



CLINICAL PRACTICE ARTICLE

Intraoperative frozen section as a reliable ancillary technique in salivary gland surgery: A cross sectional study [version 1; referees: 1 approved, 1 not approved]

Andrea Marzullo ¹, Gabriella Serio ¹, Luigi Madami², Federica Pezzuto¹, Francesco Fortarezza¹, Nicola Quaranta², Maria Luisa Fiorella², Teresa Lettini¹, Leonardo Resta¹, Massimo Marrelli³, Marco Tatullo³, Luigi Santacroce ⁴

¹Department of Emergency and Organ Transplantation – Pathology Section, Medical School, University of Bari, Bari, Italy
²Department of Neurosciences, ENT Section, Medical School, University of Bari, Bari, Italy
³Biomedical Section, Tecnologica Research Institute, Crotona, Italy
⁴Ionian Department (DISGEM), University of Bari, Bari, Italy

v1 First published: 27 Feb 2018, 7:231 (doi: [10.12688/f1000research.13043.1](https://doi.org/10.12688/f1000research.13043.1))
 Latest published: 27 Feb 2018, 7:231 (doi: [10.12688/f1000research.13043.1](https://doi.org/10.12688/f1000research.13043.1))

Abstract

Background. Salivary glands tumours are uncommon, frequently benign lesions, prevalently located in the parotid gland (80%). Surgical decision making is based on the patient’s history, examination findings, imaging and fine needle aspiration (FNA). FNA is a pre-operative method with good ability in detecting malignancy. During surgery, therefore, Frozen section (FS) can differentiate benign lesions from malignant tumours, to reduce incorrect treatments, to increase the chances of conservative surgery and to better evaluate surgical margins. The aim of our study is to demonstrate the accuracy of the FS procedure in surgery of the salivary glands and to stress the need for dedicated pathology units specialized in lesions of the oral cavity.

Methods. The study included 499 patients who underwent surgery from May 2005 and October 2014. An intra-operative frozen section procedure was done for 288 of them. All frozen sections were compared with the final results. The cases were classified by site, nature of the lesion and histotype, according to the WHO classification. Comparison was made between the intra-operative and the definitive diagnosis.

Results. Of the 288 FS procedures, 259 were for neoplastic lesions, 199 of which benign and 60 malignant, and 29 for non-neoplastic lesions. Of the 259 neoplastic FS results, 2 were shown to be false positives and 2 were diagnosed as different malignant types. Of the 29 non-neoplastic FS results, 4 were false negatives.

Conclusions. Our results showed that the accuracy of frozen section procedure is 98% for salivary glands tumors. The highest concordance between frozen section and the definitive diagnosis was for inflammatory processes (99%), pleomorphic adenoma (98%), Warthin’s tumor (97%) and malignant neoplasms (96%). In conclusion, based on these findings, frozen section of the salivary glands may be proposed as a routine procedure and should be used in decision-making.

Open Peer Review

Referee Status:

	Invited Referees	
	1	2
version 1 published 27 Feb 2018	 report	 report
1 Michele Cassano , University of Foggia, Italy		
2 Kyung-Ja Cho , University of Ulsan College of Medicine, South Korea		

Discuss this article

Comments (0)

Keywords

Salivary gland tumors, frozen section, differential diagnosis, Warthin's tumor, pleomorphic adenoma

Corresponding author: Luigi Santacroce (luigi.santacroce@uniba.it)

Author roles: **Marzullo A:** Conceptualization, Data Curation, Investigation, Methodology, Resources, Validation, Writing – Original Draft Preparation; **Serio G:** Conceptualization, Funding Acquisition, Investigation, Methodology, Supervision, Validation, Writing – Original Draft Preparation; **Madami L:** Investigation, Methodology, Supervision, Validation; **Pezzuto F:** Data Curation, Investigation, Methodology, Visualization; **Fortarezza F:** Formal Analysis, Investigation, Methodology, Visualization; **Quaranta N:** Data Curation, Investigation, Methodology, Resources, Validation; **Fiorella ML:** Data Curation, Methodology, Resources, Validation, Visualization; **Lettimi T:** Formal Analysis, Validation; **Resta L:** Formal Analysis, Funding Acquisition, Methodology, Supervision, Validation; **Marrelli M:** Formal Analysis, Methodology, Validation, Visualization; **Tatullo M:** Data Curation, Formal Analysis, Methodology, Visualization, Writing – Review & Editing; **Santacroce L:** Conceptualization, Data Curation, Formal Analysis, Methodology, Project Administration, Resources, Supervision, Writing – Review & Editing

Competing interests: No competing interests were disclosed.

How to cite this article: Marzullo A, Serio G, Madami L *et al.* **Intraoperative frozen section as a reliable ancillary technique in salivary gland surgery: A cross sectional study [version 1; referees: 1 approved, 1 not approved]** *F1000Research* 2018, 7:231 (doi: [10.12688/f1000research.13043.1](https://doi.org/10.12688/f1000research.13043.1))

Copyright: © 2018 Marzullo A *et al.* This is an open access article distributed under the terms of the [Creative Commons Attribution Licence](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. Data associated with the article are available under the terms of the [Creative Commons Zero "No rights reserved" data waiver](#) (CC0 1.0 Public domain dedication).

Grant information: The author(s) declared that no grants were involved in supporting this work.

First published: 27 Feb 2018, 7:231 (doi: [10.12688/f1000research.13043.1](https://doi.org/10.12688/f1000research.13043.1))

Introduction

Salivary gland tumors are rare lesions that occur mainly in the major salivary glands; 80% of tumors occur in the parotid gland, and these are prevalently benign.

The parotid gland histology is complex: there are *i*) abundant intralobular and extralobular adipose tissues, which increase in relative volume with age, *ii*) randomly distributed lymphoid aggregates, and *iii*) lymph nodes that occasionally contain ducts or salivary acini. Therefore, it is often difficult to distinguish a neoplastic lesion from a non-neoplastic lesion, as well as a benign lesion from a malignant lesion, especially in view of the morphological variability of salivary tumors. However, a correct differential diagnosis, safely and promptly executed, is crucial for the entire patient's management, both clinical and surgical, including possible tissue regeneration¹⁻⁵.

The clinical approach to salivary lesions is supported by imaging techniques, such as ultrasound, computed tomography or magnetic resonance imaging; these can provide greater definition of the lesion, but are not always sufficient to formulate a definitive diagnosis. Therefore, it is necessary to resort to pre-surgical techniques to better define a salivary lesion.

Fine needle aspiration (FNA), which can be performed at the time of the initial clinical consultation, can be used both as a diagnostic test and as a guideline in selecting the patient's management: surgical vs follow-up without surgery. The FNA technique demonstrates high sensitivity and specificity (80% and 97%) for benign tumors, but is not very sensitive for malignant neoplasms (sensitivity ranges from 54% to 92%; specificity 87% to 98%). False-negative rates range from 2% to 31% and false positive rates from 0% to 7%⁶⁻¹¹.

FNA is a simple, safe procedure that does not require the use of local anesthesia, and can be performed either blinded or under ultrasound guidance. It is important to remember that the methodological approach, clinical needle aspiration skills and experience of the pathologist are the elements that affect the definitive diagnosis. Frozen section (FS) is a less rapid, more invasive intraoperative diagnostic procedure, but precisely because of this, it is a guarantee of better histological results. It allows surgery to be performed in a targeted treatment continuum^{1,9-10}.

Once again, it should be emphasized that salivary tumors are a heterogeneous group of lesions that necessitate the examination of many sections. Early diagnosis is essential, to establish the correct histological type of salivary glands lesion, in order to achieve proper planning of the surgical treatment, which may also involve the regional lymph nodes, and adjacent tissues¹¹. Variable percentages of effectiveness of the FS method applied to the diagnosis of salivary glands lesions are reported in the literature. The reliability rate ranges from 40% to 100%; this variability is often attributed to the pathologist's experience or to the technician responsible for slide preparation¹²⁻¹⁶.

Objectives

In this study we assessed the diagnostic accuracy of FS; specifically, the concordance between FS and the definitive histological diagnosis, as well as verifying the importance of

being able to rely on a team of experts, to reduce false positive or negative cases.

Methods

Patients

Between May 2005 to October 2014, 499 patients (275 males and 224 females, mean age 54±17.2 years) suffering from localized masses in the salivary glands were recruited at the Complex Unit of Otolaryngology at the University of Bari (Italy).

Inclusion criteria were related to the clinical aspects of the first access diagnosis: only patients with localized masses developed in the major salivary glands regions were included. Exclusion criteria were a previous history of cancer or any suspicion of infectious disease as main noxa of the mass. Also, patients reporting smoking habits were excluded as well (Figure 1).

Data collection

We selected 288 salivary lesions (out of the total 499) operated on by the same team of surgeons and pre-analyzed with FS.

Bias

To reduce bias, in 90% of cases, the intraoperative examination was performed by the same team of pathologists. In all cases, the radiological examination posed an indication for surgical treatment and suggested a provisional diagnosis (i.e. benign vs malignant). FNA was not considered because if done at all, it was at non-dedicated centers and there was a high number of "non-diagnostic" results.

Quantitative variables

Our study was aimed to assess if and how many were the neoplastic lesions, how many were benign and/or malignant, and if some non-neoplastic lesions were also diagnosed. Our attention was also directed towards the identification of some false positive/negative results. The accuracy of FS procedure was compared, in all the reported cases, with the traditional histological assay. Our null-hypothesis was aimed to assess that FS of the salivary glands may be proposed as a routine procedure and should be used in the decision-making process.

FS process

The FS procedure, also called cryosection, is a commonly used procedure to perform rapid microscopic analysis of a specimen: it is mostly used as a first-look diagnostic tool in intraoperative oncological surgery. The protocol adopted for performing intraoperative FS was a standardized method: *i*) the biopsy was fixed and cut in a cryostat at -25°-28°C, *ii*) the samples were cut into 5 micrometers thick slices, *iii*) the samples were subjected to haematoxylin-eosin staining. (Figure 2).

The medical report was obtained, in all patient cases, within 10–15 minutes. Each sample was stored with labels detailing the biodata and the macroscopic characteristics of the excised lesion.

Results

Participants

We reported 288 patients with salivary lesions operated on by the same team of surgeons and pre-analyzed with FS, in order

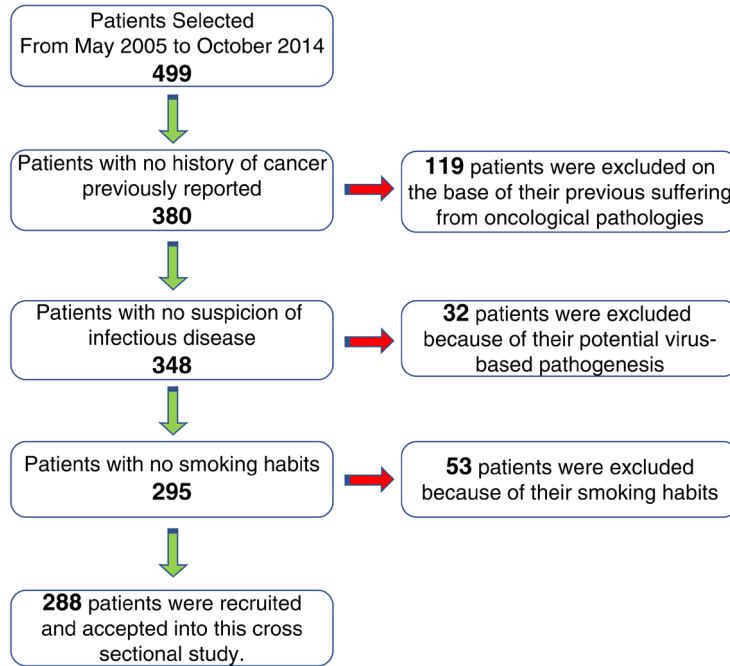


Figure 1. Flowchart of patient recruitment.

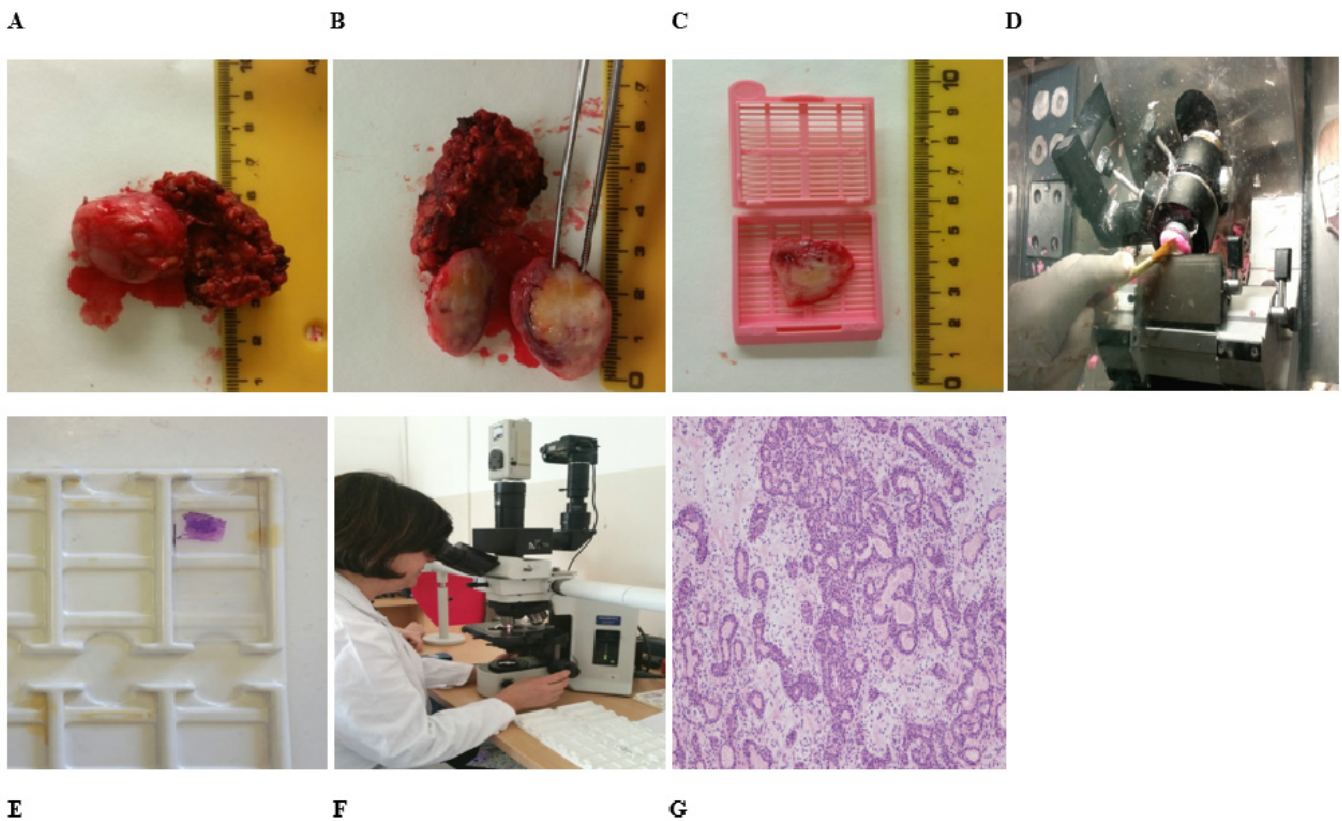


Figure 2. The intraoperative frozen section (FS) technique. (A–C) Left parotidectomy surface that includes a well-demarcated nodule measuring 3cm in diameter and showing translucent appearance of the cut surface; (D) The intraoperative frozen section (FS) technique and (E) corresponding hematoxylin-eosin section; (F–G) Pathology preliminary FS report compatible with pleomorphic adenoma.

to make a comparative analysis between the two different techniques.

Outcome data

FS was useful to indicate the correct surgical treatment of 269 nodules (93.4%) of the parotid gland, 14 nodules (4.9%) of the submandibular gland and 5 lesions (1.7%) that involved the minor salivary glands located in the palatal mucosa. Using the FS method, a correct diagnosis was obtained in 280 cases (97.2%) (Table 1).

Main results

The highest concordance between the FS and the definitive diagnosis was for pleomorphic adenoma (98%), Warthin’s tumor (97%), inflammatory processes (99%) and malignant neoplasms (96%). In 8 cases (2.8%), the “provisional diagnosis” with intraoperative FS and the definitive diagnosis were discordant. The false negative results for FS consisted of the following: 1 inflammatory process, actually diagnosed as Warthin’s tumor;

1 normal tissue, actually diagnosed as an arteriovenous malformation; 1 reactive lymphoid tissue, actually diagnosed as a non-Hodgkin’s lymphoma; and 1 sialometaplasia, actually diagnosed as a squamous cell carcinoma. Two false positive results were obtained: in both these cases squamous metaplasia in pleomorphic adenoma was interpreted as malignant (Table 2). Thus, the sensitivity and specificity for malignancy were assessed to 97%. Patients with a positive FS diagnosis underwent lymphadenectomy. The FS diagnosis of malignancy was not confirmed at histology in two cases: 1 carcinoma, actually a NH-Lymphoma; 1 NH-Lymphoma, actually a small cell carcinoma. In these cases, only one patient received overtreatment.

Other analyses

Analyzing the definitive diagnosis vs FS, the false positive cases were found to have been diagnosed by a non-dedicated pathologist, as also one case of Warthin’s cancer, defined as an inflammatory process.

Table 1. Results of cases investigated with the frozen section (FS) diagnosis.

Cases (n = 288)	FS diagnosis
Pleomorphic adenoma	124 (43.1%)
Warthin's tumor	68 (23.6%)
Adenoid cystic carcinoma	4 (1.4%)
Acinic cells carcinoma	5 (1.7%)
Lymphomas	18 (6.2%)
Adenocarcinoma, NOS	10 (3.5%)
Squamous cells carcinoma	11 (3.8%)
Carcinoma, NOS	5 (1.7%)
Mucoepidermoid carcinoma	3 (1%)
Small cells carcinoma	2 (0.7%)
Carcinoma ex-pleomorphic adenoma	2 (0.7%)
Oncocytoma	4 (1.4%)
Lipoma	3 (1%)
Inflammatory/normal tissue	29 (10.1%)

Table 2. Frozen section (FS) diagnosis vs. the definitive histological diagnosis.

	FS diagnosis	Definitive histological diagnosis	Cases (n=8)
False positive	<i>Squamous carcinoma</i>	<i>Pleomorphic adenoma</i>	2
False negatives	<i>Sialometaplasia</i>	<i>Squamous carcinoma</i>	1
	<i>Inflammation</i>	<i>Warthin's tumor</i>	1
	<i>Lymphoid hyperplasia</i>	<i>Lymphoma</i>	1
	<i>Normal tissue</i>	<i>AVM</i>	1
Discordant diagnosis	<i>Carcinoma NOS</i>	<i>N-H Lymphoma</i>	1
	<i>N-H Lymphoma</i>	<i>Small cell carcinoma</i>	1

Dataset 1. Raw data underlying the study, including final diagnosis and FS diagnosis<http://dx.doi.org/10.5256/f1000research.13043.d192935>**Discussion**

This study was conducted in order to assess the diagnostic accuracy, sensitivity and specificity of FS examination, used as a preoperative method to guide the surgeon in the choice of treatment of salivary lesions. A further aim of the study was to analyse the diagnostic accuracy of FS examination to pose the diagnosis of malignancy. Previous studies have compared the accuracy of FS examination with FNA cytology, observing a greater reliability of the FS technique¹³. In fact, FNA shows too high a number of false negatives due to factors related to: *i*) triage errors, *ii*) hypocellularity of the material, *iii*) interpretation errors^{10,11}. Some studies have demonstrated an excellent effectiveness of a FS examination, obtaining a specificity of 99% and a sensitivity of 90%, other authors have even reported maximum specificity and sensitivity, equal to 100%^{16,17}.

Key results

In our study, we excluded FNA as a perioperative examination because the number of “inadequate” tissues was too high. In our experience, FS could replace FNA, reducing the risk of inappropriate surgery, of unnecessary adjuvant radiotherapy, as well as reducing National Health System costs¹⁸.

Based on the results described in this study, the FS examination shows a high reliability in the diagnosis of salivary gland tumors. FS is particularly useful in cases of differential diagnosis between neoplastic and non-neoplastic lesions; it also shows a good reliability in the differential diagnosis between benign and malignant neoplasms.

Limitations

Despite our study limitations, related to the small sample size and related to the single unique center participating to this study,

our results are in agreement to those reported by various authors, and they also highlight the need to be able to rely on a dedicated team of clinicians and pathologists^{15–20}.

Conclusions

“Misinterpretation” was observed in those cases diagnosed by pathologists with no specific experience of head and neck tumors. Our work group experience emphasizes the importance of intraoperative examination in order to define the histotype and the cancer margins, permitting the performance of effective, predictable surgery of the salivary glands.

Ethical statement

In Italy, directors of clinical research units and those responsible for clinical services may access data records for research purposes if patients have previously signed a consent form that confirming that they allow this use of their data. Therefore, no specific ethical approval is required for this study, and all patients included in the study signed written informed consent allowing their data to be used in future research.

Data availability

Dataset 1: Raw data underlying the study, including final diagnosis and FS diagnosis. DOI, [10.5256/f1000research.13043.d192935](https://doi.org/10.5256/f1000research.13043.d192935)²¹

Competing interests

No competing interests were declared.

Grant information

The author(s) declared that no grants were involved in supporting this work.

Acknowledgements

The authors are grateful to Mary V. Pragnell, BA, for language assistance.

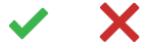
References

- Barnes L, Evenson JW, Reichart P, *et al.*: **Pathology and Genetics. Head and Neck Tumours**. WHO Classification of Tumours. Lyon, 2005.
[Reference Source](#)
- Pinkston JA, Cole P: **Incidence rates of salivary gland tumors: results from a population-based study**. *Otolaryngol Head Neck Surg.* 1999; **120**(6): 834–403.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Spiro RH: **Salivary neoplasms: overview of a 35-year experience with 2,807 patients**. *Head Neck Surg.* 1986; **8**(3): 177–844.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Lee RJ, Tan AP, Tong EL, *et al.*: **Epidemiology, Prognostic Factors, and Treatment of Malignant Submandibular Gland Tumors: A Population-Based Cohort Analysis**. *JAMA Otolaryngol Head Neck Surg.* 2015; **141**(10): 905–12.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Perniconi B, Coletti D, Aulino P, *et al.*: **Muscle acellular scaffold as a biomaterial: effects on C2C12 cell differentiation and interaction with the murine host environment**. *Front Physiol.* 2014; **5**: 354.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Ball AB, Fish S, Thomas JM: **Malignant epithelial parotid tumours: a rational treatment policy**. *Br J Surg.* 1995; **82**(5): 621–23.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Tan LG, Khoo ML: **Accuracy of fine needle aspiration cytology and frozen section histopathology for lesions of the major salivary glands**. *Ann Acad Med Singapore.* 2006; **35**(4): 242–48.
[PubMed Abstract](#)
- Megerian CA, Maniglia AJ: **Parotidectomy: a ten year experience with fine needle aspiration and frozen section biopsy correlation**. *Ear Nose Throat J.* 1994; **73**(6): 377–380.
[PubMed Abstract](#)

9. Howlett DC, Trantafyllou A: **Evaluation: Fine Needle Aspiration Cytology, Ultrasound-Guided Core Biopsy and Open Biopsy Techniques.** *Adv Otorhinolaryngol.* 2016; **78**: 39–45.
[PubMed Abstract](#) | [Publisher Full Text](#)
10. Foundakowski C, Castaño J, Abouyared M, *et al.*: **The role of indeterminate fine-needle biopsy in the diagnosis of parotid malignancy.** *Laryngoscope.* 2014; **124**(3): 678–81.
[PubMed Abstract](#) | [Publisher Full Text](#)
11. Fakhry N, Antonini F, Michel J, *et al.*: **Fine-needle aspiration cytology in the management of parotid masses: evaluation of 249 patients.** *Eur Ann Othorhinolaryngol Head Neck Dis.* 2012; **129**(3): 131–35.
[PubMed Abstract](#) | [Publisher Full Text](#)
12. Santacroce L, Luperto P, Fiorella ML, *et al.*: **[Carcinoma of unknown origin++ with latero-cervical metastasis. Diagnostic problems. Retrospective analysis of 110 cases of latero-cervical tumefaction].** *Clin Ter.* 2000; **151**(3): 199–201.
[PubMed Abstract](#)
13. Fakhry N, Santini L, Lagier A, *et al.*: **Fine needle aspiration cytology and frozen section in the diagnosis of malignant parotid tumours.** *Int J Oral Maxillofac Surg.* 2014; **43**(7): 802–5.
[PubMed Abstract](#) | [Publisher Full Text](#)
14. Hillel AD, Fee WE Jr: **Evaluation of frozen section in parotid gland surgery.** *Arch Otolaryngol.* 1983; **109**(4): 230–232.
[PubMed Abstract](#) | [Publisher Full Text](#)
15. Olsen KD, Moore EJ, Lewis JE: **Frozen section pathology for decision making in parotid surgery.** *JAMA Otolaryngol Head Neck Surg.* 2013; **139**(12): 1275–78.
[PubMed Abstract](#) | [Publisher Full Text](#)
16. Mostaan LV, Yazdani N, Madani SZ, *et al.*: **Frozen section as a diagnostic test for major salivary gland tumors.** *Acta Med Iran.* 2012; **50**(7): 459–62.
[PubMed Abstract](#)
17. Carvalho MB, Soares JM, Rapoport A, *et al.*: **Perioperative frozen section examination in parotid gland tumors.** *Sao Paulo Med J.* 1999; **117**(6): 233–37.
[PubMed Abstract](#) | [Publisher Full Text](#)
18. Arabi Mianroodi AA, Sigston EA, Vallance NA: **Frozen section for parotid surgery: should it become routine?** *ANZ J Surg.* 2006; **76**(8): 736–39.
[PubMed Abstract](#) | [Publisher Full Text](#)
19. Schmidt RL, Hunt JP, Hall BJ, *et al.*: **A systematic review and meta-analysis of the diagnostic accuracy of frozen section for parotid gland lesions.** *Am J Clin Pathol.* 2011; **136**(5): 729–738.
[PubMed Abstract](#) | [Publisher Full Text](#)
20. Tatullo M, Marrelli M, Amantea M, *et al.*: **Bioimpedance Detection of Oral Lichen Planus Used as Preneoplastic Model.** *J Cancer.* 2015; **6**(10): 976–83.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
21. Marzullo A, Serio G, Madami L, *et al.*: **Dataset 1 in: Intraoperative frozen section as a reliable ancillary technique in salivary gland surgery: A cross sectional study.** *F1000Research.* 2018.
[Data Source](#)

Open Peer Review

Current Referee Status:



Version 1

Referee Report 24 April 2018

doi:10.5256/f1000research.14142.r32697



Kyung-Ja Cho

Department of Pathology, Asan Medical Center, University of Ulsan College of Medicine, Seoul, South Korea

- This paper is recommending frozen section as a primary evaluation of salivary gland tumors, and it is considered out of date and unethical. Many salivary gland tumors share histologic features and have distinct expression or molecular characteristics. Ancillary tests are occasionally necessary. Accurate diagnosis before operation, if possible, is needed for proper patient management.
- Core needle biopsy of the salivary gland tumors is recently substituting aspiration cytology. Authors need to comment the usefulness of such preoperative diagnostic procedures.
- Three exclusion criteria from selection seem irrelevant with diagnosis of salivary gland tumors.
- The distribution of tumors appear very biased even after exclusion. Mucoepidermoid carcinoma is the most common malignancy worldwide, while squamous cell carcinoma of salivary gland is rare.
- It is difficult to be convinced that most of lymphomas, and all small cell carcinomas and carcinoma ex pleomorphic adenomas were accurately diagnosed by frozen section. Illustrations or other back-up data are needed.

Is the background of the cases' history and progression described in sufficient detail?

Partly

Are enough details provided of any physical examination and diagnostic tests, treatment given and outcomes?

Partly

Is sufficient discussion included of the importance of the findings and their relevance to future understanding of disease processes, diagnosis or treatment?

No

Is the conclusion balanced and justified on the basis of the findings?

Partly

Competing Interests: No competing interests were disclosed.

I have read this submission. I believe that I have an appropriate level of expertise to state that I do not consider it to be of an acceptable scientific standard, for reasons outlined above.

Referee Report 26 March 2018

doi:10.5256/f1000research.14142.r31226



Michele Cassano

ENT Clinic, University of Foggia, Foggia, Italy

Interesting paper about the importance of frozen section in the diagnosis of Salivary glands lesions. The casistic is very wide and the results are innovative also if many guidelines report the need of performing Fine Needle Aspiration in the preoperative setting of a salivary gland lesion and I do not think that we can actually derogate from this guideline. Anyway this paper is worthy of indexing but some points need to be reviewed:

- The first sentence of the "FS process" in method section should be moved to the introduction to complete the paragraph about the frozen section.
- In the inclusion criteria the authors report that just major salivary glands lesions were analyzed but in the outcomes data they state that 5 cases were minor salivary glands glands in the palatal mucosa. Please clarify this point.
- In the discussion the authors state that the FS is used as "pre-operative method". I think that is more correct to define it an intraoperative method.

Is the background of the cases' history and progression described in sufficient detail?

Yes

Are enough details provided of any physical examination and diagnostic tests, treatment given and outcomes?

Yes

Is sufficient discussion included of the importance of the findings and their relevance to future understanding of disease processes, diagnosis or treatment?

Yes

Is the conclusion balanced and justified on the basis of the findings?

Yes

Competing Interests: No competing interests were disclosed.

I have read this submission. I believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

The benefits of publishing with F1000Research:

- Your article is published within days, with no editorial bias
- You can publish traditional articles, null/negative results, case reports, data notes and more
- The peer review process is transparent and collaborative
- Your article is indexed in PubMed after passing peer review
- Dedicated customer support at every stage

For pre-submission enquiries, contact research@f1000.com

F1000Research