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Original Article

Pathology Section

Accuracy of Frozen Section in Diagnosis of Head and Neck Lesions

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ABSTRACT

This study was carried out in the Department of Pathology, GSVM Medical College, Kanpur. 100 neoplastic and non-neoplastic lesions from the head and neck were subjected to frozen section examination and they were compared with permanent sections on the same tissue as well as after further sampling. The specimens included lymph nodes, oral cavity lesions, thyroid lesions, and salivary gland lesions. The accuracy of frozen section in diagnosis of head and neck lesions was 94% with discordance rate of 5% and deferral rate 1%. The disagreements were the result of a gross sampling error in 3 (42.8%) cases, an interpretative error in 2 (28.6%) cases and (14.3%) a microscopic sampling error and inadequate tissue in the frozen section in 1 case each.

Key Words: frozen section, neoplastic and non-neoplastic lesions

MATERIAL AND METHOD

The specimens from Head and Neck lesions of excised malignant lesions were received from Department of Surgery and Otolaryngology, LLR and Associated hospitals, Kanpur. A proper history was taken and a detailed clinical examination was done in every case.

After a detailed gross examination and after making a gross provisional diagnosis, the representative tissue sections which measured 3–5mm were submitted for the frozen section procedure by using a rapid sectioning cryostat (LEICA–CM 1510 S). The sections were cut at a thickness of 6–10 microns. The rapid Hematoxylin and Eosin method was employed to stain the sections as per the standard protocol. The remains of the tissues which were used for making the frozen sections were subsequently processed by the paraffin embedding technique, so as to compare the diagnosis of the frozen sections with the permanent sections on the same tissues. The results were also compared with the final, permanent section diagnosis after giving more sections, so as to evaluate the sampling errors, if any.

The deferred and the incorrect cases were studied further to determine the reasons for the discrepancies. The classification of the errors which was used in the present study, was based on that which was followed by Rogers [1] et al (1987).

- **Gross sampling error:** where the lesion was contained in a portion of the tissue which was selected for the cryostat section, but was not sampled.
- **Microscopic sampling error:** where the lesion was in the tissue which was sampled, but was not revealed in the sections which were studied.
- **Interpretation error:** where the relevant tissue was on the frozen section slide, but the correct diagnosis was not made.
- **Failure of communication:** where the surgeon possessed the information that probably would have changed the frozen section diagnosis, had it been communicated to the pathologist.

RESULTS AND OBSERVATIONS

Hundred (100) cases of head and neck lesions were studied for diagnosis.

Out of the 100 cases of the head and neck region, 35 cases (35%) were lymph node lesions, followed by 30 oral cavity lesions (30%), 18 thyroid lesions (18%) and 17 salivary gland lesions (17%).

The Lymph Node Lesions [Table/Fig-1]

35 cervical lymph node specimens were received.

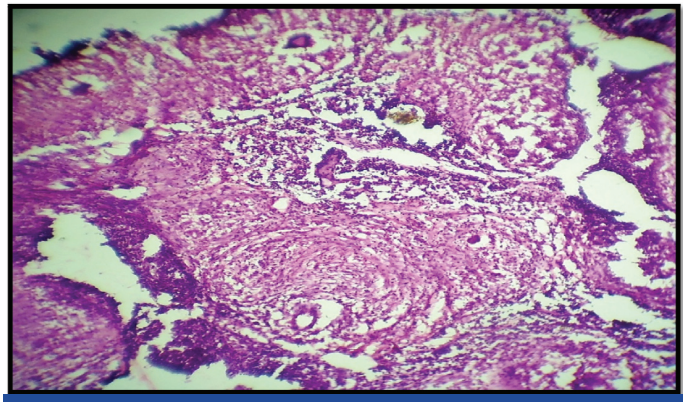
Permanent Section Diagnosis	Number of Cases	Frozen Section Diagnosis			
		Correlated		Not Correlated	
		No.	%	No.	%
Tubercular lymphadenitis	14	12	85.7%	02	14.3%
Reactive hyperplasia	12	12	100%	00	0%
Non-hodgkin's lymphoma	05	05	100%	00	0%
Metastatic	04	04	100%	00	0%
TOTAL	35	33	94.3%	02	5.7%

[Table/Fig-1]: Correlation of frozen section and permanent section diagnosis in lymph node lesions

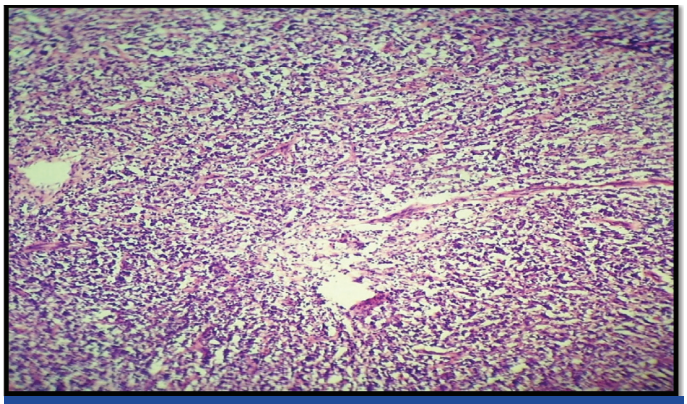
Of the 35 cases, 14 were of tubercular lymphadenitis [Table/Fig-2], 12 were of reactive lymphadenitis, 5 were of non Hodgkin's lymphoma [Table/Fig-3] were of metastatic squamous cell carcinoma.

All (85.7%), but 2 (14.3%) cases of tubercular lymphadenitis were correctly diagnosed on the frozen sections. 1 was misdiagnosed as reactive lymphadenitis (interpretative error). In 1 case, there was a microscopic sampling error.

All (100%) cases of reactive lymphadenitis were precisely diagnosed on the frozen sections.



[Table/Fig-2]: Frozen section: Photograph of tubercular lymphadenitis



[Table/Fig-3]: Frozen section: Photograph of Non Hodgkin lymphoma (low power view)

Among the 5 cases of non-Hodgkin's lymphoma, all (100%) were correctly diagnosed on the frozen sections.

4 (100%) cervical lymph node specimens were diagnosed as metastatic squamous cell carcinomas on the frozen sections, which was further confirmed on the permanent sections.

The accuracy of the frozen section in diagnosing the lymph node lesions was 94.3% and a discrepancy was found in 2 (5.7%) cases.

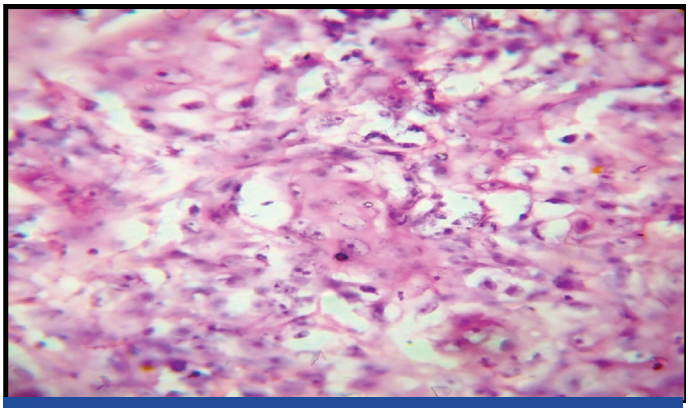
The Oral Cavity Lesions [Table/Fig-4]

30 oral cavity lesions were submitted for frozen section examination.

Permanent Section Diagnosis	Number of Case	Frozen Section Diagnosis			
		Correlated		Not Correlated	
		No.	%	No.	%
Capillary hemangioma	03	03	100%	00	0%
Carcinoma in situ	04	04	100%	00	0%
Squamous cell carcinoma	23	21	91.3%	01	8.7%
Total	30	28	93.3%	02	6.7%

[Table/Fig-4]: Correlation of frozen section and permanent section diagnosis in oral cavity lesions

Of the 30 oral cavity lesions, 3 were benign and 27 were malignant. All the benign lesions were diagnosed as capillary hemangiomas on the frozen as well as the paraffin sections. Of the 27 malignant lesions, 23 cases were squamous cell carcinomas [Table/Fig-5]. Of these, 21 were precisely diagnosed on the frozen sections. However, gross sampling errors in the 2 specimens resulted in false negative diagnoses of moderate grade dysplasia with



[Table/Fig-5]: Frozen section: Photograph of Squamous cell carcinoma oral cavity

absence of invasion on the frozen sections. All the remaining 4 cases were correctly diagnosed as carcinoma in situ on the frozen sections.

Thus, the total diagnostic accuracy in the oral cavity lesions was 93.3% and a discrepancy was found in 2 (6.7%) cases.

The Thyroid Lesions [Table/Fig-6]

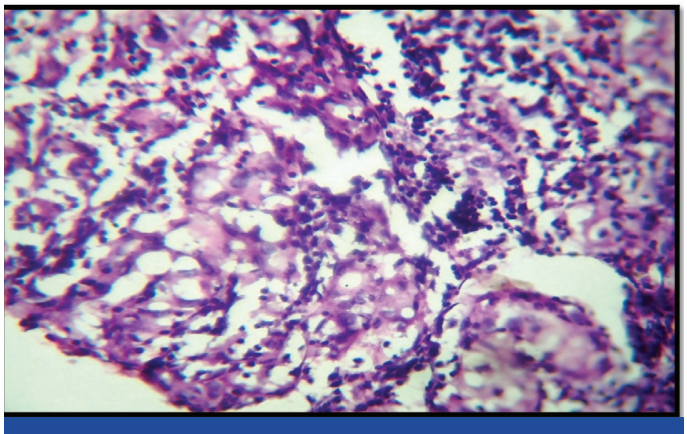
18 thyroid tissues were received for the frozen section analysis.

Permanent Section Diagnosis	Number of Case	Frozen Section Diagnosis			
		Correlated		Not Correlated	
		No.	%	No.	%
Colloid goitre	07	07	100%	00	0%
Hashimoto Thyroiditis	03	03	100%	00	0%
Follicular adenoma	05	05	100%	00	0%
Papillary carcinoma	03	03	100%	00	0%
Total	18	18	100%	00	0%

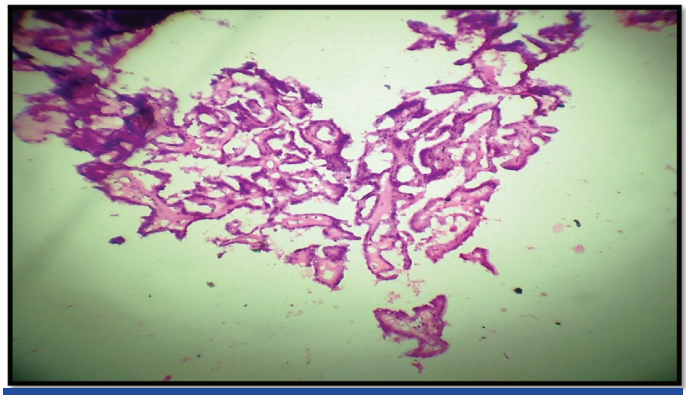
[Table/Fig-6]: Correlation of frozen section and permanent section diagnosis in thyroid lesions

Of the 18 thyroid lesions, ten (55.6%) were benign lesions, five (27.8%) were follicular neoplasms and three (16.6%) were papillary carcinomas. Of the ten benign lesions, seven were diagnosed as colloid goiter and three as Hashimoto's thyroiditis [Table/Fig-7] on the frozen as well as the paraffin sections.

All the follicular neoplasms were follicular adenomas and they were correctly diagnosed on the frozen sections.



[Table/Fig-7]: Frozen section: Photograph of Hashimoto thyroiditis (high power view)



[Table/Fig-8]: Frozen section: Photograph of Papillary carcinoma thyroid

All the 3 malignant lesions were precisely diagnosed as papillary carcinomas [Table/Fig-8] on the cryostat sections, which were confirmed subsequently on the paraffin sections. Thus, the diagnostic accuracy of the frozen sections for the thyroid lesions was 100%.

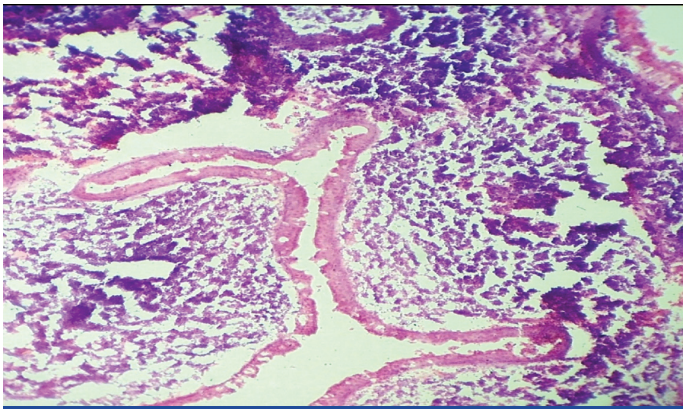
The Salivary Gland Lesions [Table/Fig-9]

17 salivary gland lesions were submitted for the frozen section study. These included 11 parotid lesions, 3 submandibular lesions, and 3 lesions from the minor salivary gland.

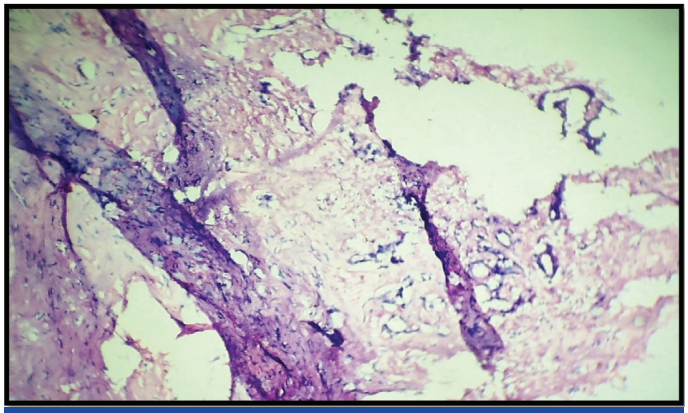
Permanent Section Diagnosis	Number of Case	Frozen Section Diagnosis			
		Correlated		Not Correlated	
		No.	%	No.	%
Chronic sialadenitis	04	04	100%	00	0%
Pleomorphic adenoma	08	08	100%	00	0%
Warthintumor	02	02	100%	00	0%
Carcinoma ex Pleomorphic adenoma	01	00	100%	01	100%
Adenoid cystic carcinoma	02	01	50%	01	50%
Total	17	15	88.2%	02	11.8%

[Table/Fig-9]: Correlation of frozen section and permanent section diagnosis in salivary gland lesions

Of the 17 salivary gland lesions, 15 were diagnosed correctly on the frozen sections. 8 of these were diagnosed as pleomorphic adenomas, 4 as chronic sialadenitis, 2 as Warthin's tumour [Table/Fig-10], and 1 as adenoid cystic carcinoma.



[Table/Fig-10]: Frozen section: Photograph of Pleomorphic adenoma



[Table/Fig-11]: Frozen section: Photograph of Carcinoma ex pleomorphic adenoma misdiagnosed as pleomorphic adenoma

An interpretative error in 1 case resulted in a false diagnosis of pleomorphic adenoma, owing to the abundant chondromyxoid ground substance and the sparse epithelial structures, which on paraffin section, was diagnosed as carcinoma ex pleomorphic adenoma [Table/Fig-11].

A gross sampling error in the other parotid specimen resulted in a false negative diagnosis of absence of tumour. The final diagnosis was deferred for the permanent section examination, which revealed an adenoid cystic carcinoma.

Thus, the overall diagnostic accuracy for the salivary gland lesions was 88.2%, a discrepancy was found in 1 (5.9%) case and a deferral was found in 1 (5.9%) case.

The Cause of the Discrepancy

Of the 6 errors which we faced, 3 (50%) were due to gross sampling errors, 2 (33.3%) were due to interpretative errors and 1 (16.7%) was due to a microscopic sampling error.

The Overall Accuracy

The accuracy of the frozen sections in the diagnosis of head and neck lesions was 94%, with a discordance rate of 5% and a deferral rate of 1%.

DISCUSSION

In the present study, 100 neoplastic and non-neoplastic lesions from various head and neck sites were subjected to the frozen section examination for diagnosis and then they were compared with the gold standard paraffin sections, in order to determine the usefulness of this technique. The diagnostic accuracy of this series was assessed by using the conventional measures of sensitivity and specificity and this was compared with the data which was obtained from the Indian and the international literature.

The Lymph Nodes

In the present series, 35 lymph node specimens were subjected to the frozen section analysis. An accuracy of 94.3% was achieved, which was slightly lower than those which were reported in most of the reviews in the literature. Ackerman [2] et al. (1959) and Holaday [3] et al. (1974) found an accuracy of 98.6% and 97.5% respectively for the lymph node frozen sections.

Ackerman [2] et al. (1959) affirmed that the evaluation of the cut surface of the lymph nodes was important, since a firm and grey appearance was likely to be microscopically unremarkable, while the fish flesh appearance of lymphoma was well known. The areas of necrosis indicated a pathological process, which may have been

due to granulomas or metastasis. The authors did not encounter any difficulty in assigning a diagnosis of lymphoma and they did not feel the necessity to subtype the lymphoma, once the patient was put in this category.

Holaday [3] et al. (1974) stated that the improper gross sampling which was done by the surgeon or the pathologist was the reason for the discrepancy.

Rogers [1] et al. (1987) came across three false negative results that were traced to the microscopic sampling, while in one case, Hodgkin's lymphoma was misinterpreted as reactive hyperplasia. Ahmad [4] et al. (2008) also conformed to a similar reason for the error.

Oneson [5] (1989) reported a high inconclusive rate of 11%, stating that a higher deferral rate was acceptable in this group, as the prime reason for the consultation had been obtained for the biopsy and for special staining.

The microscopic sampling error led to a wrong diagnosis in 1 case (2.9%) and to an interpretative error in 1 case (2.9%). The discordance which was reported by other workers was also significantly low and there was agreement in the reason for the erroneous diagnosis.

The Oral Cavity

In the present study, 30 oral cavity specimens were subjected to frozen section analysis for their diagnoses. 3 were benign and 27 were malignant. All the benign lesions were precisely diagnosed as capillary hemangiomas. Among the 27 malignant lesions, gross sampling errors in 2 specimens resulted in a false negative diagnosis of moderate grade dysplasia.

The total diagnostic accuracy in the oral cavity lesions was 93.3%, which was slightly lower, as compared to that in the other studies in the literature, which had looked at the entire head and neck lesions. The slightly lower diagnostic accuracy of the frozen section in the oral cavity lesions can be attributed to the fact that the other studies had included the entire head and neck lesions and this had to be taken in account.

The Thyroid

In the present series, 18 thyroid specimens were subjected to the frozen section analysis. Ten (55.6%) were benign lesions, five (27.8%) were follicular neoplasms and three (16.6%) were papillary carcinomas. This was similar to those in the study which was done by João Paulo [6] et al. (2009).

The overall diagnostic accuracy was 100%. Although in this study, no difficulty was experienced in this category, the place of the frozen section in the thyroid was quite controversial.

The diagnostic importance of the psammoma bodies in the papillary carcinomas was emphasized by Kraemer [7] et al. (1987), who recommended a prompt search for a nearby carcinoma in such a case. Ackerman [2] et al. (1959) reported a case where, although the psammoma bodies were seen, they were not given the importance that they deserved, leading to a missed diagnosis of a papillary carcinoma. Another unique feature regarding the frozen sections in papillary thyroid carcinomas, is the absence of optically clear nuclei, which is a fixation artifact and one of the most reliable features on the paraffin sections. Kraemer [7] (1987) considers the presence of true papillae as the most reliable feature in the frozen sections.

In the analysis which was conducted by Nakazawa [8] et al. (1968), most of the errors were the result of a faulty sampling of the multicentric tumours, while others were attributed to microcarcinomas which were harboured in the glands, which otherwise had the features of goitre. In all these cases, a correct diagnosis could be rendered only after examining multiple paraffin sections. This brought out the difficulties which were encountered in frozen sections of the thyroid. These reasons were also stated by Holaday [3] et al. (1974), who found a 0.6% false negative rate. Nakazawa [8] et al. (1968) also experienced a difficulty in a colloid rich thyroid lesion, which would crumble on cryotomy, accounting for a missed diagnosis of a malignancy in the unsatisfactory preparation.

The status of the frozen sections as a cost effective tool was questioned by workers like McHenry [9] et al. (1996), DeMay [10] (1998), and Alonso [11] et al. (2003), since fine needle aspiration biopsy is regarded as the most accurate test, except in the follicular lesions. They opined that there was a low probability of picking up the follicular carcinoma on the frozen sections. Simpson [12] (1998) however contradicted this view and advocated cytology as an adjunct to the frozen sections.

The accuracy which was achieved in the present study was 100%, which was superior to that in most of the reports in the literature. However, the fact that these reports looked at a much larger sample size, cannot be underscored.

The Salivary Gland

In the present study, 17 salivary gland lesions were subjected for the frozen section examination. Of these, 82.4% were benign and 17.6% were malignant. This was similar the picture in the study of Allen [13] et al. (1983), Granick [14] et al. (1985), Rigual [15] et al. (1986), Gnepp [16] et al. (1987) and Yong [17] et al. (1996).

15 were diagnosed correctly on the frozen sections. 8 of these were diagnosed as pleomorphic adenomas, 4 as chronic sialadenitis, 2 as Warthin's tumour, and 1 as an adenoid cystic carcinoma.

An interpretative error in 1 case resulted in a false diagnosis of a pleomorphic adenoma, owing to the abundant chondromyxoid ground substance and the sparse epithelial structures, which on the paraffin sections, was diagnosed as carcinoma ex pleomorphic adenoma.

A gross sampling error in the other parotid specimen resulted in a false negative diagnosis of absence of tumour. The final diagnosis was deferred for the permanent section examination, which revealed an adenoid cystic carcinoma.

Thus, the overall diagnostic accuracy for the salivary gland lesions was 88.2%, a discrepancy was found in 1 (5.9%) case and a deferral was found in 1 (5.9%) case. This was in concordance with the findings in the studies of Yong [17] et al. (1996) and Seethala [18] et al. (2005), while these values were slightly lower than those in other studies.

Assessment of the Discrepancy and the Errors

The primary purpose of the frozen section analysis is to determine the correct pathological process and to decide the therapeutic course of action. Therefore, when it was viewed with respect to the diagnosis of a correct pathological process, 6 out of the 100 frozen sections in the current study were found to be erroneous. This amounted to a discordant diagnosis rate of 6%, which was only due to a false negative error.

Ackerman [2] et al (1959) reported 2% errors and they believed that the errors would be reduced to a minimum if the clinical data were available and if the technique which was employed was of a high quality. However, a pathologist must refrain from getting swayed by the overwhelming clinical data and he/she must report only what he/she sees.

In the series which was undertaken by Horn [19] (1962), 1.9% lesions were labeled as false negative, while 0.6% lesions were rendered a false positive diagnosis of malignancy. The author is of the view that a false negative diagnosis may simply indicate an unlucky selection of the tissue, but in some cases, especially in those of the thyroid, where the decision requires a lengthy study of multiple sections, these problems may be virtually unavoidable. According to Horn [19] (1962), the incorrect diagnosis of a malignancy was far more important than an incorrect diagnosis of benignancy, as the former could subject the patient to a needless mutilating surgery.

Holaday [3] et al (1974) highlighted the significant differences between the frequencies of the false positives and the false negatives. Although a low figure of 1% false negatives was seen, it was still six times the occurrence of the false positives (0.15%). This analysis brought out the overall attitude of the pathologist towards the extreme conservatism.

The categorization of the errors which were found in the present study, was based on the review which was made by Rogers [1] et al (1987), with the inclusion of technical artifacts as one of the most potential sources of the errors. In their experience, interpretation errors contributed to 57% of the total errors, while microscopic sampling, gross sampling, and a lack of communication accounted for 24%, 9.5% and 9.5% of the total errors respectively. We divulged from their results, as gross sampling errors were responsible for 40% of the errors in our case.

The results of the present study were strongly congruent with those of the study which was done by Nakazawa [8] et al (1968), with respect to the extremely low false positive diagnoses as compared to the false negative diagnoses. The reasons for the discrepancy were also comparable, with gross sampling and misinterpretation being the most frequent factors.

The most significant source of the errors in the present study was a faulty gross sampling, which contributed to 50% of the total errors, which was also found by Nakazawa [8] et al (1968), Gandour-Edwards [20] et al (1993) and Ahmad [4] et al (2008) in their studies.

Accuracy of the Frozen Sections

An overall accuracy of 94% was achieved in our study, which was slightly lower than those in most of the reviews in the literature. Ackerman [2] et al (1959), Rogers[1] et al (1987), and Ahmad [4]et al (2008) reported a 100% accuracy for the head and neck frozen sections, while was comparable to that in the studies of Remson [21] et al (1984) and Gandour-Edwards [20] et al (2006), who found accuracies of 96 % and 97.7%.

The New Emerging Techniques and Their Comparison with the Frozen Sections

In the recent times, the role of frozen sections is minimal. Cytology has played a major role in avoiding an intraoperative diagnosis and if at all it is required, then a crush or an imprint smear cytology is a better option. The crush smears are more frequently used in neurosurgical specimens than in other fields.

Moreover, microwave processing of the biopsy sections is superior to that of the frozen sections. The microwave processing reduces the preparation time (2-3 hours, which includes the fixation, processing, microtomy, and the staining) and it allows the same-day tissue processing and the diagnosis of the small biopsy specimens without compromising on the overall quality of the histologic sections. In some instances, it is desirable to perform a biopsy and a definitive surgery on the same day, thereby decreasing the patients' expenses and the requirement for multiple trips between their homes and the referral centres. Reliance on frozen sections with its attendant difficulties of interpretation and greater expenses can be reduced substantially [22].

Inspite of the fact that other techniques are there which have got their own advantages, we cannot overlook the importance and the frequent use of frozen sections in surgery.

CONCLUSION

This study highlighted that inspite of the varied types of diagnoses in head and neck lesions, a reasonably good percentage of accuracy could be achieved in the frozen sections. Also, for the margin assessment like other sites, it plays an important role.

Hence, frozen sections should also be used for the diagnosis and the margin assessment in head and neck lesions.

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