Quantifying the Extent of Invasive Carcinoma and Margin Status in Partial Mastectomy Cases Having a Gross Lesion

Is a Defined Tissue Processing Protocol Needed?

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Key Words: Invasive carcinoma; Margin status; Partial mastectomy; Processing protocol; Breast carcinoma; Ductal carcinoma; Lobular carcinoma

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Abstract

Accurate estimation of disease extent and margin status is critical when evaluating partial mastectomy cases because both are predictors of recurrence. No published standards exist for processing specimens involved by invasive carcinoma, presumably because such cases have a gross lesion. We retrospectively studied 100 partial mastectomy cases and concluded that a standardized tissue mapping protocol is needed to ensure adequate pathologic examination even when a gross lesion is present. When mapped and unmapped findings were compared, 17 cases (10 with ductal and 7 with lobular carcinoma) had an increase in carcinoma size, 12 cases (9 with ductal and 3 with lobular carcinoma) had an increase in pathologic T stage, and positive margins were found in 8 cases (7 with ductal and 1 with lobular carcinoma). We describe our tissue-mapping protocol, and advocate its use as a standardized protocol for processing all partial mastectomy specimens.

Breast-conservation therapy is an acceptable option for the treatment of invasive and in situ breast carcinoma. Accurate estimation of disease extent and margin status are critical when evaluating partial mastectomy specimens because both are predictors of prognosis. Relative to ductal carcinoma in situ (DCIS), precise measurement of disease extent is challenging because DCIS involves the 3-dimensional ductal system of the breast, resulting in discontinuous areas of involvement; it often occurs with no gross lesion; and it can exist without associated calcification.1-4

Although not specifically required for assigning a pathologic stage in DCIS, the extent of disease is pivotal in patient management.5,6 Disease extent correlates with residual disease in subsequent reexcisions7-11 and risk of occult invasion and multicentricity.12 Furthermore, margin status is directly linked to risk of recurrence.9,25

Lack of standardization in specimen processing increases discrepancies in DCIS measurement and margin assessment. As such, the College of American Pathologists (CAP) has published a protocol for the examination of specimens with DCIS.5 No published standards exist for processing specimens involved by invasive carcinoma, presumably because such cases often have a grossly identifiable lesion that directs the extent of sampling. To demonstrate the need for a standardized processing protocol for partial mastectomy specimens having a gross lesion containing invasive carcinoma, we retrospectively studied 100 cases. We compared tumor size, pathologic T stage, and margin status using 2 different sampling techniques: tissue mapping vs a more focused and accepted process involving submission of grossly lesional tissue and adjacent stroma. In this report, we describe a protocol of tissue mapping for
invasive carcinoma similar to that already reported in the literature for DCIS\textsuperscript{26,27} and discuss its impact on determination of disease extent and margin status.

**Materials and Methods**

This study was approved by the institutional review board at the University of Tennessee Graduate School of Medicine, Knoxville, and a waiver of informed consent was granted. Signed out partial mastectomy cases containing a grossly identifiable lesion composed of invasive carcinoma were retrieved from our files at the University of Tennessee Medical Center at Knoxville Department of Pathology between January 2007 and April 2010. A grossly identifiable lesion for purposes of this study was defined as a clearly visible mass lesion from which a size measurement can be obtained and distance to closest surgical margins can be measured. Final reports for all potential study cases were reviewed. Descriptions of gross lesions, including their size, distance from margins, and location within the specimen, were clearly documented in the gross description of each study case. Location of unremarkable fibrous stroma relative to gross lesions was also described. The gross description of each study case also contained a detailed slide key and/or block diagram detailing the location of sections taken from unremarkable fibrous stroma and gross lesions. We selected 100 consecutive cases meeting these criteria for study, and the slides were reviewed. Cases not having the necessary components in the gross description and a detailed slide key and/or specimen block diagram were excluded from the study because precise tissue mapping and location of sections within the specimen could not be confirmed.

All partial mastectomy specimens in this study were received for gross intraoperative evaluation for margins, and specimen radiographs were performed on all cases containing a hookwire to confirm its extirpation. None of the study cases had reexcision based on the gross intraoperative findings.

These partial mastectomy specimens were processed per standard department protocol, which includes a mapping process similar to that described in the literature to confirm disease extent in cases of DCIS\textsuperscript{4,26,27} The specimen is differentially inked to delineate surgeon orientation, measured in 3 dimensions, and sequentially sectioned into slices, and slides are prepared from gross lesions and all fibrous stroma to facilitate microscopic reconstruction of the breast tissue \textsuperscript{Image 1}. Location of sections is documented in a detailed slide key and, in some cases, a specimen block diagram that accompanies the gross description.

We do not routinely submit unremarkable fat because restricting microscopic evaluation to fibrous stroma and lesional tissue optimizes the number of blocks relative to the probability of detecting significant disease, as described by other authors\textsuperscript{28} The specimen is entirely submitted if this can be accomplished in fewer than 25 to 30 blocks, as recommended by other authors.\textsuperscript{3}

All grossly detected lesions are described, measured, and documented in the gross description. Microscopic disease extent in 1 dimension is determined by reapproximation of involved slices by using calculation of average slice thickness. Microscopic disease extent in the remaining 2 dimensions is established by reapproximation of slides as puzzle pieces by using the slide key and specimen diagram \textsuperscript{Image 2}. Microscopic extent of disease is compared with gross findings to

\textsuperscript{Image 1} Each specimen is differentially inked to maintain orientation.

\textsuperscript{Image 2} Slides are reapproximated like puzzle pieces to measure the microscopic extent of disease.
confirm pathologic T stage and margin status. This information is used to construct the diagnosis as it appears in the signed out report.

For purposes of comparison, the slide key and specimen block diagram were used to segregate slides obtained from grossly lesional and adjacent tissue, including margins, to study differences between mapping and a more focused and widely accepted sampling technique described in surgical pathology textbooks for processing partial mastectomy specimens having a gross lesion. In this study, the more focused sampling approach is referred to as unmapped and is defined as slides obtained from the grossly identified lesion, the fibrous stroma surrounding the lesion, and all adjacent margins, accounting for a radius of at least 2 cm around the tumor. Tumor size, pathologic T stage, and margin status obtained from unmapped tissue were compared with the same parameters from mapped tissue as documented in the final report of each study case. The differences between the 2 sampling techniques were documented.

Results

Patients ranged in age from 38 to 91 years. Partial mastectomy specimen size ranged from 3.7 to 15.5 cm, and 10 to 76 blocks were submitted to map each case as described in the “Materials and Methods” section (average of 25 blocks). For the study, 7 to 20 slides were segregated from study cases to fulfill criteria for unmapped tissue (average of 10 slides). Study results are summarized in Table 1.

The majority of cases had a diagnosis of ductal carcinoma (93) with fewer cases of lobular carcinoma (7). In 17 cases, there was a change in the size of carcinoma, and 12 of these cases also had an increase in the pathologic T stage as a result of mapping Table 2. The average grossly described tumor size among these 17 cases was 1.38 cm, and the average size following mapping was 3.39 cm. Of the 7 cases with lobular carcinoma, 4 had a change in size owing to mapping, and 3 of them also had an increase in pathologic T stage.

In 5 total study cases, there was a history of preoperative chemotherapy, and 3 of these cases had an increase in size and pathologic T stage. In 5 cases of ductal carcinoma and 2 cases of lobular carcinoma with an increase in pathologic T stage, there also was a change in margin status in mapped vs unmapped tissue. In 4 cases with ductal carcinoma (including 1 case that also had an increase in tumor size), additional separate tumor foci were identified as a result of tissue mapping that were not identified grossly. These additional tumor foci measured between 0.2 and 0.7 cm and were up to 5 cm away from the grossly described tumor. One additional tumor focus involved an inferior margin.

A difference in margin status in mapped vs unmapped tissue was identified in 8 cases Table 3. In 5 of these cases, there were grossly negative margins greater than 1 cm. In 3 cases, close margins were described in the gross description.

Table 2 Cases With Differences in Tumor Size in Slides From Lesional vs Mapped Tissue

<table>
<thead>
<tr>
<th>Tumor Classification</th>
<th>Gross Tumor Size (cm)</th>
<th>Following Mapping Tumor Size (cm)</th>
<th>Change in Pathologic T Stage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ductal carcinoma</td>
<td>1.0</td>
<td>1.7</td>
<td>No</td>
</tr>
<tr>
<td>Ductal carcinoma</td>
<td>2.0</td>
<td>2.9</td>
<td>No</td>
</tr>
<tr>
<td>Ductal carcinoma</td>
<td>3.5</td>
<td>4.0</td>
<td>No</td>
</tr>
<tr>
<td>Ductal carcinoma</td>
<td>1.2</td>
<td>1.5</td>
<td>No</td>
</tr>
<tr>
<td>Lobular carcinoma</td>
<td>2.6</td>
<td>3.5</td>
<td>No</td>
</tr>
<tr>
<td>Ductal carcinoma</td>
<td>1.2</td>
<td>3.5</td>
<td>Yes (1c→2)</td>
</tr>
<tr>
<td>Ductal carcinoma*</td>
<td>1.2</td>
<td>3.8</td>
<td>Yes (1c→2)</td>
</tr>
<tr>
<td>Lobular carcinoma</td>
<td>1.5</td>
<td>6.0</td>
<td>Yes (1c→3)</td>
</tr>
<tr>
<td>Lobular carcinoma</td>
<td>1.8</td>
<td>4.2</td>
<td>Yes (1c→2)</td>
</tr>
<tr>
<td>Ductal carcinoma*</td>
<td>1.0</td>
<td>3.5</td>
<td>Yes (1c→2)</td>
</tr>
<tr>
<td>Ductal carcinoma</td>
<td>1.0</td>
<td>2.5</td>
<td>Yes (1c→2)</td>
</tr>
<tr>
<td>Ductal carcinoma†</td>
<td>1.4</td>
<td>3.5</td>
<td>Yes (1c→2)</td>
</tr>
<tr>
<td>Ductal carcinoma*</td>
<td>0.6</td>
<td>3.5</td>
<td>Yes (1b→2)</td>
</tr>
<tr>
<td>Lobular carcinoma</td>
<td>1.5</td>
<td>8.0</td>
<td>Yes (1c→3)</td>
</tr>
<tr>
<td>Ductal carcinoma</td>
<td>0.8</td>
<td>1.5</td>
<td>Yes (1b→1c)</td>
</tr>
<tr>
<td>Ductal carcinoma†</td>
<td>0.4</td>
<td>0.9</td>
<td>Yes (1a→1b)</td>
</tr>
<tr>
<td>Ductal carcinoma*</td>
<td>0.8</td>
<td>3.0</td>
<td>Yes (1b→2)</td>
</tr>
<tr>
<td>Average tumor size</td>
<td>1.38</td>
<td>3.39</td>
<td>—</td>
</tr>
</tbody>
</table>

* Received preoperative chemotherapy.
† With extensive intraductal component.
‡ 2 cases also had a change in margin status.
§ 3 cases also had a change in margin status.

Table 3 Summary of Findings Comparing Slides From Grossly Lesional Tissue With Mapped Tissue

<table>
<thead>
<tr>
<th>Change in Size of Carcinoma</th>
<th>Change in Pathologic T Stage</th>
<th>Change in Margin Status</th>
<th>Additional Tumor Identified</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ductal (n = 88)</td>
<td>11 7*</td>
<td>3†</td>
<td>4</td>
</tr>
<tr>
<td>Lobular (n = 7)</td>
<td>4 3†</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Ductal with extensive intraductal component (n = 5)</td>
<td>2 2†</td>
<td>3</td>
<td>0</td>
</tr>
<tr>
<td>Total (n = 100)</td>
<td>17 12</td>
<td>8</td>
<td>4</td>
</tr>
</tbody>
</table>

* 3 cases also had a change in margin status.
† Margin positive in 1 case owing to additional tumor identified.
‡ 2 cases also had a change in margin status.
§ 3 cases also had a change in margin status.
but additional positive margins not identified grossly were found only through mapping. One case had a positive margin owing to a separate focus of tumor, and 2 cases had a positive margin owing to DCIS. Reexcision was performed in 6 of 7 cases, and 4 of these cases had residual DCIS in the reexcision specimen.

**Discussion**

We describe a tissue mapping protocol and advocate its use as a standardized method for processing all partial mastectomy cases to ensure accurate pathologic staging and margin assessment for ductal and lobular carcinoma. When mapped and unmapped findings were compared, 17 cases (10 with ductal and 7 with lobular carcinoma) had an increase in carcinoma size, 12 cases (9 with ductal and 3 with lobular carcinoma) had an increase in pathologic T stage, and positive margins were found in 8 cases (7 with ductal and 1 with lobular carcinoma).

Many breast sampling methods are described in the literature. Focused histologic sampling of a gross lesion and its surrounding stroma is considered an acceptable processing method for partial mastectomy specimens and is described in standard surgical pathology textbooks. The block multiplier method entails multiplying the number of involved blocks by a multiplier of 0.3 to 0.5 to estimate disease extent. This method is prone to size underestimation, particularly as the number of involved blocks increases. The single slide method for measurement can be used when 1 block is involved. Submitting the entire specimen through a serial sectioning technique provides maximum detection but is often not cost-effective relative to time spent in interpretation and preparation. In their study of grossly benign breast biopsies, Schnitt and Wang found that it is extremely unlikely to find carcinoma in fatty tissue, and they recommend submission of fibrous stroma only for maximum cost-effective lesion detection. The CAP protocol for the examination of specimens for DCIS recommends complete submission of smaller specimens and selective sampling of larger specimens to include the lesion and fibrous stroma. This CAP protocol does not make sampling recommendations for breast resections performed for invasive carcinoma with a mass lesion.

Our study confirms that even when a gross lesion is present, accurate size measurement can be difficult in cases of lobular carcinoma. Of our 7 lobular carcinoma cases, 6 had an increase in size, and 3 of these cases also had an increase in the pathologic T stage as a result of mapping. Accurate gross measurement of lobular carcinoma is difficult, likely owing to the lack of a desmoplastic stromal reaction and its growth pattern of satellite small foci in adjacent tissue. Microscopically, tumor was present in each breast tissue slice as individual tumor cells in fat and fibrous stroma with no desmoplasia, causing a significant discrepancy between the gross and microscopic extent of disease.
that results in a larger size than suspected grossly, necessitating systematic sequential submission identical to the mapping protocol we describe.

Unlike lobular carcinoma, ductal carcinoma typically forms a discrete mass with correlation between the gross and microscopic tumor sizes. A focused sampling method that includes submission of the lesion, adjacent fibrous stroma, and all close margins is sufficient for accurately assigning pathologic stage and margin status. \(^{29,30}\) Of our 17 study cases having an increase in tumor size following mapping, 13 had ductal carcinoma, and 9 cases also had an increase in pathologic T stage. In all of these cases, tumor extended past the grossly identified mass as individual tumor cell clusters in small fibrous tissue septa \(\text{Image 5 and Image 6}\). In 4 study cases with ductal carcinoma, there were separate tumor foci that were identified in grossly unremarkable fibrous stroma, including 1 focus 5 cm away from the main tumor. The separate tumor foci measured between 0.2 and 0.7 cm, and 1 focus involved an inferior margin. These results indicate that a focused sampling technique is inadequate for even ductal carcinoma because of the potential of understaging and missing separate tumor foci. Even if stroma 2 to 3 cm around the tumor is sampled, separate tumor foci can still be missed, as evidenced by our finding a separate tumor focus 5 cm away from the grossly identified lesion, present within grossly unremarkable fibrous stroma.

In our study, we also observed that gross assessment of tumor size and margin status is imprecise in cases of ductal carcinoma after preoperative chemotherapy. In 5 of our study cases with ductal carcinoma, preoperative chemotherapy was given, and 3 of these cases had an increase in tumor size and pathologic T stage as a result of mapping. Residual tumor in these cases had a discontiguous growth pattern, including small islands of tumor cells within minimally fibrotic stroma, a growth pattern similar to that observed microscopically in lobular carcinoma \(\text{Image 7 and Image 8}\). It is this growth pattern that likely accounts for the discrepancy between gross and mapped tumor size in our cases of ductal carcinoma after
preoperative chemotherapy. As such, sequential submission is needed in these cases to ensure accurate assignment of pathologic stage and margin status.

In 8 cases, a change in margin status was found following mapping, including 7 cases of ductal carcinoma and 1 case of lobular carcinoma. Margins noted to be microscopically positive were grossly negative at greater than 1 cm. Microscopically positive margins were characterized by small tumor cell clusters tracking along fibrous tissue septa. If focused sampling techniques are used, microscopically positive margins can be missed, thus increasing the patient’s risk of local recurrence.

Our study confirms that restricting histologic sampling to grossly abnormal and surrounding stroma results in overlooking separate tumor foci and inaccurate pathologic staging and margin assessment in a significant number of cases. Two of our observations explain these findings. First, the lesion’s growth pattern, not histologic subtype, determines its likelihood of having a discrepancy between gross and microscopic findings. Cancers having small tumor clusters growing along fibrous tissue septa are likely to have a larger microscopic extent than can be identified grossly. This growth pattern is also a predisposing factor to inaccurate gross margin assessment. Second, separate small tumor foci are often too small to be detected grossly or by preoperative imaging and can be found only by systematic and thorough fibrous stroma sampling.

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References


