

Intraoperative Frozen Section Analysis of Sentinel Lymph Nodes in Breast Cancer Patients

A Meta-Analysis and Single-Institution Experience

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BACKGROUND: Accurate intraoperative pathologic examination of sentinel lymph nodes (SLNs) has been an important tool that can reduce the need for reoperations in patients with SLN-positive breast cancer. The objective of the current study was to determine the accuracy of intraoperative frozen section (IFS) of SLNs during breast cancer surgery. **METHODS:** The authors retrospectively reviewed the records of 326 patients with breast cancer who underwent IF analysis of SLNs at a single institution. Then, they conducted a meta-analysis that included 47 published studies of IFS of SLNs in patients with breast cancer. **RESULTS:** Hematoxylin and eosin (H&E) staining revealed metastasis in SLNs in 99 patients (30.4%), including 61 patients with macrometastasis (MAM) (>2 mm) (the MAM group) and 38 patients with micrometastasis (Mi) or isolated tumor cell (ITC) deposits (the Mi/ITC group). The overall sensitivity of the institutional series was 60.6% (60 of 99 patients), and overall specificity was 100% (227 of 227 true negatives). The sensitivity of IFS was significantly lower in the Mi/ITC group (28.9%) than in the MAM group (80.3%; $P < .0001$). According to the meta-analysis of published studies and data from the author's institution (47 studies, for a total of 13,062 patients who underwent SLN dissection with IFS of SLNs), the mean sensitivity was 73%, and the mean specificity was 100%. The mean sensitivity was 94% for the MAM group and 40% for the Mi/ITC group. **CONCLUSIONS:** IFS of SLNs was more reliable for detecting MAM than for detecting Mi/ITC deposits. It lacked sufficient accuracy to rule out Mi/ITC deposits. *Cancer* 2011;117:250-8. © 2010 American Cancer Society.

KEYWORDS: breast cancer, sentinel lymph node, frozen section, meta-analysis, intraoperative evaluation.

Sentinel lymph node (SLN) dissection (SLND) has become a standard technique for accurately determining axillary lymph node status in patients with breast cancer,¹⁻⁴ in whom numerous studies have confirmed the high sensitivity and specificity of SLND.⁵⁻¹⁸ Several studies have reported that SLND significantly minimized morbidity and lymphedema compared with axillary lymph node dissection (ALND).¹⁹⁻²¹

Accurate SLN analysis requires coordinated efforts by a multidisciplinary team. If the SLN is identified as positive intraoperatively, then the patient may be a candidate for immediate ALND. Intraoperative pathologic examination of SLNs is helpful to avoid reoperations for SLN-positive patients, thus sparing them an additional operation with its risks and additional costs.

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Frozen section is the most common method of intraoperative evaluation, but no pathologic method has been standardized for SLN evaluation. Multilevel sectioning has been used for pathologic evaluation of SLNs; however, institutional protocols vary widely.^{22,23} The 2005 American Society of Clinical Oncology (ASCO) guidelines for the intraoperative evaluation of SLNs indicated that, although intraoperative frozen section (IFS) risks significant destruction of potentially diagnostic tissue, it may be the most desirable method for experienced teams of surgeons and pathologists.²⁴ The objective of the current study was to determine the accuracy of IFS of SLNs during breast cancer surgery.

MATERIALS AND METHODS

Patient Selection

In this retrospective study, we reviewed the records of 326 consecutive patients who with invasive breast cancer who underwent SLND with routine IFS at a single institution (the University of California at San Francisco [UCSF]) from November 1997 through April 2003. This study was approved by the institutional review board at UCSF. No patients in this series received neoadjuvant chemotherapy.

Preoperative Lymphoscintigraphy

The decision to use preoperative lymphoscintigraphy to identify the draining axillary SLNs was at the discretion of the operating surgeon. Technetium 99 sulfur colloid was injected either intradermally above the tumor, peritumorally, or at the surgical site of the previous biopsy. Patients underwent injection of radioisotope, with or without lymphoscintigraphy, either the morning of the surgery or in the late afternoon the day before surgery.

Intraoperative Identification of Sentinel Lymph Nodes

During the operation, a hand-held gamma probe (Neoprobe Corporation, Dublin, Ohio) was used to evaluate radioactive counts. Injection of Lymphazurin dye (Hirsch Industries, Richmond, Va) was at the surgeon's discretion.²⁵ An SLN was defined as a blue lymph node and/or a lymph node with an ex vivo radioactive count $\geq 10\%$ of the ex vivo radioactive count of the hottest lymph node. In addition, any clinically suspicious, palpable lymph nodes were removed and defined as SLNs.²⁶

Pathologic Evaluation

The radioactive count of all SLNs that were removed was measured and documented. All SLNs were submitted to

the pathology department and subjected to standard frozen section evaluation with 1 or 2 hematoxylin and eosin (H&E)-stained sections.

Tumors were classified according to American Joint Committee on Cancer (AJCC) using the sixth edition of the *AJCC Cancer Staging Manual*; permanent sections were stained with H&E with or without immunohistochemistry (IHC) and consisted of a bisection of ≥ 3 levels at intervals of 40 μm to 100 μm .^{27,28} All lymph node tissue specimens that were submitted for frozen section evaluation were resubmitted for permanent-section analysis (PSPA). Our protocol included 3 levels of IHC stains and 2 levels of H&E stains.^{22,23}

Literature Review

We systematically searched the PubMed database from January 1997 through June 2008 using the following keywords: "breast cancer," "breast neoplasm," "frozen section," "sentinel lymph node," and "SLN." References from included articles were searched for additional studies that met our inclusion criteria. We included all articles that reported sufficient data for cross-tabulation of the results of IFS of SLNs against PSPA. We excluded all articles with missing statistical data (ie, sensitivity, specificity). We also excluded all articles in languages other than English that were not available in translated form. All studies that involved neoadjuvant therapy were excluded.

The data were collected on a per-patient basis rather than per lymph node examined. The objective of our meta-analysis was to evaluate the accuracy of IFS evaluation.

Statistical Analysis

Sensitivity, specificity, and accuracy were calculated using the results from PSPA. Because the numbers of patients with micrometastases (Mi) and with isolated tumor cell (ITC) deposits were low, we combined them into the Mi/ITC group and compared their results with those from the macrometastases (MAM) group. We used Mantel-Haenszel and/or chi-square statistics to compare the sensitivity and accuracy for different tumor sizes and to compare the sensitivity for detecting MAM and Mi/ITC. *P* values $< .05$ were considered statistically significant.

Because different studies in a meta-analysis may have used different thresholds to define positive results, we used summary receiver operating characteristic (SROC) curve analysis to account for such differences in our meta-analysis.^{28,29} We also used a generalized linear

Table 1. Patient Characteristics (n=326)

Variable	No. of Patients (%)
Tumor classification	
T1a	31 (9.5)
T1b	83 (25.5)
T1c	151 (46.3)
T2	57 (17.5)
T3	4 (1.2)
Histologic findings	
Invasive ductal carcinoma	278 (85.3)
Invasive lobular carcinoma	29 (8.9)
Other ^a	19 (5.8)
Surgery	
Mastectomy	85 (26.1)
Lumpectomy	241 (73.9)

^aOther histologies included 10 tubular carcinomas, 5 mucinous carcinomas, 3 micropapillary carcinomas, and 1 medullary carcinoma.

mixed model to evaluate differences in diagnostic accuracy between patient groups or methods. For all of our statistical analyses, we used the statistical software package R (version 2.6.0; R Foundation for Statistical Computing, Vienna, Austria).³⁰

RESULTS

Patient Characteristics

The characteristics of all 326 patients in our single-institution study are summarized in Table 1. All patients were women, and their mean age at diagnosis was 55.7 years (range 23.5-88.5 years). The mean tumor size was 14.9 mm (range, 1.0-82 mm). SLND identified the SLNs in all patients. The mean number of SLNs removed was 3.2 per patient.

Sentinel Lymph Node Findings by Method and Tumor Size

Of the 326 patients who underwent IFS of SLNs, 99 patients (30.4%) had positive SLNs diagnosed by PSPA. Of those 99 patients, 39 patients (39.4%) had negative results according to the IFS. IFS yielded a sensitivity of 60.6%, a specificity of 100%, and an accuracy of 88%. The positive predictive value was 100%, and the negative predictive value was 83.2%. There were no false-positive IFS results.

Macrometastases Versus Micrometastases/Isolated Tumor Cell Deposits

Of the 99 SLN-positive patients, 61.6% had MAM (Table 2), and 38.4% had Mi/ITC (Table 3). The sensi-

tivity of IFS was significantly lower in the Mi/ITC group (28.9%) than in the MAM group (80.3%; $P < .0001$). Sensitivity was not correlated significantly with tumor size in either the MAM group ($P = .22$) or the Mi/ITC group ($P = .74$).

In the Mi/ITC group, 13 patients (34.2%) had metastasis identified in an SLN when PSPA was done using H&E staining, whereas 12 patients (31.6%) had metastasis identified in an SLN using IHC. The Mantel-Haenszel statistical method to control for tumor size indicated a significant difference in the sensitivity of IFS between the MAM group and the Mi/ITC group ($P < .00001$). In the Mi/ITC group, 8 patients had only ITC deposits in SLNs, and the sensitivity was 0% (0 of 8 patients).

Meta-Analysis

In total, 47 articles (including our own data) reported sufficient data for cross-tabulation of the results of IFS of SLNs against PSPA (Table 2).³¹⁻⁷⁶ Of those articles, 2 evaluated metastasis in IFS of SLNs using H&E staining and ultrarapid cytokeratin IHC.^{36,37} In those 2 studies, we included only the results from H&E staining, because few patients underwent ultrarapid IHC. Most IFS SLNs were stained with H&E or toluidine blue. One study reported 2 methods of intraoperative SLN evaluation.⁴¹

The total number of enrolled patients in the 47 studies was 13,062; 32% of those patients had positive SLNs, and 68% had negative SLNs. When we combined the data from all 47 studies in our meta-analysis, the overall sensitivity was 68%, and the overall accuracy was 90%.

Only 18 studies reported comparing IFS results between patients with MAM and patients with Mi (Table 3). The sensitivity in those 18 studies ranged from 80% to 100% for patients with MAM and from 11% to 100% for patients with Mi. Only 5 studies reported the results from IFS of ITC deposits (Table 4), and 3 of those studies reported 0% sensitivity for detecting ITC deposits.^{31,37,70}

According to our meta-analysis, the mean sensitivity for detecting metastasis using the SROC model was 73%, and the mean specificity was 100%. The mean sensitivity for detecting Mi, however, was substantially lower at 40%, whereas the mean sensitivity for detecting MAM was 94%. The mean specificity was 99% regardless of the type or size of metastasis. Frozen section was significantly more sensitive for MAM than for Mi in our meta-analysis ($P < .0001$).

We also analyzed the sectioning and staining methods of both IFS and PSPA. For the different intraoperative sectioning methods, we observed no significant

Table 2. Studies From the Sentinel Lymph Node Meta-Analysis

Study	No. of Patients	No. of True-Positive Findings	No. of False-Negative Findings	No. of False-Positive Findings	No. of True-Negative Findings	Sensitivity, %	Specificity, %	Accuracy, %
Veronesi 1997 ⁴⁰	107	32	18	0	57	64	100	83
Flett 1998 ³⁹	56	18	3	0	35	86	100	95
Hill 1999 ⁴²	405	68	10	0	327	87	100	98
Noguchi 1999 ⁴³	62	19	9	0	34	68	100	85
Turner 1999 ⁴⁴	278	53	58	0	167	48	100	79
Van Diest 1999 ³⁸	54	21	2	0	31	91	100	96
Morgan 1999 ⁴⁵	32	5	5	0	22	50	100	84
Rahusen 2000 ⁴⁶	100	21	16	0	63	57	100	84
Weiser 2000 ⁴⁷	890	135	96	1	658	58	99.8	89
Imoto 2000 ⁴⁸	52	17	2	0	33	89	100	96
Motomura 2000 ³⁵	101	13	12	0	76	52	100	88
Gemignani 2000 ⁴⁹	375	43	34	0	298	56	100	91
Liu 2000 ⁵⁰	38	11	6	0	21	65	100	84
Gulec 2001 ⁵¹	157	18	23	0	116	44	100	85
van der Loo 2001 ⁵²	275	64	13	0	198	83	100	95
Tanis 2001 ⁵³	265	71	25	1	168	74	99.4	90
Zurrida 2001 ⁵⁴	192	55	26	0	111	68	100	86
Chao 2001 ⁵⁵	203	36	17	1	149	68	99.3	91
Nagashima 2003 ⁵⁶	124	23	6	0	95	79	100	95
Leidenius 2003 ⁵⁷	375	116	23	2	234	83	99.2	93
Liang 2003 ⁵⁸	20	5	3	0	12	63	100	85
Menes 2003 ⁵⁹	67	21	12	0	34	64	100	82
Nahrig 2003 ³⁶	30	14	3	0	13	82	100	90
Krogerus 2004 ⁴¹	102	28	0	0	74	100	100	100
(Method A)								
Krogerus 2004 ⁴¹	102	36	6	0	60	86	100	94
(Method B)								
D'Errico 2004 ⁶⁰	68	14	1	0	53	93	100	99
Lauridsen 2004 ⁶¹	121	36	29	0	56	55	100	76
Holck 2004 ⁶²	263	75	28	0	160	73	100	89
Wada 2004 ⁶³	569	133	26	0	410	84	100	95
Aihara 2004 ⁶⁴	107	23	4	0	80	85	100	96
Reitsamer 2004 ⁶⁵	328	104	24	0	200	81	100	93
Khalifa 2004 ⁶⁶	96	18	3	0	75	86	100	97
Mitchell 2005 ⁶⁷	858	153	94	0	611	62	100	89
Hung 2005 ⁶⁸	105	34	16	1	54	68	98.2	84
Cao 2005 ⁶⁹	221	66	11	0	144	86	100	95
Perez 2005 ⁷⁰	87	25	7	0	55	78	100	92
Al-Shibli 2005 ⁷¹	70	15	4	0	51	79	100	94
Grabau 2005 ⁷²	272	61	23	1	187	73	99.5	91
Broqi 2005 ⁷³	133	29	21	0	83	58	100	84
Schrenk 2005 ⁷⁴	2326	513	282	0	1531	65	100	88
Celebioglu 2006 ⁷⁵	102	36	13	0	53	74	100	87
Choi 2006 ³⁷	79	14	6	0	59	70	100	92
Leung 2007 ⁷⁶	262	63	11	0	188	85	100	96
Langer 2009 ³¹	648	149	83	0	416	64	100	87
Arora 2008 ³²	327	78	30	1	218	63	99.5	90
McLaughlin 2008 ³³	931	170	136	0	625	56	100	85
Shimazu 2008 ³⁴	301	66	27	3	205	71	98.6	90
Current study	326	60	39	0	227	61	100	88
Total	13,062	2878	1346	11	8827	68	99.9	90

differences in diagnostic accuracy measured by SROC curves (data not shown). In contrast, for the different PSPA staining methods, the combination of H&E and IHC provided the best diagnostic accuracy according to the SROC curve analysis ($P < .004$).

DISCUSSION

Although an accurate method for analyzing SLNs intraoperatively is needed to prevent a second operation for completion ALND in patients with breast cancer who have lymph node-positive disease, no universally accepted

Table 3. Macrometastasis and Micrometastasis Results: Studies From the Sentinel Lymph Node Meta-Analysis

Study	No. of Patients	Micrometastases		No. of Patients	Macrometastases	
		No. of False-Negative Findings	Sensitivity, %		No. of False-Negative Findings	Sensitivity, %
Turner 1999 ⁴⁴	64	57	11	47	1	98
Rahusen 2000 ⁴⁶	18	13	28	19	3	84
Weiser 2000 ⁴⁷	115	87	24	109	9	92
Nagashima 2003 ⁵⁶	8	4	50	21	0	100
Leidenius 2003 ⁵⁷	45	17	62	94	6	94
Menes 2003 ⁵⁹	10	8	20	23	4	83
Nahrig 2003 ³⁶	6	2	67	5	0	100
Krogerus 2004 ⁴¹ (Method A)	7	0	100	21	0	100
D'Errico 2004 ⁶⁰	5	1	80	7	0	100
Lauridsen 2004 ⁶¹	30	22	27	35	7	80
Wada 2004 ⁶³	21	16	24	77	10	87
Perez 2005 ⁷⁰	3	1	67	23	0	100
Grabau 2005 ⁷²	28	20	29	56	3	95
Broqi 2005 ⁷³	25	20	20	25	1	96
Celebioglu 2006 ⁷⁵	20	13	35	27	0	100
Choi 2006 ³⁷	4	2	50	12	0	100
Langer 2009 ³¹	68	61	11	145	3	98
Shimazu 2008 ³⁴	32	22	31	61	5	92
Total	509	366	28	807	52	94

Table 4. Isolated Tumor Cell Results: Studies From the Sentinel Lymph Node Meta-Analysis

Study	No. of Patients		Sensitivity, %
	ITC Deposits	False-Negative Findings	
Nahrig 2003 ³⁶	6	1	83
D'Errico 2004 ⁶⁰	3	0	100
Perez 2005 ⁷⁰	6	6	0
Choi 2006 ³⁷	4	4	0
Langer 2009 ³¹	19	19	0
Total	38	30	21

ITC indicates isolated tumor cell; SLN, sentinel lymph node.

method exists. Many surgeons use IFS; however, its use is controversial because its sensitivity varies widely, as our study demonstrates.

We observed that IFS was reliable for detecting MAM and could help avoid reoperations in patients with breast cancer; however, IFS typically failed to detect Mi/ITC. Multiple published studies also indicated that IFS failed to detect Mi/ITC.^{40,46,47,55} In addition to its low sensitivity for detecting Mi/ITC, IFS has other potential disadvantages, including tissue consumption, artifacts from freezing and thawing of specimens,⁷⁷⁻⁷⁹ increased surgical time, and cost. The benefit of avoiding a second surgical procedure must be weighed against the risk of

obtaining a false-negative result, which could increase the risk of an inaccurate lymph node assessment. Despite these concerns, our meta-analysis validated the routine use of IFS given its outstanding sensitivity for MAM.

IFS reduces the need for secondary operation in patients who have been recommended to undergo completion axillary dissection for a positive SLN. Recently, results reported from the American College of Surgeons Clinical Oncology Group (ACOSOG) Z0011 trial indicated that there was no benefit from ALND for women who had clinically lymph node-negative breast cancer and a positive SLN.⁸⁰ Our findings support the routine clinical use of IFS of SLNs. However, we recognize that the appropriate use of IFS will continue to be an evolving issue given findings from the ACOSOG Z0011 trial. If those findings are validated and incorporated into clinical practice, then fewer completion ALNDs may be recommended for the very patients in whom IFS previously played an important role.

Recently, a commercially available, real-time, reverse-transcriptase polymerase chain reaction (the GeneSearch BLN assay; Veridex LLC, Warren, NJ) was used to detect SLN metastasis. Viale et al⁸¹ compared results from the BLN assay with results from serial IFS in a series of 293 SLNs from patients with breast cancer. The BLN assay correctly identified 51 of 52 SLNs with MAM and 5

of 20 SLNs with Mi; its sensitivity was 98.1% for detecting MAM (94.7% for MAM >1 mm and 77.8% for MAM >0.2 mm). Those authors concluded that the sensitivity of the BLN assay was comparable to that of histopathologic examination of the entire SLN by serial sectioning at 1.5 mm to 2 mm.⁸¹ Compared with IFS, the BLN assay for intraoperative SLN analysis yielded better results than those produced by our single-institution study and our meta-analysis.

The value of imprint cytology (IC) for the intraoperative analysis of SLNs is controversial. A meta-analysis of 31 studies that evaluated intraoperative IC for SLNs by Tew et al⁸² indicated that the pooled sensitivity of IC was 63%, and its specificity was 99%. Pooled sensitivity was 81% for MAM and 22% for Mi. The authors concluded that IC is simple and rapid and has good sensitivity for MAM but not for Mi. The sensitivity of frozen section for detecting MAM (94%) and Mi (40%) in our meta-analysis was better than the sensitivity for IC in published reports.^{35,82,83}

Some have argued that tissue loss is a potential concern of IFS. The term “false-negative rate” implies a difference between IFS and PSPA, because tissue loss cannot be measured exactly in IFS. Studies that have carefully and exhaustively sampled SLNs through multiple, deeper level sections have indicated that metastatic deposits may continue to be discovered throughout the lymph node.^{84,85} Tissue loss is 1 of the factors cited by various authors who argue against routine IFS analysis of SLNs.^{77,78} Some authors contend that grossly benign, diminutive SLNs should be sent for PSPA without IFS evaluation.⁷⁹ However, a metastatic deposit so small that it would be exhausted at the time of IFS indicates a very small lymph node tumor burden that would be questionable in terms of clinical significance.

In contrast, others would answer this concern regarding tissue loss as a theoretical source of false-negative results by suggesting that pathologists examine technical variables as a source of tissue loss. Some examples of such variables would be 1) the expertise of the technician performing sectioning; 2) the variability of fatty tissue present within and around the lymph node(s), which creates challenges in achieving suitable sections; and 3) the pathologist's emphasis (whether on preserving tissue for PSPA or on rendering a definitive diagnosis by performing multiple levels of IFS).

An important limitation of both our study and our meta-analysis was the lack of any standard for defining clinically suspicious lymph nodes in need of IFS. Specht

et al⁸⁶ defined clinically suspicious lymph nodes as firm, shotty, and more prominent than lymph nodes on the contralateral side. This lack of a standard definition for clinically suspicious lymph nodes, in addition to different pathologic techniques and variable institutional experience, may explain in part the wide range of sensitivity reported for IFS.

In most situations, IFS evaluation increases the efficiency of surgical treatment for patients with early stage breast cancer. However, in select situations, it is potentially problematic (fatty lymph nodes that are difficult to section, diminutive lymph nodes that may result in substantial tissue loss if subjected to IFS). IFS generally is helpful for avoiding reoperations for complete ALND in patients who undergo SLND; like all approaches, there are some potential drawbacks.

IFS is performed readily by most pathologists without additional equipment or specialized training in cytopathology. Our meta-analysis demonstrates that, although results vary between institutions, the overall sensitivity of IFS of SLNs is excellent for MAM at 94%. Currently, no intraoperative method of SLN analysis is considered sensitive enough to rule out Mi/ITC. IFS reduces the need for secondary operations, at least for most patients who have breast cancer with identifiable positive SLNs and unequivocal evidence of positive lymph node disease.

CONFLICT OF INTEREST DISCLOSURES

The authors made no disclosures.

REFERENCES

1. Schwartz GF, Giuliano AE, Veronesi U, et al. Proceedings of the consensus conference on the role of sentinel lymph node biopsy in carcinoma of the breast, April 19-22, 2001, Philadelphia, Pennsylvania. *Cancer*. 2002;94:2542-2551.
2. Edge SB, Niland JC, Bookman MA, et al. Emergence of sentinel node biopsy in breast cancer as standard-of-care in academic comprehensive cancer centers. *J Natl Cancer Inst*. 2003;95:1514-1521.
3. Giuliano AE. Sentinel node biopsy: standard of care. *Breast J*. 2003;9:S3-S6.
4. Singh-Ranger G, Mokbel K. The sentinel node biopsy is a new standard of care for patients with early breast cancer. *Int J Fertil Womens Med*. 2004;49:225-227.
5. Veronesi U, Paganelli G, Viale G, et al. A randomized comparison of sentinel-node biopsy with routine axillary dissection in breast cancer. *N Engl J Med*. 2003;349:546-553.
6. Krag D, Weaver D, Ashikaga T, et al. The sentinel node in breast cancer—a multicenter validation study. *N Engl J Med*. 1998;339:941-946.

7. Morton DL, Thompson JF, Essner R, et al. Validation of the accuracy of intraoperative lymphatic mapping and sentinel lymphadenectomy for early-stage melanoma: a multicenter trial—Multicenter Selective Lymphadenectomy Trial Group. *Ann Surg.* 1999;230:453-465.
8. Morton DL, Wen DR, Wong JH, et al. Technical details of intraoperative lymphatic mapping for early stage melanoma. *Arch Surg.* 1992;127:392-399.
9. Thompson JF, McCarthy WH, Bosch CM, et al. Sentinel lymph node status as an indicator of the presence of metastatic melanoma in regional lymph nodes. *Melanoma Res.* 1995;5:255-260.
10. Reintgen D, Cruse CW, Wells K, et al. The orderly progression of melanoma nodal metastases. *Ann Surg.* 1994;220:759-767.
11. Giuliano AE, Haigh PI, Brennan MB, et al. Prospective observational study of sentinel lymphadenectomy without further axillary dissection in patients with sentinel node-negative breast cancer. *J Clin Oncol.* 2000;18:2553-2559.
12. Schrenk P, Rieger R, Shamiyeh A, et al. Morbidity following sentinel lymph node biopsy versus axillary lymph node dissection for patients with breast carcinoma. *Cancer.* 2000;88:608-614.
13. Burak WE, Hollenbeck ST, Zervos EE, Hock KL, Kemp LC, Young DC. Sentinel lymph node biopsy results in less postoperative morbidity compared with axillary lymph node dissection for breast cancer. *Am J Surg.* 2002;183:23-27.
14. Temple LK, Baron R, Cody HS 3rd, et al. Sensory morbidity after sentinel lymph node biopsy and axillary dissection: a prospective study of 233 women. *Ann Surg Oncol.* 2002;9:654-662.
15. Haïd A, Kuehn T, Konstantiniuk P, et al. Shoulder-arm morbidity following axillary dissection and sentinel node only biopsy for breast cancer. *Eur J Surg Oncol.* 2002;28:705-710.
16. Swenson KK, Nissen MJ, Ceronisky C, Swenson L, Lee MW, Tuttle TM. Comparison of side effects between sentinel lymph node and axillary lymph node dissection for breast cancer. *Ann Surg Oncol.* 2002;9:745-753.
17. Golshan M, Martin WJ, Dowlathshahi K. Sentinel lymph node biopsy lowers the rate of lymphedema when compared with standard axillary lymph node dissection. *Am Surg.* 2003;69:209-212.
18. Schijven MP, Vingerhoets AJ, Rutten HJ, et al. Comparison of morbidity between axillary lymph node dissection and sentinel node biopsy. *Eur J Surg Oncol.* 2003;29:341-350.
19. Peintinger F, Reitsamer R, Stranzl H, Ralph G. Comparison of quality of life and arm complaints after axillary lymph node dissection vs sentinel lymph node biopsy in breast cancer patients. *Br J Cancer.* 2003;89:648-652.
20. Lucci A, McCall LM, Beitsch PD, et al. Surgical complications associated with sentinel lymph node dissection (SLND) plus axillary lymph node dissection compared with SLND alone in the American College of Surgeons Oncology Group Trial Z0011. *J Clin Oncol.* 2007;25:3657-3663.
21. Baron RH, Fey JV, Borgen PI, Stempel MM, Hardick KR, Van Zee KJ. Eighteen sensations after breast cancer surgery: a 5-year comparison of sentinel lymph node biopsy and axillary lymph node dissection. *Ann Surg Oncol.* 2007;14:1653-1661.
22. Treseler PA, Tauchi PS. Pathologic analysis of the sentinel lymph node. *Surg Clin North Am.* 2000;80:1695-1719.
23. Pargaonkar AS, Beissner RS, Snyder S, Speights VO Jr. Evaluation of immunohistochemistry and multiple-level sectioning in sentinel lymph nodes from patients with breast cancer. *Arch Pathol Lab Med.* 2003;127:701-705.
24. Lyman GH, Giuliano AE, Somerfield MR, et al. American Society of Clinical Oncology guideline recommendations for sentinel lymph node biopsy in early-stage breast cancer. *J Clin Oncol.* 2005;23:7703-7720.
25. Leong SP, Morita ET, Treseler PA, Wong JH. Multidisciplinary approach to selective sentinel lymph node mapping in breast cancer. *Breast Cancer.* 2000;7:105-113.
26. Leong SP, Steinmetz I, Habib FA, et al. Optimal selective sentinel lymph node dissection in primary malignant melanoma. *Arch Surg.* 1997;132:666-672; discussion 673.
27. Greene FL, Page DL, Fleming ID, et al, eds. *AJCC Cancer Staging Manual.* 6th ed. New York: Springer-Verlag; 2002.
28. Singletary SE, Connolly JL. Breast cancer staging: working with the sixth edition of the AJCC Cancer Staging Manual. *CA Cancer J Clin.* 2006;56:37-47.
29. McClish DK. Combining and comparing area estimates across studies or strata. *Med Decis Making.* 1992;12:274-279.
30. R Foundation for Statistical Computing. The R Project for Statistical Computing. Vienna, Austria: R Foundation for Statistical Computing. <http://www.Rproject.org>. Accessed December 6, 2008.
31. Langer I, Guller U, Berclaz G, et al; for the Swiss Multicenter Study Group Sentinel Lymph Node in Breast Cancer. Accuracy of frozen section of sentinel lymph nodes: a prospective analysis of 659 breast cancer patients of the Swiss multicenter study. *Breast Cancer Res Treat.* 2009;113:129-136.
32. Arora N, Martins D, Huston TL, et al. Sentinel node positivity rates with and without frozen section for breast cancer. *Ann Surg Oncol.* 2008;15:256-261.
33. McLaughlin SA, Ochoa-Frongia LM, Patil SM, Cody HS 3rd, Sclafani LM. Influence of frozen-section analysis of sentinel lymph node and lumpectomy margin status on reoperation rates in patients undergoing breast-conservation therapy. *J Am Coll Surg.* 2008;206:76-82.
34. Shimazu K, Tamaki Y, Taguchi T, Tsukamoto F, Kasugai T, Noguchi S. Intraoperative frozen section analysis of sentinel lymph node in breast cancer patients treated with neoadjuvant chemotherapy. *Ann Surg Oncol.* 2008;15:1717-1722.
35. Motomura K, Inaji H, Komoike Y, et al. Intraoperative sentinel lymph node examination by imprint cytology and frozen sectioning during breast surgery. *Br J Surg.* 2000;87:597-601.
36. Nahrig JM, Richter T, Kuhn W, et al. Intraoperative examination of sentinel lymph nodes by ultrarapid immunohistochemistry. *Breast J.* 2003;9:277-281.
37. Choi YJ, Yun HR, Yoo KE, et al. Intraoperative examination of sentinel lymph nodes by ultrarapid immunohistochemistry in breast cancer. *Jpn J Clin Oncol.* 2006;36:489-493.
38. Van Diest PJ, Torrens H, Borgstein PJ, et al. Reliability of intraoperative frozen section and imprint cytological investigation of sentinel lymph nodes in breast cancer. *Histopathology.* 1999;35:14-18.
39. Flett MM, Going JJ, Stanton PD, Cooke TG. Sentinel node localization in patients with breast cancer. *Br J Surg.* 1998;85:991-993.
40. Veronesi U, Paganelli G, Galimberti V, et al. Sentinel-node biopsy to avoid axillary dissection in breast cancer with clinically negative lymph-nodes. *Lancet.* 1997;349:1864-1867.
41. Krogerus LA, Leidenius MH, Toivonen TS, von Smitten KJ. Towards reasonable workload in diagnosis of sentinel

- lymph nodes: comparison of 2 frozen section methods. *Histopathology*. 2004;44:29-34.
42. Hill AD, Tran KN, Akhurst T, et al. Lessons learned from 500 cases of lymphatic mapping for breast cancer. *Ann Surg*. 1999;229:528-535.
 43. Noguchi M, Bando E, Tsugawa K, et al. Staging efficacy of breast cancer with sentinel lymphadenectomy. *Breast Cancer Res Treat*. 1999;57:221-229.
 44. Turner RR, Hansen NM, Stern SL, Giuliano AE. Intraoperative examination of the sentinel lymph node for breast carcinoma staging. *Am J Clin Pathol*. 1999;112:627-634.
 45. Morgan A, Howisey RL, Aldape HC, et al. Initial experience in a community hospital with sentinel lymph node mapping and biopsy for evaluation of axillary lymph node status in palpable invasive breast cancer. *J Surg Oncol*. 1999;72:24-30; discussion 30-31.
 46. Rahusen FD, Pijpers R, Van Diest PJ, Bleichrodt RP, Torrens H, Meijer S. The implementation of the sentinel node biopsy as a routine procedure for patients with breast cancer. *Surgery*. 2000;128:6-12.
 47. Weiser MR, Montgomery LL, Susnik B, Tan LK, Borgen PI, Cody HS. Is routine intraoperative frozen-section examination of sentinel lymph nodes in breast cancer worthwhile? *Ann Surg Oncol*. 2000;7:651-655.
 48. Imoto S, Fukukita H, Murakami K, Ikeda H, Moriyama N. Pilot study on sentinel node biopsy in breast cancer. *J Surg Oncol*. 2000;73:130-133.
 49. Gemignani ML, Cody HS 3rd, Fey JV, Tran KN, Venkatraman E, Borgen PI. Impact of sentinel lymph node mapping on relative charges in patients with early-stage breast cancer. *Ann Surg Oncol*. 2000;7:575-580.
 50. Liu LH, Siziopikou KP, Gabram S, McClatchey KD. Evaluation of axillary sentinel lymph node biopsy by immunohistochemistry and multilevel sectioning in patients with breast carcinoma. *Arch Pathol Lab Med*. 2000;124:1670-1673.
 51. Gulec SA, Su J, O'Leary JP, Stoller A. Clinical utility of frozen section in sentinel node biopsy in breast cancer. *Am Surg*. 2001;67:529-532.
 52. van der Loo EM, Sastrowijoto SH, Bril H, van Krimpen C, de Graaf PW, Eulderink F. Less operations required due to perioperative frozen section examination of sentinel nodes in 275 breast cancer patients. *Ned Tijdschr Geneesk*. 2001;145:1986-1991.
 53. Tanis PJ, Boom RP, Koops HS, et al. Frozen section investigation of the sentinel node in malignant melanoma and breast cancer. *Ann Surg Oncol*. 2001;8:222-226.
 54. Zurrida S, Mazzarol G, Galimberti V, et al. The problem of the accuracy of intraoperative examination of axillary sentinel nodes in breast cancer. *Ann Surg Oncol*. 2001;8:817-820.
 55. Chao C, Wong SL, Ackermann D, et al. Utility of intraoperative frozen section analysis of sentinel lymph nodes in breast cancer. *Am J Surg*. 2001;182:609-615.
 56. Nagashima T, Suzuki M, Yagata H, et al. Intraoperative cytologic diagnosis of sentinel node metastases in breast cancer. *Acta Cytol*. 2003;47:1028-1032.
 57. Leidenius MH, Krogerus LA, Toivonen TS, Von Smitten KJ. The feasibility of intraoperative diagnosis of sentinel lymph node metastases in breast cancer. *J Surg Oncol*. 2003;84:68-73.
 58. Liang R, Craik J, Juhasz ES, Harman CR. Imprint cytology versus frozen section: intraoperative analysis of sentinel lymph nodes in breast cancer. *ANZ J Surg*. 2003;73:597-599.
 59. Menes TS, Tartter PI, Mizrahi H, Smith SR, Estabrook A. Estabrook A. Touch preparation or frozen section for intraoperative detection of sentinel lymph node metastases from breast cancer. *Ann Surg Oncol*. 2003;10:1166-1170.
 60. D'Errico A, Grassigli A, Gruppioni E, et al. Thorough intraoperative analysis of breast sentinel lymph node biopsies: histologic and immunohistochemical findings. *Surgery*. 2004;135:248-254; discussion 255-257, 357.
 61. Lauridsen MC, Garne JP, Sorensen FB, Melsen F, Lernevall A, Christiansen P. Sentinel lymph node biopsy in breast cancer—experience with the combined use of dye and radioactive tracer at Aarhus University Hospital. *Acta Oncol*. 2004;43:20-26.
 62. Holck S, Galatius H, Engel U, Wagner F, Hoffmann J. False-negative frozen section of sentinel lymph node biopsy for breast cancer. *Breast*. 2004;13:42-48.
 63. Wada N, Imoto S, Hasebe T, Ochiai A, Ebihara S, Moriyama N. Evaluation of intraoperative frozen section diagnosis of sentinel lymph nodes in breast cancer. *Jpn J Clin Oncol*. 2004;34:113-117.
 64. Aihara T, Munakata S, Morino H, Takatsuka Y. Comparison of frozen section and touch imprint cytology for evaluation of sentinel lymph node metastasis in breast cancer. *Ann Surg Oncol*. 2004;11:747-750.
 65. Reitsamer R, Peintinger F, Prokop E, Rettenbacher L, Menzel C. 200 Sentinel lymph node biopsies without axillary lymph node dissection—no axillary recurrences after a 3-year follow-up. *Br J Cancer*. 2004;90:1551-1554.
 66. Khalifa K, Pereira B, Thomas VA, Mokbel K. The accuracy of intraoperative frozen section analysis of the sentinel lymph nodes during breast cancer surgery. *Int J Fertil Womens Med*. 2004;49:208-211.
 67. Mitchell ML. Frozen section diagnosis for axillary sentinel lymph nodes: the first 6 years. *Mod Pathol*. 2005;18:58-61.
 68. Hung WK, Chan CM, Ying M, Chong SF, Mak KL, Yip AW. Randomized clinical trial comparing blue dye with combined dye and isotope for sentinel lymph node biopsy in breast cancer. *Br J Surg*. 2005;92:1494-1497.
 69. Cao Y, Paner GP, Rajan PB. Sentinel node status and tumor characteristics: a study of 234 invasive breast carcinomas. *Arch Pathol Lab Med*. 2005;129:82-84.
 70. Perez N, Vidal-Sicart S, Zanon G, et al. A practical approach to intraoperative evaluation of sentinel lymph node biopsy in breast carcinoma and review of the current methods. *Ann Surg Oncol*. 2005;12:313-321.
 71. Al-Shibli KI, Mohammed HA, Mikalsen KS. Sentinel lymph nodes and breast carcinoma: analysis of 70 cases by frozen section. *Ann Saudi Med*. 2005;25:111-114.
 72. Grabau DA, Rank F, Friis E. Intraoperative frozen section examination of axillary sentinel lymph nodes in breast cancer. *APMIS*. 2005;113:7-12.
 73. Brogi E, Torres-Matundan E, Tan LK, et al. The results of frozen section, touch preparation, and cytological smear are comparable for intraoperative examination of sentinel lymph nodes: a study in 133 breast cancer patients. *Ann Surg Oncol*. 2005;12:173-180.
 74. Schrenk P, Konstantiniuk P, Wolf S, et al. Intraoperative frozen section examination of the sentinel lymph node in breast cancer. *Rozhl Chir*. 2005;84:217-222.
 75. Celebioglu F, Sylvan M, Perbeck L, Bergkvist L, Frisell J. Intraoperative sentinel lymph node examination by frozen section, immunohistochemistry and imprint cytology during breast surgery—a prospective study. *Eur J Cancer*. 2006;42:617-620.

76. Leung KM, Chan KW, Yeoh GP, Chan JK, Cheung PS. Clinical relevance of intraoperative sentinel lymph node examination in breast cancer management. *Hong Kong Med J*. 2007;13:8-11.
77. Anderson TJ. The challenge of sentinel lymph node biopsy. *Histopathology*. 1999;35:82-84.
78. Pfeifer JD. Sentinel lymph node biopsy. *Am J Clin Pathol*. 1999;112:599-602.
79. Patrick T. Pathologic examination of the sentinel lymph node: what is the best method? *Breast J*. 2006;12:S143-S151.
80. Giuliano AE, McCall LM, Beitsch PD, et al. ACOSOG Z0011: a randomized trial of axillary node dissection in women with clinical T1-2 N0 M0 breast cancer who have a positive sentinel node [abstract]. Paper presented at: American Society of Clinical Oncology Annual Meeting; June 4-8, 2010; Chicago, Ill. Abstract CRA506.
81. Viale G, Dell'orto P, Biasi MO, et al. Comparative evaluation of an extensive histopathologic examination and a real-time reverse-transcription-polymerase chain reaction assay for mammaglobin and cytokeratin 19 on axillary sentinel lymph nodes of breast carcinoma patients. *Ann Surg*. 2008;247:136-142.
82. Tew K, Irwig L, Matthews A, Crowe P, Macaskill P. Meta-analysis of sentinel node imprint cytology in breast cancer. *Br J Surg*. 2005;92:1068-1080.
83. Llatjos M, Castella E, Fraile M, et al. Intraoperative assessment of sentinel lymph nodes in patients with breast carcinoma: accuracy of rapid imprint cytology compared with definitive histologic workup. *Cancer*. 2002;96:150-156.
84. Viale G, Bosari S, Mazzarol G, et al. Intraoperative examination of axillary sentinel lymph nodes in breast carcinoma patients. *Cancer*. 1999;85:2433-2438.
85. Cserni G. Metastases in axillary sentinel lymph nodes in breast cancer as detected by intensive histopathological work up. *J Clin Pathol*. 1999;52:922-924.
86. Specht MC, Fey JV, Borgen PI, Cody HS 3rd. Is the clinically positive axilla in breast cancer really a contraindication to sentinel lymph node biopsy? *J Am Coll Surg*. 2005;200:10-14.