodes were measured in vivo with a hand held Geiger counter and resection of the tumor and nodes was carried out. Sentinel nodes, all other nodes and the tumor were analyzed with standard histologic assessment. Negative sentinel nodes were also evaluated with immunohistochemistry.

Results: In this phase II trial, 8 surgeons participated (1–13 patients enrolled per surgeon), and 46 patients (out of a planned 150) were enrolled. Of these, 43 patients had cancer and an attempted complete resection, and 39 patients underwent sentinel node mapping. One or more sentinel nodes were identified in 24 of the 39 patients (61.5%). The sentinel node(s) were found to be accurate (no other nodes were positive for cancer if the sentinel node was negative) in 20/24 patients (83.3%). In the overall group the sentinel node mapping procedure was found to be accurate in 20/39 patients (51.2%).

Conclusions: Intraoperative sentinel node mapping in lung cancer with radioisotope yielded lower accrual and worse accuracy than expected. The multi-institutional attempt at validating this technique was unsuccessful.

Key Words: Sentinel node mapping, Lung cancer, Surgery.

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In the United States, lung cancer remains the number one source of cancer-related mortality in both men and women. Over 160,000 deaths will be attributed to lung cancer in 2008.[1] Complete surgical resection for localized disease is the most viable option for sustained remission or cure. Nonetheless the 5 year survival after complete resection of stage I tumors is only 60 to 70%. The patients who relapse after complete resection of stage I (lymph node negative) tumors by definition had occult disease at the time of their initial surgery.

Sentinel node mapping techniques have been applied to the resection and treatment of nearly all solid tumors with varying success and acceptance. The technique employs a lymphatic tracer (most commonly radioisotope or blue dye). The tracer is injected into the tumor and followed by visualization or gamma counter measurements of individual lymph nodes to determine the first site of efferent lymphatic drainage from a tumor. This sentinel node (SN) station should be the first site of lymphatic involvement if metastases have occurred.

SN assessment has become standard of care in breast cancer and melanoma surgery. The greatest utility of the technique in these cancers is the avoidance of nontherapeutic axillary or groin lymph node dissections and their incumbent morbidities. In lung cancer surgery, the adoption of SN identification has been slow in part because the morbidity of mediastinal lymph node dissection is not as significant as in breast and melanoma surgery. An equally important potential utility of SN mapping is the ability to direct pathologic examination and more sensitive techniques to detect occult micrometastatic disease.

The technique of radioactive tracer injection at the time of thoracotomy or thoracoscopy (video-assisted thoracoscopic surgery), in vivo and ex vivo mapping methods as described by Liptay[2] was employed in this phase II trial to investigate the feasibility of sentinel lymph node mapping in a multi-institutional protocol sponsored by the Cancer and Leukemia Group B.

In previous studies of the intraoperative technetium injection technique, several unsuccessful SN mapping efforts were originally attributed to clinically positive lobar or hilar nodes that resulted in lymphatic obstruction and tracer mi-

gration elsewhere. By using only clinically stage I patients, we had hoped to improve upon the results of single institution studies.

METHODS

Eligibility

Patients over 18 years of age with good performance status (Eastern Cooperative Oncology Group 0–2) with clinically diagnosed stage I lung cancers were considered for enrollment in this phase II feasibility trial. Patients with prior treatment for their lung cancers or thoracic radiation were excluded from enrollment. The trial opened in September 2004 with an accrual goal of 150 patients. It was closed before full accrual in November 2006.

Histologic confirmation of cancer was not required before enrollment. Mediastinoscopy was allowed but not required and when done was encouraged to be at a separate setting from the SN and resection procedure. All patients were provided with informed consent that complied with the National Cancer Institute guidelines, the Cancer and Leukemia Group B (CALGB) and local Institutional Review Boards.

Study Objectives

The primary objective of this study was to assess the feasibility and accuracy of intraoperative sentinel lymph node mapping in patients with resectable clinical stage I lung cancer in a multicenter setting by (1) SN identification rate (percentage of patients with lung cancer in whom at least 1 SN is identified) and (2) the accuracy rate, which reflects those patients with identified SNs positive for metastatic disease, and those with SNs negative for metastatic disease who do not have metastases in any other intrathoracic lymph nodes.

Secondary objectives were to evaluate the percentage of patients upstaged by the detection of micrometastases in SNs, the relationship between micrometastases and survival and the percentage of patients with skip metastases pattern (N2) SNs.

Intraoperative Technique

The intraoperative mapping technique was identical to that described by Liptay[2]. Briefly, it involved direct injection of the lung tumor with 0.25 mCi of Technetium Sulfur Colloid filtered once through a sterile 200 nanometer filter. After initial thoracotomy or thoracoscopic exploration, the tumor was injected with technetium-99m sulfur colloid (0.25 mCi) divided into 4 equal doses. Nuclear medicine personnel were present to assist in preparation of the radioisotope drawn into 4 tuberculin syringes (1 ml). The isotope was injected directly into the tumor using 27 gauge needles. During the time allowed for migration of the radioactivity through the lymphatics, care was taken to avoid disrupting the peribronchial tissues where the majority of lymphatic channels reside.

The migration of the radioisotope was tracked through the lymphatics by a hand held gamma probe (Navigator, Tyco Corporation). The tumor and nodal stations were initially surveyed in the thorax and background levels recorded within the mediastinum, distant from the primary tumor. Because the initial in vivo readings may often be confounded by shine-through effect of radioactivity of the tumor, final confirmatory readings were taken ex vivo. Visible mediastinal, hilar, and peribronchial lymph nodes were dissected and measured separately from the tumor specimen after removal from the chest (ex vivo).

Migration of the technetium sulfur colloid solution was considered successful if a specific nodal station registered counts per second (cps) greater than three times background values. All lymph nodes with ex vivo cps greater than this level were classified as SNs.

After the initial scintographic readings and standard anatomic resection with ipsilateral mediastinal node dissection was completed, repeat gamma probe examination was performed to assess for residual radioactivity and potentially overlooked lymph nodes. Reresection of nodal stations was performed based on the handheld gamma counter readings and visual inspection.

Lymphadenectomy and Pathologic Evaluation

A full hilar and mediastinal node dissection was required. A minimum of 5 lymph node stations were required based on the tumor location. Every effort was made to remove nodes in their entirety.

After SN identification, SNs were examined first with standard histology with hematoxylin and eosin (H&E) using conventional bi-valving techniques. If initial histologic examination was negative for metastases, at least 3 serial (step) sections at 30 to 40 μm intervals were evaluated. In addition, initially negative SNs were examined with immunohistochemistry using the AE1/AE3/PCK26 cytokeratin antibody (Ventana Medical Systems Inc, Tucson, AZ) according to standard protocol. Immunohistochemistry was considered positive if it demonstrated positive cell clusters or individual cells with the appropriate tumor cell morphology.

RESULTS

The planned accrual target for this study was 150 patients. The trial was terminated early based on disappointing accuracy, slower than expected accrual and reduced National Cancer Institute funding. Difficulties with radiation safety requirements and challenges to collaboration between specialties (surgery, nuclear medicine, and pathology) impaired accrual at some institutions.

Forty-six patients with suspected localized lung cancer were initially registered to participate. Of these, 44 patients were diagnosed with potentially resectable non-small cell lung cancer. Five patients did not undergo the SN procedure (Radio-isotope not available, [1] patient withdrew consent, [2] Metastatic disease found [1] and unknown reasons [1]).

The remaining 39 patients with clinical stage I tumors underwent an attempted intraoperative SN mapping procedure with the injection of technetium sulfur colloid. The demographic information on the participating patients is summarized in Table 1. There were 22 men and 17 women with a mean age of 68 (range, 46–85). Adenocarcinoma was the most commonly encountered cell type and the upper lobes were the most frequently involved lobes. All patients had

TABLE 1. Demographics/Tumor Stage

Patients M/F	22/17
Age–mean (range)	68 (46–85)
Race–White/Black	37/2
ECOG PS 0/1	35/4
Tumor cell type	N
Adenocarcinoma	22
Squamous cell	11
Large cell	3
Bronchioalveolar cell	3
Pathologic stage	
IA T1N0	16
IB T2N0	14
IIA T1N1	2
IIB T2N1	4
IIIA T1N2	2
IIIA T2N2	1
Lobe involved	
Left upper	11
Right upper	10
Right lower	10
Left lower	5
Right middle	3

M, male; F, female; ECOG, Eastern Cooperative Oncology Group; PS, performance status.

TABLE 2. Sentinel Node Accuracy by Surgeon/Location/Stage

Surgeon	N	SN ID	Accuracy
A.			
A	13	9/13	7/9
B	6	5/6	5/5
C	7	4/7	4/4
D	5	2/5	2/2
E	3	1/3	1/1
F	2	2/2	1/2
G	2	0/2	—
H	1	1/1	0/1
Totals	39	24/39	20/24
B.			
Lobe			
LUL	11	5/10	4/5
RUL	10	7/9	5/7
RLL	10	4/9	4/4
LLL	5	4/6	3/4
RML	3	2/3	2/2
C.			
Pathologic stage			
IA T1N0	16	9/16	9/9
IB T2N0	14	9/14	9/9
IIA T1N1	2	0/2	—
IIB T2N1	4	4/5	2/4
IIIA T1N2	2	1/2	0/1
IIIA T2N2	1	1/1	0/1

SN, sentinel node; LUL, left upper lobe; RUL, right upper lobe; RLL, right lower lobe; LLL, left lower lobe; RML, right middle lobe.

TABLE 3. Pathologic Nodal Status

	Number
Sentinel negative, other node(s) negative	18
Sentinel negative, other node(s) positive	4
Sentinel node positive, other node(s) negative[a]	2[a]
Sentinel node positive, other node(s) positive	0
No sentinel node identified, all nodes negative	12
No sentinel node identified, one or more nodes positive	3
Total patients	39

[a] Only positive with serial sections (1) and immunohistochemistry (1).

clinical stage I tumors. Thirty of the 39 patients had node negative stage I tumors (Table 1).

Eight surgeons participated in this national trial sponsored by the Cancer and Leukemia Group B. Surgeons enrolled between 1 and 13 patients. Before participating, surgeons were required to view an instructional video and complete and pass a quiz on the technical aspects of the intraoperative SN mapping procedure (Table 2).

Of the 39 patients injected with the radioactive tracer, at least one specific SN was identified in 24 (61.5%) (Table 2). The SNs was accurately classified in 20 of those 24 patients (83.3%). This accuracy was defined by either a positive SN, or no other positive lymph nodes found when the SN was negative for metastatic involvement. This was an overall accuracy rate of 51.2% (20 of 39 patients) (Table 3).

No specific pattern of failure to identify SNs or detect migration of the radioisotope through the lymphatics was noted in relation to specific lobar involvement or ultimate pathologic stage.

Of the 39 patients, 9 had pathologically identified positive lymph nodes. A SN was identified in six of these nine patients but was accurate (i.e.,: pathologically involved as well as the other involved nodes) in only two out of the six patients. In these two cases the SN was negative on standard H&E staining but cancer cells were identified on immunohistochemistry for epithelial markers in the nodal tissue. An overall accuracy rate was 51.2% (20 of 39).[3]

DISCUSSION

Sentinel node identification has become standard of care in melanoma and breast cancer surgical care. The idea of a first nodal station draining a tumor theoretically would allow the assessment of that node(s) to represent the state of the remaining regional nodes. The SN is used to limit potentially morbid lymph node dissections. Another benefit of the technique is directing applications of more focused pathologic or molecular staging techniques (Serial sections, Immunohistochemistry, reverse transcription-polymerase chain reaction etc.). This ability to direct a more focused search for metastatic disease in the SN rather than all of the nodes removed is a primary benefit of the technique in lung cancer.

With current indications for adjuvant chemotherapy in resected lung cancer largely based on the status of the

locoregional lymph nodes, the accurate identification of positive nodes has gained therapeutic importance.

Intraoperative Tracer Injection

Sentinel node mapping in lung cancer was first reported by Little et al.[4] in 1999. They used blue dye and found a less than 50% success rate with the main impediment being difficulty distinguishing blue dyed nodes from black anthracotic nodes. Intraoperative radionuclide mapping techniques reported improved results of SN identification,[5–7] but issues of shine through effect from the tumor and background aerosolization of radioactivity made in vivo readings often unreliable.

The first reported use of intraoperative radioactive tracer was in 2000.[6] The technique used Technitium-99 sulfur colloid filtered through a 20 μm filter. This initial single institution series demonstrated promise with consistently short migration times for the radioisotope and reasonably accurate SN identifications.[5,6] Ninety-one patients with operable lung cancer underwent the intraoperative SN technique. Mean time from injection of the radionuclide to identification of SN was 63 minutes (range, 23–170). Seventy-eight of the 91 (86%) had a SN identified and 69 of 78 SNs were classified as true-positives, with no metastases found in other intrathoracic lymph nodes without concurrent SN involvement. Thus, an accurate SN was identified in 69 of 91 (76%) patients with lung cancer mapped with intraoperative technetium radionuclide. Metastatic disease was found in 21 (27%) of the 78 identified SNs. In 9 of these 21 patients, the SN was the only positive node. In 7 (77%) of these 9 patients, the SN was negative by conventional histology and only harbored micrometastatic disease detected by either serial sectioning or immunohistochemistry.[5]

Since these initial reports, other single institutional groups have reported SN identification rates of 74% with Technitium alone[7] and 81% using a combination of technetium and blue dye.[8] In contrast, the accepted accuracy rate of SN identification in breast cancer is over 95%. While the use of radionuclide in lung cancer mapping techniques reported improved results of SN identification, issues of shine through effect from the tumor and background aerosolization of radioactivity made in vivo readings often unreliable and ex vivo readings were required for accurate SN identification.

CALGB 140203 opened for accrual in September 2004. After 2 years, 46 of the planned 150 patients were accrued. Factors affecting accrual and accuracy rates included barriers to cooperation between nuclear medicine, surgery and pathology for intraoperative use of radioactivity and intensified pathologic evaluation along with various state regulations for radioactivity handling complicating the protocol.

The disappointing 51.2% SN identification rate in our series might have been due in part to a learning curve potentially not overcome by the time of the study's termination. Eight surgeons participated in the study for an average of 5 cases and range between 1 and 13.

Preoperative Tracer Injection

Transportation and handling of the radioisotope in the operating room adds a layer of complexity to the intraoperative SN procedure in lung cancer. Preoperative transthoracic injection, usually under CT guidance, has some logistical benefits. It allows the injection to be done in radiology or nuclear medicine and avoids the intraoperative handling of radioisotope. However, the risks of pneumothorax, bleeding and seeding of tumor along needle tracts are all at least theoretical concerns.

Because of restrictions in the use and handling of radioactive substances in Japan, Japanese surgeons have been pioneers in the preoperative injection of radioactive tracer for SN mapping in lung cancer. Technetium Tin Colloid has been the most reported. With its higher mass, the migration time is prolonged enough to have the CT guided injection the day before surgery.

Nomori et al.[9] have reported the largest experience. Most recently, they report an 83% SN identification rate in 53 patients with small clinical T1N0 tumors considered for segmentectomy. Their SN guided segmentectomy resulted in positive SNs on frozen section leading to completion lobectomies in three patients.

Nonradioactive Tracers

Several groups have moved on to study new techniques to nodal mapping which do not rely on radioactive materials and have had promising results. Soltecz et al.[10] reported on the use of quantum dots that fluoresce in the near infrared spectrum. They used a camera system that simultaneously acquires color video, the near-infrared fluorescence and a merged picture to guide dissection. In pigs this technique was able to reliably identify nodal drainage and SNs. Lymphatics and nodes were able to be seen through 1 cm of solid tissue and 5 cm of lung parenchyma. A clinical trial using this technology in humans with lung cancer is nearing accrual.

Adusumilli et al.[11] have recently reported success with herpes simplex virus containing a green fluorescent protein transgene. This oncolytic herpes strain was shown to easily infect cancer cells and track lymphatic metastases within 2 to 4 hours of injection in a murine model. Fluorescent thoracoscopy could detect these nodal metastases aiding in resection and due to the oncolytic effect of the virus, a therapeutic potential was also cited.

Intraoperative FDG-PET

In a pilot study, Nwogu et al.[12] injected 10 patients with 10 mCi of F18-fluorodeoxyglucose (FDG) on the day of surgery. Using a handheld device in surgery they discovered 3 (30%) patients who had FDG positive nodes with micrometastases present. This technique differs from a standard SN mapping as the FDG is selectively taken up by tumor cells rather than tracking the lymphatic drainage. Further study on this form of ultra-staging is needed. This is the first study examining a technique to actually identify metastases and not the SN that may harbor disease.

The suboptimal results of this phase II trial demonstrate the limitations of the intraoperative radiotracer technique requiring collaboration between nuclear medicine, surgery and pathology disciplines as well as a learning curve that may not have been completed before its termination.

Accurate assessment of lymph node status in operable lung cancer remains critical. This multi-institutional study sponsored by the CALGB failed to demonstrate the feasibility of an intraoperative SN mapping technique with a radioactive technetium-99 tracer. New techniques for both diagnostic SN mapping potentially coupled with the delivery of therapeutic agents are under investigation. With the aid of new technologies and innovation, the surgeon will continue to lead the diagnosis and treatment of early stage lung cancer.

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