

## Imprint Cytology and Frozen Section in The Diagnosis of Breast Lesions

การตรวจเซลล์โดยวิธีตัดแปะและวิธีการตรวจชิ้นเนื้อแช่แข็ง

ในการวินิจฉัยรอยโรคของเต้านม

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### Abstract

**Introduction:** Frozen section has been accepted as a high reliable procedure for intraoperative diagnosis. The accuracy varies with the type of tissue and experienced pathologists. Recently, the utility of intraoperative imprint cytology becomes widely used because of its simplicity, rapidity and economy. Its accuracy is high, comparable to frozen section. The Department of Anatomical Pathology, Saraburi hospital has employed the two techniques on breast tissue submitted for intraoperative diagnosis. **Objective:** To compare the accuracy of frozen section and imprint cytology with permanent section for diagnosis of breast lesion. **Materials and methods:** From April 1997 to August 2003, one hundred and twenty three breast specimens submitted to department of Anatomical Pathology, Saraburi Hospital for intraoperative diagnoses were included into the study. All specimens, imprint and frozen section were carried out and the results were compared with the permanent sections. Those finding were used to find out accuracy, sensitivity, specificity, positive predictive value and negative predictive value of frozen section and imprint cytology. **Results:** Imprint cytology, as compared to permanent section, had 94.9% accuracy, 94.1% sensitivity, 96.9% specificity, 98.7% positive predictive value and 91.4% negative predictive value, while respective corresponding data for frozen section were 96.5%, 95.5%, 100%, 100%, and 91.8%. When intraoperative imprint cytology and frozen section were used together, it yielded 98.4% accuracy, 97.8% sensitivity and 100% specificity. **Conclusion:** Both the frozen section technique and imprint cytology showed high accuracy rates. The imprint cytology conjunction with frozen section examination are ensuring accuracy in rapid tissue diagnosis of breast with more rapidity and more simplicity.

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**บทคัดย่อ:** การตรวจเซลล์โดยวิธีตัดแปะและวิธีการตรวจชิ้นเนื้อแช่แข็งในการวินิจฉัยรอยโรคของเต้านม  
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**ภูมิหลัง:** วิธีการตรวจชิ้นเนื้อแช่แข็งเป็นวิธีการที่ได้รับการยอมรับว่ามีความแม่นยำสูงในการวินิจฉัยโรคระหว่าง  
 ผ่าตัด ความแม่นยำของการตรวจชิ้นเนื้อแช่แข็งขึ้นกับประเภทของชิ้นเนื้อที่ส่งตรวจและพยาธิแพทย์ที่มีประสบการณ์  
 ในปัจจุบันได้มีการนำการตรวจเซลล์โดยวิธีตัดแปะมาใช้วินิจฉัยโรคระหว่างผ่าตัดมากขึ้น เนื่องจากความรวดเร็ว  
 เตรียมง่าย และประหยัด มีความแม่นยำในระดับใกล้เคียงกับการตรวจชิ้นเนื้อแช่แข็ง กลุ่มงานพยาธิวิทยาภาควิภาค  
 โรงพยาบาลสระบุรี ได้นำวิธีการทั้งสองมาใช้ในการวินิจฉัยโรคระหว่างผ่าตัด **วัตถุประสงค์:** เพื่อเปรียบเทียบความ  
 แม่นยำของการวินิจฉัยรอยโรคของเต้านมด้วยการตรวจชิ้นเนื้อแช่แข็งและวิธีตัดแปะ เปรียบเทียบกับการตรวจชิ้น  
 เนื้อตามปกติ **วิธีการศึกษา:** ศึกษาชิ้นเนื้อของเต้านมจำนวน 128 รายที่ส่งตรวจระหว่างผ่าตัด ณ กลุ่มงานพยาธิวิทยา  
 ภาควิภาค โรงพยาบาลสระบุรี ระหว่างเดือนเมษายน 2540 ถึงเดือนสิงหาคม 2546 โดยชิ้นเนื้อทุกรายได้รับการตรวจ  
 ชิ้นเนื้อตามปกติ, การตรวจชิ้นเนื้อแช่แข็งและตรวจเซลล์โดยวิธีตัดแปะ จากนั้นนำผลการวินิจฉัยที่ได้มาหาค่าความ  
 แม่นยำ ความไว และความจำเพาะ **ผลการศึกษา:** ความแม่นยำ ความไว และความจำเพาะของการตรวจเซลล์โดย  
 วิธีตัดแปะ เปรียบเทียบกับการตรวจชิ้นเนื้อตามปกติเป็นร้อยละ 94.9, 94.1 และ 96.9 ตามลำดับ ขณะที่การตรวจ  
 ชิ้นเนื้อแช่แข็งเป็นร้อยละ 96.5, 95.5 และ 100 ตามลำดับ เมื่อใช้ทั้งสองวิธีร่วมกันได้เป็นร้อยละ 98.4, 97.8 และ 100  
 ตามลำดับ **สรุป:** การตรวจชิ้นเนื้อแช่แข็งและการตรวจเซลล์โดยวิธีตัดแปะของรอยโรคที่เต้านมมีความแม่นยำสูง  
 และทั้งสองวิธีจะช่วยให้การวินิจฉัยโรคได้แม่นยำมากขึ้น

## INTRODUCTION

The frozen section technique was first performed by Welch in 1891 and developed for intraoperative pathologic diagnosis by Wilson in 1905<sup>(1)</sup>. Following the development of the cryostat in 1960, the intraoperative frozen section examination was accepted as a highly reliable procedure for rapid histologic diagnoses of tissue specimens during surgery. This technique required a well-trained pathologist who had experience and knowledge of clinical medicine and pathology<sup>(2)</sup>. Most tissues can be submitted for frozen section and the accuracy also varies with the type of tissues. The frozen section

diagnosis in breast lesions remains a high accurate procedure; the false positive rate is essentially zero, the false negative rate is around 1.5% and the number of deferred diagnoses is around 5%<sup>(3-9)</sup>.

The imprint cytological diagnosis was favourably reported by Dudgeon and Patrick in 1927<sup>(10)</sup> and numerous subsequent studies confirmed the utility of this method. During the last few years, this technique has grown and is regarded as a useful adjunct to the frozen section with proven acceptable accuracy rates<sup>(10-22)</sup>. Its widespread utility may be due to its simplicity, rapidity and high degree of reliability, com-

parable to frozen section technique, when performed by well-trained and experienced individuals<sup>(23)</sup>.

At the Department of Anatomical Pathology of Saraburi hospital, the standard service for intraoperative consultation is frozen section and the author had performed imprint cytology from surgical specimens submitted for intraoperative diagnosis since 1997.

Among various origins of tissue received for frozen section, breast specimens have been the most frequent. Breast disease is extremely common and breast cancer remains one of the most common causes of female cancer death in Thailand. Early detection plays a most important role in improving the death rate from breast malignancy<sup>(24)</sup>. Toward achieving this goal, immediate diagnosis during operation has been requested by many surgeons for making therapeutic decisions. Because of these main reasons, the author conducted this study to compare the accuracy of frozen section and imprint cytology with permanent section of breast lesions.

## MATERIALS AND METHODS

From April 1997 to August 2003, imprint cytologic smears were obtained from 128 breast specimens submitted for frozen sections and were prepared by cytotechnologists at Department of Anatomical Pathology, Saraburi hospital. The specimen received was macroscopically examined and then sectioned. A glass slide was gently pressed on to the freshly cut surface of the specimen, avoiding a gliding movement, which will distort the shape of the cells. The imprint slide was immediately fixed in 95% ethyl alcohol for 1 minute and then stained with

Papanicolaou stain. On average, two imprints per case were prepared.

The frozen section was prepared by freezing the specimens, sectioned and stained with Hematoxylin & Eosin. On average, two or three slides per cases were prepared. The rest of the specimen was then fixed in 10 % formalin and was transferred for permanent histological technique.

The diagnoses of frozen and permanent sections were made immediately by either one of two pathologists at Saraburi hospital. All imprints were examined and analyzed subsequently by the author without prior knowledge of the results of frozen sections or permanent sections.

In each specimen, the imprint and frozen section diagnoses were compared with final pathological reports on permanent sections. From these results the accuracy, sensitivity, specificity, positive predictive and negative predictive values were calculated.

## RESULTS

**Permanent section.** Out of 128 breast lesions with final pathological diagnoses on permanent sections, 35(27.3%) of specimens were benign and 93(72.7%) were malignant. All diagnoses were reviewed in Table I.

**Frozen section.** Table II showed the comparison of frozen section and permanent sections. By excluding four deferred cases, 122 cases were analyzed. In 34/37 specimens (91.8%), frozen sections were correctly diagnosed as benign and while all 85 specimens (100%) were correctly diagnosed as malignant mammary neoplasm. False negative result was 3 cases

**Table I** Distribution of diagnoses for 123 breast specimens according to final permanent histologic diagnoses

Diagnosis	No.	Percentage
<b>Malignant</b>	<b>93</b>	<b>72.7</b>
- Invasive ductal carcinoma	81	
- Intraductal carcinoma	6	
- Mucinous carcinoma	4	
- Others	2	
<b>Benign</b>	<b>35</b>	<b>27.3</b>
- Fibrocystic disease	13	
- Fibroadenoma	6	
- Inflammatory lesion	8	
- Fibroadenosis	2	
- Gynecomastia	2	
- Others	4	

(2.3%), whose permanent sections revealed malignant phyllodes tumor in one case and invasive ductal carcinoma in two cases. On reviewing these slides, the causes of mistakes were found, the first case was freezing artifact and the others were sampling error. The number of deferred cases was 6 cases (4.6%)

**Table II** Comparison between frozen section and permanent histologic diagnosis of breast lesion

	Frozen section	
	Malignant	Non-malignant
<b>Permanent section</b>		
Malignant	85	3
Non-malignant	0	34

**Table III** Comparison between imprint cytology and permanent histologic diagnosis of breast lesion

	Imprint cytology	
	Malignant	Non-malignant
<b>Permanent section</b>		
Malignant	81	5
Non-malignant	1	32

whose correct diagnoses made on permanent section were shown.

**Imprint cytology.** Table III demonstrated comparison between imprint cytology and permanent sections. By excluding seven deferred cases, 119 cases were analyzed. In 32/37 cases (86.4%), imprint smears were correctly diagnosed as benign and 81/82 breast specimens (98.7%) were correctly reported as malignant tumors. False positive result was one case (0.8%). False negative result was noted in five cases (4.2%), whose the permanent sections showed invasive ductal carcinoma in four cases and sarcoma in the other one. On subsequently review, the interpretative errors were caused by insufficient cells.

Table IV gave the overall comparative analysis of frozen section and imprint cytology results. The diagnostic accuracy, sensitivity, specificity of frozen section technique in diagnosis of breast lesions were 97.5%, 96.5% and 100% respectively while those of imprint cytology were 94.9%, 94.1% and 96.9% respectively. The overall combined diagnostic accuracy was 98.4%, with a sensitivity of 97.8% and specificity of 100%.

**Table IV** Comparative analysis of frozen section and imprint cytology

	Frozen section (%)	Imprint cytology (%)	Combined frozen section and imprint cytology (%)
Accuracy	97.5	94.9	98.4
Sensitivity	96.5	94.1	97.8
Specificity	100	96.9	100
Positive predictive value	100	98.7	100
Negative predictive value	91.8	86.4	94.5

## DISCUSSION

The major indication for the use of frozen section in breast lesions is to encourage the surgeon to make an immediate therapeutic decision, possibly sparing the patient from the second operation. Frozen section diagnosis may have serious consequences for the treatment of the patient, so a high degree of accuracy is necessary. The previously published accuracy of frozen section technique of breast lesions had varied from 90-99%<sup>(2,4-9)</sup> and was confirmed by this study. The false positive rate was reported to be lower than 0.3% and the false negative rate was around 1.5%<sup>(3-6,19)</sup>. In this study, no false positive was made, the three false negative results (2.3%) were caused by freezing artifact and sampling error. They were proved to be malignant phyllodes tumor and invasive ductal carcinoma on permanent section. The previously documented number of deferred diagnoses was ranging from 0.5-5.4%<sup>(3-6)</sup>. Deferred diagnosis in this series was 6 cases (4.6%). Five cases were proved to be malignant and one was fibroadenosis. These cases are difficult to interpret.

Intraoperative cytology diagnosis of breast lesions by imprint method is of great value, as has been stressed by many authors. Several studies have demonstrated the validity of this technique, varying from 87.5-97%. In the series of Sakai and Lauslahti<sup>(25)</sup>, the results of cytodiagnosis and frozen section during operation in 400 cases, 196 of which were breast lesions. There was no false positive or false negative diagnosis whereas 15 diagnoses were deferred. Helpap and Tschubel<sup>(26)</sup> compared the results of imprint cytology with permanent section in 700 breast biopsies and showed the diagnostic accuracy of imprint cytology of 95%. They also mentioned that proliferating fibroadenomas, papillomas and inflammatory lesions might lead to false positive results in the imprint. Dutta et al<sup>(27)</sup> published imprint cytology diagnoses on 51 breast lesions. The diagnostic accuracy was 94.1% and no false negative result. Koshi et al<sup>(28)</sup> studied imprint cytology from core biopsy of 150 breast lesions which 59/62 lesions (95.1%) were correctly diagnosed.

Suen and associates<sup>(10)</sup> reported the imprint

cytological diagnoses in 1,258 cases, 473 of which were breast lesions. There was no false positive diagnosis but 20 false negative diagnoses. Sources of error included carcinoma with paucicellularity, tubular carcinoma and lobular carcinoma. The accuracy for breast lesions was 95.7%.

One of the largest series in the literature reporting results of intraoperative cytology was that of Scucchi et al<sup>(12)</sup>. They reported the diagnostic accuracy for breast imprint cytology of 99% as compared with frozen section among 2,250 cases, of which 1,197 were breast specimens. There was no false positive diagnosis. Three false negative diagnoses (0.25%) were correctly reported as in situ lobular carcinoma, tubular carcinoma and myxoid liposarcoma.

In this study, the diagnostic accuracy for imprint cytology was 94.9%. There was one false positive diagnosis (0.8%) which was due to hypercellular smear. Five false negative diagnoses (4.2%) were due to carcinoma with paucicellularity. On subsequent review of 9 deferred cases, the main reason for delayed diagnoses was inadequate cellularity.

Scucchi et al<sup>(10)</sup>, Shabaik et al<sup>(29)</sup>, and Suen et al<sup>(12)</sup> mentioned the advantages of imprint cytology as compared with frozen section as follows (1) rapidity of preparation with similar accuracy; (2) possibility of identifying malignancy confined to small specimens or small areas of large specimens; (3) a simple and inexpensive tool, not requiring any special instrument; (4) excellent preservation of cellular details, devoid of freezing artifact; (5) preservation of tissue from freezing alterations in definite sections in case of very small samples.

The limitations of imprint cytology employment may be in following situations; (1) it often does not allow one to distinguish in situ from infiltrating carcinoma; (2) in the presence of some lesions, such as the distinction which is based mainly on the presence of stromal infiltration and in case of large bowel adenomatous polyps which are not always related to cytologic atypia<sup>(10,12)</sup>.

The imprint cytology performed in this paper is not proposed as a substitute for intraoperative frozen sections but wished to study the accuracy of this method as previously stated by many authors and to be used as a learning tool toward fine needle aspiration cytology for cytotechnologists. The author strongly recommended the employment of the two methods in combination because of higher percentage of correct intraoperative diagnosis obtained than achievement from each method alone as documented by many published studies<sup>(10,12,26,27,29-32)</sup>. In this study, the combination of imprint cytology and frozen section yielded 98.4% accuracy thus proving their value in using together.

## SUMMARY

This study revealed the high diagnostic accuracy of frozen section as many previous reports. It also showed a good correlation between the imprint cytology diagnosis and corresponding final histology. Although several advantages of imprint cytology technique were mentioned and high accuracy was stated, total replacement for frozen section should be carefully considered due to some important limitations. The author emphasized the use of imprint cytology

examination and frozen section in combination because the value of the method is enhanced when it is used with frozen section diagnosis.

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