

RESEARCH ARTICLE

A Comparison between the Diagnostic Accuracy of Frozen Section and Permanent Section Analyses in Central Nervous System

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Abstract

Objective: Using diagnostic pathological methods during surgery is a valuable means of determining the appropriate management for patients. Application of Frozen Section in CNS surgeries might face challenges due to friability of brain tissue and its relative inaccessibility. Various studies have evaluated the diagnostic acuity of frozen section compared to gold standard but results have been quite inconsistent. We conducted the present study to evaluate the accuracy of cryostat in diagnosing central nervous system tumors compared to the Gold Standard method. **Methods:** In this descriptive retrospective study, patients with definite diagnosis of central nervous system tumors made through histopathological evaluations were identified by reviewing the archives of pathology reports during 1996-2013. Demographic data, clinical history, radiologic findings and results of pathologic evaluations were extracted from the medical records and entered into SPSS statistical software v.22 for analysis. **Results:** A total of 405 patients diagnosed with CNS tumors were identified, of which 16 patients were not eligible and eventually 389 patients were included in the study. Regarding tumor category, subtype and grade, the results of the two methods were totally compatible in 303 patients (77.9%) and discrepant in 22.1% of cases. The tumors located in the middle fossa ($p=0.031$; OR=2.27; 95% CI: 1.08-4.79) and the posterior fossa ($p=0.021$; OR=2.46; 95% CI: 1.15-5.26) and the tumors biopsied using the stereotactic method ($p=0.050$; OR=2.42; 95% CI: 1.001-5.83) were associated with an increased chance of discrepant results between the two methods. **Conclusion:** Frozen section can correctly diagnose and affect the management of CNS lesions in 77.9% of cases. Finding ways to increase the sensitivity and specificity of this method and providing surgeons with more definite and exact intra-operative diagnosis can improve management of central nervous system lesions to a considerable degree.

Keywords: Central nervous system tumors- frozen section analysis- permanent section analysis- compatibility

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Introduction

Application of intra-operative histopathological evaluations is a valuable method that can guide the treatment of patients (Somerset and Kleinschmidt-DeMasters, 2011). Frozen section analysis was first introduced in 1891 by a pathologist in Johns Hopkins hospital and is currently used all over the world. Application of this method became much easier by the improvements made in cryostat devices mainly during 1950-1960 (Shrestha et al., 2009). Frozen section provides the surgeons with a primary diagnosis that can compensate for the shortcomings of pre-operative diagnostic assessments (Somerset and Kleinschmidt-DeMasters, 2011).

In the extensive study conducted by the College of American Pathologists (CAP) on more than 90,000 diagnoses made through frozen section analysis in 461 different medical centers, the diagnostic accuracy of frozen

section was calculated to be 98.52% (Shrestha et al., 2009).

Compared to other intra-operative diagnostic methods, the tissue structure is better preserved in frozen section analysis and this method better visualize the structural arrangement of cells, stroma and the tumor (Di Stefano et al., 1998; Sharma and Deb, 2011). Frozen section can provide determinant information in a very short period of time (Khalid and AUI, 2004) and can spare patients the need for a re-operation (Mitra et al., 2010).

Intra-operative histopathological evaluations play a key role in diagnosis of Central Nervous System (CNS) tumors since the clinical presentation and radiologic findings in these lesions do not provide sufficient information and a definite diagnosis relies upon the histopathological findings (Jaiswal et al., 2012). Frozen section analysis can precisely diagnose CNS tumors (Plessec and Prayson, 2007); however, sometimes it is performed only to determine the grading of the tumor

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(Khalid and AUI, 2004).

An intra-operative diagnostic method should be able to provide the diagnosis within minutes, preserving a considerable proportion of the specimen for analysis by the gold standard method. Therefore, requiring a relatively large specimen for frozen section analysis is one of the main shortcomings of this method (Goel et al., 2007), becoming more eminent in the lights of the improvements made in stereotactic biopsy methods (Khamechian et al., 2012).

Accordingly, we aimed to assess the compatibility between results of frozen section analysis and permanent section analysis in patients diagnosed with CNS tumors in Shohada-e-Tajrish Hospital during 1996-2013.

Materials and Methods

In this descriptive retrospective study, patients with definite diagnosis of a CNS tumor made through histopathological evaluations were identified by reviewing the archives of pathology reports documented in the department of pathology of Shohada-e-Tajrish Hospital during 1996-2013. Of these patients, subjects who had undergone a frozen section analysis of the tumor and their diagnoses were recorded based on the findings of the gold standard method were included as the study population. A total of 405 patients diagnosed with CNS tumors were identified, of which 16 patients were excluded because making a definite final diagnosis was not possible by accessible methods and so further immunohistochemical study was requested or their data were not sufficient for inclusion. Eventually 389 patients were included in the study.

The slides of these patients had been evaluated and reported by seven different pathologists during these years, who were interested in the field of neuropathology with at least five years of experience in this field. For every frozen section analysis, most often two pathologists evaluated the slides and reached a consensus on frozen section diagnosis.

In order to assess the sources of discrepancies between permanent and frozen section analyses, an experienced pathologist interested in neuropathology, blinded to previous diagnoses, re-evaluated the available slides. The diagnoses made by this pathologist were compared to previous results of both frozen section and permanent analyses and accordingly the possible sources of discrepancies were identified. Since the included patients had been initially assessed during a wide period of time (1996-2013), the slides of 64 out of 86 patients with discrepant results were available.

Age, gender, radiological findings, location, biopsy method, result of frozen section analysis and the definite diagnosis of the gold standard method were extracted from the medical records and entered into SPSS statistical software v.22 (SPSS, 2013) for analysis. Comparison between frozen section analysis and the results of gold standard method was done according to the World Health Organization classification (Bosman et al., 2013). The study protocol was evaluated and approved by the Ethics Committee of Shahid Beheshti University of Medical

Sciences. The extracted data were used anonymously and the patients' information was regarded confidential throughout the survey.

Descriptive analysis of the data was performed presenting the results as frequency and percent for qualitative variables and as mean and standard deviation for age. The relation between qualitative variables was evaluated by Chi-squared test and Fisher's Exact test if needed. The method proposed by Hosmer and Lemeshow for purposeful selection of variables in logistic regression analysis (Bursac et al., 2008) was used in this survey in order to assess the independent risk factors for discrepancy between the results of frozen section and permanent section analyses. Accordingly, univariate analyses were performed on all the factors evaluated in the study and the ones correlated to discrepant results with a p value of less than 0.2 were selected as candidates for multivariable analysis. Subsequently all the selected factors were included in the multivariable regression model and the results were presented as odd ratios and 95% confidence intervals. So the final multivariable model included only variables found to be significant univariate predictors with a p value < 0.2, less than 20% missing data and no significant collinearity.

Results

Data gathered from a total of 389 eligible patients diagnosed with CNS tumors were included in this study. Table 1 presents the demographic characteristics and clinical information of the sample population. The mean age of the subjects was 34.52 ± 18.39 years with a minimum of 1 and a maximum of 77 years old. Thirty seven patients (9.2%) were aged 0-9 years old, 144 patients (35.6%) were aged 10-19, 144 (35.6%) were aged 20-39, 118 (29.2%) were aged 40-59 and 40 (9.9%) patients were aged 60-90 years old. A male preponderance was observed in the sample population with 231 males (57.0%) and 174 females (43.0%) and a male/female ratio of 1.3. The average age of male patients was higher than females but the differences were not statistically significant (35.8 ± 18.7 vs. 32.8 ± 17.9 , $p=0.101$).

Tumors were located in the brain in 356 subjects (87.9%) and in the spinal cord in the 49 (12.1%) remaining patients. Based on their radiological findings the lesion was solid in 337 patients (83.2%), cystic in 20 patients (4.9%) and was composed of both solid and cystic components in 48 (11.9%) subjects. No enhancement was observed in 303 cases (74.8%), lesions in 87 subjects (21.5%) were heterogeneous and homogeneously enhanced and ring enhancement was reported in 15 patients (3.7%). Calcification was visualized in only 16 patients (4.0%). The majority of lesions were intra-axial (61.0%) and extradural tumors had the lowest prevalence (2.5%). Considering their anatomical location, most lesions were purely located in the middle fossa (28.4%) followed by the anterior fossa (26.7%) and posterior fossa (26.4%). As the least common locations, thoracolumbar region was involved in only one patient and another subject had simultaneous involvement of anterior, middle and posterior fossa. The majority of patients had undergone

open biopsies (91.6%) and stereotactic method was applied in only 34 subjects (8.4%).

Regarding the microscopic findings, necrosis was reported in 71 cases (17.5%), mitosis in 77 (19.0%), pleomorphism in 128 (31.6%) and endothelial proliferation in 44 patients (10.9%).

The most common diagnosis made by frozen section analysis was low grade astrocytoma reported in 46 patients (11.4%) followed by meningioma diagnosed in 39 subjects (9.6%), ependymoma in 24 (5.9%) and GBM also in 24 cases (5.9%). As for the permanent section analysis, low grade astrocytoma was similarly the most common diagnosis established for 53 patients (13.1%) followed by meningioma in 40 cases (9.9%),

ependymoma in 33 (8.1%) and GBM in 29 patients (7.2%). Of the tumors diagnosed by permanent section analysis to be low grade astrocytoma, 79.2% were correctly identified by frozen section analysis. This figure for meningioma, ependymoma and GBM was found to be 92.5%, 63.6% and 82.8%, respectively.

Compatibility between the two methods was assessed considering the main diagnosis, its typing details, grading of the tumor and differential diagnoses proposed. Accordingly, the results of the two methods were concordant in 303 patients (77.9%) while they were not compatible in the remaining 86 cases (22.1%). The most common diagnosis in compatible cases was low grade astrocytoma found in 42 patients (13.86%) followed

Table 1. Demographic Characteristics and Clinical Information of the Study Population

Variables	Frequency	Percent	
Age Groups	0-9	37	9.20%
	10-19	65	16.10%
	20-39	144	35.60%
	40-59	118	29.20%
	60-90	40	9.90%
Gender	Male	231	57.00%
	Female	174	43.00%
Components	Solid	337	83.20%
	Cystic	20	4.90%
	Solid and Cystic	48	11.90%
Enhancement	No Enhancement	303	74.80%
	Ring Enhancement	15	3.70%
	Total Enhancement	87	21.50%
Calcification	No Calcification	389	96.00%
	With Calcification	16	4.00%
Involved Organ	Brain	356	87.90%
	Spinal Cord	49	12.10%
Location	Intra-axial	247	61.00%
	Extra-axial	110	27.20%
	Extradural	10	2.50%
	Intradural-Extramedullary	13	3.20%
	Intramedullary	25	6.20%
Tumor Site	Anterior Fossa	108	26.70%
	Middle Fossa	115	28.40%
	Posterior Fossa	107	26.40%
	Anterior and Middle Fossa	15	3.70%
	Middle and Posterior Fossa	10	2.50%
	Cervical	19	4.70%
	Thoracic	13	3.20%
	Lumbar	10	2.50%
	Sacral	3	0.70%
	Thoracocervical	3	0.70%
Thoracolumbar	1	0.20%	
Type of Biopsy	Open	371	91.60%
	Stereotactic	34	8.40%

Table 2. Discrepancies between Permanent Section and Frozen Section Analysis

Type of Discrepancy	Frozen Section	Permanent Section	Frequency	Total		
Defer (Wait for Permanent)	Defer	Low grade Astrocytoma	1	7 (1.8%)		
		Craniopharyngioma	1			
		Hemangioma	1			
		Lymphoma	1			
		Meningioma	1			
		Metastatic Carcinoma	1			
		Unremarkable Brain Tissue	1			
Reactive Versus Neoplastic Discrepant	Encephalitis Ganglioglioma Neoplastic Tissue Reactive Gliosis Granulation Tissue	Lymphoma	1	9 (2.3%)		
		Granulation Tissue	1			
		AVM	1			
		Embryonal Carcinoma	1			
		Low grade Astrocytoma	2			
		Lymphoma	1			
		Schwannoma	1			
Unremarkable Brain Tissue		Metastatic Papillary Adenocarcinoma	1			
Only Malignancy Confirmed	Malignant Neoplasm	Anaplastic Ependymoma	1	13 (3.3%)		
		GBM	3			
		Germ Cell Tumor	1			
		Malignant Chordoma	1			
		Medulloblastoma	1			
		Malignant Meningioma	1			
		Metastatic Carcinoma	2			
		Anaplastic Oligodendroglioma	1			
		Pineoblastoma	1			
		PNET	1			
		Branch of Neoplasm Discrepant or not Identified	Craniopharyngioma		Germ Cell Tumor	1
Pituitary Adenoma	2					
Ependymoma	Meningioma		1			
	Neurocytoma		1			
Germ Cell Tumor	PNET		1			
	Lymphoma		1			
Glioma	Low grade Astrocytoma		1			
	Ependymoma		1			
Lymphoma	Germinoma		1			
	Ependymoma		2			
Neoplastic Tissue			Gliosarcoma	1		
			Neurocytoma	1		
			Oligodendroglioma	1		
			Optic Nerve Glioma	1		
			Pilocytic Astrocytoma	3		
			Pineocytoma	1		
			Pituitary Adenoma	3		
			Schwannoma	2		
			Neovascularity	Metastatic Carcinoma	1	
			Neurocytoma	Ependymoma	1	
		Neurofibroma	Pilocytic Astrocytoma	1		
		Teratoma	Chordoma	1		
		Ependymoma	Ependymoma	1		
Subtype and Grade of Glioma Discrepant or not Identified	Glioma	Anaplastic Astrocytoma	4	12 (3.1%)		
		Low grade Astrocytoma	4			
		Ependymoma	2			
		GBM	1			
		Pilocytic Astrocytoma	1			
Grading of Glioma Discrepant	Anaplastic Astrocytoma	Low grade Astrocytoma	1	8 (2.1%)		
		Pilocytic Astrocytoma	1			
	Low grade Astrocytoma	Anaplastic Astrocytoma	3			
		GBM	1			
	High grade Glioma	Low grade Glioma	1			
		High grade Glioma	1			
Subtyping of Glioma Discrepant or not Identified	Astrocytoma Ependymoma Low grade Glioma	Ependymoma	1	7 (1.8%)		
		Low grade Astrocytoma	1			
		Low grade Astrocytoma	1			
		Ependymoma	2			
		Pilocytic Astrocytoma	1			
		Ependymoma	1			
Total				86 (22.1%)		

Table 3. Multivariable Logistic Regression Analysis Evaluating the Risk Factors for Discrepancy in Results of Frozen Section and Permanent Section Analysis

Variables	Odds Ratio	95% C.I	P value
Gender			
Male	Reference		
Female	1.557	(0.935-2.590)	0.089
Calcifications	2.5	(0.849-7.364)	0.096
Site of Tumor			
Anterior Fossa	Reference		
Middle Fossa	2.274	(1.079-4.791)	0.031*
Posterior Fossa	2.457	(1.146-5.265)	0.021*
Anterior and Middle Fossa	3.431	(0.979-12.022)	0.054
Middle and Posterior Fossa	2.616	(0.555-12.335)	0.224
Cervical	3.223	(0.964-10.770)	0.057
Thoracic	3.803	(0.838-17.249)	0.083
Lumbar	2.102	(0.391-11.295)	0.386
Sacral	4.459	(0.366-54.346)	0.241
Thoracocervical	4.459	(0.366-54.346)	0.241
Thoracolumbar	-	-	-
Type of Biopsy			
Open	Reference		
Stereotactic	2.416	(1.001-5.834)	0.050*
Pleomorphism	1.683	(0.990-2.860)	0.054

*, Factors with significant p values are presented in bold, which indicate the higher risk of discrepant results between permanent and frozen section analyses in tumor located in middle and posterior fossa and the lesions biopsied via stereotactic method.

by meningioma diagnosed in 37 cases (12.21%). Unremarkable brain tissue was reported in 9 subjects (2.97%).

Based on the various discrepancies between the results of the two methods, 7 subcategories were described which included (Table 2).

1. Defer (wait for permanent)

In 7 patients (1.8%) frozen section analysis was not able to make any suggestions while permanent section analysis established diagnoses for the subjects such as low grade astrocytoma.

2. Reactive versus neoplastic discrepant

Reactive versus neoplastic discrepancy was observed in 9 cases (2.3%). For example a sample diagnosed as granulation tissue by frozen section analysis was reported by the permanent section to be low grade astrocytoma.

3. Only malignancy confirmed

For 13 subjects (3.3%) frozen section could only identify the malignant nature of the tumor while permanent section analysis provided a specific diagnosis. As an example, a tumor that the pathologist could only identify its malignant nature was reported by the permanent section analysis as a medulloblastoma.

4. Branch of neoplasm discrepant or not identified

The branch of the neoplasm was not identified or incompatible in 30 subjects (7.7%). For instance, a lesion diagnosed as ependymoma by frozen section, was found

to be a meningioma based on permanent section analysis.

5. Subtype and grade of glioma discrepant or not identified

In 12 patients (3.1%) both the subtype and the grade of tumor were discrepant between the two methods. For instance a tumor recorded as glioma by the frozen section, was reported as low grade astrocytoma by the permanent section. Although to assess these incompatibilities the exact request of the surgeon from the pathologist should be evaluated. They might have merely asked whether the tumor is a glioma or not.

6. Grading of glioma discrepant

The grading of glioma was discrepant in 8 patients (2.1%). As an example, a patient diagnosed with anaplastic astrocytoma by frozen section analysis was to have a low grade tumor of this nature.

7. Subtyping of glioma discrepant or not identified

The subtyping of glioma was discrepant in 7 patients (1.8%). For instance, a low grade astrocytoma lesion identified by frozen section analysis was reported by permanent section analysis as an ependymoma.

Exploratory univariate analysis found gender (p=0.156), presence of calcifications (p=0.130), tumor site (p=0.112), type of biopsy (p=0.124) and visualization of pleomorphism in microscopic evaluations (p=0.085) as potential covariates for multivariable analysis. The results of the multivariable regression analysis is presented in Table 3. Based on the presented results, the likelihood of a discrepant result in the tumors located in the middle fossa

is 2.27 times that of lesions located in the anterior fossa ($p=0.031$; $OR=2.27$; 95% CI: 1.08-4.79). Similarly the chance of a discrepant result is 2.46 times in the lesions of posterior fossa compared to anterior fossa ($p=0.021$; $OR=2.46$; 95% CI: 1.15-5.26). The chance of a discrepant result in tumors biopsied using the stereotactic method was also found to be 2.42 times that of the lesions openly biopsied ($p=0.050$; $OR=2.42$; 95% CI: 1.001-5.83).

Discussion

Frozen section analysis is a common histopathological diagnostic method used during surgical procedures performed on patients with CNS tumors. One of the reasons for its extensive use is that frozen section analysis is a fast method that can provide surgeons with determinative information for the treatment of patients during the surgical procedure, which is utmost important particularly in high risk surgeries such as craniotomies (Plesec and Prayson, 2007). Moreover, while sending the specimens for frozen section analysis, the pathologist can evaluate the remainder of the specimen for later permanent section analysis, which becomes more prominent with the improvements of stereotactic biopsy methods in CNS tumors and if the pathologist declares insufficiency of the remaining specimen, the surgeon can take more biopsies from the tumor in the same session.

Previous studies have reported similar results on the diagnostic accuracy of frozen section analysis; however, considering the incompatibilities between the results of frozen section analysis and gold standard methods along with the improvements in intra-operative diagnostic methods, the practical diagnostic value and accuracy of this method is being questioned (Khalid and AUI, 2004). In the study conducted by Shrestha et al. (2009) in BPKMCH, 404 cases were evaluated during 2003 to 2007 and the diagnostic accuracy of frozen section analysis was reported to be 94.6% (Shrestha et al., 2009). However, they evaluated tumors of different organs in the body and did not limit their study population to patients with CNS tumors.

In the present study, 389 patients with definite diagnosis of CNS tumors were included from the patients treated in the referral center of Shohada-e-Tajrish Hospital during 1996-2013. In 22.1% of cases the results of frozen section and permanent section were incompatible to some extent and in 77.9% the results of the analyses were exactly the same.

We applied the diagnostic classification proposed by the WHO. Classification method of the diagnoses is an important factor in measuring the diagnostic accuracy of frozen section analysis method. Moreover, finding a suitable method to classify cases in order to compare the accuracy of the two diagnostic measures in this setting is very challenging. In a similar attempt Khoddami et al. (2015) aimed to assess the accuracy of frozen section results in 273 CNS lesions by categorizing the cases into three groups of complete concordance, partial concordance and discordant (Khoddami et al., 2015). Such classification probably yields a higher accuracy rate. However, we tried to classify the patients in greater

details with the expense of yielding a lower accuracy figure in order to reach more comprehensive results, from which sources of diagnostic errors could be extracted. Accordingly, in 77.9% of the patients evaluated, the results of frozen section analysis were entirely compatible with that of the permanent section analysis. Among the remaining cases (22.1%) with incongruent results, seven distinct patterns of incompatibility were identified and subjects were classified accordingly.

Various factors might lead to an inaccurate diagnosis by frozen section analysis which include sampling errors, difficulties in histologic determination of the cell types or errors in identifying the grade of the lesion (Folkerth, 1994). Sampling error seems to be an inevitable compromise in exchange for the rapid intraoperative diagnosis, the importance of which becomes more prominent in assessment of tumors such as gliomas that are heterogeneous and require a thorough precise sampling. On the other hand, few of these errors might be improvable such as inaccurate grading of the lesions or incorrect diagnoses made by the pathologist (Plesec and Prayson, 2007).

As mentioned, the diagnoses made by an experienced pathologist blinded to previous results were assessed and accordingly the possible sources of discrepancies were detected. These identified causes can be listed in order of frequency between discrepant cases: sampling error, technical error, inadequate/inaccurate clinical information and pathologist error.

Sampling error was one of the main causes of discrepancies between diagnoses in a way that in almost all discrepant categories some cases of sampling error were identified. For instance in one of the cases deferred during frozen section analysis, the sample included bone trabeculae and since it could not be processed appropriately during frozen section, the pathologist had to defer the result, while in permanent section analysis the specimen was diagnosed as hemangioma. In another case, the sample for frozen section analysis was obtained from the necrotic area of the tumor and so the result was deferred. However, permanent section analysis of the resected tumor suggested the final diagnosis of metastatic carcinoma. Sampling error had caused discrepancy in diagnosis of glial tumors as well. For example, the biopsy for frozen section analysis was taken from the low grade areas of the tumor and so the pathologist reported the specimen as low grade astrocytoma, while examination of the tissue sent for permanent section analysis revealed the high grade characteristics of the tumor as well and the diagnosis was changed to anaplastic astrocytoma. In fact, the tumor had transformation in different parts which highlights the importance of a proper sampling.

Another source of discrepancies between permanent and frozen section analyses was found to be technical errors. For example, in one case the sections in frozen analysis led the pathologist to make a diagnosis of ependymoma while the further complete sections during permanent analysis revealed a final diagnosis of neurocytoma. Technical errors had also been identified as the source of misdiagnosis in glial tumors. For instance, a diagnosis of glioma without further identifying the subtype

and grade was made by frozen section analysis due to unavailability of crush smear, and the final diagnosis was found to be low grade astrocytoma.

The other source of discrepancies between permanent and frozen section analyses were insufficient or even false clinical information provided for the pathologist during frozen section analysis. As an example, the pathologist could only identify the benign nature of the neoplasm in a pituitary adenoma because the suprasellar location of the mass was not mentioned to the pathologist for frozen section analysis. If the pathologist was provided with the required information, a more precise diagnosis would have been reported. In another case, based on the information recorded in the patient's medical history indicating the location of the lesion as intra-axial, the pathologist made the diagnosis of ependymoma in frozen section analysis. However, the final diagnosis was found to be meningioma and the radiologic information provided in the history was false and misleading.

The third possible cause of discrepancy was the pathologist error which was typically the source of discrepancy in the grading and subtyping of glial tumors; although, it was suspected to be the reason for other misdiagnoses as well. For instance, in the case with a frozen section diagnosis of encephalitis and a permanent section diagnosis of lymphoma, the blinded pathologist in the present study could make the diagnosis of lymphoma based on the frozen slides. Therefore, this discrepancy could be attributed to the pathologist error. Nevertheless, considering the process in our pathology department through which the final diagnoses were made (participating two pathologists in each frozen section analysis), we could not include the experience of the corresponding pathologist as an independent variable in our analyses and assess its effects on the results.

In a similar survey, Regragui et al., (2003) assessed the compatibility between results of intraoperative frozen section analysis with permanent diagnoses in 1,315 cases and reported the concordance in categorizing tumors as neoplastic or non-neoplastic to be 96.6% and in identifying the malignant nature of the tumor to be 92.6%. The rate of precise histologic compatibility was also reported at 87.6%. Gliomas, metastases and hemangioblastomas were found to be associated with the highest chance of diagnostic errors. Similarly, in the present survey gliomas had the highest chance of diagnostic errors and more specifically, ependymomas followed by low grade astrocytomas; however, the differences between various types of tumors were not statistically significant. The errors were made mostly on establishing the subtype or grade of these lesions.

Reactive versus neoplastic discrepancy, which has been mentioned in previous studies as well, was observed in 9 cases (2.3%). For example a patient with a report of granulation tissue by frozen section analysis was found by the permanent section to be a low grade astrocytoma. In the study conducted by Plesec et al., (2007) on 2,156 CNS cases evaluated, 2.7% had discrepant diagnoses, of which 14.0% involved errors in differentiating reactive from neoplastic processes.

One of the most difficult diagnostic challenges in

neuropathology is differentiating between a low grade glial tumor and a reactive gliosis. Reactive astrocytes in gliosis lead to an evenly distributed hypercellularity as opposed to the uneven distribution of neoplastic cells in gliomas. The nuclear and cytoplasmic characteristics of the cells can also be used in differentiating the two, since reactive astrocytes present with slightly enlarged eccentric nuclei within eosinophilic cytoplasm with stellate processes while tumor cells have nuclei with irregular contours, hyperchromasia and heterogeneous chromatin. Nuclear-cytoplasmic ratio is lower in reactive gliosis while increased in astrocytoma cells. Presence of other features such as mitotic and microcystic changes, microcalcifications and clustering of cells around vessels or neurons are also suggestive of a non-reactive lesion and can help the diagnosis (Plesec and Prayson, 2007).

In an earlier publication, Oneson et al., (1989) reported their assessment of 1000 intraoperative consultations. Lesions of the nervous system were present in 91 samples, among which diagnostic errors by intraoperative frozen section analysis were made in 13 (14.3%) cases. In 11 (12.1%) of these cases, the diagnosis was deferred to permanent section analysis. Although in the present study the rate of diagnostic errors was higher than their survey (22.1% vs. 14.3%), but the percentage of cases deferred to permanent section analysis was considerably lower (1.8% vs. 12.1%). It seems that the pathologists in their study were more conservative and preferred to defer to permanent section assessment rather than taking risk of making diagnostic errors.

Geramizadeh et al., (2010) compared the overall accuracy of frozen section analysis with permanent section and reported that CNS lesions had the highest rate of discrepancy. They attributed these diagnostic errors to incorrect interpretation, sampling error and the insufficient clinical information provided for the pathologist. As other possible factors, we assessed the effects of tumors' location, their radiologic characteristics and method of biopsy. Based on the results of multivariable regression analysis tumors located in the middle fossa or the posterior fossa and the lesions biopsied using the stereotactic method were found to be associated with higher chance of their frozen section analysis being incompatible with the permanent section analysis. It can be attributed to the fact that access to the tumors located in the middle fossa and posterior fossa might be difficult for the surgeons which can affect the quality of samples obtained from these lesions. This might also be due to the difference in the types of tumors occurring in each location in the CNS.

In a more comprehensive study on stereotactic brain biopsy method, Brainard et al., (1997) assessed the frozen section analysis of 188 CNS specimens obtained via stereotactic biopsies. Based on their findings, they suggested that frozen section analysis at the time of surgery should be routinely performed to confirm the adequacy of obtained specimen, since 33% of their cases could not be diagnosed by the first stereotactic biopsy. They also reported an increase in the diagnostic yield from 67% to 89% by taking up to four biopsies. Their results are somehow congruent with our findings, indicating a higher chance of incompatibility in cases biopsied using

stereotactic method.

Overall it seems that as an important intra-operative diagnostic method, frozen section analysis provides the surgeon with vital information during the surgery and can successfully guide the treatment.

One of the limitations of this survey was the missing data in the medical records which was inevitable considering the duration of time passed from the hospitalization of the patients. Another problem was the changes in classification methods during these years which made the comparisons very difficult in some cases. Consultation with an experienced pathologist was used to solve the discrepancies caused by these changes. It should be mentioned that the clinical course of patients included in this survey was not affected by the errors made in frozen section analyses, since the results of their permanent section assessments were always ready within the duration of their hospitalization and the mistakes made in their final diagnosis were corrected. This survey merely aimed to acquire information on the primary diagnostic errors and provide suggestions on their possible sources.

Determining the diagnostic accuracy of frozen section analysis based on the results of gold standard method in CNS tumors is a challenging issue which can be attributed to inaccessibility of these lesions and their fragility. In the present study, frozen section analysis successfully guided the treatment of patients in 77.9% of cases.

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