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Prediction of Nipple Areolar Complex Involvement in Breast Cancer

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Abstract

Background: **The role of nipple sparing mastectomy as an alternative to standard mastectomy is becoming available. Due to the varied reported incidence of nipple-areolar complex (NAC) involvement (6-54%) in breast cancer with no reliable preoperative clinico-pathological predictors, the patient selection for nipple sparing mastectomy is challenging.**

This study aimed to determine the incidence of NAC involvement in Thai breast cancer patients with clinically-uninvolved nipples. The predictive value of frozen section of subnipple tissue and other clinico-pathological parameters in determining NAC involvement were systematically examined.

Research Design: **Prospective descriptive analysis in a tertiary care hospital**

Materials and Methods: **Patient selection: All breast cancer patients who underwent standard mastectomy were included, excluding those who had clinically nipple involvement, neoadjuvant chemotherapy, Paget's disease and pre-excisional biopsy.**

Study Procedure: **From mastectomy specimen the subnipple tissue was collected for frozen section to compare with serial permanent section of NAC. The clinical data, mammographic or sonographic finding and pathological report were collected for analysis.**

Results and Analysis: **Forty-six patients were recruited between July 2003 to May 2004. Forty-five patients had ductal carcinoma and one had lobular carcinoma. The NAC involvement was found in 13/46 specimens (28%). The frozen section of subnipple tissue showed 84.8% specificity, 84.6% sensitivity and 84.8% accuracy. There was no statistically significant correlation between other clinico-pathological parameters or radiological parameters.**

Conclusions: **Frozen section of subnipple tissue can predict the NAC involvement of breast cancer. Nipple sparing mastectomy in selected patients with favorable clinical predictors could be considered in negative frozen section group.**

Carcinoma of the breast is the most common site specific cancer in woman worldwide (32% of all female cancer). It is the leading cause of death from cancer for females 40-44 years of age which is responsible for 19% of the cancer related death in woman.[1] In Thailand,it is the second most common cancer which falls behind those of cervical carcinoma.[2]

Early breast cancer (Stage I, II, and IIIA) often requires a multimodality approach of treatment. The choices of surgical management for breast cancer depends on several factors including tumor location, size, staging, grading, lymph node status and patients' satisfaction. Total mastectomy is generally respected for the control of breast cancer loco-regional recurrence. But the disadvantages include psychological effects on individuals and the limited cosmetic result. Breast conservation therapy (BCT)is introduced for the treatment of breast cancer, based on the same concept of achieving the loco-regional control of the disease with better cosmetic result. It still has the controversies in many surgical techniques including how the breast should be conserved. Reconstructive surgery may be applied to improve the cosmetic results. It may be done at the time of the mastectomy (immediate reconstruction) or later (delayed reconstruction). Breast contour can be restored by the artificial implant or a rectus muscle or other myocutaneous flaps.[3-6]

The Nipple Areolar Complex (NAC), routinely excised during mastectomy to prevent the recurrence, is proved to be the drainage pathway of breast lymphatic especially the subareolar lymphatic plexus.[3,5,7] Very few lymphatic drainage is found via breast subdermal lymphatic vessel compare to the subareolar lymphatic plexus.[8]

Restoration of nipple and areola is still a major problem in limited breast surgery because there is no other unique tissue which can ideally replace the nipple areola complex (NAC). The survival of "free nipple graft" during breast reconstruction or breast conservative surgery were reported with very good results and few complications.[9,10] The necrosis of the nipple can be prevented by restoration of the subdermal plexus. The challenge now is not how to preserve the NAC but to prove that the NAC is safe to be preserved.

The incidence of the NAC involvement in breast cancer was studied and reported in several literatures. Approximately 6-54% of the NAC were involved by the malignancy.[7] The incidence depended on various predictors (eg. tumor size, tumor staging, clinical appearance of nipple and areola, the presence of nipple discharge, the distance between tumor and the margin of nipple areolar complex, the location of the tumor, the histological type, etc.).[7,11,12]

We decided to sample the tissue from sub-nipple area which is believed to be the major final drainage pathway of those lymphatic channel from breast to NAC. The sub-nipple tissue could represent the tumor involvement of NAC in breast cancer patients

This sub-nipple tissue was defined as the tissue lying at the base of the nipple, the depth is under the dermis and subdermal plexus. It consists of part of the terminal mammary ducts, lymphatic plexus and ducts, smooth muscle and the connective tissue of the subcutaneous layer. (Figure 1)

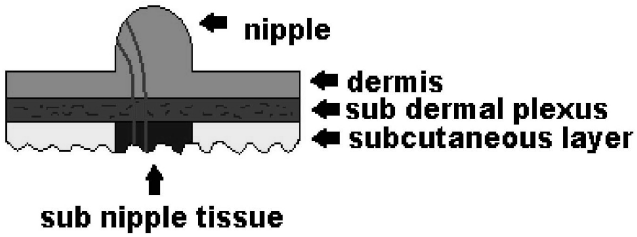

Figure 1 The sub-nipple tissue

The NAC is vascularized from the perforator vessels and from subdermal plexus. The sub-nipple tissue could be taken without resulting in NAC necrosis because the blood supply are from those preserved subdermal plexus. The autologous nipple graft during breast reconstruction or breast conservative surgery provide satisfactory results and has few complications.[9,10]

In most studies the standard H&E histological section is mainly used to describe the involvement of the NAC. The frozen section is more practical in determining the nipple sparing mastectomy procedure. The correlation between a frozen section and a permanent section has been reported with high satisfaction.[13-15] The frozen section can be interpreted by trained pathologist in any center with nearly the same results.

With high correlation of the frozen section to the permanent section,[13-15] we decided to use the frozen section of the sub-nipple area to determine the involvement of NAC in breast cancer patient who undergo total mastectomy. This study will prove the reliability of frozen section from sub-nipple area in determining the involvement of NAC and will report various predictors of the tumor involvement of NAC in breast cancer patients. Those selective patients with negative frozen section will have benefit from nipple sparing mastectomy.

Research Design

Analytical study in a tertiary care hospital and diagnostic test

Materials and Methods

Study population : we included all operable breast cancer (total mastectomy) but excluded those who had 1. clinical nipple involvement (skin ulcer, nipple discharge, nipple retraction, skin change such as lymphedema) 2. neoadjuvant chemotherapy 3. pre-excisional biopsy 4. Paget's disease 5. Phylloides tumor.

Study Procedure

The informed consent was obtained from the patient. The case record forms were completely filled with various data (eg. tumor size, tumor staging, physical examination, the presence of nipple discharge, the distance between tumor and the margin of nipple areolar complex, the quadrant of the tumor, the histological type, etc.) The distance of the tumor was measured from the nearest margin of the tumor to the nearest margin of the nipple in the radial axis (Figure 2). The measurement was performed from mammogram by trained breast radiologists.

Surgical Methods

The patients underwent mastectomy (including simple mastectomy, total mastectomy with sentinel lymph node dissection or modified radical mastectomy) under general anesthesia. The specimens were immediately delivered to the specimen table outside the operating room.

Then the NAC flaps were raised subcutaneously beneath subdermal layer by sharp dissection. The adequate exposure of sub-nipple area was obtained. The small piece of entire sub-nipple tissue was collected by sharp dissection (Figure 3).

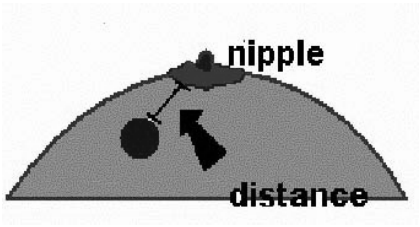

Figure 2 The distance between tumor and the margin

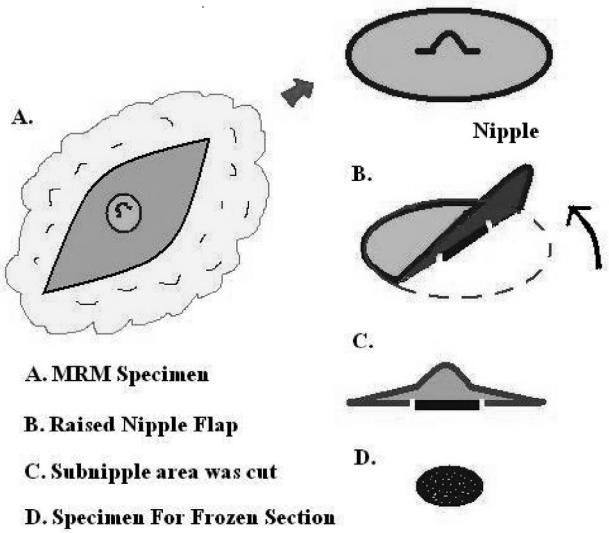

Figure 3 Collection of sub-nipple tissue

The remaining subnipple specimen was inked on the base of the subnipple tissue before collected into cassette box.

A specimen which was adequate for frozen section (less than 5 mm thickness) and covered entire subnipple area was sent for pathohistology examination.

The remaining mastectomy specimen was processed for routine pathological examination.

Pathological Methods

Subnipple specimen The tissue was embedded in CRYOMATRIX (active ingredient: 10% polyvinyl alcohol and 4% polyethylene glycol) and was immediately frozen in cryostat at -20°C. Two consecutive 5μ thick section in horizontal plane will be stained with H&E for frozen section examination.

Mastectomy specimen The followed mastectomy specimen was processed for H&E stain and evaluated routinely according to the current protocol of Department of Pathology, Siriraj Hospital. These included gross description and diagnosis of specific type of carcinoma, differentiation, size, location, calcification, vascular invasion, perineural invasion, multi or unicentricity, NAC and skin involvement, margin and axillary lymph node status.

The involvement of the NAC was determined by diagonally serial sectioning of the NAC (serially 2-3 mm) and submitted all sections for microscopic examination. (Figure 4) The frozen sections and permanent sections were evaluated by two pathologists without knowing the result of the frozen sections or permanent sections.

The histopathology report of the result of subnipple tissue (frozen section and permanent H&E section) and NAC were in term of positive and negative. We classified the positive group of the gold standard as having either the tumor present in permanent section of subnipple tissues or in the permanent serial section of NAC. The positive cell types included invasive ductal carcinoma, ductal carcinoma in situ, Paget's disease, angiolymphatic invasion of malignant cell and atypical intraductal hyperplasia; and the negative group as having no tumor presented in permanent section of subnipple tissues or in the permanent serial section of NAC. The histology of the negative group were normal epithelium and ductal hyperplasia

Sample Size Estimation

This study was aimed at determining the accuracy (i.e. sensitivity and specificity) of frozen section of subnipple area as compared to the gold standard of permanent section of entire NAC. However, the main interest was on the validity of negative frozen section compared to negative permanent section of the entire NAC. That is, the primary objective of this study was to estimate the specificity of frozen section of subnipple area among patients with negative NAC. Sample size estimation was then based on the determination of 95% confidence interval (CI) of specificity. A sample of 35 patients with negative NAC would be enough to get a 95% CI for specificity of 90% with 10% error.

Data Analysis

1. Specificity and its 95% CI was calculated from those analysed data.

2. To evaluate the association between NAC involvement and various predictors, including type of carcinoma, differentiation, size, location, calcification, vascular and perineural invasion, multi or unicentricity and axillary lymph node status. Chi-square analysis (or Fisher's exact test) was used for qualitative predictors and unpaired t-test or Mann-Whitney U test

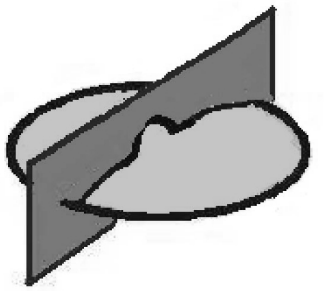

Figure 4

Table 1 Comparing Frozen section of subnipple tissue and the involvement of NAC

	Involvement of NAC (gold standard)		Total
	Negative	Positive	
Frozen of Subnipple			
Negative	28	2	30
Positive	1	15	16
Total	29	17	46

Table 2 Tumor Characteristics

Data	Method of Measurement		Mean ± SD		2-sided	
			Positive	Negative	p-value*	
Tumor size (mm.)	Specimen examination	Diameter	29.0 ± 10.9	21.1 ± 7.8	0.012	**
	Ultrasonography#	Diameter	26.8 ± 10.6	20.7 ± 7.7	0.028	**
	Mammography†	Diameter	31.1 ± 10.3	21.8 ± 8.3	0.005	**
	Pathological report specimen	Diameter	39.2 ± 13.0	28.9 ± 16.9	0.007	**
Tumor Distance (mm.)	Specimen examination	Tumor to Areola	17.0 ± 12.1	24.2 ±16.9	0.166	
		Tumor to Nipple	22.3 ± 13.7	30.5 ± 18.8	0.124	
	Mammography	Tumor to Nipple	26.9 ± 19.4	35.3 ± 20.4	0.141	
Ratio Size/Distance	Mammography	Length / Tumor to Nipple	1.92 ± 1.77	0.91 ± 0.74	0.009	**

* Mann-Whitney U test (Exact method)
** Statistically significance
# 2 unavailable datas and † 6 unavailable datas

Table 3 Tumor Characteristics

Predictor	NAC			2-sided p-value	Odd Ratio	95%CI*
	Positive	(%)	Total			
Location (Quadrant)						
Peripheral	16	36.4	44	1.000	1.75	0.10, 29.9
Central	1	50.0	2			
Cell type						
Non-DCIS	9	29.0	31	0.192	2.79	0.77, 10.01
DCIS	8	53.3	15			
Axillary Lymph node status						
No Metastasis	5	45.5	11	1.000	0.92	0.21, 3.91
Metastasis	10	43.5	23			
Angiolymphatic invasion						
Absent	9	33.3	27	0.757	1.45	0.43, 4.88
Present	8	42.1	19			
Perinueral invasion						
Absent	14	41.2	34	0.489	0.476	0.10, 2.08
Present	3	25.0	12			
Calcification						
Absent	11	36.7	30	1.000	1.03	0.29, 3.63
Present	6	37.5	16			
Multicentricity						
Absent	15	38.5	39	1.000	0.64	0.11, 3.72
Present	2	28.6	7			
Estrogen receptor						
≥ 25 %	9	40.9	22	1.000	0.91	0.25, 3.28
< 25 %	7	38.9	18			
Progesterone receptor						
≥ 25 %	4	25.0	16	0.188	3.00	0.75, 1.99
< 25%	12	50.0	24			
HER-2						
Present	2	33.3	6	1.000	1.00	0.13, 7.45
Absent	5	33.3	15			

*Fisher's exact test

for quantitative. All P values were two sided, with statistical significance evaluated at the .05 alpha level.

Results

Forty-six female patients were recruited during July 2003 to May 2004. Mean age of the group was 52.98 (34-76 years, SD 10.57). Twenty tumors were located in the left breast and twenty-six in the right breast. 29 (63.0%) tumors were in upper outer quadrant, 8 (17.4%) in upper inner quadrant, 5 (10.9%) in lower outer quadrant, 2 (4.3%) in lower inner quadrant and 2 (4.3%) in central quadrant. Modified radical mastectomy was done in 34 specimens, total mastectomy with sentinel lymph node dissections in 11 specimens and simple mastectomy in 1. The average tumor size was 32.74 mm (7-80 mm, SD 16.28) T1 = 14 cases, T2 = 26 cases and T3 = 6 cases. The pathological nodal status were pN0 in 24 cases, pN1 in 11 cases pN2 in 6 cases pN3 in 4 cases.

The histological cell types of the tumor were invasive ductal carcinoma (IDC) in 29 (63.0%), IDC with ductal carcinoma in situ (DCIS) in 7 (15.2%), IDC + mucinous component in 2 (4.3%), IDC + DCIS + Paget's disease in 1 (2.2%) and invasive lobular carcinoma and lobular carcinoma in situ in 1 (2.2%).

The size of subnipple tissues were 15.7 ± 5.7 mm in length, 9.3 ± 3.6 mm in width and 3.0 ± 1.7 mm in thickness. The pathological reports revealed 16.7 ± 8.1 major mammary ducts.

The subnipple tissues showed tumor involvement in 34.8%(16 specimens) by frozen section. The permanent serial sections of subnipple and NAC (gold standard) demonstrated 37.0% (17 specimens) tumor involvement.

The frozen section of subnipple tissue showed specificity of 96.6% (95%CI 82.2, 99.9), sensitivity of 88.2% (95%CI 63.6,98.5) and accuracy of 93.5% (95%CI 82.1, 98.6). The positive predictive value was 93.8% and negative predictive value was 93.3%.

The potential predictive factors of NAC involvement which included tumor size (by preoperative ultrasonography, mammography, specimen examination and pathological report specimen), tumor distance (tumor to areola and tumor to nipple), tumor location, histological cell type, axillary lymph node status, angiolymphatic invasion, perinueral invasion, calcification, multicentricity, estrogen receptor, progesterone receptor and HER-2 status were included for univariable analysis as shown in Table 2 and 3.

Discussion

The overall incidence of the NAC involvement was approximately 6 - 54%.[7] Simmons et al[7] reported 10.6% overall NAC involvement in 217 patients and reduced to 6.7% in a subgroup of patients with tumors <2 cm, peripheral tumors, and with two positive nodes or less.

Vyas et al[11] predicted a 16% (22/140) NAC involvement, while in all of those positive cases the tumour was within 2.5 cm of the areola. Crowe et al[16] took core specimen from NAC in fifty-four nipple sparing mastectomies and revealed neoplastic involvement in 6 of 54 (11.1%) frozen section analysis. Santini et al[12] studied 1,291 specimens and found 12% overall NAC involvement and 7% in those patients with early invasive stage I or II breast carcinoma.

The presentation of 37% tumor involvement makes surgeons uncomfortable to preserve nipple in breast cancer patients. However, in a selected subgroup with confirmation of negative frozen section, nipple preservation may be possible.

We found that tumor size and tumor size/distance ratio were strong predictor of nipple involvement (p = 0.005 and 0.009 respectively), as presented in Table 2. The tumor size measured by either preoperative ultrasound or mammogram could be used as a predictor of NAC involvement as an alternative to pathological measurement, but the distance from nipple to the nearest edge of tumor was not practical by ultrasound.

Other clinico-pathological factors such as tumor location, histological cell type, axillary lymph node status, angiolymphatic invasion, perinueral invasion, calcification, multicentricity, estrogen receptor, progesterone receptor and HER-2 status were not significant in predicting NAC tumor involvement (Table 3).

In our study, most tumor were in stage I, IIa and IIb in which the incidence of NAC involvement should be less. Meticulous serial section examination of NAC may increase detection rate of NAC involvement.

In our series, the frozen section of subnipple tissue showed specificity of 96.6% (95%CI 82.2, 99.9), sensitivity of 88.2% (95%CI 63.6, 98.5) and accuracy of 93.5% (95%CI 82.1,98.6). The data will provide more

information for intraoperative decision in the NAC preserving mastectomy procedure.

The gold standard in this study is the serial permanent section of NAC. The positive group included invasive ductal ccarcinoma, ductal carcinoma in situ, Paget's disease, angiolymphatic invasion of malignant cell and atypical intraductal hyperplasia (AIDH). Hence, AIDH is not a malignancy but reported as having higher risk for malignant change[17] and considered as risk factor of breast cancer.[18] Further study showed same molecular and genetic basis of AIDH and ductal carcinoma.[19] Then AIDH is categorized as positive cell type and should not be preserved in the NAC preserving mastectomy procedure. Negative group consisted of normal ductal cell and ductal hyperplasia without atypical cell.

Most tumors were located in upper outer quadrant (29 or 63%). The tumor in central quadrant was reported to have higher incidence of NAC involvement which was found in 1 of 2 cases (50 %) in our report.

Various clinicopathological predictors were collected and statistically analysed. The location, histopathological cell type, axillary lymph node status, angiolymphatic, perineural invasion, calcification, multicentricity and receptor status were not statistically significant. Tumor with component of DCIS cell type tended to have NAC involvement (7 in 15 cases) more than those without DCIS component (6 in 31 cases). Simmons et al[7] agreed that axillary lymph node status was one of the predictor for determining NAC involvement but it was not statistically significant in our study.

The tumor size and distance were established as significant predictors in several literatures. Suehiro S[20] reported 24 of 65 (36.9%) involvement of NAC which related to patient younger than 50 years and the tumor distance to the nipple was less than 4 centimeters. Simmons et al,[7] Vyas et al[11] proposed the same conclusion for nipple distance. Cense HA[21,22] found nipple involvement in up to 58% and suggested nipple sparing mastectomy for small tumour (T1) at a large distance (>4-5 cm) from the nipple.

Our study showed the significance of these two factors; tumor size by the specimen examination, ultrasonography, mammography and pathological report specimen (p value = 0.012, 0.028, 0.005 and 0.007)and Mammographic Size / Distance Ratio (p value = 0.009)

Conclusion

Frozen section of subnipple tissue and pre-operative measurement of tumor size and distance can predict the NAC involvement of breast cancer. Nipple sparing mastectomy in selected patients with favorable clinical predictors could be considered in negative frozen section group.

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