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Lymphoseek: A Molecular Radiopharmaceutical for Sentinel Node Detection

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Background: Lymphoseek is a new radiopharmaceutical that accumulates in lymphatic tissue by binding to a receptor that resides on the surface of macrophage cells. We conducted a phase I clinical trial in which Lymphoseek was compared with filtered [^{99m}Tc]sulfur colloid (fTcSC) for sentinel node detection in patients with breast cancer.

Methods: Twelve women (42–71 years) with breast cancer were randomly assigned to a 3-hour imaging protocol with peritumoral/subdermal injections (.5 mCi) of either Lymphoseek (1 nmol; molecular weight, 28 kDa; diameter, .007 μm) or .2 μm of fTcSC. Serial images were acquired for 180 minutes. Sentinel nodes, excised within 4.2 to 7.3 hours of administration, were assayed in a dose calibrator.

Results: The receptor-binding agent, Lymphoseek, exhibited a significantly ($P = .0025$) faster injection site clearance (rate, .255 ± .147/hour; fTcSC rate, .014 ± .018/hour); the mean Lymphoseek clearance half-time was 2.72 ± 1.57 hours compared with 49.5 ± 38.5 hours for fTcSC. The primary sentinel node uptake of Lymphoseek (range, .02%–1.12%; mean, .55% ± .43%) and fTcSC (range, .00%–1.93%; mean, .65% ± .63%) did not differ ($P = .75$). Lymphoseek exhibited a lower mean number of sentinel nodes per study (n = 1.3) than fTcSC (n = 1.7) and a higher concordance with Lymphazurin.

Conclusions: The molecular receptor-binding agent Lymphoseek demonstrated faster injection site clearance and equivalent primary sentinel node uptake when compared with fTcSC.

Key Words: Sentinel lymph node biopsy—Radiopharmaceutical—[^{99m}Tc]DTPA-mannosyl-dextran—Lymphoseek.

Technetium-99m sulfur colloid, the standard radiopharmaceutical for sentinel node imaging in the United States, is a large-diameter particle composed of sulfur and gelatin. Introduced in 1966,[1,2] the agent was designed for hepatic imaging. Filtration[3,4] has recently been used to enhance biodistribution for lymph node mapping; however, this was not its original Food and Drug Administration–approved use. An ideal sentinel node agent is one that allows rapid injection site clearance, so as to not interfere with sentinel node identification, but that also has nodal binding properties, so as to limit drainage to distal nodes that are not anatomically sentinel. Neither unfiltered nor filtered [^{99m}Tc]sulfur colloid (fTcSC), albumin-based colloids used in Europe, or [^{99m}Tc]antimony trisulfide colloid, used in Australia, exhibits these properties,[5–7] and as such their use often leads to cases in which either the sentinel node is not properly identified or in which multiple distal nodes are visualized, causing more than the necessary sentinel nodes to be removed.

Lymphoseek is a new radiopharmaceutical[8] that accumulates in lymphatic tissues by binding to a receptor that resides on the surface of macrophage cells. The receptor, a mannose-binding protein,[9] recognizes and binds macromolecules with carbohydrate side-chains that terminate with a mannose glycoside. Lymphoseek (Fig. 1), previ-

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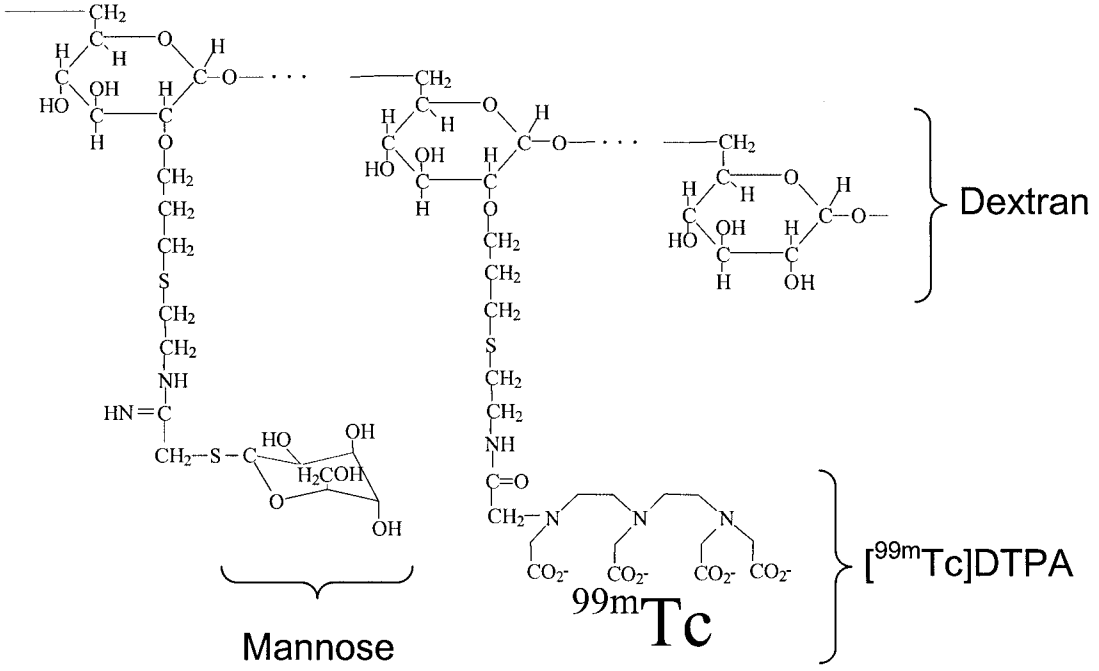

FIG. 1. The chemical structure of Lymphoseek, also known as [^{99m}Tc]diethylenetriamine pentaacetic acid (DTPA)-mannosyl-dextran. The average molecular weight was 28,200 g/mol. The mean molecular diameter was 7.0 nm. The mannose units are substrates for receptor recognition and binding. The DTPA units serve as attachment sites for labeling the molecule with ^{99m}Tc.

ously ^{99m}Tc-diethylenetriamine pentaacetic acid (DTPA)-mannosyl-dextran, is composed of a dextran backbone to which multiple units of mannose and DTPA are synthetically attached. The dextran, having a molecular weight of 10,000 g/mol, satisfies the receptor's requirement for a macromolecule. The mannose units are substrates for receptor recognition and binding. The DTPA units serve as attachment sites for labeling the macromolecule with ^{99m}Tc.

Previous preclinical studies[8] with Lymphoseek demonstrated the properties required for sentinel lymph node imaging. The agent exhibited high affinity for the receptor; the equilibrium binding constant K_D was .12 nM. In rabbits, Lymphoseek exhibited significantly faster injection site clearance than fTcSC and significantly lower distal node uptake.

In this article, we present the injection site clearance and sentinel node uptake data from the first 12 subjects of the phase I study. Six subjects received fTcSC, and six subjects received a 1-nmol dose of Lymphoseek.

MATERIALS AND METHODS

Institutional Review

A phase I clinical trial of Lymphoseek was conducted at the Thornton Hospital of the University of California, San Diego Medical Center, La Jolla. The protocol received the consent of the Division of Medical Imaging and Radiopharmaceutical Drug Products of the US Food and Drug Administration as a physician-sponsored investigational new drug. The protocol and the informed consent form were approved by the University of California, San Diego, Office for Human Research Protection, Cancer Center Protocol Review Monitoring Committee, Human Exposure Review Committee, and Radioactive Drug Research Committee.

Patient Selection

Patients with breast cancer who would normally be offered sentinel node biopsy per University of California, San Diego guidelines participated in this study. The need to have follow-through axillary node dissection was determined by pathologic outcome of the sentinel node and did not affect this study. Twelve qualifying and consenting subjects were randomly selected to receive one of the two imaging agents: Lymphoseek or fTcSC. Pregnant and lactating women, male patients, patients with known metastatic breast cancer, and patients currently enrolled in another protocol were excluded from this study. The subjects ranged in age from 43 to 71 years (Table 1). One patient (subject 7) had bilateral disease and, therefore,

TABLE 1. *Subject and radiopharmaceutical summary*

Subject No.	Age (y)	Diagnosis	Size (cm)	Quadrant	SN status	Radiopharmaceutical	Radiochemical Purity (%)
1	48	IDC and DCIS	.8	Lower-outer	Negative	fTcSC	93.0
2	48	IDC	1.3	Nipple	Negative	Lymphoseek	99.2
3	70	IDC and DCIS	.8	Nipple	Negative	fTcSC	97.0
4	55	IDC	2.5	Upper-outer	Positive	fTcSC	93.4
5	71	IDC	1.5	Lateral	Negative	Lymphoseek	98.0
6	58	IDC and DCIS	.9/1.8	Upper-inner	Negative	Lymphoseek	97.7
7[a]	57	DCIS	1.5	Lower-outer	Negative	fTcSC	95.5
7[a]		DCIS/ICP	2.6	Nipple	Negative	fTcSC	95.5
8	55	IDC	1.6	Lateral	Negative	fTcSC	98.8
9	53	IDC	2.5	Upper-inner	Negative	Lymphoseek	98.4
10	46	DCIS	4.0	Upper-inner	Negative	fTcSC	98.3
11	42	Infiltrating lobular	2.5	Upper-outer	Negative	Lymphoseek	97.7
12	43	DCIS	4.0	Upper-mid	Negative/positive[b]	Lymphoseek	97.7

IDC, infiltrating ductal carcinoma; DCIS, ductal carcinoma-in-situ; ICP, intracystic papillary carcinoma; fTcSC, filtered [^{99m}Tc]sulfur colloid.
[a] Bilateral disease.
[b] On the basis of immunohistology.

has two entries in Table 1. Tumor size ranged from .8 to 4.0 cm. One sentinel node was positive on frozen section, and one was negative on frozen section but positive on immunohistochemistry.

Agent Preparation

Lymphoseek was synthesized as previously described.[8] Clinical-grade dextran (Amersham-Pharmacia Biotech, Piscataway, NJ), having an average molecular weight of 10,500 g/mol, was tested for antigenic impurities by USP 24 and was used as the molecular backbone for the agent. The average molecular weight of the Lymphoseek preparation used in this study was 28,200 g/mol. The average DTPA and mannose densities were 2.1 and 42 mol of DTPA and mannose per mole of dextran. The mean molecular diameter was 7.0 nm. Formulation, ^{99m}Tc labeling, and quality control were performed as previously described.[10] The radiochemical purity (Table 1) ranged from 97.7% to 99.2%. The radiopharmaceutical was administered within 1 hour of radiolabeling. The chemical structure of the radiopharmaceutical is depicted in Fig. 1.

Sulfur colloid labeling kits were purchased from CIS-US (Bedford, MA). The CIS-US labeling protocol as described by the package insert was used and followed by filtration through a .22-μm syringe filter (Millex-GV; Millipore Corp., Bedford, MA). Quality control[11] of the filtered product included instant thin layer chromatography with Whatman 31ET chromatography paper (Whatman Ltd., Maidstone, UK) as the stationary phase and 85% methanol as the mobile phase, measurement of pH via paper indicator, and visual inspection for color and particulate matter. The fTcSC was administered within 1 hour of filtration; the radiochemical purity (Table 1) ranged from 93.0% to 98.3%. The fTcSC colloid ranges in size from 5 to 50 nm;[4] the unfiltered colloid has a bimodal size distribution of 5 to 50 nm and 200 to 650 nm.[7]

Nuclear Imaging

Each patient received four 1.0-mL, 125-μCi injections of either Lymphoseek (.25 nmol) or fTcSC at the 3-, 6-, 9-, and 12-o'clock position surrounding the breast lesion by using a peritumoral/subdermal technique. Each subject was monitored for any sign of an allergic reaction, such as the occurrence of a rash, hives, edema, or other cutaneous manifestations. Electrocardiograms were obtained at baseline, immediately after injection, and 30 minutes after injection. Blood pressure, heart rate, respiratory rate, temperature, and oxygen saturation were obtained at baseline; immediately after injection; at 15 and 30 minutes after injection; and 1, 2, and 24 hours after administration. Clinical chemistry (21-panel), hemogram, and urinalysis panels were obtained at baseline, 4 hours, and 24 hours. All subjects were subjectively evaluated on arrival for the study and were monitored for 24 hours during an overnight hospital stay. Patients were additionally seen 1 week later in clinic follow-up and either were contacted by phone 30 days after the procedure for a subjective evaluation or were seen in the clinic.

Sentinel node imaging of the Lymphoseek and fTcSC studies used the same imaging protocol. With a large–field-of-view gamma camera fitted with a high-resolution collimator, images of the administration site were acquired immediately after the injection and at 15-minute intervals for 1 hour and at 2 and 3 hours after injection. At each time point, oblique views of the chest

(which included the axilla and breast) and anterior views that included the liver were acquired. An imaging standard of a known dilution of injectate was placed within the field of view of each image. All images were acquired for 3 minutes and stored on an image-processing computer as a 256 × 256 × 16 matrix. Whole-body scans (anterior and posterior views) were performed at 1.0, 3.0, and 12 hours after injection. Blood was sampled at 15 minutes and 1, 2.5, 4, and 6 hours after injection. Hematocrits for each sample were measured, and the plasma and blood cells were separated by centrifugation. Known volumes of plasma and whole blood were assayed for radioactivity.

The clearance of Lymphoseek and [^{99m}Tc]sulfur colloid from the injection site was quantified in the following manner. For each of the serial images acquired from 15 minutes through 3 hours, regions of interest were drawn around the injection site and the imaging standard. By using standard nuclear medicine software, the total number of counts within each region of interest was calculated. The logarithm of these values was plotted as a function of time; a linear regression (JMP; SAS Institute, Cary, NC) yielded a straight line with slope m. The clearance rate constant k_c was defined as the absolute value of m. If the regression yielded a positive slope, the value was set to 0. The clearance half-life was calculated as $-.693/m$.

Sentinel Node Detection and Measurement

Sentinel node biopsy was performed with standard technique. At the start of the surgical procedure, isosulfan blue (Lymphazurin 1%; US Surgical Corp., Norwalk, CT) was injected in the same manner as the radiopharmaceutical. The injection site was massaged for several minutes. Also during this time, a handheld gamma probe (Neoprobe 2000; Neoprobe Corp.) was used to scan the skin surface to localize the underlying sentinel node base on radioactivity. If the sentinel node was visualized by nuclear imaging, the scintigrams were used to guide the search. After marking the skin at the site of the highest count rate, the patient was prepped for surgery, and an incision was made at the marked location. Dissection was performed with the gamma probe to the hot lymph node, to the lymphatic taking up the blue dye, or both. The lymph node was isolated, removed, and placed on the tip of the gamma probe. The count rate of the excised lymph node was recorded and defined as the target activity. A background measurement was made by placing the tip of the gamma probe on the skin surface at least 20 cm from the injection site. Verification that the node contained radioactivity at least 10 times background was performed before it was sent to pathology. Frozen-section analysis was then performed to identify metastases. Sentinel nodes were defined by having a node-background ratio of at least 3 or an ex vivo count rate greater than a minimum value defined by the following equation, which uses the injected dose in millicuries:

$$\text{Minimum count rate} = 150 \text{ cps} \times \text{injected dose}/1 \text{ mCi} \quad (1)$$

Finally, the gamma probe was placed back within the nodal basin to ensure that no significant residual radioactivity remained. If the sentinel node was histologically positive, the axillary lymph node dissection was completed.

The percentage of injected dose in the sentinel node ($\%ID_{SN}$) was calculated in the following manner. All sentinel nodes were assayed for radioactivity by using a dose calibrator located adjacent to the operating room. This assay required approximately 10 minutes, after which the biopsy sample was submitted for pathologic examination. The mean of at least six measurements was normalized by counting .20 mL of the injectate. The result yielded the $\%ID_{SN}$. The SE of $\%ID_{SN}$ was calculated as the quadratic sum of the measurement errors.[12]

Statistical Methods

Tests of statistical significance used the unpaired Student's t-test with JMP software. A P value of $<.05$ was considered statistically significant. For subject 7, with bilateral injections, calculation of the mean $\%ID_{SN}$ used the average of both breasts.

RESULTS

Lymphoseek exhibited a significantly ($P < .0025$) faster injection site clearance than fTcSC. The clearance rate constants for Lymphoseek ranged from .114 to .444/hour (Table 2); k_c for fTcSC ranged from .0 to .045/hour. The mean injection site clearance rate constant k_c for Lymphoseek equaled .255 ± .147/hour (Table 3), which was an order of magnitude higher than the mean fTcSC clearance rate constant of .014 ± .018/hour. These values translated to clearance half-times of 2.72 ± 1.57 hours for Lymphoseek and 49.5 ± 38.5 hours for fTcSC. The calculated percentage of injected dose within the injection site at the time of excision ranged from 6% to 56% for Lymphoseek and 82% to 100% for fTcSC. These values depend on the time of sentinel node excision, which ranged from 4.2 to 7.3 hours after administration (Table 2). There were no abnormal variations in vital signs or electrocardiogram tracings from the preprocedure evaluations. Clinical chemistry, hemogram, and urinalysis deviations were those commonly

TABLE 2. *Injection site clearance and sentinel node uptake*

Subject No.	Radiopharmaceutical	Clearance: Rate constant, k_c/h[a]	Clearance: Half-life, T_c (h)[a]	% Dose at excision, $\%ID_{IS}$ (%)	Excision time (h)	$\%ID_{SN}$[a]: Primary (%)	Second (%)	Third (%)	Fourth (%)
1	fTcSC	.045 ± .015	15.4 ± 5.10	82	4.2	.19 ± .04			
2	Lymphoseek	.215 ± .030	3.23 ± .47	21	7.3	.84 ± .09			
3	fTcSC	.018 ± .004	38.2 ± 9.2	91	4.8	.56 ± .11			
4	fTcSC	0	–	100	5.4	.37 ± .05	.36 ± .01		
5	Lymphoseek	.434 ± .091	1.60 ± .34	14	4.5	.02 ± .03	0[b]		
6	Lymphoseek	.172 ± .008	4.03 ± .18	30	7.0	.07 ± .16	0[b]		
7[c]	fTcSC	0	–	100	5.0	1.93 ± .04	.13 ± .40[d]	.14 ± .31[d]	.11 ± .08[d]
7[c]	fTcSC	0	–	100	5.5	.25 ± .21			
8	fTcSC	0	–	100	4.5	0			
9	Lymphoseek	.444 ± .044	1.56 ± .16	6	6.3	.54 ± .45			
10	fTcSC	.019 ± .002	36.5 ± 3.71	91	5.0	1.67 ± .15			
11	Lymphoseek	.149 ± .014	4.65 ± .44	36	6.8	.70 ± .07	.20 ± .10[d]		
12	Lymphoseek	.114 ± .009	6.08 ± .50	56	5.5	1.12 ± .08	.53 ± .09		

Primary, hottest lymph node; fTcSC, filtered [^{99m}Tc]sulfur colloid; $\%ID_{IS}$, percentage of injected dose at injection site at time of excision; $\%ID_{SN}$, percentage of injected dose in the sentinel node.
[a] Mean ± SD.
[b] Lymph node activity equivalent to background.
[c] Bilateral disease.
[d] Nonblue lymph node.

associated with surgery. Any increases were rechecked and showed resolution. No adverse outcomes related to the agent were noted.

Figure 2 illustrates the clearance of radioactivity from the administration sites of two sentinel node studies. The data composed of diamonds are from a sentinel node study (subject 2) that used Lymphoseek. Linear regression analysis gave a slope of .215 ± .030/hour. This value is equivalent to a half-life of 3.23 ± .47 hours. The $\%ID_{SN}$ at 7 hours after administration of Lymphoseek was .84% ± .09%. The data composed of triangles are from a study that used fTcSC (subject 1). The linear regression yielded a slope of .045 ± .015/hour, which is equivalent to a half-life of 15.4 ± 5.10 hours; the $\%ID_{SN}$ at 4 hours after administration was .19% ± .04%. Three studies produced data sets that yielded a linear regression with a positive slope; the k_c values were, therefore, set to 0. A positive slope can randomly occur in decay-corrected time-activity data obtained from tissue that exhibits no accumulation or clearance of a radiotracer.

The primary sentinel node uptake was extremely variable (Table 2); the range for the receptor-binding agent was .02% to 1.12%, and the range for the fTcSC was from no uptake to 1.93%. With Lymphoseek, the mean value for primary sentinel node uptake ($\%ID_{SN}$) was .55% ± .43% of the injected dose. This value was not significantly different ($P = .75$) from the mean sentinel node uptake of .65% ± .63% for fTcSC.

TABLE 3. *Injection site clearance and sentinel node uptake of Lymphoseek and filtered [^{99m}Tc]sulfur colloid*

Radiopharmaceutical	n	Clearance[a]: Rate constant, k_c(/h)	Clearance[a]: Half-life, T_c[c] (h)	% Dose at excision, $\%ID_{IS}$ (%)[a]	$\%ID_{SN}$[b]: Mean[d] ± SD (%)	Range (%)
Lymphoseek	6	.255 ± .147	2.72 ± 1.57	26.6 ± 16.8	.55 ± .43	.02–1.12
Filtered [^{99m}Tc]sulfur colloid	6[ad]	.014 ± .018	49.5 ± 38.5	94.9 ± 15.7	.65 ± .63	.00–1.93
P value		.0025		<.0001	.75	

[a] Mean ± SD.
[b] Primary only.
[c] On the basis of the mean rate constant k_c.
[d] Subject 7, average of both injections.

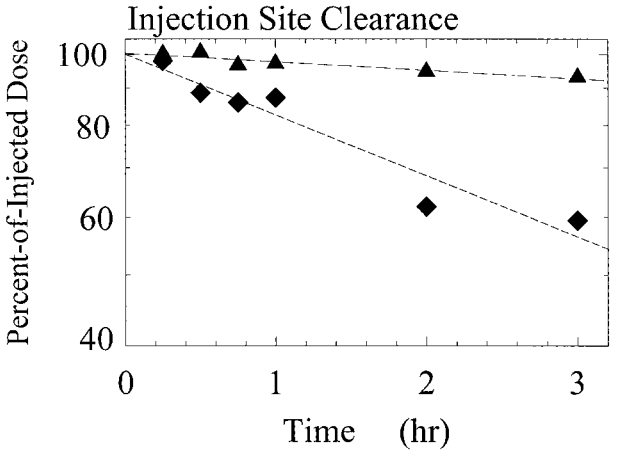

FIG. 2. Clearance of radioactivity from the injection site of two sentinel node cases. Lymphoseek (◆, subject 2) exhibited a half-life of 3.23 ± .47 hours. Filtered [^{99m}Tc]sulfur colloid (▲, subject 1) exhibited a half-life of 15.4 ± 5.10 hours.

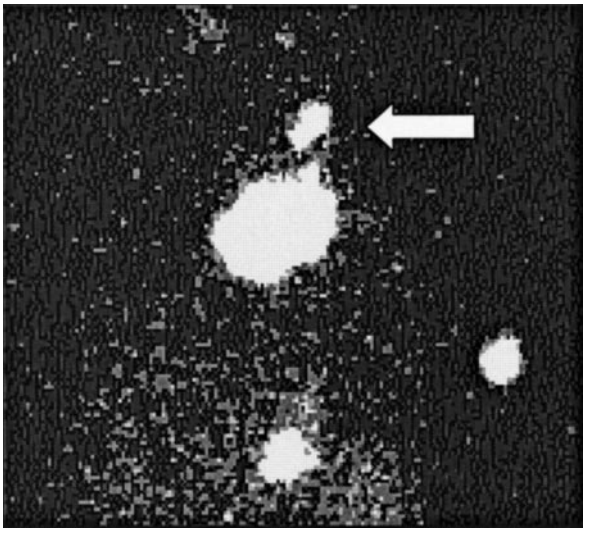

FIG. 3. An image acquired 3 hours after injection of Lymphoseek (subject 2). When measured at 7.3 hours, the sentinel node (arrow) accumulated .84% ± .09% of the dose. On the basis of the clearance half-life, the injection site at 7.3 hours contained 21% of the dose. The two foci of activity in the lower right quadrant of the image are imaging standards for calibration.

Figure 3 is an image acquired 3 hours after injection of Lymphoseek (subject 2). The two foci of activity in the lower right quadrant of the image are the imaging standards. When measured at 7.3 hours, the sentinel node (arrow) accumulated .84% ± .09% of the dose. On the basis of the clearance half-life of 3.23 hours, 21% of the Lymphoseek dose remained at the injection site.

All of the studies that used Lymphoseek exhibited a sentinel node by gamma probe detection. The mean number of sentinel nodes detected by Lymphoseek per patient was 1.3. The mean number of sentinel nodes detected by Lymphazurin per patient was also 1.5. Of the six Lymphoseek studies, a secondary sentinel node was detected by the gamma probe during two studies (Fig. 4A). Of these two lymph nodes, only one was blue. A third study (subject 6) yielded a blue distal node that did not achieve the minimum activity requirement and was not defined as a sentinel node. An additional study yielded a blue lymph node distal to the sentinel node. This blue lymph node had a target-background ratio <3 and was, therefore, not classified as a sentinel node. Concordance of the primary sentinel nodes between Lymphoseek and Lymphazurin was 100%; the concordance of all sentinel nodes was 70%.

One of the seven studies using fTcSC failed to detect a sentinel node (Fig. 4B), resulting in an 87% success rate. Of the six remaining studies, one study detected a secondary sentinel node by the gamma probe and visualization by Lymphazurin, and another study detected three additional lymph nodes that were not stained blue but qualified as sentinel nodes by gamma detection. The mean number of sentinel nodes per patient detected by fTcSC and Lymphazurin was 1.7 and 1.1, respectively. Concordance of the primary sentinel nodes between fTcSC and Lymphazurin was 86%; the concordance of all sentinel nodes was 64%.

DISCUSSION

The molecular receptor-binding agent Lymphoseek demonstrated rapid and superior injection site clearance, equivalent primary sentinel node uptake, and fewer sentinel nodes per study when compared with fTcSC. The purpose of a phase I clinical trial[13] of a new medical imaging agent is an evaluation of its radiopharmacokinetics and a preliminary assessment of its biological safety. None of the 12 subjects exhibited an adverse effect or abnormal change in any laboratory parameter.

The faster injection site clearance of Lymphoseek produced a significant effect on the intraoperative detectability of the sentinel node. This is illustrated in Fig. 5, which plots the mean injection site clearance of Lymphoseek and fTcSC. At 5 hours after administration, there was a 3.8-fold difference in the target–injection site ratio. Because the sentinel node uptake for both agents is similar, the higher target–injection site ratio enhances the use of the intraoperative probe during studies, especially when the sentinel node is near the administration site. We expect this ratio to be even more pronounced if a

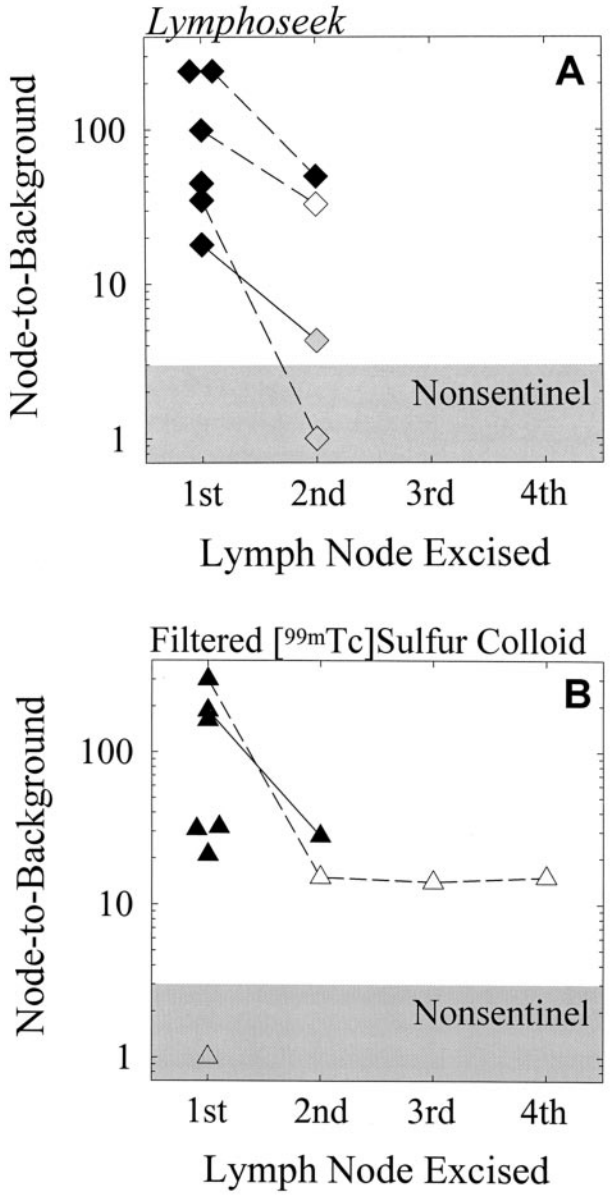

FIG. 4. Lymph node–background ratio of the lymph nodes excised after administration of (**A**) Lymphoseek and (**B**) filtered [^{99m}Tc]sulfur colloid. The open symbols indicate lymph nodes that were not stained blue. Nodes that did not qualify as sentinel have gray-filled symbols. Symbols connected by lines represent multiple nodes from the same subject.

2-day protocol is used. If the same Lymphoseek clearance rate is maintained beyond our initial observations, the percentage of injected dose in the breast would be approximately 2.5% at 15 hours after administration. If Lymphoseek and fTcSC maintain a sentinel node uptake of .5% at 15 hours, the injection site–sentinel node ratio for Lymphoseek would be 5 vs. 50 for the fTcSC. On the

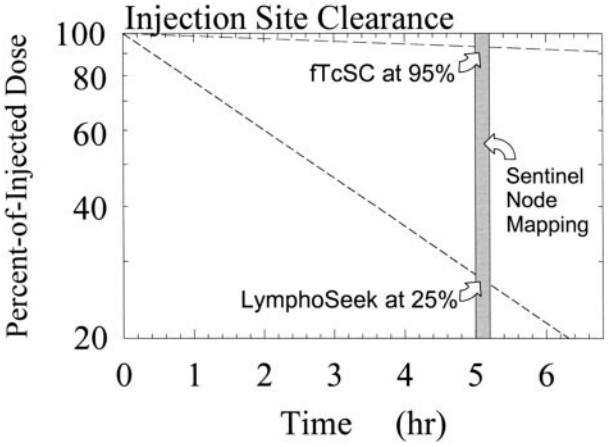

FIG. 5. The faster injection site clearance of Lymphoseek produces a 3.8-fold difference in the target–injection site ratio. Because the sentinel node uptake for both agents is similar, the higher target–injection site ratio enhances the use of the intraoperative probe during studies when the sentinel node is near the administration site. fTcSC, filtered [^{99m}Tc]sulfur colloid.

basis of the percentage of injected dose data of Gulec et al.,[14] we estimate the injection site–sentinel node ratio for unfiltered [^{99m}Tc]sulfur colloid to be approximately 2000.

The mean number of sentinel nodes per study detected by Lymphoseek (n = 1.3) was lower than the fTcSC value obtained in this study and significantly lower than values reported in the literature. Both fTcSC and unfiltered [^{99m}Tc]sulfur colloid yielded similar values for mean sentinel nodes per study. Using filtered colloid, McMasters et al.[15] reported mean values of 2.3, 2.5, and 2.6 for peritumoral, subdermal, and dermal injections, respectively. Using peritumoral injections of unfiltered colloid, Krag et al.[16] reported a mean of 2.4 sentinel nodes removed per study; of his group of 30 successful mapping studies, 13 subjects had ≥3 nodes excised. The lower value for the fTcSC studies of this work can be explained by our requirement that the preparation be administered within 1 hour of preparation. Commercial preparations of filtered sulfur colloid typically stand for 6 hours before use, during which time the percentage of colloid bound activity can decrease to 85% of total.[8]

Although the mean number of sentinel nodes for Lymphoseek was lower than for fTcSC, the difference was not statistically significant. A test of statistical significance will require a larger sample size. The sample size of this study was designed to test differences in injection site clearance rates, which is a pharmacokinetic measurement and the focus of a phase I radiopharmaceutical trial.

The value of 1.3 nodes per study may have multiple explanations. Radiopharmaceutical explanations of distal node uptake are (1) a 1 nmole injection is saturating all of the available receptor sites within the sentinel node or (2) the affinity of the radiopharmaceutical is not high enough for complete extraction by the receptors within the sentinel node. There are remedies for both conditions: (1) inject fewer molecules of the radiopharmaceutical and (2) increase the receptor affinity of the radiopharmaceutical by increasing the substrate density.[17] The radiochemical technology of this radiopharmaceutical will permit these solutions. An alternate explanation, independent of the radiopharmaceutical, is that three of the six subjects studies actually had two sentinel lymph nodes.

Our definition of sentinel node was based on Krag et al.,[16] who, using a 1-mCi dose of unfiltered [^{99m}Tc-]sulfur colloid, defined the sentinel node as any lymph node with radioactivity 3-fold greater than background and at least 150 counts per second. Our minimum value corrects for the lower administered activity used in this study. There are other operative definitions. Veronesi et al.,[18] using ^{99m}Tc-labeled colloidal human serum albumin, defined the sentinel node as the node with the highest radioactivity. Albertini et al.,[19] using fTcSC, defined the sentinel node as the lymph node with at least 10 times the radioactivity of the neighboring lymph nodes. Mariani et al.[7] recommend histopathology of any lymph node with counting rates at least 20% of the hottest node.

Although difficult to project, the cost per patient dose of Lymphoseek should equal that of a typical radiopharmaceutical. A patient dose of fTcSC costs approximately $150. A single sulfur colloid kit can provide four patient studies. The current Lymphoseek kit yields 10 patient studies. The reagents required for synthesis will not determine the cost of the radiopharmaceutical; 10 g of dextran, the major constituent of Lymphoseek, can provide more than a million patient doses.

CONCLUSION

The molecular receptor-binding agent Lymphoseek demonstrated rapid and superior injection site clearance, equivalent primary sentinel node uptake, and fewer sentinel nodes per study when compared with fTcSC.

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The acknowledgments are available online at www.annalssurgicaloncology.org.

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