

Touch Impression Cytology Versus Frozen Section as Intraoperative Consultation Diagnosis

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The efficacy of Touch Imprint Cytology in the diagnosis of various pathological processes was evaluated and compared with Frozen Section in intraoperative consultation. We examined 60 cases from various sites of the body that were referred for intraoperative consultation. All cases were analyzed by Touch Imprint Cytology followed by Frozen section. We found diagnostic accuracy of both procedures comparable. The diagnostic accuracy in distinguishing benign from malignant lesions by combined procedures was 100%. There were no false positive or false negative cases. For diagnosing specific subtypes of malignancy, the diagnostic accuracy of each method alone was 96.6% with a sensitivity of 86% and specificity of 100% and the combined sensitivity 90%. Touch Imprint Cytology provided better cellular morphology and fewer artifacts. On the other hand Frozen Sections provided more tissue architectural details but frequently hampered by freezing artifacts. We conclude that both Touch Imprint Cytology and Frozen Sections are dependable intraoperative consultation diagnostic modalities. Touch Imprint Cytology alone may provide a correct diagnosis in vast majority of cases with minimal expense and without the need of sophisticated cryostat machine thus making it quite suitable for many hospitals where cryostat machines are not available.

Key words: Touch Imprint Cytology; Frozen Section; Intraoperative Consultation Examination.

Introduction

Intraoperative Consultation Examinations are required by surgeons from pathologists for immediate important decisions regarding the optimal extent of surgery. Surgeons particularly want to know whether a lesion is malignant or not. Both Frozen Section (FS) and Touch Impression Cytology (TIC) serve this purpose well.¹⁻¹² Both provide accurate results in minutes while the patient is under anesthesia. Surgeon then modifies his surgical plan based on the Intraoperative Consultation from Pathologist. While FS tissue architecture closely approximates permanent histology sections, enabling a degree of comfort, TIC provides better and crisp cellular details and even some tissue architecture. However there is still some reluctance on part of some pathologists most likely due to their inadequate experience to render a definite diagnosis on TIC alone. In this study we have compared the accuracy of TIC and FS using permanent histopathology sections as a gold standard.

Patients and Methods

We evaluated and compared TIC and FS in 60 Intraoperative Consultation Examinations. The

diagnosis on permanent paraffin histopathological sections was considered final against which both TIC and FS diagnosis were compared.

The study comprised a representative sample of cases from different sites submitted to the Surgical Pathology Laboratory of Pakistan Institute of Medical Sciences (PIMS) Islamabad for Intraoperative Consultation Examinations during August 99 to August 2002. Patient's samples with suspected tuberculosis were not included in this study. The reason being that the contamination of the cryostat leads to temporarily cessation of frozen section services for 2-3 days requiring defrosting, decontamination and again starting the machine which would reach the desired temperature of -20°C in 1-2 days. It should also be noted that tuberculosis is rather easily diagnosed on TIC alone. Patients in whom Hischsprung's Disease was suspected were evaluated only by FS and TIC was not performed as ganglion cells and thickened nerve fibers do not come out on slides. These cases were also, therefore, excluded.

Touch Imprint Cytology: The fresh tissue submitted for frozen section is first used for TIC. The excess of

fluid i.e. blood, saline water, cyst contents, was gently blotted with dry gauze to facilitate adhesions of the cells to the surface of glass. The tissue being held in forceps simply touched on the glass slide at several points without undue pressure or lateral movement. The slides were immediately immersed in absolute alcohol for at least 10 minutes. The slides were later stained with standard H & H stain.

Frozen Section: All fresh tissues submitted for Intraoperative Consultation Examinations were examined grossly and representative areas were sectioned and embedded in Optimal Cutting Temperature Compound [OCTC].⁸ The tissues were frozen in cryostat at -20 Celsius. The tissue sections are cut 5-6 um thick and picked up on glass slides. These were stained with routine H & E stains.

Study Design: All frozen section slides were assessed by a postgraduate resident followed by the consultant pathologist. Relevant clinical data was made available at the time of initial surgical procedure. All slides were assessed for quality preparation and degree of diagnostic accuracy by assigning scores as follows.

Specimen Quality Assessment: A point score was assigned to each FS and TIC as follows:

0 point...The preparation exhibited such poor quality that the diagnosis had to be deferred. The poor quality implied severe frozen section artifacts, excessive tissue folding by frozen section, marked cellular degeneration, or inadequate volume of cellular material on cytological examination.

1 point...The tissue sections were distorted or TIC smears were suboptimal due to borderline cellularity

2 point...The tissue section and smears with well preserved cellular details with minimal or no distortion.

Diagnostic Accuracy Assessment: The following points were allocated to denote degree of diagnostic accuracy.

0 point...The diagnosis was incorrect, with respect to benign versus malignant

1 point...Diagnosis was correct regarding benign versus malignant diagnosis but incorrect with respect to the final specific diagnosis i.e. carcinoma vs. lymphoma. OR

The diagnosis was deferred until examination of permanent section.

2 points...The diagnosis was essentially correct i.e. correct regarding benign versus malignant status and in terms of classification as carcinoma,

sarcoma, inflammation, but not absolutely specific e.g. carcinoma instead of lobular carcinoma of breast, sarcoma not otherwise specified, instead of leiomyosarcoma OR

The correct specific diagnosis was included in the differential diagnosis (e.g. a case of adenocarcinoma is reported "adenocarcinoma vs. large cell undifferentiated carcinoma"

3 points...The diagnosis was correct and specific, for example Non-Hodgkin's Lymphoma, leiomyosarcoma

Each case was assigned cytology and frozen section score based on the diagnostic accuracy and specimen quality.

Results

Our study included 31 [51.66%] malignant and 29 [48.33%] benign cases (Table I). The benign cases included neoplastic and non-neoplastic lesions. They represented wide spectrum of tissues and lesions. In 2 [3.3%] cases the diagnosis was deferred till permanent sections. The diagnostic accuracy in distinguishing benign from malignant lesion by combined intraoperative cytology and frozen section was 100%. There were no false positive or false negative cases. For diagnosing specific sub-types the diagnostic accuracy of each method alone was 96.6% with a sensitivity of 86% and specificity of 100%. The combined sensitivity of TIC and FS was 90%. When all 60 cases were considered cumulatively [$\chi^2 = p > .05$] there was no significant difference between accuracy of TIC and FS diagnosis

Scores for Accuracy of Diagnosis

Zero Score: There was no such case in our study.

1 Score: In these cases the diagnosis was correct in terms of benignity or malignancy but inaccurate in terms of specific type of lesion. There were 10 such cases. Two of the cases were deferred till permanent sections. Among the remaining 8 cases there was overlap in four cases as these had one point on both cytology and frozen section. Among the remaining four cases three cytology cases and one frozen section had one point.

There were only two cases in which cytology had a score of one while frozen sections carried score 2. These cases were those of gastric leiomyoma and abdominal fibromatosis. There was one case of retroperitoneal papillary tumor in which FS had a score of one while cytology had two points. In one case of Metastatic adenocarcinoma to omentum, the cytology had a score of one while frozen section was

diagnostic with a score of 3.

Table 1: Final Intraoperative Consultation Results According to Organ of Origin

Site	Cases	Percentage
Breast	21	35
Thyroid	10	16.6
Lymph node	8	13.3
Parathyroid	4	6.6
Mediastinal masses	3	5
CNS	2	3.3
Resection margin	2	3.3
Urinary Bladder	1	1.6
Ovary	1	1.6
Bone	1	1.6
Gastric	1	1.6
Retroperitoneal Masses	3	5
Omentum	1	1.6
Mesenteric Tissue	1	1.6
Abdominal mass	1	1.6
Total cases	60	100

2 Scores: A score of 2 was assigned to the cases where diagnosis was correct in terms of benignity or malignancy as well as to the broad range of specific type, however the subtype of the lesion i.e. adenocarcinoma versus squamous cell carcinoma could not be determined although possibility suggested in the differential diagnosis. There were 10 such cases. Among these, FS had nine while TIC had five cases. Two cases on TIC had a score of three (Chronic Granulomatous Inflammation of breast and Rhabdomyosarcoma of urinary bladder) while FS had two. In another two cases, TIC had a score of one as described above while FS had two. One case of retroperitoneal papillary tumor had a score of 2 on cytology and a score of one on frozen section.

3 Scores: 43/60 cases earned a score of 3. Out of these

vast majority (40/ 60) had 3 points on both cytology smears and frozen section. In remaining three cases, two frozen sections rated a point of 2 and cytology had 3 points. In one case (metastatic carcinoma), the cytology had a score of one while the frozen section had 3 points.

Here we present organ wise brief break down of the cases:

Breast: All twenty-one cases were correctly diagnosed. There were thirteen malignant and eight benign lesions. Both neoplastic and non-neoplastic lesions were included in the benign lesions. There were four cases of fibroadenoma and mastitis each. Of the total twenty one cytology diagnosis recorded in this study, all were correctly diagnosed; there were no false positive or false negative diagnosis or deferrals. Among the frozen section diagnosis on the same cases all were correct, with no false positive or false negative diagnosis.

Thyroid: The ten cases included seven cases of adenomatous goiter and three malignant lesions (Two cases of anaplastic carcinoma and one case of papillary carcinoma). There was no difficulty in making diagnosis in all the seven benign cases and in one case of papillary carcinoma both on cytology smears and frozen sections (Fig. 2a & 2b). In two cases, the diagnosis of "malignancy" was rendered on TIC and FS. The specific diagnosis of anaplastic carcinoma was made on permanent sections.

Parathyroid: Parathyroid glands were identified and easily differentiated from adipose, lymphoid and thyroid tissues in all cases, both on imprint preparation and frozen sections.

Lymph Nodes: There were 8 cases; five of these were submitted for evaluation of metastasis. Two of these lymph nodes showed metastatic pancreatic adenocarcinoma. The remaining three were free of tumor. Three lymph nodes were submitted for suspicion of lymphoproliferative disorder. Two Lymph nodes (cervical) were found to be reactive. One case of abdominal lymph node both on frozen section and touch preparation cytology showed malignancy. However it was difficult to determine the type of lymphoma. The permanent section provided the final diagnosis.

CNS: There were two cases of CNS. Both were correctly diagnosed on touch cytology and frozen section. One was a posterior fossa space-occupying lesion diagnosed correctly as medulloblastoma. In the other case metastatic malignancy was suspected and was confirmed on permanent section.

Mediastinum: Among the three mediastinal sections, while in the third case lymphoma was lymphomas in two cases the diagnosis was deferred and final diagnosis was rendered on permanent

Table 2: Diagnostic Accuracy Scores of Less Than 3

Cytology Scores	F.section Scores	Sites	Final diagnosis	Cases
1	1	Hilar lymph node	NHL	2
1	1	Abdominal mass	Follicular Lymphoma	1
1	1	Mesenteric tissue	Fibrosis	1
1	1	Thyroid	Anaplastic carcinoma	2
1	2	Abdominal mass	Fibromatosis	1
1	2	Gastric Growth	Leiomyoma	1
2	1	Retroperitoneal mass	Papillary tumor	1
1	3	Omentum	Metastatic Adenocarcinoma	1
2	2	Lymph node	Metastatic Adenocarcinoma	2
2	2	Brain Tumor	Metastatic adenocarcinoma	1
2	2	Ovary	Mucinouscyst adenocarcinoma	1
2	2	Retroperitoneal mass	Ganglioneuroblastoma	1
3	2	Urinary bladder	Rhabdomyosarcoma	1
3	2	Breast	Chronic Granulomatous Inflammation	1

Diagnostic accuracy scores: 0: wrong regarding malignant vs. benign, 1: correct regarding malignant vs. benign but not specific 2: Diagnosis close or included in the differential 3: Correct and specific diagnosis

diagnosed both on touch preparation and frozen section

Genitourinary Tract: This included one case of rhabdomyosarcoma of urinary bladder that was accurately diagnosed both on touch and frozen section.

Female Genital Tract: There was one case of ovarian tumor that was evaluated by touch and frozen section. A diagnosis of cyst adenocarcinoma was made. The mucinous nature was confirmed on permanent sections.

Bone: One case of Giant Cell tumor from the iliac crest was accurately diagnosed both on touch and frozen section (Fig. 3a & 3b)

Resection Margins: In two cases in which the resection margins were evaluated for malignancy, a diagnosis "Free of tumor" was made accurately both

on touch and frozen section. These two cases included squamous cell carcinoma of oral cavity and a fibrous histiocytoma of buttock. These findings were confirmed on permanent sections.

Miscellaneous: There were seven cases in this group including three of retroperitoneal tumors. One case of non-Hodgkin's lymphoma accurately diagnosed both on touch and frozen sections, while the second case of ganglioneuroblastoma was diagnosed as neuroblastoma on both techniques. The third case on TIC and FS as well as on permanent section was considered to be either a papillary adenoma or papillary hyperplasia of mesothelium.. The fourth case of gastric tumor on TIC and FS was considered either leiomyoma or fibrosis. On permanent section the definitive diagnosis of leiomyoma was made. The fifth case (A huge 20x20 cm mesenteric mass) on TIC and

FS was considered leiomyoma vs. benign fibrous tumor. On permanent sections the diagnosis of mesenteric fibrosis was rendered. The sixth case (mesenteric tissue), on touch preparation and frozen section was diagnosed as reactive fibrosis that was confirmed on permanent section. The seventh case (Omental tissue), on touch and frozen section was considered metastatic carcinoma. The permanent sections confirmed the diagnosis.

It was observed that the cellular details were much better preserved in cytology slides while architecture was better seen in frozen section. Overall TIC of Lymph node and CNS were of much better quality than FS. In 93% cases the cytology specimens were quite adequate and satisfactory. While in 83.5% cases frozen section preparations were considered satisfactory.

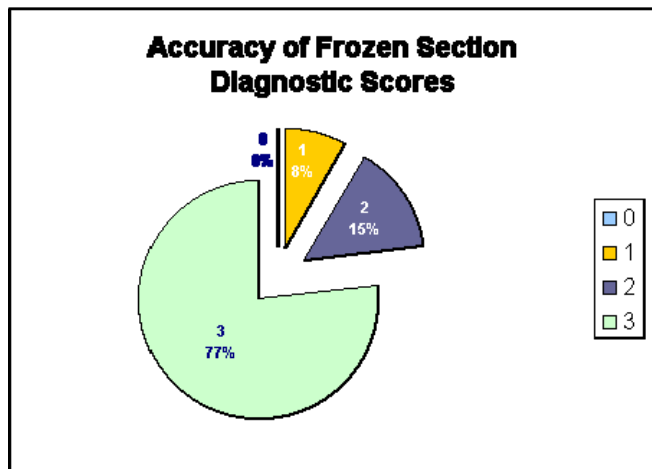
Points	Cytology cases		Frozen section cases	
	No.	Percentage	No.	Percentage
0	Nil	0	Nil	0
1	4	7	10	17
2	56	93	50	83

0: Material not adequate to distinguish between benign and malignant diagnosis 1: Tissue section or cytology smears were so distorted that diagnosis was rendered with difficulty

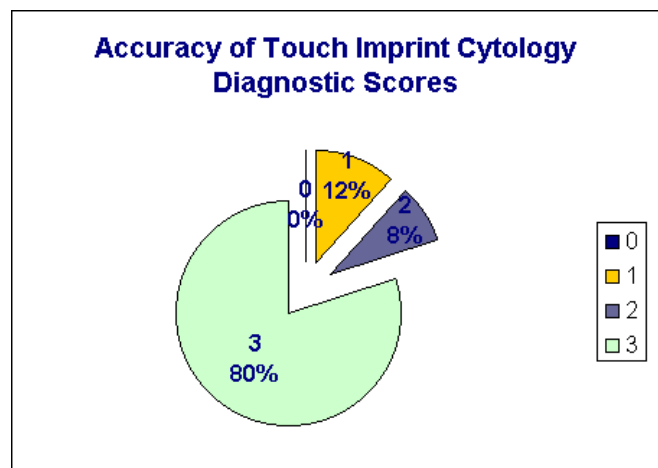
2: Material was adequate and satisfactory

Discussion

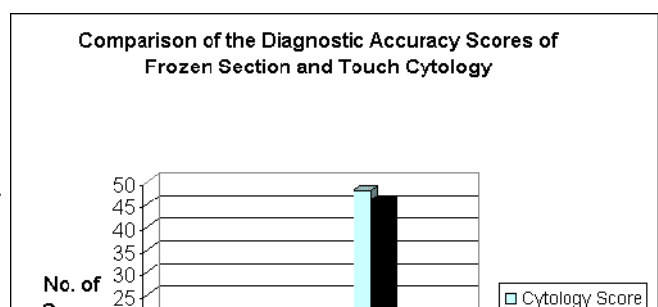
There are several reports in the world literature confirming the utility and efficacy of TIC as Intraoperative Consultation Diagnosis. Liu et al have investigated the utility of intraoperative touch preparation with comparison of frozen section in 122 cases.⁹ The rate of correct diagnosis for touch preparation was 88.5% as compared to 86.1% for frozen section. The rate of incorrect diagnosis for touch preparation was 4.1% as compared to 2.5% for frozen



Graph 1: Diagnostic Accuracy Scores: 0: Wrong regarding malignant vs benign. 1: Correct regarding malignant vs benign but not specific. 2: Diagnosis close or included in the differential. 3: Correct and specific diagnosis.



Graph 2: Diagnostic Accuracy Scores: 0: Wrong regarding malignant vs benign. 1: Correct regarding malignant vs benign but not specific. 2: Diagnosis close or included in the differential. 3: Correct and specific diagnosis.



Graph 3: Scores of specimen quality
0: Material not adequate to distinguish between benign and malignant diagnosis 1: Tissue section or cytology smears were so distorted that diagnosis was rendered with difficulty 2: Material was adequate and satisfactory

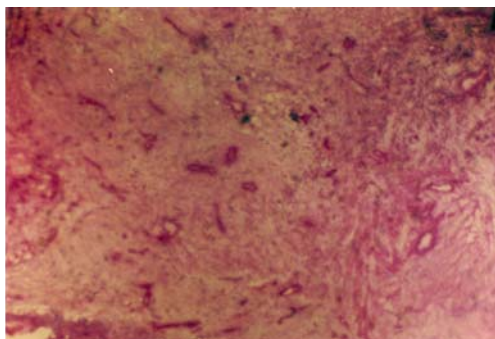
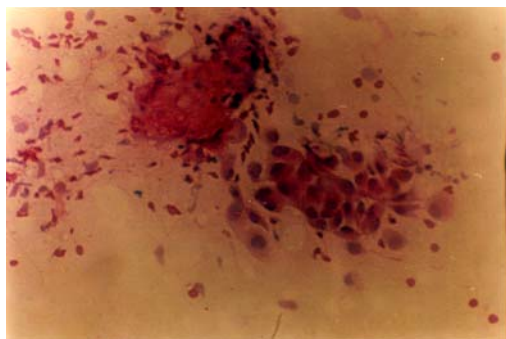


Fig. 1: a) TIC of Fibroadenoma of Breast (H & E X 100). b) FS of Fibroadenoma of Breast (H & E X 100)

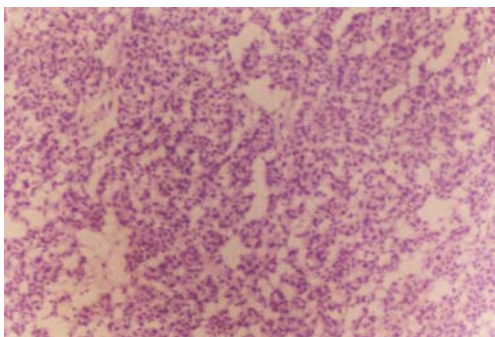
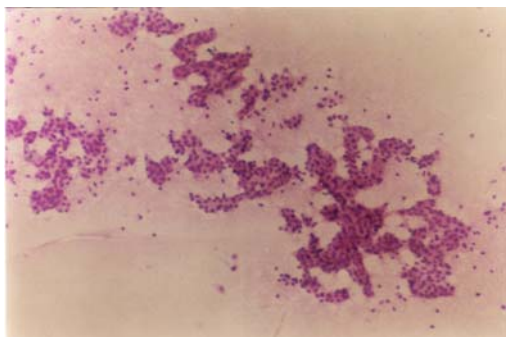


Fig. 2: a) TIC Papillary Carcinoma of Thyroid. (H & E X 100) b) FS. Papillary Carcinoma of Thyroid (H & E X 100)

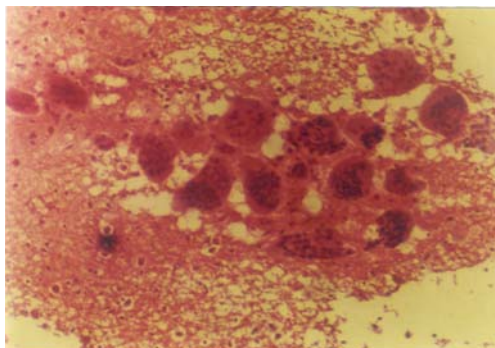
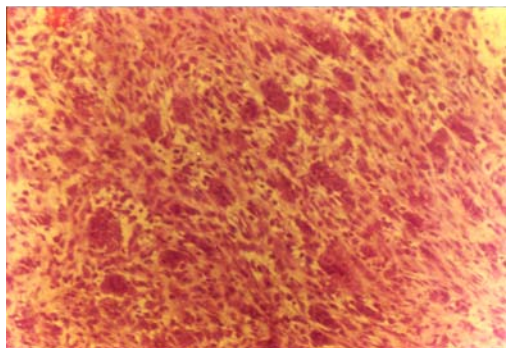


Fig. 3: a) TIC of Giant Cell Tumor. (H & E X 100) b) FS of Giant Cell Tumor (H & E X 100)

section while the undetermined diagnosis were 7.4% for touch preparation and 11.6% for frozen section.

For pathologists, with considerable experience of cytology, both touch and frozen section diagnostic accuracy rates were similar. The pathologist who lacked this experience had lower touch diagnostic accuracy. All pathologists who had cytological experience requested a frozen section only for cases with an undetermined touch preparation. In correct TIC diagnosis a frozen section was requested in 46.3% cases. Therefore touch and frozen section were complementary and both provided accurate diagnosis. Some pathologists consider that touch preparation may replace frozen section completely in over 50 % of cases.

It is well recognized, however, that the freezing and sectioning techniques of frozen section results in unavoidable distortions and artifacts, rendering diagnosis difficult in many instances. The greatest advantage of cytology examination is of not having such artifacts, resulting in superb nuclear and cytological details. Minimal tissue is needed for cytology examination. The diagnosis of very small lesions is therefore facilitated and tissue is saved for permanent section. Certain tissues that cannot be studied by frozen section i.e. bone, necrotic tissue and fat etc. give accurate results on touch preparations.

Scucchi et al compared 2,250 intraoperative cytology with frozen section with the final diagnosis achieved on paraffin sections.¹⁰ In 18 cases the diagnosis were deferred until the paraffin section at the time of Intraoperative consultations. The diagnostic accuracy in distinguishing benign from malignant lesions by combined intraoperative cytology and frozen section was 99.2%. The accuracy rate is higher than reported in large series based on frozen section alone. Sensitivity and specificity were respectively 98.2% and 100%. The diagnostic accuracy of each technique alone was 94.9%. For frozen section the sensitivity was 89.9% and specificity 97.9% as compared to the touch cytology, which had a sensitivity of 94.9%, and specificity of 96.8%.

Guarda carried out a comparative study of the two techniques and found the accuracy of cytology and frozen section 98.4% and 99.2% respectively.¹¹ The cytology was found to be more valuable in the field of neuro-pathology, lymph node and most of the epithelial tumors. It also provided better morphological preservation and details than frozen sections. Mair et al studied 206 cases from various body sites and found the accuracy of cytological examination comparable to that of frozen section analysis. The quality of cytological preparation was

significantly superior to that of frozen section. The combination of the techniques provided 99.5% accuracy rate.¹² Leesab and Haque also confirmed the utility of touch imprint cytology and its diagnostic accuracy comparable to frozen section.¹³

In our study the sensitivities of both methods were comparable. There were no false positive or false negative cases. For diagnosing specific subtypes of malignancy, the diagnostic accuracy of each method alone was 96.6% with a sensitivity of 86% and specificity of 100% and 90% combined sensitivity of touch imprint and frozen section. Touch cytology provided better cellular details and fewer artifacts. The diagnostic accuracy in distinguishing benign from malignant lesion by combined touch imprint cytology and frozen section was 100%. Both these methods complemented each other. As frozen section gives good architectural details, cytology provides excellent cellular details. The experience of the pathologist in interpretation of the touch preparations, is the mainstay in improving the diagnostic accuracy when both techniques are combined. Two cases were deferred and these were both mediastinal masses in which both cytology and frozen section were not able to make the definitive diagnosis.

The Usefulness of TIC is not limited to simple differentiation between benign and malignant lesions. TIC has been found quite reliable and useful in determination of surgical resection margins^{14, 15}, sentinel lymph nodes¹⁶, adenomatous goiter^{17, 18} and confirmation of parathyroid tissue.¹⁹⁻²¹ TIC has special role in brain and spinal cord lesions as these tissues are quite soft and various types of gliomas, meningiomas and other lesions yield cellular smears.²²⁻²⁴ In gynecology and obstetrics TIC is quite useful in differential diagnosis of ovarian tumors.²⁵⁻²⁶ In basal carcinomas of skin where a lot of frozen sections were needed to evaluate margins heavily taxing the time and energy of pathologist, TIC has provided satisfactory results without that much pain and sweat.²⁷

TIC has also been shown to be quite reliable in diagnosis of various soft tissue tumors. Its utility and accuracy, for instance has been reported in alveolar soft part sarcoma.²⁸ Lymph nodes are very difficult to interpret by FS, however TIC provide much more crispier and preserved cellular details enabling to render a confident diagnosis of lymphomas in most cases.²⁹ Finally in cases of metastatic tumors, TIC provides accurate diagnosis.³⁰

In conclusion both FS and TIC provide

accurate diagnoses. The accuracy is quite high in both, particularly in differentiating benign from malignant lesions. TIC reveals crisper cytological details. TIC has further advantage of being inexpensive, simple and quicker than frozen section.

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