Ethiopian Medical Journal 2024, 62(3)
Open access articles distributed in terms of
Creative Commons Attribution Licence [CC by 4.0]
ISSN 0014-1755 eISSN 2415-2420 Debebe et al.

Original Article

Practicality of Frozen Section at Tikur Anbessa Specialized Hospital: A Pilot Study

Melat Debebe[1*], Wondwossen Ergete[1], Bethelhem Shiferaw[3], Veronica Afework[2], Senait Ashenafi[1], Amanuel Damie[1]

[1]Department of Pathology, School of Medicine, College of Health Sciences, Addis Ababa University
[2]Department of Surgery, School of Medicine, College of Health Sciences, Addis Ababa University
[3]Department of Gynecology, School of Medicine, College of Health Sciences, Addis Ababa University

Corresponding authors*: melat.debebe@aau.edu.et

Abstract

Background: *Frozen section is an intraoperative rapid diagnostic tool that could direct a patient's intra- or post-operative therapy. Given the importance of frozen section in the management of surgical patients and the lack of published research in Ethiopia, it is prudent to evaluate its efficacy in our setting.*

Methods: *A prospective diagnostic accuracy study was done on 39 samples from June 2022 to December 2022. Cases were selected based on suspicion of a neoplasm and where there was an indication to perform frozen section. Data analysis was conducted using SPSS version 23. Findings were demonstrated using text, charts and diagrams.*

Results: *The accuracy, sensitivity, specificity, positive predictive value, and negative predictive value of frozen section as opposed to histopathological diagnosis using paraffin-embedded samples was found to be 92.3%, 88% and 95.2%, 94.1%, 90.9%, respectively. The mean turnaround time for frozen section was found to be 23 minutes.*

Conclusion: *The accuracy, ability to detect true positive cases, and ability to exclude false-positive cases of frozen section diagnosis in this research are consistent with most worldwide quality assurance data for frozen section. Despite its constraints, frozen section is a highly dependable method when carried out by experienced personnel, in the study context.*

Keywords: *Frozen section, Intra-operative Consultation, Accuracy, Sensitivity, Specificity, Ethiopia*
Citation: *Debebe M, Ergete W, Shiferaw B et al Practicality of Frozen Section at Tikur Anbessa Specialized Hospital: A Pilot Study Ethiop Med J 62 (3) 181-188*

Submission date : *23 February 2023 Accepted: 03 June 2023 Published: 01 July 2024*

Introduction

The practice of examining tissue removed by surgery pathologically did not become standard until the late 19th and early 20th centuries (1). Clinicians had limited resources aside from the patient's medical history and physical evaluation to assist in identifying neoplasms. Over time the use of x-ray and CT scan have helped in the diagnosis of malignant tumors. Despite advanced technologies, a definitive diagnosis would still need histopathology (1).

Assessing tissue obtained after operation following formalin fixation of the excised tissue usually takes days. In specific situations, surgeons need immediate histopathologic diagnosis during surgery to make intra -operative decisions. As a result, they order frozen section diagnoses (2, 3, 4).

Frozen section (FS) is an intraoperative examination of a tissue, which aims at histologically assessing small fragments of tissue in which there is a diagnostic doubt (5). FS offers quick diagnoses for directing patient care during surgery, including identifying unknown pathological conditions, assessing surgical margins, detecting lymph node metastases, and determining tissue type (4). FS should not be treated as an emergency procedure due to the practical aspects of the procedure that require preparation and the availability of the technician at the time of the operation. Therefore, an appointment at least a day before the operation needs to be made with the pathologist (4). Periodic reassessment of FS diagnosis by comparing it with the final diagnosis is valuable for pinpointing potential errors and preventing

their recurrence (3). The correlation of intra-operative FS diagnosis with final diagnosis on permanent section, formalin fixed paraffin embedded (FFPE) tissue, is an integral part of quality assurance in surgical pathology (6). Most studies indicate a frozen section accuracy of over 90%, with variation based on the specific organ being analyzed (5). Reasons put as the main causes for discrepancies were either misinterpretation of the original FS (31.8%), absence of diagnostic tissue in the frozen material but present in the un-sampled tissue or in the corresponding permanent section (31.4%) (6,7). FS serves as a rapid tool for guiding intra- or peri-operative patient management, but it does not supplant FFPE tissue technique. Its restrictions, such as limited sampling and technical challenges in obtaining high-quality tissue, affect the pathologist's interpretation of the section (1). The method remains dependable when performed by skilled practitioners. In Ethiopia, the use of FS as intraoperative consultation is near to zero. The Department of Pathology at School of Medicine, College of Health Sciences (CHS) is in preparation to start this procedure at Tikur Anbessa Specialized Hospital (TASH). This pilot investigation seeks to establish the practicality and accuracy of frozen section analysis in Ethiopia, as there is significant lack of research on this topic in the country. It will be the first research of its kind and serve as a foundation for future studies in this area.

Methods and materials

Study design

A pilot study on the diagnostic accuracy of FS carried out at the Department of Pathology, CHS at TASH, Addis Ababa, Ethiopia. A prospective diagnostic accuracy pilot study of intraoperative
FS specimens sent to the department from June 2022 to December 2022 were included. The project was initiated at the Department of Pathology, CHS, TASH, Addis Ababa, Ethiopia as it receives pathology specimens from all over the country providing large-scale pathology-based data (8).

Study population

Patients undergoing surgery having an indication for frozen section due to suspicion of a neoplasm were further selected, after which study population was chosen according to the eligibility criteria.

Eligibility criteria

All specimens sent to the pathology department for FS analysis were included in the study. Samples with inadequate tissue for diagnosis, samples with no follow up resection specimen, cases from emergency operation, and patients with age less than 18 years due to difficulty of obtaining parental consent for a practicality study were excluded from the study.

Sample size determination and sampling technique

In determining the sample size for this study,the rule of thumb was utilized, taking into consideration the fact that pilot studies need a minimum of 30 samples or greater to estimate a parameter (9). Samples were collected based on suspicion of a neoplasm and an indication to conduct FS including clinical features, the presence of potential contributing factor or result of preceding test (10).

Data collection procedure

Intraoperative FS specimens sent to the department from June 2022 to December 2022 were included. Intraoperative tissue was transferred to the pathology department in normal saline, and the time of sample receipt was recorded. Representative sample was taken to the cryostat machineand frozen at -25°C. Slides were prepared for evaluation by two senior pathologists and an expert pathologist. Findings were reported to the surgeon while still in surgery. After the frozen section procedure, both frozen and any remaining non-frozen tissue were preserved in a 10% formalin solution and forwarded to thehistopathology lab for creating permanent paraffin-embedded sections. FFPE slides were again further evaluated by two senior pathologists and one expert pathologist (Flow diagram 1). Upon comparing the FS report provided during surgery with the final histopathology report of the FFPE sections, the accuracy rate, sensitivity, and specificity of the FS reporting were evaluated (2,11). Using SPSS 23, data were statistically analyzed, and evaluation of sensitivity, specificity, positive and negative predictive values of the test was conducted.

Ethical considerations

The study received ethical approval from the institutional review board (IRB) (028/22/Patho), confirming that ethical principles were followed. Patient anonymity was maintained throughout the research by using coded identifiers to ensure patient privacy. To protect data, strict safeguards were put in place, such as limiting access to authorized researchers, storing physical and electronic records securely, and anonymizing participant identification during data analysis and reporting.

Results

A total of 39 samples were collected. The age of patients ranged from 29 years to 68 years. Mostpatients (46.1%) were between the ages of 36 and 45 (Table 1). 35 (89.7%) patients were femalewhile the rest 4 (10.3%) of the patients were male (Table 1).

Table 1: Patient demography and turnaround time for FS, Addis Ababa, Ethiopia (n=39)

		Count
Age groups	26-35 years	10
	36-45 years	18
	46-55 years	6
	56-65 years	4
	>66 years	1
Sex	Female	35
	Male	4
Turnaround time	≤20 minutes	19
	21-35 minutes	12
	26 -30 minutes	7
	31-35 minutes	1

Table 2: Specimen body sites and indication for FS, Addis Ababa, Ethiopia

		Indication for frozen section				**Total**
		Primary diagnosis	LN metastasis	Local Metastasis	Margins	
Specimen body	Ovary	7	0	1	0	8
	Omentum	0	0	5	0	5
	Lymphnode	0	10	0	0	10
	Breast	0	0	0	3	3
	Colon	1	0	0	1	2
	Fallopian tub	0	0	1	0	1
	Cervix	3	0	0	0	3
	Uterus	6	0	0	0	6
	Mesentery	1	0	0	0	1
Total		18	10	7	4	39

Assessment of primary diagnosis was the most common reason for FS accounting for 46.2% (18) of cases, followed by assessments of lymph node metastasis(10) (25.6%), local metastasis (7) (17.9%), and margin evaluation (4) (10.3%).

Among the collected specimens, the majority were lymph nodes (10) (25.7%), followed by ovary, parametrium, omentum, cervix, surgical margin, colon, fallopian tube, and mesentery (Table 2).

The entire FS procedure took anywhere between 20 and 35 minutes, from the time the specimen was out of the operation room to the time the FS diagnosis was relayed to the attendingsurgeon. In most of the cases (19) (47.5% of cases), the time it took to reach diagnoses was 20 minutes (Table 1). In general, the diagnosis of FS took an average of 23 minutes.

Of all FS samples evaluated, 51.3% (20) were negative for neoplasm and 43.6% (17) of samples were positive for neoplasm. Two samples had indeterminate results (5.1%) as it was difficult to give conclusive diagnosis due to poor staining and sampling. (Flow diagram 1)

Negative for neoplasm samples were further classified as reactive lymph node (8) (40%), unremarkable omentum (5) (25%), free margin (4) (20%), unremarkable ovary (2) (10%), and unremarkable mesentery (1) (5%).

The most common type of neoplasm detected was Leiomyoma (6) (35.2%), followed by benign ovarian cyst (4) (23.5%). Two samples (11.7%) were diagnosed as suspicious for malignancy, two lymph nodes (11.7%) as secondaries of carcinoma, one sample (5.8%) as mucinous neoplasm, one sample (5.8%) as sex cord stromal tumor and one cervical punch biopsy (5.8%) as Invasive squamous cell carcinoma.

Of the 39 FS samples, 3 (7.7%) were discordant while the rest 36 (92.3%) samples were concordant with FFPE section diagnosis. From the discordant samples, one case diagnosed as positive for malignancy ended up being negative for malignancy during FFPE sectioning, which was counted as false positive. The two indeterminate results were classified as false negative as FFPE section slides showed positive for malignancy; the diagnosis for both was invasive squamous cell carcinoma. This classification of putting indeterminate result as false negative was based on a conservative approach assuming 'worst case scenario' (12).

Table 3: Cross tabulation of FS and FFPE section results of samples sent, Addis Ababa, Ethiopia (n=39)

FS diagnosis	**FFPE tissue diagnosis**	
	Positive for Neoplasm	Negative for Neoplasm
Positive for Neoplasm	16	1
Negative for Neoplasm	2	20

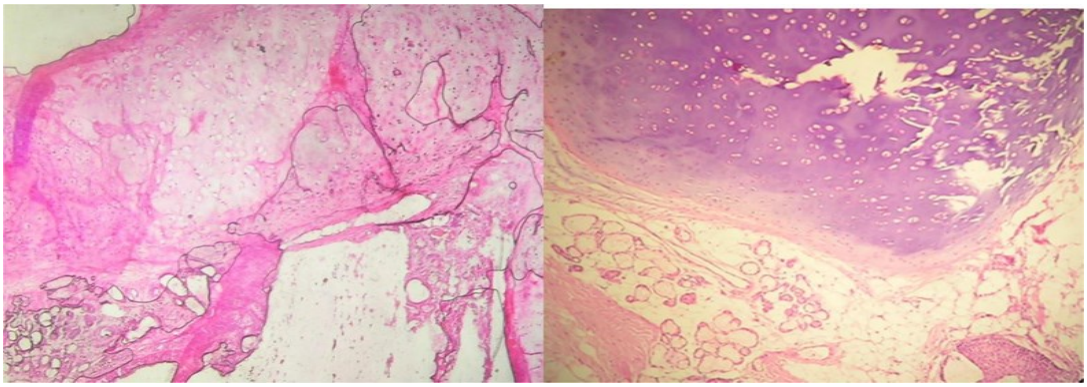
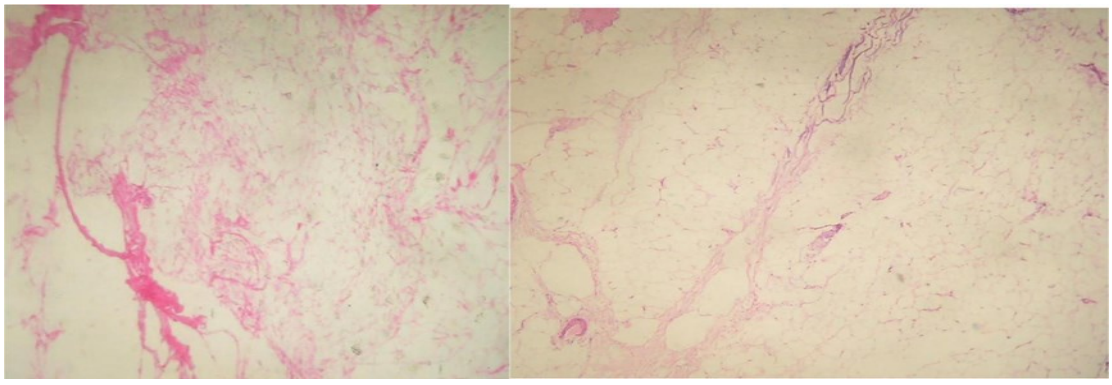
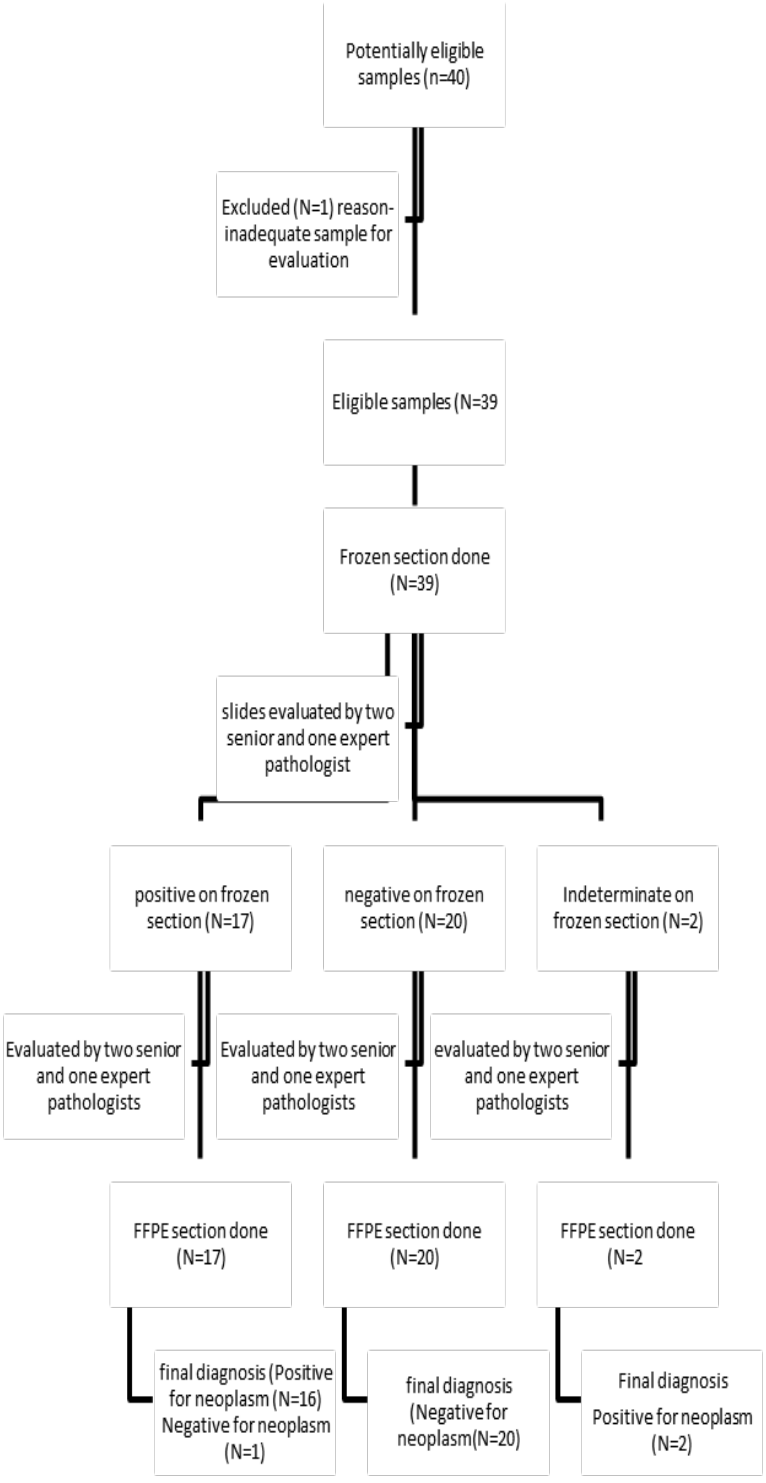


Figure 1: A. (Left, 20x magnification). FS (HE stained slide) of a mature teratoma. B. (Right, 20x magnification)



FFPE (HE stained slide) of a mature teratoma.



Flow Diagram 1: Sample identification and processing workflow

Overall concordance rate was 92.3% (N=36) with 3 (7.7%) discordant cases. All the discordant cases were due to technical error, specifically bloating of cells on FS and poor staining quality. It was determined that FS has a sensitivity of 88% and a specificity of 95.2%. Findings showed that FS had a 90.9% positive predictive value and a 94.1% negative predictive value when it came to identifying neoplasms.

Table 4: Comparison of various studies with the present study on overall accuracy, sensitivity and specificity

Studies	Country	Number of specimens	Accuracy	Sensitivity	Specificity
Present study	Ethiopia	39	92.3%	88%	95.2%
Tangde A et al. (3)	India	83	91.57%	85.7%	97.9%
Gudeli Vahini etal.(11)	India	21	91%	90%	-
Hatami H et al. (13)	Iran	306	97.9%	92.9%	99.5%
Laila C et al. (14)	Morocco	14,000	95%	-	-
Santana RP et al. (5)	Brazil	1,226	96.3%	-	-
Chandramouleeswar i K et al. (15)	India	51	92%	-	-

Discussion

This study looks at common indications for FS, turnaround time, accuracy, and causes ofdisagreement in FS diagnosis. The findings show that primary diagnosis and lymph node involvement are the two main reasons for FS diagnosis request in our setting. The frequently sampled sites are lymph nodes and ovaries. The study also highlights the importance of a timely turnaround time in FS diagnosis. The accuracy rate of 92.3% aligns with previous research, supporting the reliability of FS. Discrepancies between FS and final histopathological reports areattributed to technical errors, which was also the case in other similar studies. Most common indication for FS in our study (Table 2) was similar to other studies (2,3,16,17). Both pathologists and operating surgeons should be fully aware of the indications for FS to guarantee that the right requests are fulfilled. Errors can only be decreased after that (14,18,19). In this study lymph nodes and ovaries were determined to be the anatomical areas most frequently sent for FS diagnosis (46.1%) (Table 2), which was also the case in most other studies (5,16).

The turnaround time, described as the span of time from the moment the sample was received until the surgeon received the report, should not exceed 30 minutes. In our study 97.4% of samples were diagnosed within the proposed turnaround time, which was similar to studies donein India (16,20). One sample, a lymph node, which was the first one processed in this study, took 35 minutes to process, which could be explained by the time needed to become familiar with the cryostat system. This finding shows that it is feasible to do FS in our setting within the proposed time.

Our study showed an accuracy rate of 92.3%. This finding aligns with similar studies (Table 4), Based on the kinds of cases they examined, most centers reported accuracy rates ranging from 92% to 98% (21). Accuracy rate has shown to be more than 90% (2). Several studies have found a clear relationship between sample size and FS accuracy (Table 4). The accuracy of FS appears to rise as sample size increases, which is why a plan to do a larger study in the same setup is necessary. In this study, sensitivity was found to be 88%, while specificity was found to be 95.2%.We found that the sensitivity was lower than the specificity when it comes to identifying neoplastic cases, which is consistent with most researches (13,15,22). Higher specificity can be explained by the fact that the rationale for FS in our investigation was to rule out the existence of malignancy. Consequently, this study demonstrated that FS can be used to exclude malignancy, hence limiting the extent of the operation.

Technical error was determined to be the possible reason for the discrepancy between theintraoperative FS and the final FFPE histopathological report. This finding agrees with other studies which showed that causes for discordance were broadly classified as sampling error, technical error, and interpretative error in various studies (3,11,17,21). Sample error encompasses inadequate tissue sampling as well as inappropriate tissue selection subsequent to grossing.

A substandard section, Xylene/freezing artifact, bloated cell structure, and poor staining were among the technical issues (11). In our study bloated cell morphology and poor staining were the main causes which resulted in discordant cases. This finding can be explained by the relative inexperience in doing FS in the department. Hence, refining technical aspects of FS is believed to increase the accuracy rate.

Since this was the department's first attempt at implementing FS, there have been technical obstacles and a learning curve involved with the work. Ongoing training and quality improvement efforts may aid in

overcoming these restrictions. Due to the nature of this pilot study, which primarily focused on assessing the feasibility of FS, long-term follow-up data including patient outcomes and treatment success rates were not included. This limitation arises from ethical considerations concerning the decision -making process during surgical operations. Thus, surgeons were specifically instructed to base treatment decisions on their clinical judgment rather than only on the FS diagnosis provided. The relationship between FS diagnosis and patient outcomes would need to be examined in further research with a bigger sample size and longer follow-up.

Conclusion

FS was found to have 92.3%, 88%, and 95.2% accuracy, sensitivity, and specificity when compared to FFPE tissue diagnosis which are comparable with most international studies on FS. Given that this is a pilot study, a larger investigation should be carried out to use these findings as a benchmark. As the turnaround time was within the standard, FS results, particularly in institutions with trained pathologists, can be used to guide the type and scope of operation.

Increased diagnostic accuracy can be achieved through more precise and larger sampling, avoidance of sectioning errors, accurate interpretation, and knowledge of clinical history and presentation. We recommend forming and communicating with a multidisciplinary team that includes operating surgeons, on-call pathologists, and technicians, so as to decrease diagnostic errors. Despite its technical errors, the FS approach is very dependable and efficient in the right hands.

Acknowledgments

We have received funding from the Health Professionals Education Partnership Initiative (HEPI) for this research. Thus, we would like to acknowledge HEPI for funding this pilot study. We would also like to thank the Department of Pathology and the Department of Anatomy for helping to get this project started.

Authors contribution

MD primarily led the conception and design of the research in addition to conducting the statistical analysis and preparing the initial manuscript. SA and AD played vital roles in contributing to the study's conception and providing valuable input during manuscript review. WE carried out the pathologic interpretation of the collected samples, ensuring accurate data interpretation. BS and VA were responsible diligent collection of research data. Throughout the process, all authors actively participated in revising the manuscript, incorporating feedback and suggestions, and ultimately approving the final version.

Competing interests

The contributors assert to have no known conflicts of interest.

Funding

The study received external funding (Health Professional Education Partnership Initiative). The funders had no role in the initiation or design of the study, collection of samples, analysis, interpretation of data, writing of the paper, or the submission for publication. The study and researchers are independent of the funders.

Data availability

All necessary data is obtainable upon plausible request.

References

1. James R, Wright Jr. The Johns Hopkins University Press. 1985 [cited 2022 Jan 26]. p. 295–326 THE DEVELOPMENT OF THE FROZEN SECTION TECHNIQUE, THE EVOLUTION OF SURGICAL BIOPSY, AND THE ORIGINS OF SURGICAL PATHOLOGY on JSTOR. Available from: https://www.jstor.org/stable/44441868
2. Ray M, Mohanty P, Agarwal S, Mohapatra D. Accuracy of intra-operative frozen section consultation of gastrointestinal biopsy samples in correlation with the final histopathological diagnosis. Int J Curr Res Rev. 2021 Jan 1;13(1):41–6.
3. Tangde A, Shrivastava V, Joshi A. Analysis of frozen section in correlation with surgical pathology diagnosis. Int J Res Med Sci. 2019 May 29;7(6):2312.
4. Jaafar H. INTRA-OPERATIVE FROZEN SECTION CONSULTATION: CONCEPTS, APPLICATIONS AND LIMITATIONS.
5. Santana RP, Morais NS, Samary YRS, Bezerra ALR, Takano DM. Evaluation of the accuracy of frozen section in different anatomical sites. J Bras Patol Med Lab. 2018 Sep 1;54(5):319–24.
6. Shrestha S, Mc L, Basyal R, Pathak T. comparative Study of Frozen Section Diagnoses with Histopathology. Postgraduate Medical Journal of NAMS. 2009.
7. Zarbo RJ, Hoffman GG, Howanitz PJ. Interinstitutional comparison of frozen-section consultation. A College of American Pathologists Q-Probe study of 79,647 consultations in 297 North American institutions. Arch Pathol Lab Med. 1991 Dec;115(12):1187–94.
8. Gemechu T, Lako W, Deyessa N. The Relative Frequency and Patterns of Solid Malignant Tumors in Tikur Anbessa Specialized Teaching Hospital: A 10 Year Histopathology Review. Scholars Journal of Applied Med-

ical Sciences. 2018 Jan 30;
9. Whitehead AL, Julious SA, Cooper CL, Campbell MJ. Estimating the sample size for a pilot randomised trial to minimise the overall trial sample size for the external pilot and main trial for a continuous outcome variable. Stat Methods Med Res. 2016 Jun 19;25(3):1057–73.
10. Mathes T, Pieper D. An algorithm for the classification of study designs to assess diagnostic, prognostic and predictive test accuracy in systematic reviews. Syst Rev. 2019 Dec 3;8(1):226.
11. Vahini G. Intraoperative Frozen Section -A Golden Tool for Diagnosis of Surgical Biopsies. Int Clin Pathol J. 2017 Jan 9;4(1).
12. Cohen JF, Korevaar DA, Altman DG, Bruns DE, Gatsonis CA, Hooft L, et al. STARD 2015 guidelines for reporting diagnostic accuracy studies: explanation and elaboration. BMJ Open. 2016 Nov 14;6(11):e012799.
13. 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):295–9.
14. Chbani L, Mohamed S, Harmouch T, Fatemi H El, Amarti A. Quality assessment of intraoperative frozen sections: An analysis of 261 consecutive cases in a resource limited area: Morocco. Health N Hav. 2012;04 (07):433–5.
15. P. Arunalatha JBAR. 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. Devi J. Intraoperative Frozen Section Diagnosis in Surgical Pathology – Our Experience At A Tertiary Care Centre. International Journal of Contemporary Medical Research. 2020;7(9):16–9.
17. Ahmad Z, Barakzai M, Idrees R, Bhurgri Y. Correlation of intra-operative frozen section consultation with the final diagnosis at a referral center in Karachi, Pakistan. Indian J Pathol Microbiol. 2008;51(4):469–73.
18. White VA, Trotter MJ. Intraoperative Consultation/Final Diagnosis Correlation Relationship to Tissue Type and Pathologic Process [Internet]. Vol. 132, Arch Pathol Lab Med. 2008. Available from: www.cebm.
19. McIntosh ER, Harada S, Drwiega J, Brandwein-Gensler MS, Gordetsky J. Frozen section: guiding the hands of surgeons? Ann Diagn Pathol. 2015 Oct 1;19(5):326–9.
20. S Rima, A Santhosh, Sanjeet Roy. A Retrospective Study on Turnaround Time for Frozen Sections- A Tertiary Care Centre Experience from Southern India. Journal of Clinical and Diagnostic Research. 2022 May 1;16(5):EC42–5.
21. Jaafar H. Intra-operative frozen section consultation: concepts, applications and limitations. Malays J Med Sci. 2006 Jan;13(1):4–12.
22. Maheshwari A, Gupta S, Kane S, Kulkarni Y, Goyal LCBK, Tongaonkar HB. Accuracy of intraoperative frozen section in the diagnosis of ovarian neoplasms: Experience at a tertiary oncology center. World J Surg Oncol. 2006 Dec 24;4(1):12.