Fig 5. Long-term anterior view of the breast after breast conserving surgery.

Irradiation Group and Non-irradiation Group

Irradiation of the conserved breast was performed for 165 patients. We avoided irradiation for 101 of patients at low risk of local recurrence. Low risk group was defined as pathologically n0, ly (− or 1+), without an extended intra-ductal component, with negative margins (cancer free within 5 mm of the surgical margin), without multi-centric lesions and without a contra-lateral lesion.

Incidence of Local Recurrence

We performed breast conserving surgery for 266 patients from Sep. 1990 to Dec. 2003. Fifteen patients (5.6%) had local relapse. The median observation period was 72 months. Irradation of the conserved breast was performed for 165 patients. We avoided irradiation for 101 low risk patients as noted above. In the non-irradiated patients the incidence of local relapse was 6.9%. The incidence of local relapse in the group undergoing irradiation was 4.8%. There was no statistically significant difference between irradiated and non-irradiated cases.

Cosmesis

Fig 5 shows the cosmetic features of BCT with volume replacement using LTF for the right breast. There is no conspicuous difference between either the breast. The operation scar is not seen from the front.

Discussion

We started breast conserving surgery in 1989. One of the advantages of using LTF for volume replacement is long-term maintenance of the graft. LTF does not shrink like a musculo-cutaneous flap (MC flap) using the latissimus dorsi, which shrinks because of disuse atrophy.

Volume replacement with LTF is a minimally invasive breast reconstruction technique compared with breast reconstruction with a saline bag.

The incidence of local recurrence is not inferior to that of ordinary breast conserving surgery. In our department, local recurrence is found in early stages without distant metastasis in almost all cases by careful and frequent follow up by mammography and ultrasonography. For most patients, local recurrence occurs within 5 years, we perform ultrasonography every 3-6 months for 5 years. Annual examination by mammography is also performed. The incidence of local recurrence is 5.6%. This is not inferior to the incidence reported in western countries[1, 6-9]. This is due to volume resection of the mammary gland sufficient to achieve negative margins and appropriate irradiation after estimation of risk. As Voogd *et al.* reported that an age younger than 35 years, extended intraductal component, and vascular invasion are the factors that contribute to local recurrence[10], our data suggests that appropriate risk evaluation can avoid irradiation in the low-risk group. On the other hand, local recurrence in high-risk cases with is well controlled by irradiation. We were able to reduce the rate of local relapse to as low as 4.8%. Recently, we estimated tumor extension by CT and MRI[3, 4]. When there is very limited extent of tumor, we employ partial mastectomy. These modalities also assist us in improving negative margin rates.

With breast conserving surgery followed by volume replacement with LTF, blood flow of the LTF should be well maintained. We preserve communicating branches from the latissimus dorsi and lateral thoracic vessels when we prepare the LTF. Careful lymph node dissection should be done with complete removal of lymph nodes and adipose tissue around the lateral thoracic vessels when we preserve lateral thoracic vessels so as to completely remove cancer cells (Fig 3).

The long term volume stability of the LTF is good, because adipose tissue does not shrink due

to disuse atrophy.

Breast conserving surgery with primary volume replacement with LTF has advantages because, 1) patients can avoid foreign prosthesis like silicone bags, 2) LTF maintains its volume for a long period and 3) patients can avoid poly-surgery. The disadvantage of volume replacement with LTF is that the thickness of the LTF depends on the thickness of subcutaneous adipose tissue. It does not always meet the demand for the thickness of breast tissue. In cases with thin LTF for volume replacement, the central area of the breast under the nipple should be covered by existing mammary gland to maintain thickness with a partial suture and the peripheral area would be covered by LTF. Using this method, the difference in volume between the breasts is not conspicuous.

In conclusion, volume replacement with LTF is a reasonable and useful method to achieve both local control and good cosmesis in primary surgical breast cancer treatment.

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Case Reports

MRI Accurately Depicts Underlying DCIS in a Patient with Paget's Disease of the Breast Without Palpable Mass and Mammography Findings

Goro Amano[1,3], Mioko Yajima[2], Yasunori Moroboshi[1], Yoshiki Kuriya[1] and Noriaki Ohuchi[3]

[1]Department of Surgery and [2]Department of Pathology, Sakata Municipal Hospital, Sakata, Yamagata and [3]Department of Surgical Oncology, Tohoku University School of Medicine, Sendai, Japan

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Breast-conserving therapy must be carefully indicated among patients with Paget's disease of the breast, because the disease is often associated with an underlying *in situ* or invasive carcinoma, even when there are no palpable mass or mammography findings. We report a 52-year-old woman who complained of skin color change of her right nipple for 11 months. No mass was palpable in her breasts, and mammography did not show any density or calcification. Nipple biopsy revealed Paget's disease of the breast with ductal carcinoma *in situ* (DCIS) in the breast epithelium just beneath the nipple. Magnetic resonance imaging (MRI) of the breast demonstrated diffuse segmental enhancement in two different quadrants. According to the pattern of enhancement, the lesions depicted by MRI were diagnosed as an extensively spreading type of DCIS. Based on informed consent, the patient received a total mastectomy. The histopathological examination demonstrated non-invasive ductal carcinoma with comedo-necrosis. The histological mapping with subserial sectioning demonstrated an extent of the lesions that corresponded accurately to the lesions defined by MRI. We conclude that MRI may play an important role in selecting candidates for breast-conserving therapy out of those patients with mammary Paget's disease with no clinical evidence of an underlying breast carcinoma.

Key words: Paget's disease – breast carcinoma – MRI – ductal carcinoma in situ

INTRODUCTION

The treatment for patients with Paget's disease of the breast is controversial. The standard treatment has been mastectomy (1,2). However, some studies have proposed the use of breast-conserving therapy for patients with Paget's disease in whom an underlying breast cancer cannot be located (3,4).

Nevertheless, other investigators reported that wide local excision of the nipple–areola complex and underlying breast tissue (cone excision) would have been insufficient surgery in 40% of their cases with no palpable mass and a normal mammogram, because of the multicentricity of the disease (5). Therefore, candidates for breast-conserving therapy must be selected carefully on an individual basis among those patients who have Paget's disease of the breast (6–8).

Here we report a case of a mammary Paget's disease patient who did not present either palpable mass or mammography findings. Magnetic resonance imaging (MRI) of the breast was very useful for making a decision on the appropriate surgical procedure.

CASE REPORT

A 52-year-old woman visited our hospital in January 2004 for an 11 month history of skin color change of her right nipple. She had also worried about the exudates from the nipple for 3 months. Physical examination showed the flattening and scaling of the nipple (Fig. 1). No mass was palpable in her breasts, and mammography did not reveal any density or calcification. There was also no abnormal finding on ultrasonography. Exfoliative cytology of the nipple demonstrated Paget's cells. To determine the surgical procedure of choice, MRI of the right breast was investigated. There were diffuse segmental enhancements existing in the upper and lateral quadrants (Fig. 2). According to the pattern of enhancement,

For reprints and all correspondence: Goro Amano, Department of Surgery, Sakata Municipal Hospital, 2-3-20, Sengoku-cho, Sakata 998-8585, Japan. E-mail: gamano@hospital.sakata.yamagata.jp

Figure 1. Flattening, scaling and color change of the right nipple were noted.

the lesions depicted by MRI were diagnosed as an extensively spreading type of ductal carcinoma *in situ* (DCIS). In an attempt to excise all the suspicious lesions, we recommended mastectomy to the patient, to which she gave her consent.

Prior to mastectomy, nipple biopsy was performed to confirm the histopathlolgical diagnosis of Paget's disease (Fig. 3A). Additional DCIS was also demonstrated in the breast epithelium just beneath the nipple (Fig. 3B).

On March 1, 2004, modified radical mastectomy was performed. The specimen was subserially sectioned in 7 mm thick slices, and every block was examined histopathologically. Non-invasive ductal carcinoma with comedo-necrosis was present (Fig. 4). Cytonuclear grade was grade 2 and estrogen/progesterone receptor status was negative. HER-2 was strongly (3+) immunoreactive. These cytological and immunohistochemical results were similar to those of Paget's cells in the nipple biopsy specimen. Lymph node metastasis was not

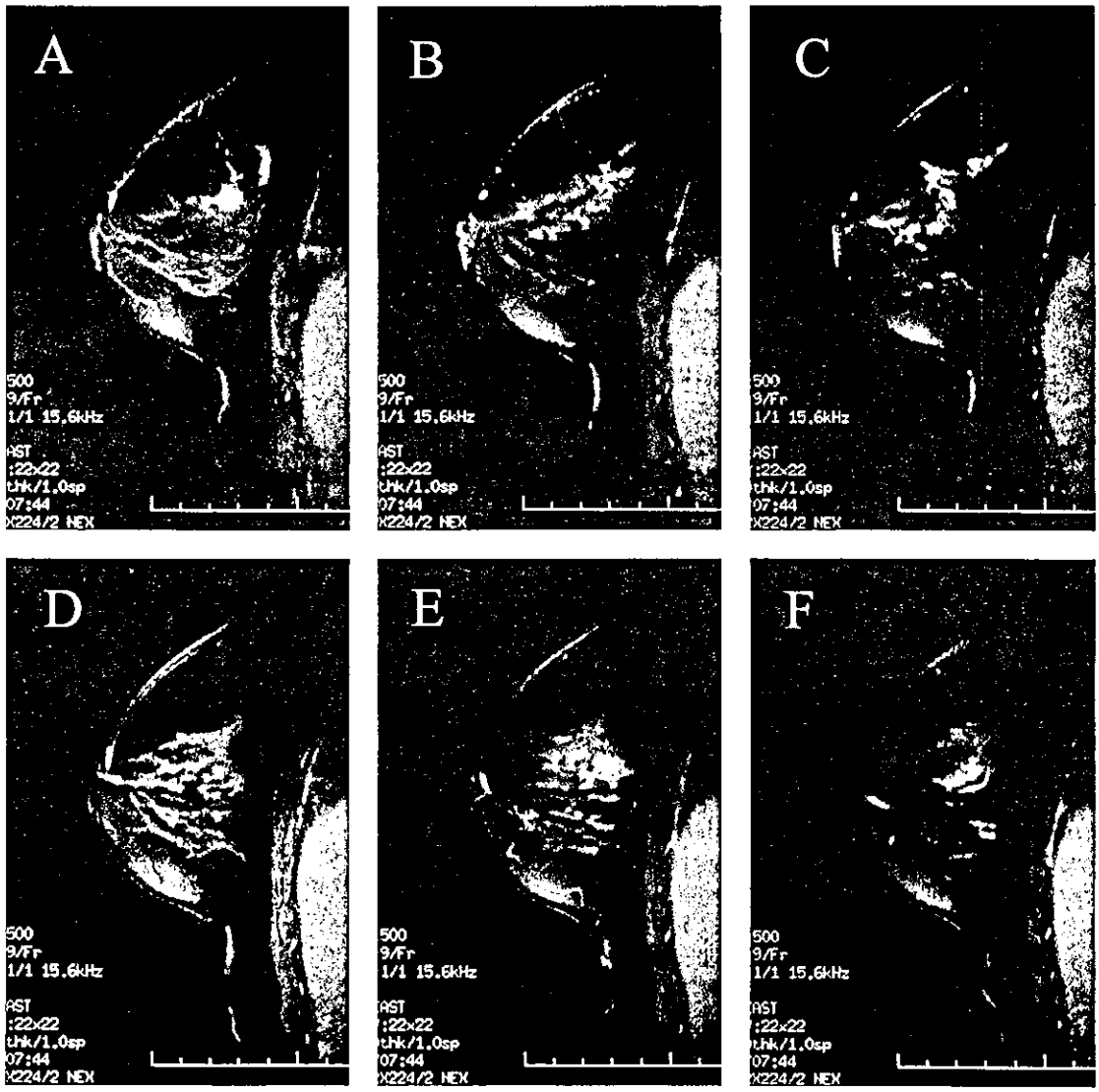

Figure 2. Fat-saturated, contrast-enhanced MRI of the right breast in the sagittal plane. From (A) to (F), a more lateral plane is shown at 6 mm intervals. (A–C) A diffuse segmental enhancement was apparent in the upper quadrant. (D–F) Another segmental enhanced lesion was shown in the lateral quadrant. The scale is graduated in centimeters.

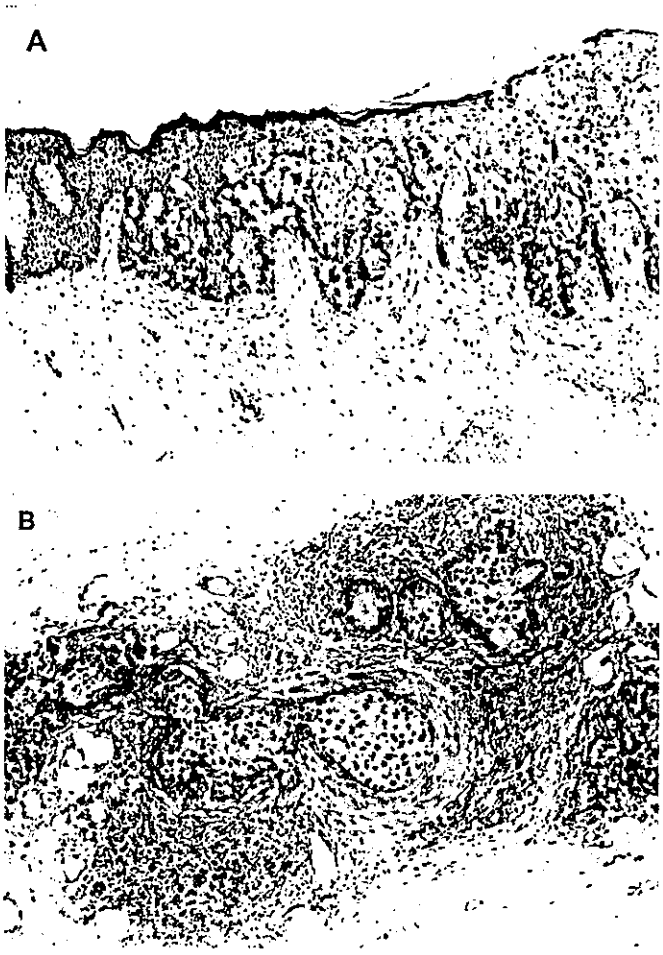

Figure 3. (A) A photomicrograph of a cross-section of the nipple biopsy specimen shows the Paget's disease. Large, round or ovoid intraepidermal Paget's cells with abundant clear cytoplasm and enlarged pleomorphic nuclei are present (H&E, objective: ×10). (B) A photomicrograph of a cross-section of the nipple biopsy specimen shows additional DCIS in the breast epithelium just beneath the nipple (H&E, objective: ×10).

Figure 4. A photomicrograph of a cross-section of the mastectomy specimen shows non-invasive ductal carcinoma with comedo-necrosis (H&E, objective: ×4).

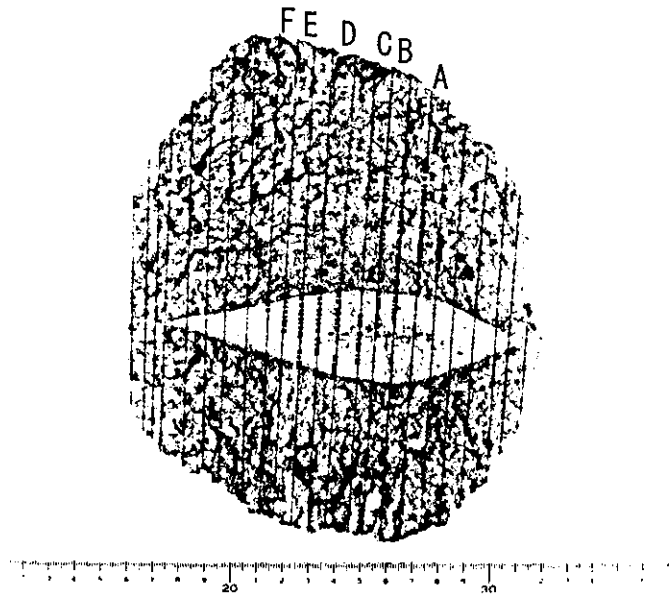

Figure 5. A histopathological cancer map of the mastectomy specimen, with the red marks denoting DCIS, reveals the extent of the lesions that are spreading in the upper and lateral quadrants. A–F indicate the location of the sliced specimen shown in Fig. 6. The scale is graduated in centimeters.

detected. According to the cancer map, DCIS were demonstrated extensively in the upper and lateral quadrants (Figs 5 and 6), accurately corresponding to the lesions shown by MRI (Fig. 2).

DISCUSSION

Paget's disease of the breast is a rare malignancy of the nipple–areola complex, comprising 0.5–5% of all breast cancer (1,7,9). It is manifested by progressive eczematoid changes of the areola with persistent soreness or itching (1,2,7). There have been debates about the histogenesis of this disease. According to the fact that this disease has been reported to be associated with an underlying breast carcinoma in 87–100% of cases (1–3,5–10), the epidermotropic theory, which postulates that Paget's cells are ductal cells that have migrated from an underlying breast carcinoma to the epidermis of the nipple, seems acceptable. The present case, where there existed underlying DCIS spreading extensively, is also compatible with the epidermotropic theory.

The treatment for patients with Paget's disease of the breast is controversial. Those patients with a palpable mass have a much greater incidence of invasive cancer, multifocal diseases, lymph nodal involvement and poor prognosis (1,2,6–8,10). Therefore, modified radical mastectomy is often the most appropriate treatment for patients with Paget's disease with a palpable lesion. On the other hand, for patients with Paget's disease who present no palpable mass, some investigators have proposed the use of breast-conserving therapy. Bijker et al. demonstrated that cone excision and radiotherapy is a feasible alternative for patients with Paget's disease and a limited extent of underlying DCIS (3). They reported a 5-year local

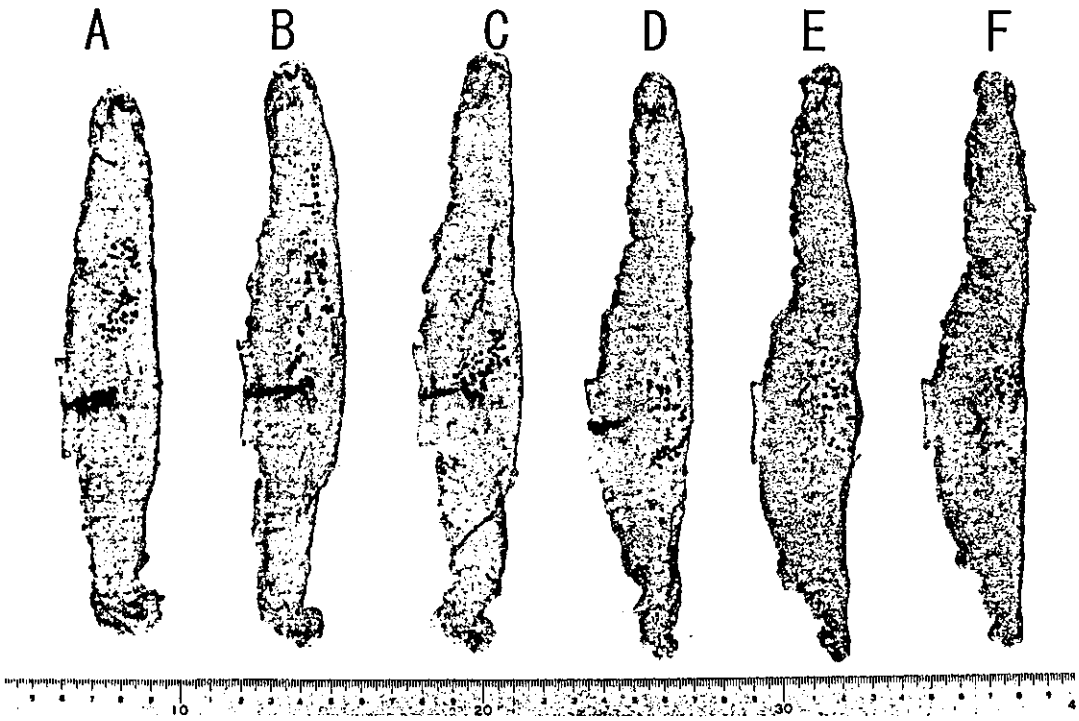

Figure 6. A more precise cancer map drawn on the cut surface of the mastectomy specimen. From (A) to (F), a more lateral plane is shown, and the location of the sliced specimen is indicated in Fig. 5. Note that the extent and distribution of DCIS, denoted by the red marks, correspond accurately to the enhanced lesions depicted by MRI in Fig. 2.

recurrence rate of 5.2%. Marshall et al. also recommended local excision and definitive breast irradiation as an alternative to mastectomy for patients with Paget's disease presenting no palpable mass or mammographic density (4). In their report, 5- and 10-year local control rates are 91 and 83%, respectively.

On the contrary, Kothari et al. warned against using breast-conserving therapy (5). They retrospectively reviewed the cases of 70 women with a clinical diagnosis of Paget's disease. Despite the fact that only one-third of women presented with a palpable mass, the malignancy was frequently extensive, being confined to the retroareolar region in only 25% of cases. They also demonstrated that the true extent of the disease was underestimated by mammography in 43% of cases. Of the 10 patients with no palpable mass and a normal mammogram, 40% had multicentric or multifocal carcinoma which would have been incompletely excised by cone excision of the nipple. Fu et al. described that of eight patients with no palpable mass who had been treated by quadrantectomy, two (25%) patients had recurrence (8). They concluded that even if the patient has no palpable mass, conservative surgery should be selected cautiously because of a higher recurrence rate and multifocal lesions.

In an era when breast-conserving surgery is sometimes recommended even for advanced infiltrating breast tumors, it seems quite reasonable to propose breast-conserving therapy for patients with Paget's disease with no definitive underlying breast cancer. However, as is already widely known, Paget's disease of the breast has a very high incidence of being accompanied by an underlying invasive or *in situ* carcinoma, even when there is no palpable mass (1–6,8–10). Mammography often fails to demonstrate the true extent of the disease (3–6,9). A more accurate and reliable imaging modality is necessary to select candidates for breast-conserving therapy more safely from among the patients who have Paget's disease of the breast.

Clinical utilization of MRI for breast cancer diagnosis has been under investigation since the late 1970s. With advances in surface coil technology and new imaging protocols using intravenously administered gadopentetate dimeglumine, MRI of the breast can now detect invasive cancer with 98–100% sensitivity (11,12). Amano et al. demonstrated that MRI can also detect the extensively spreading type of DCIS, that is often occult clinically and mammographically, as a pattern of diffuse segmental enhancement (13). In their study, the sensitivity of MRI to detect the extensively spreading type of DCIS was calculated to be up to 100%, and the specificity was estimated as high as 95%. The role of MRI in determining the extent of breast cancer is now well established (14). In the present case, no mass was palpable in her breasts, neither was there any abnormal findings on mammography or ultrasonography. There seemed a chance for her right breast to be treated conservatively, and for that reason we investigated it by MRI. There were diffuse segmental enhancements in the upper and lateral quadrants, strongly indicating the extensively spreading type of DCIS. Post-operative histopathological examination demonstrated non-invasive ductal carcinoma

with comedo-necrosis in the upper and lateral quadrants. The histological mapping with subserial sectioning demonstrated an extent of the lesions that corresponded accurately to the lesions defined by MRI. We conclude that MRI may play an important role in selecting candidates for breast conserving therapy out of those patients with mammary Paget's disease with no clinical evidence of an underlying breast carcinoma.

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Review Article

Ductal Carcinoma *in situ* and Related Lesions of the Breast: Recent Advances in Pathology Practice

Takuya Moriya*[1], Hisashi Hirakawa*[2], Takashi Suzuki*[3], Hironobu Sasano*[1,3], and Noriaki Ohuchi*[4]

*[1]*Department of Pathology, Tohoku University Hospital,* *[2]*Department of Surgery, Tohoku Kousai Hospital,* *[3]*Department of Pathology, Tohoku University Postgraduate School of Medicine, and* *[4]*Department of Surgery, Tohoku University Postgraduate School of Medicine, Japan.*

The incidence of ductal carcinoma in situ (DCIS) of the breast has increased significantly in Japanese women. It comprises 14.1% (172/1216) of all primary breast cancers at our institute, and nowadays this histological type is familiar to the surgeons and pathologists of any institute.

Several subclassifications have been published recently. Most based on nuclear atypia and the presence of comedonecrosis, and sometimes on the structures of the involved glands. These classifications are correlated with the biological behavior, tumor extent and the risk for local recurrences. The diagnostic accuracy of minimally invasive procedures (aspiration biopsy cytology/core needle biopsy) may differ between subclasses.

Atypical ductal hyperplasia (ADH) and microinvasive ductal carcinomas are lesions which resemble but deviate from the DCIS spectrum. The incidence of ADH seems to be lower than in Western countries. Patients with ADH may have a risk for subsequent breast cancer, because ADH is frequently associated with contralateral breast carcinomas. Microinvasion should be treated with caution, but we could not find any metastatic foci in microinvasive ductal carcinomas (T1mic). Tentatively, ADH may be treated similarly to non-comedo (low-grade) DCIS cases, according to our limited clinical experience.

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Key words: Breast, Ductal carcinoma *in situ*, Atypical ductal hyperplasia, Intraepithelial neoplasia, Microinvasive carcinoma

Recently, the incidence of ductal carcinoma *in situ* (DCIS) of the breast has increased, probably because of the early detection of cancer, especially by screening mammography. Nowadays, it is well known that DCIS is a heterogeneous group of diseases, both morphologically and biologically. Thus, further subclassification is recommended after this tumor is diognosed. Moreover, recent advances in less invasive diagnostic procedures has increased the chances for diagnostic pathologists to diagnose problematic intraductal lesions. These include questionable lesions suspicious for carcinoma, based on either fine needle aspiration cytology (FNAC) or core needle biopsy (CNB) results, and lesions that are definitely carcinoma of ductal origin, the invasiveness of which however cannot be determined by CNB. Additionally, the concept of intraductal proliferative lesions has been advanced to stratify lesions that pose different risks for the development of subsequent invasive carcinoma[1]. We will review recent advances in the field and the current situation in Japanese women.

Reprint requests to Takuya Moriya, Department of Pathology, Tohoku University Hospital, 1-1 Seiryo-machi, Aoba-ku, Sendai 980-8574, Japan.
E-mail: moriya@patholo2.med.tohoku.ac.jp

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Incidence of DCIS

DCIS was not frequent several decades ago. Since the 1980's its incidence has progressively increased, especially in western countries. In the USA, CIS (most of them were DCIS) incidence rates increased between 1973 and 1997 (under 50 years old, white 146%, black 283%; and over 50 years old, white 308%, black 349%)[2]. In 1997 CIS accounted for 16.4% of all breast carcinomas in white women, and 18.6% in black women[3]. In Los Angeles County, the average annual age-matched

incidence of CIS increased more than 5-fold between 1972-1981 and 1997-1998 (3.8 to 17.7/100.000 women), and the ratio of increase was higher than that of invasive carcinomas (86.1 to 111.5). The ratio of CIS among all breast carcinomas has increased from 4.2% (1972-1981) to 15.0% (1997-1998)[4]. National surveillance in Japan from 1980 to 1990 revealed that the ratio of CIS was between 2 to 4%, and had gradually increased[4]. The ratio of CIS among non-palpable breast carcinomas was more than 50% in most of series in Japan[4]. Considering the development of mammographic detection of breast carcinoma during the past two decades, it is not surprising that the ratio of DCIS has increased substantially.

It is necessary to note the methods of pathologic examination used to diagnose DCIS, because more precise examination may detect a minute focus of invasive carcinoma. Invasiveness is associated with the ability to metastasize. It is desirable to examine the lesions as closely as possible. However, this precision will be limited in routine practice. If the diagnoses of DCIS is made, the manner of the pathological examination is very important. We usually examined resected specimens from lumpectomy or larger operations by serial sectioning every 3-5 mm of the whole specimen (for cases of breast conserving surgery) and parenchyma (for cases of total mastectomy). By these methods, we detected 172 DCIS cases (14.1%) among 1,216 breast carcinomas between December 1998 and March 2002. The significance of DCIS diagnosed by CNB will be discussed later.

Subclassification of DCIS

DCIS represents a spectrum of disease, and the main purpose of subclassification is to stratify the risk of subsequent invasive carcinoma and/or local recurrence[5]. A rare exception is the classification by growth pattern (microfocal, diffuse, and tumor forming) according to mammographic or gross findings[6].

Previously, the microarchitecture was thought to be the most reliable feature[7,8]. Any architectural pattern may present with any nuclear grade, with or without necrosis. The simplest ways of subclassification is separating comedo from non-comedo lesions. Comedo type DCIS cases may have higher nuclear grade (by definition in many articles), larger tumor size, aggressive biological marker expression, and a higher risk of the (micro) invasion[9-13]. Close to 90% of palpable, pre-mammographic DCISs were reported as high grade comedo type lesions. In contrast, nearly 60% of mammographically detected lesions are the non-comedo subtype[14]. The incidence of the latter group is increasing. Architectural patterns other than comedo included cribriform, papillary, micropapillary (low papillary), solid, and mixed subtypes[5,10].

One of the problems using the term "comedo" DCIS is that the definition is not uniform. Variable criteria have been employed according to the proportion of comedonecrosis, architecture, nuclear grade, and a combination of these characteristics for the same category[15]. Additionally, it is not easy for pathologists to fit each DCIS into an architectural classification. In such cases, the term mixed subtype may be used, but many DCIS cases may therefore be classified as mixed. Thus, the employment of architectural subdivision may not always be reliable. Various pathologic subclassifications, using other than architectural features, have since been proposed.

Both nuclear grade and necrosis are thought to be more reliable predictive factors than architecture by some authors[9,10,13], and some of the new subclassifications employ mainly nuclear atypia (nuclear grade), or a combination of nuclear atypia and necrosis[17-20]. Some studies enhanced inter-observer reproducibility by using subclassifications devoid of architecture[21-25]. However, the architecture may be correlated with nuclear atypia, and some classifications still recommend describing the architecture along with stypia. Table 1 shows the relationship between architecture, nuclear atypia, and other findings of DCISs published previously[9], and the current van Nuys classification[16]. The predominant architectural patterns may correlate relatively well with the new grading system, although it is devoid of architecture. Additionally, there is some evidence that the micropapillary architectural type, when present in its pure form, is more commonly associated with more extensive, multifocal and multicentric disease[7,15].

One of the classifications employing an architectural description is the proposed classification of DCIS by the study group of the Japanese Breast Cancer Society (Table 2)[5]. The new WHO classification described three tier grading (low/intermediate/high grade), mainly according to nuclear features. The presence and absence of necrosis,

Table 1. Predominant Architecture of DCIS and their Comparison with the van Nuys Classification

Predominant architecture	COM	C + N	M + N	S + N	CRB	MCP	SOL	OTH	Total (or average)
No. of cases	7	3	7	6	22	19	9	12	85
NG 3	7								7
2		3	7	6	8	6	8	9	47
1					14	13	1	3	31
van Nuys Group	3	2	2	2	1	1	1	any	
mitotic counts; marked	4/7	2/3	1/7	2/6	0/22	2/19	1/9	2/12	17/85
mean No. of duct profiles involved	846	71	156	34	53	88	133	108	140
Maximum diameter (cm)	1.6	0.8	1.1	0.6	0.6	1.1	1.1	0.9	0.9

Van Nuys Group 3: high grade nuclei, Group 2: non-high grade nuclei with necrosis, Group 1: non-high grade nuclei without necrosis, COM: comedo, defined by high-grade nuclei, with solid nests and central necrosis, C + N: cribriform with necrosis, M + N: micropapillary with necrosis, S + N: solid with necrosis, CRB: cribriform, MCP: micropapillary, SOL: solid, OTH: others or mixed types

Table 2. Intraductal Proliferative Lesions: Different Terminology used in the Different Classification and their Relationships

Traditional terminology	Proposal by the study group, Japanese Breast Cancer Society, 2000	WHO classification 2003
Usual ductal hyperplasia (UDH)	Proliferative ductal lesions without atypia (mild/moderate-florid)	Usual ductal hyperplasia (UDH)
Flat epithelial atypia	Proliferative ductal lesions with atypia	Ductal intraepithelial neoplasia, grade 1A (DIN 1A)
Atypical ductal hyperplasia (ADH)	Proliferative ductal lesions with atypia (including ADH)	Ductal intraepithelial neoplasia, grade 1B (DIN 1B)
Ductal carcinoma in situ, low grade (Grade 1)	DCIS, HG 1 (low grade)	Ductal intraepithelial neoplasia, grade 1C (DIN 1 C)
Ductal carcinoma in situ, intermediate grade (Grade 2)	DCIS, HG 2 (intermediate grade)	Ductal intraepithelial neoplasia, grade 2 (DIN 2)
Ductal carcinoma in situ, high grade (Grade 3)	DCIS, HG3 (high grade)	Ductal intraepithelial neoplasia, grade 3 (DIN 3)

HG; histological grade

architectural feature, size of the lesions, and other characteristic features are also explained together[1]. If the different grade lesions are admixed within the same tumor, the description of their proportion is recommended. In any classification, the three-tier subdivision is always used, and the interrelationships between classifications are obvious. We employ the van Nuys classification currently, but we believe that it could be translated directly into the WHO classification in most cases.

Table 3 shows our recent experience of 82 cases of DCISs. The operative procedures consisted of 21 total mastectomies, 12 quadrantectomies, 37 wide excisions, 4 duct-lobular segmentectomies and 8 local excisions. All the cases were diagnosed by pathological examination of the whole tumor using 3-5 mm slices. By definition, all of the high-nuclear grade cases were classified into Group 3. The Group 1 cases were either of low or intermediate nuclear grade, but all of the Group 2 cases showed intermediate nuclear grade. Characteristically, the Group 3 cases showed a lower incidence of positive hormone receptor status ($p < 0.001$), and a higher incidence of HER-2 positivity ($p < 0.001$) compared with non-Group 3 cases. The results imply that nuclear grade will correlate well with hormone receptor/HER-2 neu status in DCIS cases. Additionally, although some authors reported a few cases (incidence 1-2%) of node-positive DCIS[15], we did not encounter any in our

Table 3. van Nuys Classification of DCIS and the Relationships between other Clinicopathological Features (Tohoku University Hospital 2002.6-2003.11)

Van Nuys Group	1	2	3	Total
Definition	non-high grade nuclei without necrosis	non-high grade nuclei with necrosis	high grade nuclei with/without necrosis	
No. of cases	39	30	13	82
NG 1/2/3	20/19/0	0/30/0	0/0/13	20/49/13
Age (average)	33-78 (54.4)	42-79 (54.1)	40-75 (59.2)	33-79 (55.0)
ER positive cases	33/34 (97.1%)	23/27 (85.2%)	2/10 (20.0%)*	58/71
PR positive cases	31/34 (91.2%)	22/27 (81.5%)	4/10 (40.0%)**	57/71
HER2 positive cases	2/32 (6.3%)	5/26 (19.2%)	7/10 (70.0%)***	15/68
Cases with lymph node positive	0/14	0/17	0/6	0/37

NG: Nuclear grade, ER: estrogen receptor, PR: progesterone receptor
*, **: significantly less frequent than non-Group 3 cases ($p < 0.001$)
***: significantly more frequent than non-Group 3 cases ($p < 0.001$)

series.

The unusual, rare subtypes include apocrine, mucinous, signet-ring cell, solid & papillary, spindled, neuroendocrine, Pagetoid, squamous, and clear. Most of these are classified according to their characteristic cell differentiation, rather than their architecture. Flat type DCIS, previously called clinging DCIS, may be a unique variant, which may resemble blunt duct adenosis on scanning magnification[26]. These lesions are malignant based on their genetic alteration[27], but are practically very difficult to diagnose accurately, especially low-grade lesions. More experience as well as further investigations will be necessary.

Differential Diagnoses of DCIS and Benign/Atypical Lesions

There are several lesions confused with DCIS in routine practice. They range from benign or borderline (atypically proliferating) intraductal lesions to minimally (micro-) invasive ductal carcinoma. Lobular neoplasia (atypical lobular hyperplasia and lobular carcinoma *in situ*) is another consideration.

Minimal Requirement to Diagnose DCIS

Low grade DCIS should be differentiated from benign and borderline (atypically proliferating) intraductal lesions. There have been several studies and proposals, including two famous studies by Page and colleagues, and Tavassoli[28, 29]. The new WHO classification employed both morphological and size criteria, probably according to these articles[1]. Morphologically, a monotonous cell population, high nuclear/cytoplasmic ratio, round nuclei, and hyperchromasia are necessary, combined with some architectural patterns. The evaluation of size has not been universally accepted. The entire involvement of 2 spaces, or cross section(s) exceeding 2 mm, are used for the minimal size.

The pathologist should pay great attention in routine practice to differentiate low-grade DCIS from intraductal hyperplasia or atypical ductal hyperplasia (ADH). A consensus conference on the classification of DCIS at Philadelphia in 1997[15] did not mention the precise distinction between DCIS and ADH, because they said it is difficult. Interobserver variability is sometimes problematic when intraductal lesions are diagnosed[30-32].

Intraductal Proliferative Lesions

Previously, proliferative disease of the breast was a form of epithelial hyperplasia usually seen with fibrocystic changes. Recently, the significance of these lesions has been enhanced, because they are related to carcinomas (whether directly or indirectly has not been proved, however), and early detection of low-grade carcinoma by mammography has also raised the incidence of precancerous proliferative lesions as well.

Currently, a new concept has emerged that describes intraductal proliferative lesions as a continuous disease entity, ranging from benign through atypical (ADH) to malignant disease

Table 4. Expression of Various Immunohistochemical Markers for Various Lesions (Including Intraductal) of the Breast

	UDH	ADH	DCIS		IDC	
			low grade	high grade	low grade	high grade
Average age	44.0	42.8	51.3	56.8	52.7	50.4
ER	5.7	6.7	6.4	5.0	5.8	3.3
PR	4.3	6.2	4.6	1.8	4.5	2.8
Ki-67 LI	3.7	4.5	9.5	9.4	21.3	35.9
p53	0/22	0/26	7/40			
c-erbB-2	0/22	0/26	8/40			

ER: estrogen receptor, PR: progesterone receptor, LI: labeling index, UDH: usual ductal hyperplasia, ADH: atypical ductal hyperplasia, DCIS: ductal carcinoma *in situ*, IDC: invasive ductal carcinoma (ER and PR were scored according to Allred DC et al. Mod Pathol 11: 155-168, 1998)

(DCIS)[29]. Even the concept of intraepithelial neoplasia (mammary intraepithelial neoplasia, MIN) has been adopted by some investigators[30]. The common loss of heterozygosity that occurrs with synchronous atypical ductal hyperplasia (ADH), DCIS and invasive ductal carcinoma (IDC) may suggest a stepwise progression from ADH to IDC[33]. However, smaller lesions are often removed by excision, and their real natural history is unknown. In practice, they are generally accepted to confer an increased risk for the subsequent development of invasive carcinoma, the magnitude of which varies according to the degree of proliferation and/or atypia[1]. In Japan, the study group of the Japanese Breast Cancer Society examined the interval to subsequent invasive carcinoma in the same quadrant of the breast after biopsy, and this was shorter in the cases showing a higher degree of intraductal proliferative lesions[5]. Table 2 shows the classification proposed by the the study group and its relationship between the new WHO classification[1, 5]. Table 4 shows the expression of some biomarkers analyzed immunohistochemically[33, 34].

Atypically proliferative lesions (atypical ductal hyperplaia-ADH/proliferative disease with atypia) are ductal proliferative lesions, which should be differentiated from DCIS histologically by the presence of structural and/or cytological atypia along with proliferative disease without atypia. This category may include the lesions with increased relative risk for subsequent invasive carcinomas, but their biological behavior and clinicopathological significance is uncertain, at least currently. Thus the diagnosis of "atypia" should be made with caution, and not used so frequently. If one uses this word on the pathological report, the reasons for the term "atypia" should be mentioned. For example, a description of the extent of the lesions, degree of epithelial proliferation, structural atypia, nuclear atypia or number of mitoses is recommended[5].

Atypical Ductal Hyperplasia (ADH)

ADH probably comprises the majority of "atypical" lesions but is also relatively rare. ADH may be diagnosed when one suspects but hesitates to diagnose DCIS, because of incompleteness of monotony (either structural or cytological) or limited extent. In any case, sufficient discussion with surgeons, and close follow-up is necessary.

At least in Japan, the diagnosis of ADH has not been widely accepted. We use this terminology in routine practice, according to the criteria of Page and colleagues[28]. They said that almost 3.5% of the biopsy specimens are diagnosed as ADH, however, we think that the incidence is much lower. This may be due to differences between Japanese and western populations or interobserver variability. We had a chance to review a biopsy series in which fibrocystic change was initially diagnosed, and found that the incidence of ADH was 1.2% by re-examination[35]. Table 5 shows the clinicopathological features of ADH cases in our laboratory. Only 21 cases were diagnosed as solitary ADH out of almost 1,000 primary breast cancer cases (cases with synchronous, ipsilateral carcinomas were eliminated, and consultation cases were not included). The patients were relatively young, as shown in Table 5. Interestingly, at least 5 cases

Table 5. Atypical Ductal Hyperplasia (ADH): Experience at The Pathology Department of Tohoku University Hospital from December 1998 to June 2002

- 21 case (cases with synclonous, ipsilateral carcinomas were eliminated)
- Background: 995 primary breast cancers, including 206 DCISs during the same period
- Age & gender: 28-56 (average 46.2) years old, all female
- Contralateral breast cancers: At least 5 cases (2 synclonous, 3 subsequent, follow-up period up to 5 years)
- Associated lesions: 3 were intermingled with papilloma, 1 with mucocele-like lesion, 1 within fibroadenoma
- Fine needle aspiration cytology: negative 5, indeterminate 6, suspicious for malignancy 3
- Diagnostic procedure: Local excision 15, Duct lobular segmentectomy 5, Core needle biopsy 1

showed contralateral breast cancer. This implies that ADH may be a relative risk for developing invasive breast carcinoma even in a population with a low incidence of ADH.

Microinvasive Ductal Carcinoma

The upper end of the DCIS spectrum is the borderline between DCIS and carcinoma with minimal stromal invasion. If invasion exists, there is a chance for metastasis. Variable definitions for "microinvasion" have been proposed previously. The cases with an invasive focus less than 1% of the total[1], or an invasive focus less than 1 mm (T1mic)[37] are relatively widely accepted to represent microvasion. They will show an apparent foci of infiltration into "interlobular" stroma. We have encountered 28 T1mic cases among 1,216 primary breast cancers (2.3%), and about 1/6 of DCIS cases (172 cases during the same period)[38]. Most were composed of small cell nests, or of single cells, but tongue-like projections with reactive stroma may be seen. There may be multiple foci (1-7 foci, average 3, in our series). These cases may show higher nuclear grade, tend to be associated with comedonecrosis, and more severe stromal reactions (lamellar fibrosis and/or chronic inflammatory cells) around the intraductal carcinomas.

Microinvasive carcinomas express a relatively low risk for lymph node metastases, and the prognosis is considered to be extremely good[39, 40]. None in our series expressed axillary lymph node metastases on serial sectioning of the whole carcinoma[38]. However, follow-up data using universally accepted procedures and/or criteria will be necessary to reach the final conclusions.

Diagnosis of DCIS and ADH by Minimally Invasive Procedures

Core needle biopsy (CNB) under stereo- or ultrasound guided procedures has been widely accepted. Thus, there has been an increased chance to diagnose earlier carcinomas (including low-grade DCIS) and borderline lesions. One of the problems of using this method is the specimen does not always include minute foci of invasion. If DCIS was diagnosed by CNB, there is still a chance for invasive carcinoma in the residual parenchyma. About 30% of DCIS diagnosed by CNB were truly invasive carcinoma in one study using a 14-gauge core[41], but the incidence fell to 10% with an 11-gauge vacuum-assisted procedure[42]. Similarly, if ADH is seen by core needle biopsy (CNB), 12-33% of the cases showed DCIS on excisional biopsy[43]. Some of the cases may be DCIS with invasion (IDC) but this situation is usually related to the number of foci (4 or more) of ADH on CNB[44].

Ultrasound-guided fine-needle aspiration biopsy cytology (FNABC) for dilated ducts may be performed. Any intraductal proliferative lesions, if correctly sampled, may show abundant, three-dimensional epithelial cell nests[45]. Our experiences revealed that the diagnostic accuracy of DCIS was 62.5%, lower than that of invasive ductal carcinomas (more than 80%)[46]. The cytological diagnosis of atypical hyperplasia is much more difficult, because most are small (less than 2 mm) and require sampling of the appropriate cells[17]. Some authors used the grading/scoring system for benign and malignant intraductal proliferative lesions[47, 48]. The author would like to recommend that if the dilated ductal lesion can be detected by ultrasound, US-guided FNABC may be performed, however, if the lesion is mammographically calcified and thought to be an intraductal lesion, CNB is recommended[49].

Pathological Factors of DCIS other than Grading, and their Significance

In addition to the methods for evaluating and grading DCIS, the extent of the lesions (size) and the surgical margins (if breast conserving surgery is performed) should be described. The Van Nuys

Prognostic Index (VNPI), which is the sum of grade, extent, and margin status, correlates well with the risk for local recurrence[50]. However, the overall survival, at least 5- or 10-years later, were not significantly different. Additionally, the ratio of invasive recurrences among all recurrent cases (almost half of the cases, as in similar results in Japanese patients[51]) was not be influenced by the VNPI.

Other pathological parameters are detected by special techniques, but they are not always performed in routine practice. The Ki-67 index, hormone receptor status (ER/PgR), HER-2/neu status, and p53 expression status are shown in Table 4. Many articles have examined the overexpression of c-erbB-2 (HER-2/neu) in high-grade DCIS, and the frequency of overexpression was higher than that of invasive ductal carcinomas.

Finally, lymph node metastases of DCIS are reported in almost 0%, or 1-2% of the cases, as mentioned previously. The method of pathological examination, rather than underestimation of microinvasion, may be the cause of the unexpected metastasis. Most node positive DCISs are higher-grade lesions.

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Pathological Evaluation of Sentinel Lymph Nodes for Breast Cancer

Takuya Moriya,[1] Shin Usami,[1,2] Hiroshi Tada,[2] Atsuko Kasajima,[1] Kazuyuki Ishida,[1,2] Yoshiyuki Kariya,[1] Noriaki Ohuchi[2] and Hironobu Sasano,[1] Departments of [1]Pathology and [2]Surgery, Tohoku University Hospital, Sendai, Japan.

Recently, sentinel lymph node (SLN) biopsy has been employed to avoid unnecessary lymph node dissection, because SLN negativity for carcinoma metastases may imply an extremely low possibility of non-SLN involvement. Pathological evaluation is essential, but standardized procedures have not yet been determined. Intraoperative consultation, either by frozen section (multiple slices are desirable) or touch imprint cytology, are usually very useful. Their accuracy, however, is variable and depends on the procedures used, but specificity is characteristically 100%, and the missed metastatic focus is always quite minute. After fixation, multiple sections, immunohistochemistry, and their combination will be able to detect small metastatic foci more frequently. The clinical significance of small or submicro- or occult metastases have not yet been clarified, and further investigations are needed. If the SLN is positive for carcinoma metastases, both the procedure for detection and the size of the metastatic focus should be clarified on the pathological reports. [*Asian J Surg* 2004;27(4):256–61]

Introduction

The occurrence of breast carcinoma has increased in Japan,[1] and currently, about one in 30 Japanese women will suffer from breast cancer during her life. This increase is mainly due to changes in lifestyle, especially in eating habits, and to the development of mass screening programmes for breast cancer. The detection of early-stage cancers by screening mammography has led to an increasing incidence of non- or early invasive carcinomas. Additionally, recent advances in breast cancer treatment enable us to perform breast-conserving surgery for early-stage cancers.

Pathological analysis of regional lymph nodes is crucial for tumour staging, which is a prognostic indicator.[2] However, total removal of axillary lymph nodes may cause significant morbidity, including limb oedema, loss of sensation and disturbances in limb motion. Sentinel lymph node biopsy (SLNB) is a new trend in breast-conserving surgery. If the sentinel lymph node (SLN) is negative for carcinoma, additional dissection may be avoided, because SLNB is considered a sensitive and specific procedure for predicting regional lymph node status.

General features of lymph node metastases

In Osaka Prefecture, 32% of breast carcinomas were positive for lymph node metastasis in 1996–1998,[3] which is about average in Japan. The proportion of node-positive cancers decreased from 46% in 1975–1977. As nodal involvement is more frequent in larger tumours,[4] the increasing incidence of early-stage cancer may lead to a decrease in the incidence of positive nodes. Many cancers may be truly node negative, but some may be positive with metastatic foci too small to find in routine practice. Multiple sectioning of the lymph nodes for histopathological analysis may improve the detection rate for small metastases.[5] Details of methods for multiple sections or other alternative procedures, as well as their clinical significance, will be discussed later.

Address correspondence and reprint requests to Dr. Takuya Moriya, Department of Pathology, Tohoku University Hospital, 1-1 Seiryo-machi, Aoba-ku, Sendai 980-8574, Japan.
E-mail: moriya@patholo2.med.tohoku.ac.jp • Date of acceptance: 3 November, 2003

Lymph node metastases mostly occur via lymphatic vessels and, rarely, blood vessels. Small foci of metastatic tumour cells are frequently seen in the subcapsular (marginal) sinus, and these are thought to be the initial evidence of node metastasis. They are either floating within the sinus or attached to the capsule,[6] and may extend to the sinusoids and invade the node parenchyma. Therefore, pathologists must seek the area just beneath the capsule in routine practice if extensive metastases are not seen in low-power view. Additionally, one study suggests a higher probability of metastatic breast carcinoma at the inflow junction of the afferent lymphatic vessels.[7]

Usually, small foci of metastasis do not enlarge the lymph node. However, it is well known that a significant proportion (approximately 20–25%) of clinically node-negative patients will have metastatic foci pathologically.[8] Additionally, negative nodes may show extensive enlargement, caused by accompanying reactive processes such as sinus histiocytosis or reactive hyperplasia of the lymph follicles.

Pathological staging for metastatic carcinoma

Small metastatic foci are currently divided into two categories:[9] isolated tumour cell(s), defined as single cells or small clusters of cells not greater than 0.2 mm in largest dimension, usually floating on the sinus, without any proliferation or stromal invasion and no evidence of malignant activity; and micrometastasis, found in tumour deposits of 0.2–2.0 mm in largest dimension, that may proliferate and destroy the stroma and may have malignant activity. If these small metastases are detected by procedures such as immunohistochemistry or reverse transcriptase-polymerase chain reaction (RT-PCR), they are recommended to note how to detect the metastatic focus.[9] Submicrometastases are metastases that can only be detected by immunohistochemistry and are usually isolated tumour cells.[10] Occult metastases are foci missed by initial screening and identified on subsequent screening, or metastases identified at additional evaluation using paraffin-embedded tissue blocks.[8] Occult metastases are not defined by size but are no larger than micrometastases.

Pathological examination of sentinel lymph nodes

Specimen handling for intraoperative diagnosis

Procedures and guidelines are available for pathological analysis.[4,10] It is strongly recommended that most pathology analysis for SLNs be performed intraoperatively, by frozen-section diagnosis, a combination of frozen section and cytology or supplementary immunohistochemistry, because these results will immediately affect further surgical procedures. Some authors consider that frozen-section diagnosis is not reliable,[11] but it may be effective if technicians and pathologists are well trained and experienced. Frozen-section diagnosis is a safe method, even if radioactive materials are used for SLN detection,[12] as pathologists and technicians suffer only minimal exposure.

In general, as the number of examined slides increases, the rate of detection of micrometastases increases. We believe that one haematoxylin and eosin (H&E)-stained section along the long axis of the node is not sufficient, especially if the node is large. However, serial or step sections across the whole node is time-consuming and costly, and should only be used in research.[13] In routine practice, pathologists should measure the size of the node and then cut it into almost 2 mm thick sections, then perform careful gross examination to detect focal lesions. Cutting along the long axis may be a standard, but it is also possible to cut along the short axis, according to the shape and size of the node. It is desirable that three levels of frozen sections are made for each slice.[10] After surgery, frozen sections should be fixed in formalin and permanent sections made to confirm the intraoperative diagnosis.

Cytology by touch preparation of the cut surface is another procedure for detecting micrometastases. It does not waste tissue by sacrificing for frozen sections and does not suffer from freezing artefacts. It is also easier and faster than frozen-section cutting. However, the evaluation of the specimen is not always easy, and may potentially lead to indeterminate or deferred diagnoses.[4] Pathologists require cytology training, including screening and cell interpretation. The advantages and disadvantages of cytology and frozen-section diagnosis are shown in Table 1.

Accuracy of intraoperative consultation

In SLN analysis, it is very important that pathologists detect metastatic carcinoma accurately. However, the procedure for treating removed node(s) has not been standardized. Therefore, it is not easy to compare different studies to assess the accuracy of each procedure.

The accuracy of frozen-section diagnosis is compared with that of paraffin sections in Table 2.[14–19] Both types of section were evaluated by H&E staining only. There is significant variation in the sensitivity (52–100%) and false-negative rate (0–48%) of frozen-section diagnosis. These discrepancies are probably due to differences in the handling process (including

Table 1. Advantages and disadvantages of frozen section and imprint cytology for intraoperative sentinel lymph node analysis

	Frozen section	Imprint cytology
Advantages	- Interpretation of nodal architecture available - More specific diagnosis possible - Size of metastatic focus measurable - Rapid immunostains available	- Simple/cheap/rapid - Interpretation of cytological/nuclear details available - Avoids tissue loss
Disadvantages	- Relatively time-consuming - More expensive - Freezing artefacts - Tissue loss (by sacrificing)	- Size and area of metastatic focus not detectable - Indeterminate/deferred diagnoses - Need special training to interpret - Sampling error may occur

Table 2. Studies of intraoperative frozen-section diagnosis for sentinel lymph nodes

Authors	No./interval of H&E sections	*N*	Accuracy, %	Sensitivity, %	Specificity, %	False-negative rate, %
Canavese et al (1998)[14]	3 (both sides)	96	96	86	100	14
Schneebaum et al (1998)[15]	Not described	47	98	91	100	9
Koller et al (1998)[16]	3 consecutive	107	83	64	100	36
Imoto et al (2000)[17]	Not described	52	96	89	100	11
Noguchi et al (2000)[18]	2	38	79	60	100	40
Noguchi et al (2000)[18]	> 3	45	93	85	100	15
Noguchi et al (2000)[18]	2 mm interval	26	100	100	100	0
Motomura et al (2000)[19]	1	101	88	52	100	48

H&E = haematoxylin and eosin.

the number and interval of slices, gross inspection and procedures for microscopic slide preparation), and procedures for final pathological evaluation. Other possible influences on the detection rate of metastatic foci are differences in the characters of primary tumours. The size of the primary cancer influences the results of frozen-section diagnosis.[20]

Most metastatic foci missed by frozen-section analysis are either micrometastases or isolated tumour cells. This argues for an awareness that small metastatic foci may be missed at routine intraoperative examination. It is interesting that the specificity of frozen-section diagnosis was 100% in all the studies listed. It is unlikely that trained pathologists will miss foci of carcinomas on microscopic examination. Thus, it is possible that the accuracy of frozen-section diagnosis may be improved either by multiple slices or step/serial sectioning, if the bias due to the skill of the pathologists is ignored. Veronesi and colleagues analysed SLNs by frozen sections every 100–500 μm, but the false-positive rates were 36% and 32% in two studies.[11,21] Therefore, they examined 15 levels of frozen sections at intervals of 50 μm using immunohistochemical analysis. The false-positive rate was reduced to 6%.[11] The accuracy of their last proposal was confirmed by Viale et al.[13] Although this procedure gives good results, it may be too complex and time-consuming for routine practice in most institutions. The significance of immunohistochemistry will be discussed later.

Imprint cytology is compared with permanent H&E-section diagnosis in Table 3. The procedure is quite simple and as accurate as tissue sections. Accuracy and sensitivity are good, and specificity was almost 100%, similar to frozen-section diagnosis.[19,22-26] It is unlikely that benign cells (i.e. histiocytes, lymphocytes) will erroneously be interpreted as carcinoma metastases in most cases. However, we have had some experience of atypical cells on the smear being tentatively described as carcinoma. In such cases, experience is necessary and, if the situation allows, the combination of both frozen section and imprint cytology will be useful.[22]

Multiple levels of H&E sections

The average diameter of ductal carcinoma cells is 20 μm. Theoretically, to detect tumour cell nests of 20–30 cells, it is necessary to make step sections at intervals of 250 μm.[27]

The results of multiple levels of H&E sections are summarized in Table 4. The rate of node-positive patients is increased (4-18% of patients upgraded) by various multiple-section procedures.[28-30] These methods are not always employed at the time of frozen-section diagnosis because they are time-consuming for technicians.

Immunohistochemical analysis of SLNs

As microscopic analysis is somewhat subjective, there are some limitations to detecting metastatic foci on routine staining, even by skillful pathologists. To make examinations more accurate, immunohistochemistry has been used as an adjunct to routine stains, both intra- and postoperatively (Table 5).[30-36] Moreover, if suspicious cells are found on H&E sections, additional immunohistochemistry will be a strong tool for confirmation. Rapid immunohistochemistry using imprint cytology has also been used.[37] Usually, detection of cytokeratin is used in both histology and cytology; 2-20% of patients are upgraded by this procedure. The combination of multiple H&E sections with either single immunohistochemistry or

Table 3. Studies of intraoperative imprint cytology for sentinel lymph node examination

Authors	No./interval of sections	*N*	Accuracy, %	Sensitivity, %	Specificity, %	False-negative rate, %
Moriya et al (1994)[22]	1	286	99	95	100	5
Rubio et al (1998)[23]	1	124	99	96	100	5
Ratanawichitrasin et al (1999)[24]	2	55	98	93	100	7
Motomura et al (2000)[19]	2 mm interval	101	96	91	99	9
Henry-Tillman et al (2002)[25]	> 1	479	99	94	100	6
Karamlou et al (2003)[26]	1	446	-	75	100	5

Results were compared with permanent haematoxylin and eosin sections of the same level; studies with immunohistochemical analysis were eliminated.

Table 4. Studies on multiple levels of haematoxylin and eosin (H&E) sections for sentinel lymph node examination

Authors	*N*	Study design	Patients positive by standard methods, *n* (%)	Patients upgraded by alternative methods, *n* (%)
Turner et al (1999)[28]	52	2 H&Es at 40 μm interval vs additional 2 H&Es at 160 μm interval	10 (19)	2 (5)
Nahrig et al (2000)[29]	40	1 H&E vs 4 additional H&Es at 150 μm intervals	18 (45)	4 (18)
Torrenga et al (2001)[30]	250	1 H&E vs additional 4 H&Es at 250 μm intervals	69 (28)	8 (4)

Table 5. Studies of immunohistochemical staining (IHC) for sentinel lymph node examination

Authors	*N*	Study design	Patients positive by standard methods, *n* (%)	Patients upgraded by IHC, *n* (%)
Czerniecki et al (1999)[31]	41	1 H&E vs 4 levels of IHC	12 (29)	3 (7)
Noguchi et al (1999)[32]	62	1 H&E vs IHC	24 (39)	1 (2)
Pendas et al (1999)[33]	478	1 H&E vs IHC	93 (19)	41 (9)
Kowolik et al (2000)[34]	33	2 H&Es vs IHC	8 (24)	4 (12)
Mann et al (2000)[35]	51	1 H&E vs IHC	10 (20)	10 (20)
Wong et al (2001)[36]	973	1 H&E vs 2 levels of IHC	104 (11)	58 (6)
Torrenga et al (2001)[30]	250	1 H&E vs IHC	69 (28)	5 (2)
Torrenga et al (2001)[30]	250	1 H&E vs 4 levels of IHC	69 (28)	17 (7)

H&E = haematoxylin and eosin.