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Original Article

Pathology Section

Diagnostic Accuracy of Frozen Section in Surgical Diseases with Critical Evaluation of Disconcordant Reports

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ABSTRACT

Introduction: The primary work of frozen section is to provide intraoperative rapid diagnosis, essential for patient management. The indications are to determine nature of lesion, margin status and staging of malignant tumours.

Aim: To find the accuracy of frozen section as compared to paraffin section. Detailed analysis of discordant cases so as to find the common errors responsible for discrepancy. Suggest ways to improve the overall accuracy of frozen section.

Materials and Methods: In this study, results of 172 consecutive cases of frozen sections were compared with the final paraffin section diagnoses in tertiary care, teaching hospital of Gujarat, India, over a period of one year and six months. Data were entered in Microsoft Excel sheet; diagnostic accuracy along with sensitivity and specificity were analysed using Medcalc statistical software, version 13.2, Belgium.

Results: Out of 172 cases, two cases were deferred for paraffin section (deferral rate 1.16%). The overall accuracy of frozen section was 95.3%. Amongst the discordant cases, interpretation error was responsible in 50%, block sampling error and specimen sampling error in 25% of cases each. The overall sensitivity and specificity was 93.4% and 98.44% respectively.

Conclusion: Frozen section is a reliable, cost-effective and useful technique if its limitations and indications are kept in mind. Detailed gross examination with multiple sections in suspicious malignant cases, broad diagnosis of benign/malignant in case of uncertainty and good interdepartmental communication helps to improve the accuracy.

Keywords: Intraoperative consultation, Paraffin section

INTRODUCTION

Frozen Section (FS) – Intraoperative pathology consultation has a great impact on surgeon's decision for surgical management of patient. The classical indication for frozen section examination is the need for an immediate decision during a surgical procedure in differentiating between benign and malignant neoplasms [1-3]. Frozen section is indicated for tissue identification with or without pathological process, ancillary test requirement like special stains or enzyme histochemistry, status of margins and/or identification of lymph node metastases in wide lesion excision/radical surgical specimens [4,5].

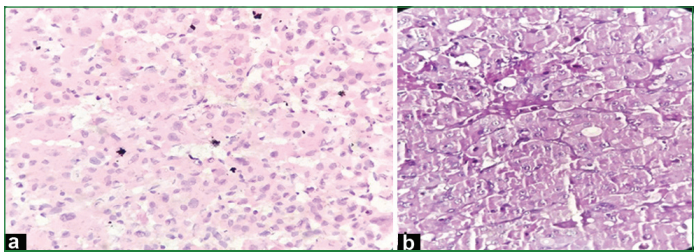
Frozen section is mainly intended for guiding the operating surgeon to decide intraoperatively about extent and/or adequacy of surgery thereby reducing the need for repeat surgery [6].

Frozen section and paraffin section diagnoses should be compared periodically as an internal quality indicator in histopathology [7]. Evaluating discrepancies, identifying deficiencies and resolving the underlying problems can improve the accuracy of frozen section [8]. Technical, sampling or interpretation errors are the three main reasons for diagnostic discrepancies between FS and paraffin section [9].

In this study, authors compared the results of FS and paraffin section with critical analysis and discussion about the reasons for discordance.

MATERIALS AND METHODS

In this hospital-based cross-sectional study, 172 cases of frozen sections received in the Department of pathology of a tertiary care hospital of Ahmedabad, Gujarat, India, over a period of one year and six months from various surgical departments were included after approval from Institutional Ethical Committee (GCSMC/EC/TRIAL/APPROVE/2017/18). All the clinical details and epidemiological data were taken from patient's files. Inclusion criteria were: All specimens sent for frozen section to diagnose presence of any pathological lesion, status of surgical margins in known malignant lesions, staging of advanced malignant lesions, detection of residual disease in post-lumpectomy breast specimens. Exclusion criteria were: tissue with extensive haemorrhage and necrosis and indeterminate frozen section diagnosis. Representative tissues taken during surgery were sent to Department of pathology without fixation. Tissues were examined grossly, selective bits taken and frozen in tissue freezing medium manufactured by Leica Biosystems and cut with YSI-119 Yorco Cryostat microtome – automatic in 4-5 µm-thick slices with disposable razors. The tissue sections are then picked up on glass slides with frosted edges and stained with Haematoxylin and Eosin (H and E) [Table/Fig-1a] (frozen section). The slides were then reported by pathologists using Nikon E 200 Light Microscope. The results were then communicated to the operating surgeon verbally as well as through printed hard copy as early as possible (within 30 minutes). Remaining tissues were fixed in 10% buffered formalin, processed for routine paraffin embedding, 4 µm thick sections were cut by Leica RM 2125 rotatory manual microtome and stained by H and E for routine histological examination and diagnosis [Table/Fig-1b] (paraffin section).



[Table/Fig-1]: Hurthle cell adenoma, thyroid gland (400x, H and E stain, frozen section (a) and paraffin section (b).

Frozen section diagnosis was compared with the paraffin section diagnosis. Frozen section results were grouped according to the anatomical site and indication of frozen section and then finally

categorised as concordant, discordant and inconclusive. Cases were considered as concordant if the frozen section and paraffin section diagnoses were similar, and discordant if diagnoses were different. Cases, where no definitive diagnosis was given on frozen section, were taken as inconclusive and not included for the calculation of accuracy.

STATISTICAL ANALYSIS

Diagnostic accuracy was calculated according to STARD 2015 guidelines [10]. Frozen section was considered as index test and paraffin section was considered as reference standard. Presence of pathological lesion was considered as presence of disease. Cases showing presence of disease in both frozen and paraffin section were considered as True Positive, absence of disease in both was taken as True Negative. Data were entered in Microsoft Excel sheet, analysed and accuracy of diagnosis along with sensitivity, specificity, positive predictive value and negative predictive value were calculated by using Medcalc stastistical software, version 13.2, Belgium.

RESULTS

Out of 172 patients, 2 (1.16%) cases were inconclusive and not included in the calculation for accuracy. Both cases were from oral cavity and were deferred due to inadequate tissue for reporting. Among 170 cases, 68 were male and 102 were female. The age range was wide and varied from 12-85 years. The most common indications for frozen section encountered in the institution in decreasing order of frequency were: (i) 104 cases for tissue diagnosis (ii) 55 cases for assessment of margins mostly in breast and oral malignancies (iii) 11 cases for assessment of the nodal status. Distribution of frozen section diagnoses according to anatomical site and indications are shown in [Table/Fig-2]. Oral cavity comprised 61 (35.88%) cases with 96.7% accuracy [Table/Fig-3]. Most common lesion was squamous cell carcinoma (57.14%) on both frozen and paraffin sections. Only two cases were discordant (false negative) where tumour were detected in deeper paraffin sections (block sampling error) [Table/Fig-4].

Anatomical sites	Total cases (N)	Diagnosis of lesion	Evaluation of margins
Oral cavity	61	21	40
Ovary	41	41	
Breast	25	18	7
Lymph node	11	11	
Uterus and cervix	11	9	2
Urinary tract	7	1	6
Liver and intestine	5	5	
Thyroid	3	3	
Salivary gland	2	2	
Others	4	4	
Total	170	115	55

[Table/Fig-2]: Distribution of frozen section examination according to anatomical sites and indications (N = 170).

In 41 (24.11%) ovarian cases, accuracy of frozen section was 95.1%. Most of the lesions were benign (73.17%), most common being serous cystadenoma followed by mucinous cystadenoma. A total of 21.9% (09/41) cases were malignant amongst which six cases were of serous adenocarcinoma and one case each of granulosa cell tumour, dysgerminoma and undifferentiated carcinoma. There were two discordant cases; one was diagnosed as mucinous cystadenoma on frozen section, later diagnosed as borderline on paraffin sections (specimen sampling error), other was diagnosed as mature cystic teratoma on frozen section whereas paraffin section showed histology of mucinous cystadenoma (misinterpretation error).

Anatomical site/organ	Number of cases	Concordant cases	Discordant cases	Accuracy (%)
Oral cavity	61	59	2	96.7
Ovary	41	39	2	95.1
Breast	25	24	1	96
Lymph node	11	11		100
Uterus and cervix	11	10	1	90.9
Urinary tract	7	7		100
Liver and intestine	5	4	1	80
Thyroid	3	3		100
Salivary gland	2	1	1	50
Others	4	4		100
Total	170	162	8	95.3

[Table/Fig-3]: Diagnostic accuracy of frozen section according to anatomical site.

Anatomical site	Frozen section diagnosis	Paraffin section diagnosis	Reason for discrepancy
Tongue	Dysplasia	Squamous cell carcinoma, superficially invasive	Block sampling error
Breast	Margins-free	Margins-Involved	Specimen sampling error
Liver biopsy	Atypia	Hepatocellular carcinoma	Misinterpretation
Parotid salivary gland	Pleomorphic adenoma	Malignant Peripheral nerve sheath tumour	Misinterpretation
Ovary	Benign cystic teratoma	Mucinous cystadenoma	Misinterpretation
Ovary	Benign mucinous cystadenoma	Borderline mucinous cystadenoma	Specimen sampling error
Uterus	Endometrial intraepithelial neoplasia (EIN) Grade 4	EIN 3	Misinterpretation
Mucosa – Lip	Dysplasia	Squamous Cell carcinoma, superficially invasive	Block sampling error

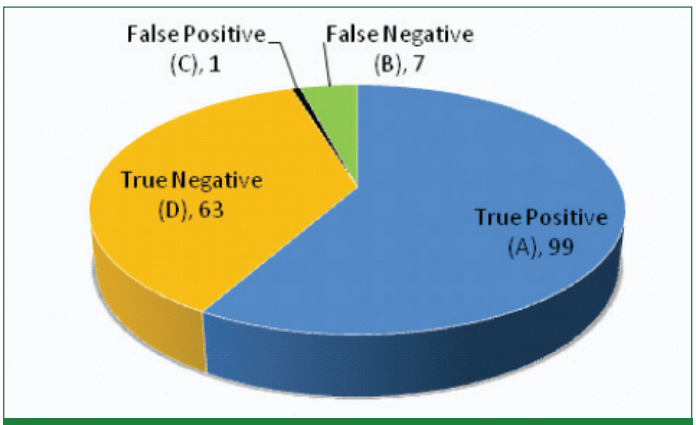
[Table/Fig-4]: Details of discordant cases.

Breast lesions comprised of 25 (14.7%) cases. Invasive ductal cell carcinoma comprised of 15 cases out of total 18 breast carcinoma cases (83.3%) and was the most common lesion diagnosed both on frozen as well as paraffin sections. In only one case false negative diagnosis was made in which negative margin on frozen turned out to be positive on paraffin section (specimen sampling error). This gave an accuracy rate of 96%.

Eleven lymph nodes were subjected for frozen section to assess the nodal status in cases of malignancy. There was 100% concordance between frozen section and paraffin section diagnosis. Out of 11 (15.71%) cases of uterine cavity and cervix, one was discordant – false positive; overdiagnosed as Endometrial Intraepithelial Neoplasm (EIN) Grade 4 on frozen and as EIN Grade 3 on conventional histology. Overall accuracy in Uterine cavity and cervix was found to be 90.9%. One out of five cases from Liver and alimentary tract and one out of two cases of salivary gland were discordant (false negative) due to interpretation errors. This gave an accuracy rate of 80% and 50% respectively. There were only seven cases of urinary tract and three cases of thyroid gland with 100% accuracy for the detection of lesion.

Out of 170 cases, diagnoses of 162 cases were concordant whereas eight cases were discordant; overall accuracy was 95.3%. Amongst the discordant cases, interpretation error was responsible in 50%, block sampling error and specimen sampling error in 25% of cases each. Frozen Section was able to identify the presence of pathological lesion in 99 cases (True Positive) and the absence of disease in 63 cases (True Negative). There was only one false positive case and

seven were false negative. [Table/Fig-5]. The overall sensitivity and specificity was 93.4% and 98.44%. The positive predictive and negative predictive value was 99.0% and 90.0% respectively.



[Table/Fig-5]: Showing comparison of Frozen section with Paraffin section. Sensitivity = A/A+B = 93.4%; specificity = D/C+D = 98.4%; PPV = A / A+C = 99.0%; NPV = D / B+D = 90.0%; Diagnostic Accuracy =A+D/A+B+C+D = 95.3%

DISCUSSION

Frozen Section (FS) is a rapid diagnostic tool for the intra- or peri-operative management of patient. It also guides the surgeon about the pathologic process, adequacy of resection, evaluation of margins, lymph node status with tissue identification [9].

The Standards for the Reporting of Diagnostic Accuracy Studies (STARD) statement provides guidelines to improve the quality of reporting of diagnostic accuracy studies [10]. In present study, authors have included most of the essential items of STARD 2015 guidelines to improve the completeness and transparency of diagnostic accuracy of frozen section. Overall accuracy of FS was 95.30% with 93.40% sensitivity, 98.44% specificity and 1.16% were of deferred diagnosis. This is comparable to results of other published studies [6, 11-16]. [Table/Fig-6]

Name of Study	Country	Study Period	No. of cases	Accuracy	Sensitivity	Specificity
Chbani L et al., [11]	Morocco	01 Year	261	95%	-	-
Hatami H et al., [12]	Iran	06 Years	306	97.96%	92.95%	99.55 %
Shrestha S. et al., [6]	India	05 Years	404	94.6%	94.56%	94.55 %
Abbasi F et al., [13]	Iran	07 Years	200	96.5%	93.1%	97.7 %
Ahmad Z et al., [14]	Pakistan	01 Year	356	97%	-	-
Chandramouleeswari K. et al., [15]	India	01 Year	51	92%	-	-
Patil P et al., [16]	India	02 Years	100	96.96%	97.23%	96.30 %
Present Study	India	1.5 years	170	95.30%	93.40%	98.44%

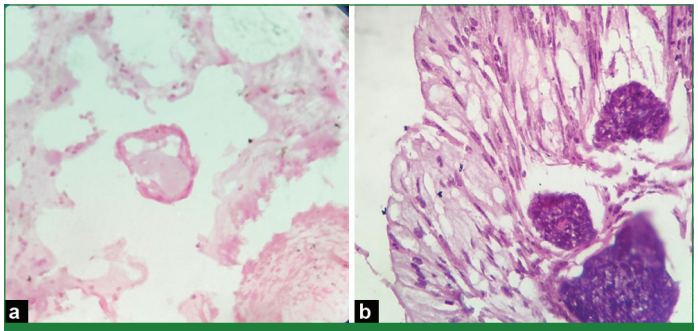
[Table/Fig-6]: Comparison of various studies with present study on overall accuracy, sensitivity and specificity.

Deferred rate in the present study was 1.16% which is comparable to that reported in various published studies i.e. 0.04%-6.7% [11]. Deferral rates may vary according to clinical expertise and also clinical setting and the type of specimens encountered [12]. The pathologist should also know the limitations of frozen section and the ability of his or her technicians in the respective laboratory. The Pathologist should not be afraid to defer in his or her diagnosis when the situation warrants it. Whenever a deferred diagnosis is made, it should be ignored and the surgeon should proceed as though the frozen section had never been performed [17].

Discordant rate was 4.7% in the present study and the causes of error were interpretation error (50%), gross sampling error (25%) and block sampling error (25%).

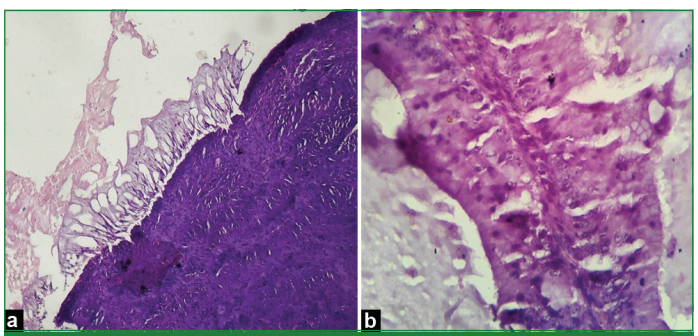
Most common cause of discordant diagnosis was interpretation error (50%). Interpretation errors may result from artefacts of the freezing procedure and rarity of the lesion or the inexperience on the part of the pathologist [6]. In various studies published interpretation errors comprised as the main cause for discrepancy ranging from 40% to 66% which is comparable to the present study [6,8,12,16].

Specimen from ovary demonstrated misinterpretation of one case as benign cystic teratoma and later diagnosed as mucinous cystadenoma on paraffin section. Tumour was predominantly cystic with thin wall and microscopically showed columnar epithelium as well as flattened epithelium misinterpreted as squamous epithelium at places. This led to a discrepant diagnosis of Mature Cystic Teratoma on FS [Table/Fig-7a,b]. Other case of Borderline Mucinous cystadenoma of ovary was reported as Benign Mucinous Cystadenoma on FS due to specimen sampling error [Table/Fig-8a,b]. Small foci of mild to moderate nuclear atypia or nuclear stratification involving only a few low power fields (≤5-10% of tumour) can be seen in mucinous cystadenoma [18]. Criteria to type the borderline tumours of ovary include stratification of lining epithelium (1-4 layers), nuclear atypia and atypical mitosis. These may be missed due to distribution of multiple focal lesions and freezing artefacts causing difficulty in identifying nuclear atypia and mitoses on FS. Coffey D et al., state that the accuracy is lower in mucinous tumours of ovary [19]. Generous sampling, particularly from solid portion of ovarian tumour, is required to rule out invasion in mucinous tumour [19].



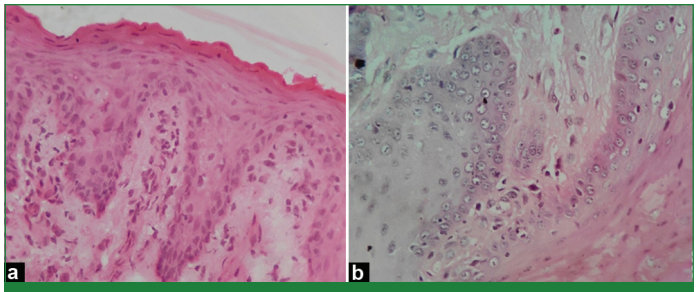
[Table/Fig-7]: (a) Frozen section of ovarian cystic mass showing cysts filled with mucin, lined by columnar epithelium with areas showing compressed lining epithelium misinterpreated as Squamous epithelium (right lower corner) leading to report as Benign Mature Teratoma (400x, H and E stain). (b) Benign Mucinous cystadenoma of ovary showing tall columnar mucinous epithelium with psuedostratification. (400x, H and E stain, paraffin section).

In the two discordant oral cavity lesions, one belonging to tongue and other to lip mucosa were underdiagnosed as dysplasia on FS and turned out to be superficially invasive squamous cell carcinoma [Table/Fig-9a,b,10a,b]. This may be due to deep seated nature of lesion, thus going unnoticed on FS and detected by deep sectioning on paraffin section. Also, dysplasia or well differentiated malignancies of oral cavity are associated with dense inflammation making it difficult to identify invasion on FS leading to false negative result. The lesions that most often lead to diagnostic disagreements are well differentiated malignant tumours that can be confused with proliferative conditions, poorly differentiated benign tumours and malignant lesions with associated inflammatory processes [2].

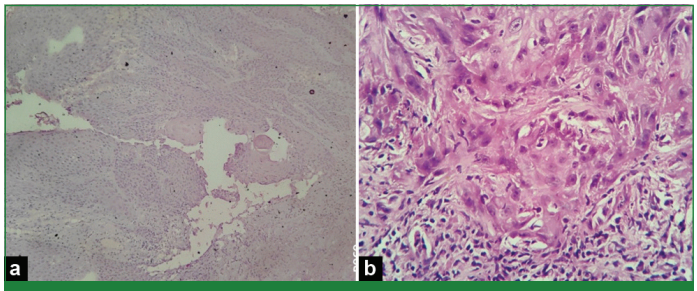


[Table/Fig-8]: (a) Mucinous cystadenoma of ovary (400x, H and E stain, Frozen Section) (b) Borderline mucinous cystadenoma of ovary, areas showing stratification of lining epithelium with nuclear atypia. (400x, H and E stain, paraffin section).

Margin status of a Breast specimen was given a false negative diagnosis due to specimen sampling error. This was because of poor



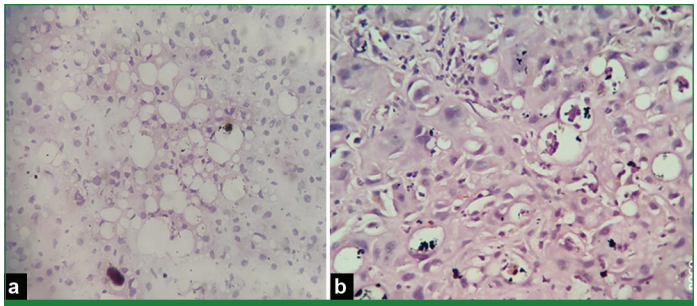
[Table/Fig-9]: (a) Squamous epithelium (tongue) showing moderate dysplasia. (400x, H and E stain, Frozen Section) (b) Superficial microinvasive Squamous cell carcinoma, tongue. (400x, H and E stain, Paffin Section).



[Table/Fig-10]: (a) Squamous epithelium (labial mucosa) showing moderate dysplasia with well defined basal epithelial layer and focal intraepithelial pearl formation. (400x, H and E stain, Frozen Section) (b) Well Differentiated Squamous Cell Carcinoma (labial mucosa) with pushing borders. (400x, H and E stain, Paffin Section).

selection of representative tissue in a large specimen and lack of communication between the pathologist and the operating surgeon.

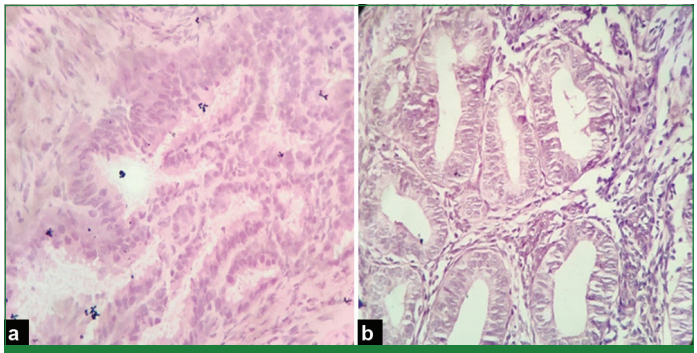
In a case of liver biopsy, hepatocellular carcinoma was misinterpretated as atypia on frozen section [Table/Fig-11a,b]. The factors affecting were lack of clinical information, small size of tissue and freezing artefacts.



[Table/Fig-11]: (a) Frozen section of biopsy from liver mass showing fatty change with moderate dysplastic hepatocytes (400x, H and E stain) (b) Paffin section of biopsy from liver mass showing thick trabeculae with sinusoidal pattern of atypical hepatocytes – Hepatocellular Carcinoma (400x, H and E stain).

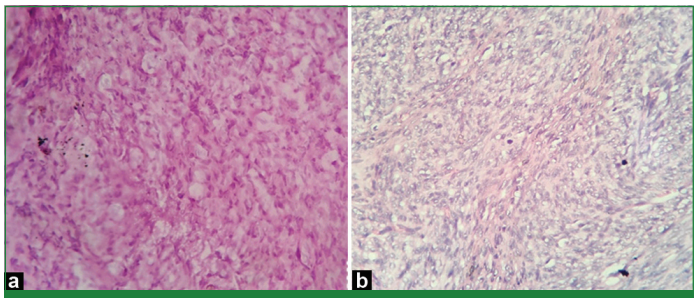
Only one false positive case in the present study was reported and was of Endometrium, Uterus.

An EIN 3 lesion was misinterpretated as EIN 4 on FS [Table/Fig-12a,b]. This could be due to drying artefacts and bloated cell morphology due to water retention in frozen section.



[Table/Fig-12]: (a) Frozen section of endometrium showing complex architecture of endometrial glands with marked nuclear atypia reported as EIN 4 with no evidence of invasion. (400x, H and E stain) (b) Paffin section of endometrium showing complex architecture of endometrial glands with moderate nuclear atypia reported as EIN 3 with no evidence of invasion (400x, H and E stain).

In salivary gland, a case of malignant peripheral nerve sheath tumour of parotid gland was underdiagnosed as pleomorphic adenoma on FS due to interpretation error [Table/Fig-13a,b]. The contributory factors were rare site of presentation, lack of clinical information, difficulty in assessing mitotic counts on FS and heterogeneity of tumour. Coffey D et al., [19] state that the accuracy of FS is lower in mesenchymal tumours. Generous sampling particularly from solid portion is required for the mitotic counts in mesenchymal tumour.



[Table/Fig-13]: (a) Frozen section of parotid mass showing spindle cells mimicking myoepithelial cells with myxoid like areas misinterpretated as pleomorphic adenoma. (400x, H and E stain) (b) Paffin section of parotid mass showing fascicles of malignant spindle cells with atypical mitosis – Malignant Peripheral Nerve Sheath Tumour. (400x, H and E stain).

In institutes where rotatory posting of pathologists for reporting frozen section, periodical review by a senior pathologist is recommended. This helps to maintain the quality and improves the diagnostic accuracy of frozen section [20].

LIMITATION

Accuracy of frozen section varies with the anatomical site, in this study we assessed the overall accuracy as total sample size was small and also site wise calculation was not feasible as cases for sites like thyroid, salivary gland, liver, intestine, skin and brain were very less. Our Institute is a comparatively new, upcoming referral centre and this could be the cause of small sample size. There were no sources of potential bias or statistical uncertainty.

Clinical implications with intended use of frozen section: Frozen section provides rapid gross and microscopic diagnosis but should not be used as a replacement to paraffin section as it has its own limitations. It is useful for identification of tissue, histological diagnosis along with lymph node and margin status. FS should be an elective procedure, not an emergency one due to difficulty of procedure and availability of trained technician. The pathologist should report with overall clinical correlation and should give only information necessary for the surgical management like benign vs malignant. The surgeon should not give undue emphasis to histomorphological diagnosis as it does not affect the surgical resection.

CONCLUSION

Frozen section is a reliable, cost-effective and useful technique if its limitations and indications are kept in mind. Good interdepartmental coordination with communication is recommended to reduce deferral rates, discordant diagnoses and for rapid diagnosis necessary for optimum patient care. In case of clinical suspicion of malignancy, multiple sections must be examined and in case of uncertainty during interpretation, opinion of second pathologist should be taken. A broad diagnosis of benign or malignant without subtyping or a judicious deferral is the best option in uncertain cases and serves the patient's best interest. Continuous monitoring with quality assessment by a senior pathologist helps to improve the accuracy of FS and reduce the errors.

REFERENCES

[1] Sienko A, Allen TC, Zander DS, Cagle PT. Frozen section of lung specimens. Arch Pathol Lab Med. 2005:129(12):1602-09.
[2] Saltzstein SL, Nahum AM. Frozen section diagnosis: accuracy and errors; uses and abuses. Laryngoscope.1973;83(7):1128-43.

[3] Makay O, Icoz G, Gurcu B, Ertan Y, Tuncyurek M, Akyildiz M, et al The ongoing debate in thyroid surgery: should frozen section analysis be omitted? Endocr J. 2007; 54(3):385-90.
[4] Dehner LP, Humphrey PA, Pfeifer JD. Chapter 52. In: Hull ME, Humphrey PA, Pfeifer JD editors. Washington Manual of Surgical Pathology. 2nd edition. Missouri: Lippincott Williams and Wilkins; 2012.
[5] Susan CL .Manual of Surgical Pathology. Second edition. Elsevier; 2006, p 49-69.
[6] Shrestha S, Basyal R, Pathak TSS, Lee M, Dhakal H, Pun C, et al. Comparative Study of Frozen Section Diagnoses with Histopathology. Post Graduate Medical of NAMS. 2009;9(02):01-05.
[7] Geramizadeh B, Larijani TR, Owji SM, Attaran SY, Torabinejad S, Aslani FS, et al., Accuracy of intraoperative frozen section consultation in south of Iran during four years. Indian J Pathol Microbiol. 2010;53(3):414-17.
[8] Mahe E, Ara S, Bishara M, Kurian A, Tauqir S, Ursani N, et al. Intraoperative pathology consultation: error, cause and impact. Can J Surg. 2013;56(3):E13-18.
[9] Jaafar H. Intra-operative frozen section consultation: concepts, applications and limitations. Malays J Med Sci. 2006;13(1):04-12.
[10] Bossuyt PM, Reitsma JB, Bruns DE, Gatsonis CA, Glasziou PP, Ilrwig L, et al STARD 2015: an updated list of essential items for reporting diagnostic accuracy studies. BMJ. 2015;351:h5527.
[11] Chbani L, Mohamed S, Harmouch T, El Fatemi H, Amarti A. Quality assessment of intraoperative frozen sections: An analysis of 261 consecutive cases in a resource limited area: Morocco. Health. 2012;4(7):433-35.
[12] Hatami H, Mohsenifar Z, Alavi SN. The diagnostic accuracy of frozen section compared to permanent section: A single center study in Iran. Iran J Pathol. 2015;10(4):295-99.
[13] Abbasi F, Yekta Z, Aryan A. Accuracy of frozen sections. Iranian Journal of Pathology. 2012;7(1):03-08.
[14] Ahmad Z, Barakzai MA, Idrees R, Bhurgri Y. Correlation of intraoperative frozen section consultation with the final diagnosis at a referral center in Karachi, Pakistan. Indian J Pathol Microbiol. 2008;51(4):469-73.
[15] Chandramouleeswari K, Yogambal M, Arunalatha P, Bose JC, Rajendran A. Frozen and paraffin sections- Comparative study highlighting the concordance and discordance rates in a tertiary care centre. IOSR Journal of Dental and Medical Sciences. 2013;12(5):26-30.
[16] Patil P, Shukla S, Bhake A, Hiwale K. Accuracy of frozen section analysis in correlation with surgical pathology diagnosis. Int J Res Med Sci.2015; 3:399-04.
[17] Khoo JJ. An audit of intraoperative frozen section in Johor. Med J Malaysia. 2004;59(1):50-55.
[18] Christopher D.M. Fletcher. ch-Tumours of the female genital tract, part A ovary. In: Zaloudek CF editor. Diagnostic Histopathology of Tumours. 3rd edn.Vol 1, Missouri: Elsevier; 2007: p567-51.
[19] Coffey D, Kaplan AL, Ramzy I. Intraoperative consultation in gynecologic pathology. Arch Pathol Lab Med. 2005;129:1544-57.
[20] Raab SS, Tworek JA, Soures R, Zarbo RJ. The value of monitoring frozen section . permanent section correlation data over time. Arch Pathol Lab Med. 2006;130:337-42.

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