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Correlation of Squash Cytology with Routine Histopathological Findings in Intraoperative Diagnosis of Glial Tumors

Dr Kirti Jetapariya, Dr Meena Patel*, Dr Hansa Goswami**
Third Year resident, Assistant Professor*,Professor & Head**,
Pathology Department, B.J medical College, Ahmedabad*,**

***Corresponding Author:**
Dr Kirti Jetapariya
Third Year resident, Pathology Department, B.J medical College, Ahmedabad

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Abstract

Introduction: Squash smear preparation is now well recognized and most commonly used in routine neurological practice. It helps in rapid intraoperative diagnosis within couple minutes of time, so it helps the surgeon to decide extent of surgery and any modifications in the surgery if required.

Materials and Methods: Study was conducted in the Department of Pathology in Tertiary Health Care center from March-2019 to October 2020. Total 68 cases of CNS glial tumors, who had both squash cytology and histopathological evaluation had been selected and studied retrospectively.

Results: Out of total 68 cases, 55(80.88%) cases were concordant while 5 (7.35%) cases were partial discordant and 8 (11.77%) cases were totally discordant. So diagnostic accuracy was of 80.88% but, out of 13 differed cases 5 cases were differed only due to grading deviation so it raised the diagnostic accuracy to 88.23%.Out of total 68 cases,40(58.82%) were male while 28(41.18%) were female. Most common age group to be involved was 41-60 year(39.7%).

Conclusion: Squash cytology is cost effective, reliable & rapid method with high accuracy on comparison to final Histopathological evaluation of CNS neoplasm. With attention towards causes of discrepancies, we can improve diagnostic accuracy of squash cytology.

Keywords: Glial tumor, Histopathological evaluation, Squash cytology

INTRODUCTION

Squash smear preparation is now well established and widely used in neurological practice. It helps in rapid intraoperative diagnosis within couple minutes of time, so it helps the surgeon to decide extent of surgery and any modifications in the surgery if required.[(1)] Squash cytology is simple, inexpensive, rapid, accurate and reliable tool for rapid intraoperative diagnosis of neurological biopsies.[(2)]

Glial tumors form the largest category of tumor modalities in CNS tumors. According to the, World Health Organization and American Brain Tumor Association[(3)], the most common grading system uses a scale from grade I to grade IV to classify benign and malignant tumor types. Among these, Grade I and II are benign slow growing, low grade glioma while Grade III(Anaplastic Astrocytoma) and IV(Glioblastoma Multiforme) are malignant, high grade variety.[(4)] Low grade tumors are curative under complete surgical excision but if left untreated turned out in malignant variety. While Malignant tumors require chemotherapy/ radiotherapy/ both. By squash cytology, quick evaluation of cytological features from prepared smear technique is done which gives preliminary diagnosis and helps the surgeon to differentiate between high grade and low grade glioma and to determine surgery accordingly.[(5)] Hence, this study was undertaken to correlate cytological features to that of histopathological

examination of CNS Glial tumors and to determine the diagnostic accuracy of squash cytology.

MATERIALS AND METHOD:

Study was conducted in the tertiary health care center from March-2019 to October 2020. Total 68 cases of CNS glial tumors, who had both squash cytology and histopathological evaluation had been selected and studied retrospectively. Tissue obtained during the time of surgery was sent to histopathology laboratory in normal saline where immediately squash smear prepared from it and stained with H&E and promptly evaluated. Relevant clinical and radiological data also noted. Remaining tissue bits or sometimes another specimen in 10% formalin were processed for routine histopathological evaluation. Then final comparison between histopathological findings and squash cytology was done.

RESULTS:

Out of total 68 cases, complete correlation was obtained in 55(80.88%) cases while 13(19.12%) cases were differed in diagnosis on squash cytology and histopathological examination. So diagnostic accuracy was of 80.88% but out of 13 differed cases, 5 cases were differed due to grading deviation so it raised the diagnostic accuracy to 88.23%. Out of total 68 cases, 40(58.82%) cases were male while 28(41.18%) cases were female.With regard to age, age ranging from 3 year to 67 years in our study. Age distribution is given in table 4 .Incidence is most common in 41-60 year of age group that is 39.7%. Most common tumors seen in children(<15 year) were of pilocytic astrocytoma while in adults(>45 year) most common tumors were of Glioblastoma multiforme. So in glial tumors, incidence of high grade tumors were increased with age while low grade tumors like pilocytic astrocytoma were decreases. Out of total 68 cases, 63(92.65%) were from brain while 5(7.35%) were from spinal cord. According to site, supratentorial tumors were 53(77.94%) while 15(22.06%) were of infratentorial in origin.

DISCUSSION:

Intraoperative consultation of CNS lesions are considered to be an important preliminary diagnostic tool to distinguish neoplastic lesions from non neoplastic conditions and it helps surgeon to modify the approach at surgery. Goal of pathologist in intraoperative setting is not to diagnose each and every case definitively, rather it's to provide sufficient preliminary information to optimize the surgery. [6,7]

In our study, out of 68 cases 55 were concordant and 13 were discordant and so diagnostic accuracy was 80.88% which is comparable with other studies. In Study done by P.Tejaswi and K.Shirisha, out of total 52 cases, 24 were glial tumor among which 19 were concordant while 5 were discordant and so diagnostic accuracy was 79.16%.[8] In Study done by Kalpana Deshpande and Sanjay Surase, out of total 250 cases, 96 cases were of glial tumors of which 81 were concordant while 15 were discordant , so diagnostic accuracy was 84.3%.[9] In Mitra S, Kumar M study, out of total 96 reported cases, 53 cases were of glial tumors out of which 45 were concordant, while 8 were discordant and diagnostic accuracy was 84.9%.[10]

A precise diagnosis requires a good correlation of clinical, radiological and histopathological data. The good correlation of above details could be the reason of high concordance in our study. Among various CNS lesions, Gliomas offered more diagnostic difficulties due to diversity of cells and sometimes difficulties in distinguishing between neoplastic and reactive nature of lesion. Improper grading of astrocytic neoplasm on cytological preparations can occur. Astrocytoma may vary significantly in grade from one area to another area in a single tumor.[11] In small biopsies, due to small tissue size, sampling might have failed to show both less and more aggressive components and only one of them is obtained on smear, so small biopsies are sometimes not suitable for grading of Gliomas.[12] In Squash cytology under grading of lesion is also a problem. The necrotic tissue, one of the key components of a high grade glioma is deliberately left behind during smear preparation and that results in under grading of tumor. So Multiple biopsies from different areas might helpful to decrease this false positive and false negative results.[13]

A case of pilocytic astrocytoma, which is a Grade I tumor, may be overgraded and misdiagnosed due to vascularity and nuclear atypia.[14] But this is seen quite less. In many cases of mixed glioma smaller component of oligodendriglioma or other component was missed, partly due to sampling error or also

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because of dense fibrillary background of astrocytic components. Differentiation of astrocytic elements from oligodendrial component is also important because immediate fixation of these biopsies decrease the diagnostic oligodendroglial artefacts from formalin fixation, which may be responsible for final histogenic and grading misinterpretation.[15] Nuclear features that include a salt-pepper appearance are features of neurocytoma, but in some cases cellular areas with alternating acellular fibrillary neuropil-rich areas may look like or be easily misinterpreted as ependymal rosettes.[16] But nuclear details of ependymomas can distinguished from those of neurocytomas by the presence of nuclear grooves and nuclear inclusions and stippled chromatin. Acellular perivascular fibrillary pseudorosettes, composed of round-shaped, monotonous, cohesive tumor cells are found in Ependymoma.[17] Thus it has provided diagnostic distinguishing points with round cell tumor. Sometimes there is difficulties in reactive and neoplastic nature of the cells. Few cases of low grade glioma may be misdiagnosed as gliosis since both have similar cytological features.[18] Reactive astrocytes had more abundant, well defined cytoplasm and prominent, numerous, long symmetrical cytoplasmic processes, whereas neoplastic astrocytes showed high cellularity, increased cell size along with increased pleomorphism. However these cases have not been included in the present study.

Technical errors in squash smear preparation can also limit the diagnostic process. Crushing and overstretching artifacts can occur in squash preparation from applying excessive pressure along with rapid pulling apart of the smears. Smears that are too thick to evaluate result from using too much tissue or using very firm or hard tissue fragments that resist spreading.

So Causes of Discrepancies in results may be due to one of the following reason:

1) Sampling error

2) Error in smear preparation

3) Staining error

4) Diagnostic error

CONCLUSION :

Squash smear is less expensive, reliable & rapid method with high accuracy on comparison to final Histopathological evaluation of CNS neoplasm. With attention towards causes of discrepancies,we can improve diagnostic accuracy of squash cytology. Further studies with larger sample size is recommended to explore this technology further.

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Tables

Table 1: Distribution according to age group

Age group	No of cases
0-20	13 (19.12%)
21 - 40	20 (29.42%)
41- 60	27 (39.70%)
>60	08 (11.76%)
Total	68

Table 2 : Distribution according to sex

Sex	No of cases
Male	40 (58.82%)
Female	28 (41.18%)
Total	68

Table 3: Distribution according to site

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Site		No of cases
Supra tentorial	Cerebral	44 (64.70%)
	Thalamus	02 (2.95%)
	Hypothalamus or 3rd ventricle	04 (5.88%)
	Lateral ventricle	02 (2.95%)
Infratentorial	Brainstem	04 (5.88%)
	Cerebellar or 4th ventricle	07 (10.29%)
Spinal cord		05 (7.35%)
Total		68

Table 4: Distribution according to diagnostic discrepancies

Glial tumor category	No of cases
Total concordant cases in diagnosis	55 (80.88%)
Partial concordant cases in diagnosis	05 (7.35%)
Total discordant cases in diagnosis	08 (11.77%)
Total cases	68

Table 5: Complete correlation between Squash cytology and Histopathological Examination

Glial tumor type	Squash Cytology (SC)	HP Examination (HPE)
Pilocytic Astrocytoma I	06	06
Fibrillary Astrocytoma II	08	08
Anaplastic Astrocytoma III	01	01
Glioblastoma Multiforme IV	08	08
Low grade glioma(I/II)	06	06
High grade glioma(III/IV)	15	15
Oligodendroglioma II	02	02
Anaplastic Oligodendroglioma III	02	02
Ependymoma	06	06

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Subependymal giant cell astrocytoma	01	01
Total	55	55

Table 6: Partial correlation due to grading deviation.

Squash cytology	Histopathological Diagnosis	Total cases
Glioblastoma Multiforme	Anaplastic Astrocytoma III	02
Low grade glioma	High Grade glioma	02
High grade glioma III	Low grade astrocytoma II	01
Total		05

Table 7: Total discordant cases

Squash cytology	HP Diagnosis	Total cases
Pilocytic astrocytoma I	Ependymoma II	01
Fibrillary astrocytoma II	Oligodendroglioma II	04
High Grade glioma	Ependymoma II	01
Oligodendroglioma	Fibrillary Astrocytoma	01
Ependymoma	Malignant round cell tumor	01
Total		08

Table 8: Comparison with other studies.

Study name	Total cases	Corcodant cases	Percentage
P. Tejaswi[8]	24	19	79.6%
Kalpana Deshpande [9]	96	81	84.3%
Mitra S, Kumar M[10]	53	45	84.9%
Present study	68	55	80.8%

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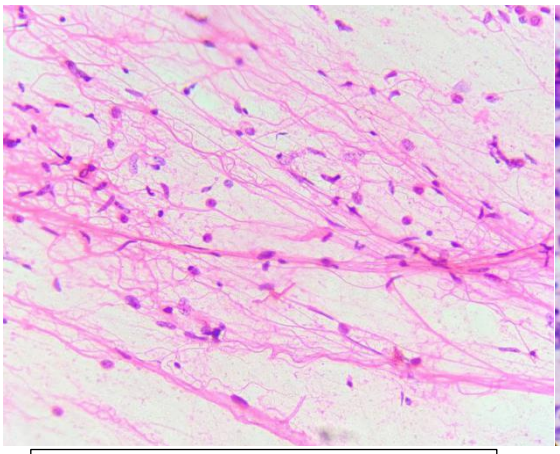

Figure :1 Pilocytic Astrocytoma
(40 x magnification)

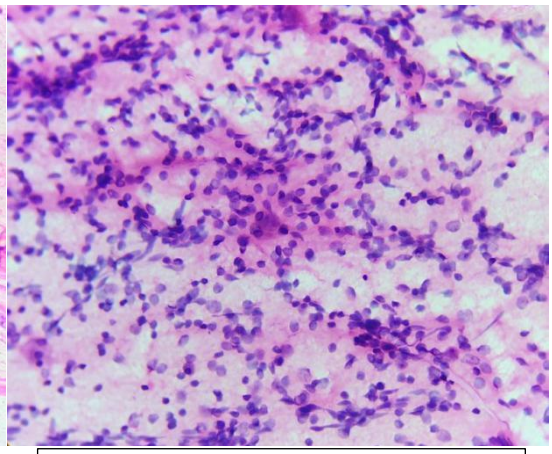

Fig:2 Diffuse Astrocytoma
(40 x magnification)

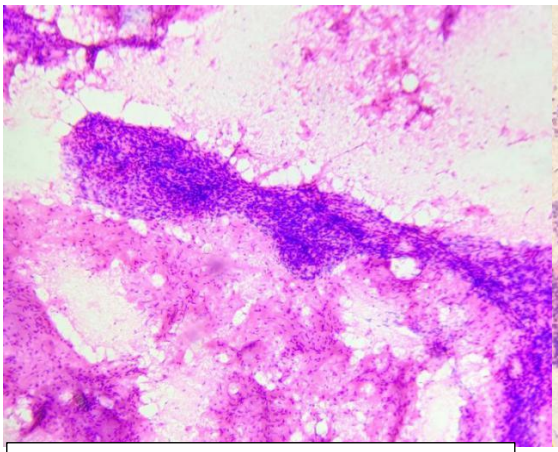

Figure :3 Glioblastoma Multiformes
(10 x magnification)

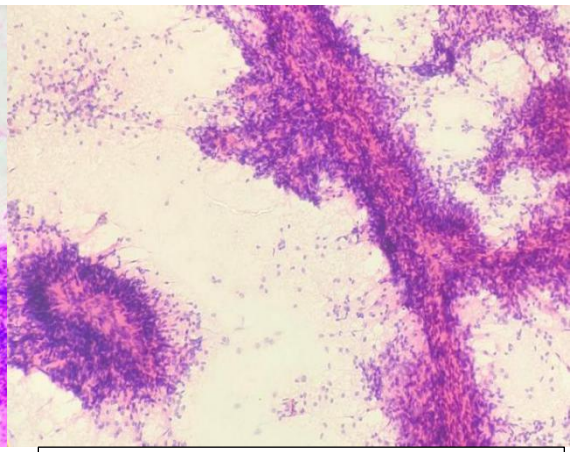

Figure :4 Ependymoma
(10 x magnification)

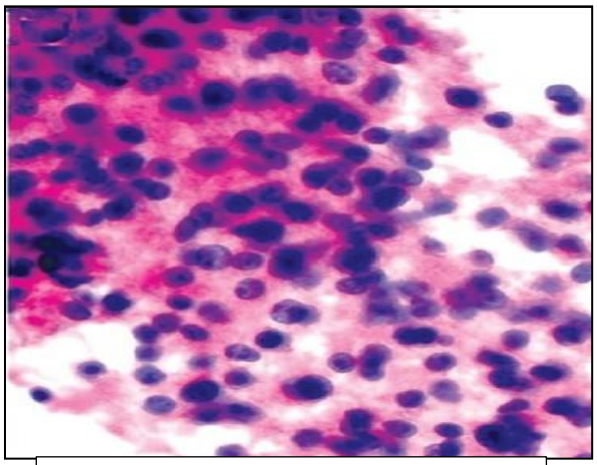

Figure : 5 Oligodendroglioma
(40 x magnification)

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