



UTILITY OF SQUASH SMEAR CYTOLOGY IN INTRAOPERATIVE DIAGNOSIS OF CENTRAL NERVOUS SYSTEM LESIONS

Pathology

Sachin R. Karanjekar

Department of Pathology, Superspeciality Hospital, Government Medical College Nagpur, Maharashtra

Sanjay N. Parate*

Department of Pathology, Superspeciality Hospital, Government Medical College Nagpur, Maharashtra *Corresponding Author

ABSTRACT

INTRODUCTION- The accurate diagnosis of disease tissue is elementary to the precise diagnosis and proper management of central nervous system lesions. Although histopathology is gold standard, in neurosurgical practice intraoperative cytological diagnosis is now well established and emerge as a dependable standalone diagnostic tool. The rapidity at which sufficient preliminary information for optimal surgery is provided to neurosurgeons, help him take crucial decision regarding patients management. Overall soft nature of CNS lesions, best suited for smear cytology, which in fact is the major restriction while performing intraoperative Frozen section consultation. So the present study was done to assess the utility of squash cytology in intraoperative diagnosis of CNS lesions.

MATERIAL AND METHODS- In this prospective study 142 patients with clinical diagnosis of CNS lesions were studied. Patients clinical and radio imaging findings were taken into account while evaluating squash smear preparation. Squash diagnosis was latter correlated with final histopathological diagnosis.

RESULTS- Out of 142 cases, cytological diagnosis was offered in 136 cases. In 6 cases no opinion was possible on cytology. Cyto-histological correlation was seen in 127 out of 136 cases, 9 cases remained discordant. Thus overall diagnostic accuracy of squash cytology in intraoperative diagnosis of CNS lesions in our study was 93.38%.

CONCLUSION- Intraoperative squash cytology is easy, rapid, reliable and cost-effective technique for neurosurgical consultation with fairly high accuracy. Knowledge of clinicoradiological details help to further improve the diagnostic accuracy.

KEYWORDS

Squash cytology, CNS lesions, intraoperative diagnosis

INTRODUCTION

Lesions within CNS consist of diverse entities, ranging from inflammatory conditions to several neoplastic lesions.¹ FNAC of central nervous system lesions is difficult due to its closed architecture. The intraoperative pathological diagnosis play deciding role in neurosurgery.² It helps neurosurgeon to formulate the treatment plan and to decide the extent of surgical resection.³

Squash smear cytology and Frozen section are the mainstay for intraoperative CNS lesions diagnosis. The role of squash preparation has increased along with the development of stereotactic biopsy as a effective resort for intraoperative evaluation of CNS lesions.² Very tiny tissue sample is obtained by stereotactic biopsy, for pathological evaluation, this small amount of tissue poses difficulty in tissue processing during Frozen section examination.^{4,5,6} The overall soft consistency of CNS tissue is not ideal for Frozen section, in fact soft nature of most CNS lesions is the major restriction for opting Frozen section as diagnostic modality. In addition, ice crystal processing artifacts may make difficulty in morphological interpretation during frozen section evaluation.^{4,5,7}

In early 1930 Eisenhard and Cushing for the first time, proposed smear technique as a mean for obtaining diagnosis for neurosurgical biopsies.⁸ As most of CNS lesions have soft and gelatinous consistency, smear preparation can be made easily often providing a high cellularity and superb cytological details.^{4,5,6,7} Squash smear display abnormal cellularity, fine nuclear and cytoplasmic details and even tissue architecture in some cases. Squash cytology can be applied for intraoperative evaluation of both neoplastic and non neoplastic CNS lesions.⁹ It is a rapid method whereby intraoperative diagnosis can be offered within few minutes of receiving biopsy material.^{6,10,11,12} It is cost effective and prevent contamination of instruments from slow virus diseases by fresh unfixed tissues used. It can be done on small amount of tissue available by stereotactically obtained biopsies and also allow to preserve some sample tissue for further processing and ancillary techniques to assist in the final diagnosis.⁷ In addition the biggest advantage of cytological diagnosis is, it is rapid which further helps the surgeon to formulate and/or modify treatment plan and to decide the extent of surgery accordingly.¹³

The current study was undertaken to assess the utility of intraoperative

consultation by squash smear cytology for rapid diagnosis of CNS lesions and its correlation with final histopathological diagnosis.

MATERIAL AND METHODS

The present study was carried out in department of Pathology, Superspeciality Hospital GMC Nagpur from May 2015 to May 2018. 142 cases of CNS lesions were sent for intraoperative smear cytology. After proper labelling the biopsy material was sent to department of Pathology in sterile bottle containing saline solution to prevent dehydration of tissue sample. Patient's clinical and radiological details were acquired prior to smear cytology evaluation.

Technique originally described by Adams AJ¹⁴ was used for smear cytology technique. Tissue piece was cut into 1-2 mm tissue bits and taken on one end of slide. With the help of another glass slide pressure applied over tissue to crush and was smeared on it. About 5-7 smears were prepared. Smears were stained with rapid H & E staining. Remaining slides were stained with Toluidine blue and/or MGG if and when needed. The remaining tissue was fixed in formalin and processed for paraffin embedding.

The diagnosis offered on squash smear cytology was compared with its final histopathological diagnosis. Cases are considered concordant when the cytological diagnosis and histological diagnosis were same including the grade in malignant tumors. And in cases where intraoperative cytological diagnosis doesn't confirmed with histological diagnosis were considered as discordant cases. In discordant cases smear slides were reviewed retrospectively to find out the possible reason for misdiagnosis.

RESULTS

In a period of 36 month total 142 cases were referred to department of Pathology from Neurosurgery department for squash smear cytology consultation and followed by subsequent histopathology examination. The age of patients ranged from 1 yrs to 74 years. Maximum number of cases were in the age group ranges from 31-40 yrs. Only 2 patients were above 70 years of age. We found a slight male preponderance in CNS lesions with total 80 patients (56.33%) being males and remaining 62 patients (43.66%) being females with a male/female ratio of 1.33 : 1. (Table 1)

Table no 1 : Age Distribution in the study. Total cases : 142

Sr. No.	Age group (yrs)	No. of cases	%
1)	0-10	18	12.67
2)	11-20	20	14.08
3)	21-30	30	21.12
4)	31-40	32	22.53
5)	41-50	15	10.56
6)	51-60	17	11.97
7)	> 60	10	7.04
	Total	142	100

Clinically the patients were presented with variable symptoms .Headache being the most common presenting symptom. Weakness of body parts, seizures ,diminution of vision and decreased hearing were the other most common symptomatology patients were presented with. According to radiological finding majority of cases i.e 127 cases (89.43%) were situated in the cranium whereas only 15 cases (10.56%)

were situated in spino-vertebral location.

On squash cytology 129 cases were neoplastic ,7 cases were benign non neoplastic and 6 cases were remains inconclusive. Of the 129 neoplastic cases 48 cases were benign and 81 cases were diagnosed as malignant neoplasm. Overall high grade astrocytoma and meningioma were found to be the most common lesions on cytology, 21 cases (15.44%) of each. This is followed by low grade astrocytoma 17 cases (12.5%) as second most common lesion. Metastatic carcinomas with 13 cases (9.56%) found as third most frequent diagnosed lesion.

On histopathological examination all 142 cases were being diagnosed. Of the 142 cases 136 cases were neoplastic and 6 cases were non neoplastic. Six cases which remained inconclusive on cytology were found to be all neoplastic with 2 cases of meningiomas, 1 case each of schwannoma ,oligodendroglioma ,haemangioblastoma and metastatic lesion. These six inconclusive cases were excluded from the study.

Table 2: Comparative report of histopathological examination and squash cytology.(excluding inconclusive cases) n = 136

CNS lesions	Histopathology Diagnosis	% of cases	Squash Diagnosis	Correlation %
Pilocytic astrocytoma	12	8.82	12	100 %
Astrocytoma (low grade)	14	10.29	13	92.86 %
Astrocytoma (high grade)	23	16.91	20	86.96%
Oligodendroglioma	7	5.15	6	85.71 %
Anaplastic oligodendroglioma	4	2.94	4	100%
Ependymoma	12	8.82	11	91.67%
Anaplastic ependymoma	4	2.94	4	100%
Central Neurocytoma	1	0.73	1	100%
Meningioma	21	15.44	20	95.24%
Papillary meningioma	1	0.73	1	100%
Schwannoma	10	7.35	9	90%
Medulloblastoma	2	1.47	2	100%
Pituitary adenoma	2	1.47	2	100 %
Choroid plexus papilloma	2	1.47	2	100%
Craniopharyngioma	1	0.73	1	100%
Pinealoblastoma	1	0.73	1	100%
Metastatic carcinoma	13	9.56	13	100%
Inflammatory/ Tuberculoma	6	4.41	5	83.34%
Total	136	100%	127	93.38%

Of the 136 cases ,on histology astrocytomas were the most common lesions (36.03%) ,of which combined astrocytoma grade III and glioblastoma constitute the most frequent found entity with 23 cases. Among non glial lesions meningiomas were the most frequently found entity with 21 cases. Among non neoplastic lesions tuberculoma was the most frequent entity consisting of 3 cases out of total 6 non neoplastic lesions.

Cytological diagnosis was correlated with histopathological diagnosis. Overall diagnostic accuracy of squash cytology was 93.38%, excluding the six inconclusive cases. Diagnostic accuracy for neoplastic lesions was higher (93.85%) in contrast to benign non neoplastic lesions (83.33%). There was disagreement between cytology diagnosis and histopathology diagnosis in 9 cases. Of the 9 cases of disagreement 5 cases of gliomas showed partial correlation.

Table no 3 : Cyto-histopathological correlation in the study. Total cases : 136

Cytological Diagnosis	Total cases	Histological diagnosis				Correct Histopath Diagnosis
		Consistent	% of concordant	Inconsistent	% of discordant	
Neoplastic Cases						
Pilocytic astrocytoma	12	12	8.82	-	-	
Astrocytoma (low grade)	17	13	9.56	4	2.94	1- Anaplastic astrocytoma 1- Glioblastoma 1- Ependymoma 1- Oligodendroglioma
Astrocytoma (high grade)	21	20	14.70	1	0.73	1-Diffuse astrocytoma Grade II
Oligodendroglioma	6	6	4.41	-	-	-
Anaplastic Oligodendroglioma	4	4	2.94	-	-	
Ependymoma	11	11	8.09	-	-	
Anaplastic Ependymoma	4	4	2.94	-	-	
Central Neurocytoma	1	1	0.73	-	-	
Meningioma	21	20	14.70	1	0.73	1-schwannoma
Anaplastic Meningioma	1	1	0.73			
Nerve Sheath tumour	10	9	6.62	1	0.73	1-Fibrous Meningioma
Medulloblastoma	2	2	1.47			
Pituitary adenoma	2	4	1.47	-	-	-
Choroid plexus papilloma	2	2	1.47			
Craniopharyngioma	1	1	0.73	-	-	-
Pinealoblastoma	1	1	0.73	-	-	-
Metastasis	13	13	9.56	-	-	-

Reactive Gliosis	1	-	-	1	0.73	1-High grade Astrocytoma
Non-neoplastic cases						
Inflammatory lesion	2	1	0.73	1	0.73	1-Tuberculoma
Tuberculoma	2	2	1.47	-	-	
Osteomyelitis	1	1	0.73	-	-	
Epidermal cyst	1	1	0.73	-	-	
TOTAL	136 (100%)	127	93.38%	9	6.62%	

Table no 4: Distribution and diagnostic accuracy of cases according to glial/glioneuronal , non-glial and non neoplastic origin.

Category	Glial /Glioneuronal n= 77*	Non-glial n= 53	Non Neoplastic n= 6	Total cases n= 136
Disagreement	6	2	1	9
Complete agreement	71	51	5	127
Overall accuracy	92.21%	96.23%	83.33%	93.38%

* _ 1 case of high grade Astrocytoma was misdiagnosed as reactive gliosis in our study

The lesions were divided into neoplastic and non neoplastic lesions, of which neoplastic lesions were further divided into glial/glioneuronal neoplasms and non glial neoplasms. Excluding the 6 inconclusive cases, 77 were glial/glioneuronal tumors (76 glial neoplasm and one case of neurocytoma), 53 were non glial tumours and 6 cases were benign non neoplastic lesions. The diagnostic accuracy for glial/glioneuronal neoplasms were 92.21% ,for non glial neoplasms were 96.23% and for non neoplastic lesions it was 83.33%. The majority of glioneuronal neoplasms were of astrocytomas ,this is followed by ependymoma ,oligodendrogliomas and a single case of central neurocytoma. Among the non glial neoplasms meningiomas ,schwannomas ,medulloblastomas ,choroid plexus papillomas ,pituitary adenomas ,metastatic carcinomas and one case each of craniopharyngioma and pinealoblastoma were included. In the non neoplastic category maximum number of cases were of tuberculoma (3 cases) ,and one case each of epidermal cyst ,dermoid cyst and inflammatory lesion.

Good correlation was seen in pilocytic astrocytomas , anaplastic oligodendrogliomas ,anaplastic ependymomas ,medulloblastomas ,pituitary adenomas, metastatic lesions and meningiomas. Poor correlation was seen cases of high grade gliomas ,grading of gliomas, oligodendroglioma and ependymoma.

Our intraoperative squash smear diagnosis help the neurosurgeon in the managing 4 unexpected lesions ,where diagnosis of those lesions were not given even on the broad differential diagnosis provided on radiological investigation. On discussion with neurosurgeon our squash smear diagnosis helped the neurosurgeon in deciding and executing treatment plan in 7 out of 20 correctly diagnosed cases of meningiomas and in a case of pilocytic astrocytoma in the infratentorial location.

The results were statically analysed using Galan and Gambian methods .Sensitivity ,Specificity , positive predictive value and negative predictive value of squash smear cytology were 98.78%, 100%, 100% and 98.18% respectively. Kappa statistics for agreement between cytology and histopathology for diagnosing malignancy shows p value <0.001 which indicates high significance.

DISCUSSION

Over the years various studies proved the importance of squash smear diagnosis in rapid intraoperative consultation of CNS lesions.^{7,15} In this era of modern stereotactic biopsy technique, smear cytology is now gaining more worldwide importance.¹³ It can provide rapid and accurate diagnosis to operating surgeon even with small neurosurgical biopsies. Smear cytology is easy ,inexpensive ,requires no technical expertise, allows preservation of tissue sample for further processing, has ability to show abnormal cellularity and give better cytological details.^{7,11} High water content ,soft nature and innate fragility of CNS tissue lead to freezing artifacts and poor quality frozen sections. On the other hand overall soft and consistency of most CNS lesions best suited for squash smear cytology preparation.⁷

Table no 5 : Split-up of inadequate cases. Total cases : 6

Sr. No.	Site	Tumour	No. of cases	%
1)	Intracranial			
		Meningioma	1	16.66
		Metastatic lesion	1	16.66
		Haemangioblastoma	1	16.66
		Oligodendroglioma	1	16.66
2)	Spinal			
		Meningioma	1	16.66
		Schwannoma	1	16.66
Total			6	100%

Of the 142 lesions, which we had evaluated intraoperatively by squash cytology a cytological diagnosis could not be done in 6 cases. When followed on histopathology these lesions were found to be 2 cases of meningiomas, 1 case each of schwannoma, oligodendroglioma, metastatic lesion and haemangioblastoma. Increased fibrous content, resistance to desegregation and rich stromal content contributed for no opinion in cases of meningiomas, schwannoma and haemangioblastoma. In case of metastatic lesion abundant haemorrhagic and necrotic material obtained which obscured the smear morphology. In oligodendroglioma case only cystic fluid was obtained showing cyst macrophages. Thus increased fibrous content, inflammation and necrosis obscuring morphology , biopsy from cyst wall and resistance to desegregation were the various causes, lead to no opinion on cytology. Other authors also have attributed similar causes for no opinion on smear cytology.^{16,17}

The median age of presentation of patients in our study was 30-35 years and a M :F ratio of 1.33: 1, which corresponds to the studies by others authors such as K Deshpande et al and Bharti Zaa et al who have reported the median age of about 30-35 years. We got a male preponderance in our study with a M :F ratio of 1.33:1. A higher incidence of CNS lesions in male has been reported in by many other authors.^{16,18}

Of the 136 cases, for which cytological diagnosis could be offered, on final diagnosis, there were 6 cases of non neoplastic and 130 cases of neoplastic. Of 136 cases on cytological evaluation 81 cases were found malignant, 54 cases were benign and one false negative case on cytology in malignant cases was seen in present study which correspond with studies by other authors.^{17,5}

The diagnostic accuracy attained in present study was 93.38% and it was comparable with various other studies which showed diagnostic accuracy ranging from 85-96 % .Also sensitivity and specificity of cytology in our study was found to be 98.78% and 100% respectively and consistent with study done by other authors.^{13,15,16,17,19,20}

We reported 12 cases of pilocytic astrocytomas. Smears showed elongated piloid cytoplasmic processes and Rosenthal fibres .Few cases also showed nuclear hyperchromasia and vascular endothelial proliferation ,which is according to literature are inherent properties of these tumors on long standing. All the cases were correlated with histopathology and we got 100% correlation.

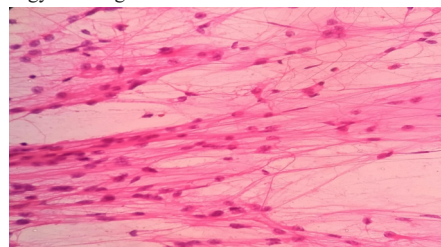


Figure 1:(Pilocytic astrocytoma)-Smear showing bipolar astrocytes, containing hairlike (piloid) cytoplasmic processes (H & E ,40x)

Low grade astrocytoma contributed 14 cases (10.29%) in our study. Smears showed moderate cellularity, cells showing minimal anisocytosis and finely to coarsely granular chromatin . The cytoplasm was scanty and showed variable cytoplasmic processes against a fibrillary background. Out of 14 cases histological correlation was

obtained in 13 case , thus we got 92.86% correlation in this group of tumor. Our 92.86%, diagnostic accuracy seems to be fairly good when compared with diagnostic accuracy obtained in various other studies as shown in table below

Table no. 6: Comparison of cyto-histological correlation of astrocytomas with other study

Author	No.of cases studied	Correlated with HPE	Accuracy %
Shukla et al21	140	135	96.42
Padma et al22	11	9	81.8
Brommeland et al23	153	140	91.5
Mertinez et al24	100	95	95.0
Nori et al25	14	12	85.71
Present study	14	13	92.86

There were 23 cases (16.91%) of astrocytoma grade III-IV tumor in our study. Smear showed marked increased cellularity , hyperchromatic and pleomorphic cells showing numerous mitotic figures, and marked vascular endothelial proliferatio and/or necrosis. We got histological correlation in 20 cases out 23 cases. Thus we got a diagnostic accuracy of 86.96% in this group of tumor.

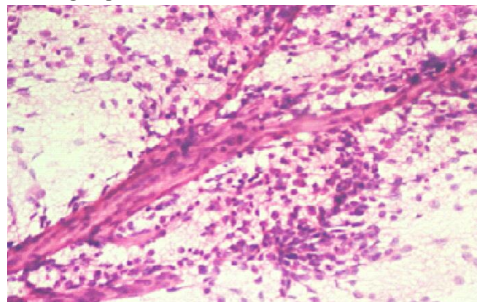


Figure 2 -Glioblastoma showing vascular endothelial proliferation. (H &E,40x)

Oligodendroglioma contributes 7 cases (5.15%) in our study. 6 out of 7 cases correlated with histopathology, thus 85.71% was the diagnostic accuracy obtained in this group. Smear showed moderately cellular neoplasm containing uniform round cells with distinct cell margins ,spread in a discohesive pattern. Cell have clear cytoplasm and background is granular. Thin walled blood vessels and foci of calcification was also noted in most of the cases. We had reported 4 cases of anaplastic oligodendroglioma and all cases correlated with histopathology. Thus obtained a diagnostic accuracy of 100% in this group. Smear showed marked cellularity ,highly pleomorphic cells showing mitotic activity and background in most cases show necrotic material in addition to it's usual granular appearance

Ependymoma contributed 12 cases (8.82%) in our study. Smears showed high cellularity , cells in layers were arranged in a papillary pattern around blood vessels stroma .Cells had scanty cytoplasm and round to oval nuclei with finely granular chromatin. The background was fibrillary. Perivascular pseudorosettes were prominent .Out of 12 cases histological correlation was obtained in 11 cases, thus we obtained a diagnostic accuracy of 91.67% in this group of tumor. Anaplastic ependymoma contributed 4 cases in our study and all of which correlated with histopathology. We reported a 100% accuracy in this group of tumor. Smears showed cells in less cohesive manner forming loose clusters ,cells had scant cytoplasm ,hyperchromatic pleomorphic nuclei showing frequent clefts and grooves.

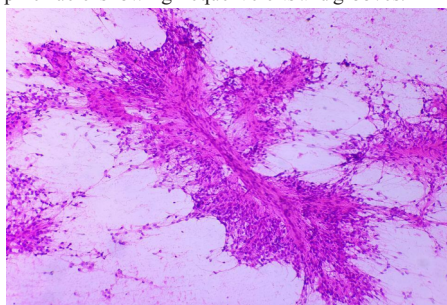


Figure 3:(ependymoma)-ependymal cells forming papillary pattern around vascular stroma with fibrillary background.(H & E, 40x)

Schwannoma contributed 10 cases (7.35%) in our study .Smears showed mostly spindle cells with elongated nuclei arranged mainly in fascicular pattern. 9 case out of 10 were correlated with final histopathological diagnosis. Thus a diagnostic accuracy of 90% obtained in this group of tumor Meningioma contributed 21 cases (15.44%) in our study. Smears in most cases except fibrous variant showed uniform round meningothelial cells in cluster, showing copious ,wispy cytoplasm .In cluster cells are seen arranged in syncytial pattern. Epithelial whorls and psammoma bodies were the most prominent findings seen in most cases. We obtained correct histological correlation in 20 out of 21 cases .Thus a diagnostic accuracy of 95.24% was obtained in this group of tumor. We reported 1 case of papillary meningioma where the slightly pleomorphic cells were arranged in papillary pattern around vascular stroma. Our smear diagnosis was correlated with histopathology diagnosis in that case.

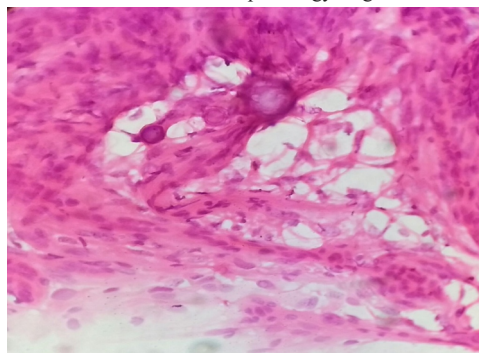


Figure 4(Meningioma) –cluster of Meningothelial cells and whorling.(H&E,40x)

Medulloblastoma contributed 2 cases (1.47%) in our study. Smears showed small dark round cells arranged diffusely in a non cohesive manner. Occasional neuroblastic rosettes formation was also seen. We obtained 100% diagnostic accuracy in this group of tumor.

Pituitary adenomas contributed 2 cases (1.47%) in our study. Smears was evenly spread ,showing monomorphic cells having round to oval nuclei with fine chromatin and acidophilic to slightly basophilic cytoplasm. We got a 100% diagnostic accuracy in this group of tumor.

We reported one case (0.73%) of Craniopharyngioma ,which was correlated with histopathological diagnosis. Smear was difficult to spread and showed squamous cells in small cohesive sheets Choroid plexus papillomas contributed 2 cases (1.47%) in our study ,and both were correlated with final histopathological diagnosis. Smear showed fine papillae. Papillae were composed of cuboidal to columnar cell in two to three layers arranged around fibrovascular core. Background was clear. We reported 100% diagnostic accuracy in this group of tumor.

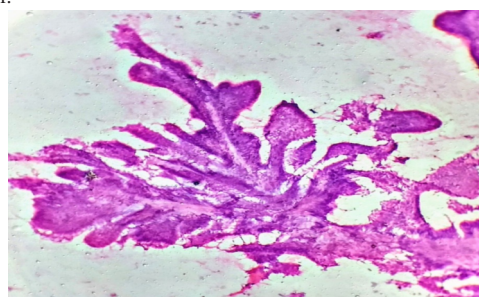


Figure 5:(Choroid plexus papilloma) -fine papillae made up of cuboidal cells arranged around fibrovascular core, clear background. (H & E, 40x)

We reported one case (0.73%) of central neurocytoma. Cytological diagnosis correlated with final histopathological diagnosis. Smear showed high cellularity with round small cells arranged diffusely .Cell free neutrophil area was also seen.

We reported 13 cases (9.56%) of metastatic carcinomas. Smear showed malignant cells having high N:C ratio ,with high mitotic activity arranged in either sheets ,follicles, papillary or glandular architecture. Background necrosis was also seen in most of cases. We

obtained a diagnostic accuracy of 100% in metastatic tumor group in our study. Clinical history contributed a major role in achieving this high diagnostic accuracy rate.

We reported one case (0.73%) of pinealoblastoma on cytology which was correlated correctly with final histopathological diagnosis. Smear showed cells with round to angulated primitive looking nuclei .

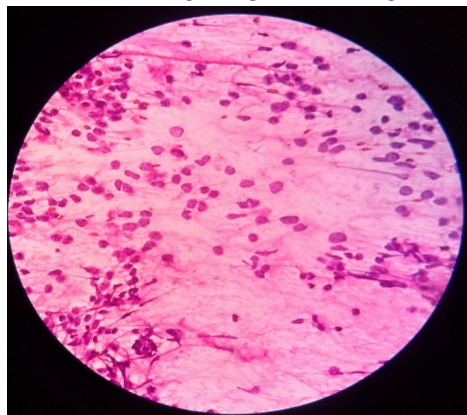


Figure 6 (Pinealoblastoma), cells with hyperchromatic angulated nuclei ,primitive looking cells. (40x H&E).

We had 3 cases(2.21%) of tuberculosis in our study. We diagnosed 2 cases correctly on squash smear cytology, thus accounting for a diagnostic accuracy of 66.67 % in tuberculous lesions. Smears showed well formed granuloma composed of epithelioid cells .Multinucleate giant cells and chronic inflammatory cells also seen. 2 smear show prominent caseous necrotic background.

We had one case of chronic inflammatory lesion at spinal cord location ,which was secondary to prolapse disc. We diagnosed that case correctly on squash smear cytology .Here smear showed mainly chronic inflammatory cells.We had one case of osteomyelitis of jaw and one case epidermal cyst. Clinical suspicion and radiological finding strongly favours the diagnosis. We correctly diagnosed those cases on squash cytology.

Table 7 : Discordant cases on cyto- histology correlation

HPE diagnosis	Cytology diagnosis	No.cases	%
Neoplastic Lesions			
Anaplastic astrocytoma	1)astrocytoma low grade 1) Reactive gliosis	2	22.22%
Glioblastoma	Astrocytoma low grade	1	11.11%
Astrocytoma grade II	Glioblastoma	1	11.11
Oligodendroglioma	Astrocytoma low grade	1	11.11%
Ependymoma	Astrocytoma low grade	1	11.11%
Fibrous meningioma	Schwannoma	1	11.11%
Schwannoma	Meningioma	1	11.11%
Non neoplastic lesion			
Tuberculosis	Inflammatory lesion	1	11.11%
Total		9	100%

In our study out of 130 neoplastic cases ,we failed to diagnose 8 cases and out of 6 non neoplastic lesions we failed to diagnose 1 case on cytology. The cyto-histological concordance rate was 93.85% in case of neoplastic lesions and 83.33% for non neoplastic lesions. Our concordance rate for neoplastic and non neoplastic lesions was closely correspond with study done by K Sanjeev et al.¹⁷

1 case of Oligodendroglioma was misdiagnosed as diffuse astrocytoma WHO grade II ,the error was probably due to the lack of uniformly appearing nuclei and seemingly apparent cytoplasmic processes simulating astrocytoma. Surgical cautery may cause spindling and wrinkling of round nuclei of oligodendroglioma, making it difficult to give a correct diagnosis as had happened with our case. Similar cause was attributed for misinterpretation in study carried out by N Krishnani et al²⁰ and S.Aacharya et al²⁶

1 case of Ependymoma was misdiagnosed as astrocytoma Grade II mostly because the tumor was moderately cellular and the cellular background showed processes simulating astrocytoma and because of the lack of ependymal rosettes in that case. Roessler et al also reported

the absence of rosettes on cytology leading to erroneous diagnosis.¹⁸

1 case of fibroblastic meningioma was misdiagnosed as schwannoma in squash preparations due to the presence of elongated spindly cells and lack of cellular whorls in smears . Kobayashi²⁷ and S.Mitra⁷ also described similar misinterpretation in their results.

1 case of schwannoma was misdiagnosed as meningioma on cytology due to thick smear and due to similarity in cytomorphology of both the lesions that is spindle cells in fascicles. Similar diagnostic difficulty is faced by K. Deshpande¹⁶ and shalini Rao et al²⁸ in their studies.

Improper grading of astrocytic neoplasms on cytology preparation can occur. As it occurred in 3 cases in our study. It may occur due to heterogenous nature of astrocytomas.²⁰ Astrocytomas are known to vary significantly in grade from one area to another within same tumor.

1 case of anaplastic astrocytoma was undergraded as diffuse astrocytoma WHO grade II , as there are areas of both less and more aggressive astrocytomas and the cytological sampling fail to show the anaplastic component. Similar pitfall is stated by N.Pawar et al⁸ in his study.

1 case of glioblastoma was underdiagnosed as diffuse astrocytoma WHO grade II .This was attributed to sampling error resulting in obviation of characteristic necrotic material while sampling which is important for diagnosis of GBM. Alternatively it may be due to the fact that necrosis cannot be recognised because it sticks poorly to the slides. Similar pitfall is experienced by S.Sharma⁵ , S.Patil²⁹ and Marshall et al.³⁰

1 case of diffuse astrocytoma grade II was overgraded as GBM ,this misdiagnosis was probably due to technical error. Surgical cautery induced slight cell morphology changes and necrotic debris was misinterpreted as necrosis, which leads us to thought erroneous diagnosis of high grade astrocytoma in that case. Similar grading discrepancy was described by S Mitra et al.⁷ In such situations it is better to correlate smear assessment with radiological finding and any discrepancy if found should be analysed objectively.

1 case of high grade astrocytoma was misdiagnosed as reactive gliosis. It was due to the non-representative sampling from the rective tumour margin instead of tumor mass proper.

One case of tuberculosis of spine was misdiagnosed as inflammatory lesion ,as no necrosis or well form granuloma or giant cells were seen on smear cytology ,only inflammatory cells were seen and thus misdiagnosed as inflammatory lesion. Similar erroneous misinterpretation was also described by S Narang et al³¹ (2015) in their study.

Overall diagnostic accuracy obtained in our study was 93.38% and 9 cases were misdiagnosed on cytology .As most of the lesions misdiagnosed on cytology were included in the category of nearly same therapeutic procedure, deleterious adverse effect of misdiagnosis on neurosurgical treatment outcome was not occurred.

CONCLUSION

Squash smear diagnosis is fairly accurate ,simple ,safe and reliable diagnostic method for rapid intraoperative diagnosis of CNS lesions. It is preferred methods because of it's ability to display fine cytological details and as it can be performed on small amount of sample tissue .We also feel that as intraoperative diagnosis of CNS lesions play a deciding role in neurosurgical management, smear cytology evaluation exercise if performed with taking clinical and radiological finding into consideration will lead to further improvement in diagnostic accuracy and thus better patients management.

REFERENCES

- 1) Sundaram C. Diagnostic utility of squash [smear] technique in the inflammatory lesions of central nervous system. Indian Journal of Pathology & Microbiology. 2003;46(4):569-72.
- 2) Powell SZ. Intraoperative consultation, cytologic preparations, and frozen section in the central nervous system. Arch Pathol Lab Med 2005;129:1635-52..
- 3) Wilson I. A method for the rapid preparation of fresh tissues for the microscope. JAMA. 1905;45:1737.
- 4) Jha B, Patel V, Patel K, Agarwal A. Role of squash smear technique in intra operative diagnosis of CNS tumors. Int J Med Sci Public Health 2013;2:889-92.
- 5) Sharma S, Deb P. Intraoperative neurocytology of primary central nervous system neoplasia: A simplified and practical diagnostic approach. J Cytol 2011;28:147-58.
- 6) Jaiswal S, Vij M, Jaiswal AK, Behari S. Intraoperative squash cytology of central nervous system lesions: A single center study of 326 cases. Diagn Cytopathol

- 2012;40:104-12
- 7) Mitra S, Kumar M, Sharma V, Mukhopadhyay D. Squash preparation: A reliable diagnostic tool in the intra operative diagnosis of central nervous system tumors. *J Cytol* 2010;27:81-5
 - 8) Pawar HN, Deshpande AK, Surase SG, Deosta GF, Balgi SS, Goel AD. Evaluation of squash smear technique in the rapid diagnosis of CNS tumours: A cytomorphological study: *The Internet Journal of Pathology* 2010; 11(1).
 - 9) Torres LF, Noronha LD, Gugelmin ES et al. Accuracy of smear technique in the cytological diagnosis of 650 lesions of the central nervous system. *Diagn Cytopathology* 2001;24:293-95.
 - 10) Savargaonkar P, Farmer PM. Utility of intra-operative consultations for the diagnosis of central nervous system lesions. *Ann Clin Lab Sci* 2001;31:133-9.
 - 11) Nigam K, Nigam N, Mishra A, Nigam N, Narang A. Diagnostic accuracy of squash smear technique in brain tumors. *JARBS*. 2013;5:186-90
 - 12) Shah AB, Muzumdar GA, Chitale AR, Bhagwati SN. Squash preparation and frozen section in intraoperative diagnosis of central nervous system tumors. *Acta Cytol* 1998;42:1149-54.
 - 13) Roessler K, Dietrich W, Kitz K. High Diagnostic Accuracy of Cytologic smears of Central Nervous Tumors. A 15- year experience based on 4172 patients. *Acta Cytol* 2002; 46:664-7.
 - 14) Adams HJ, Graham DI, Doyle D. Brain Biopsy. The smear technique for Neurosurgical Biopsies. Philadelphia: JB Lippincott, 1981.p 2-124.
 - 15) Firlik KS, Martinez AJ, Lunsford LD. Use of cytological preparations for the intraoperative diagnosis of stereotactically obtained brain biopsies: a 19- year experience and survey of neuropathologists. *Journal of Neurosurgery*. 1999;91(3):454-58.
 - 16) K Deshpande et al. Accuracy and diagnostic yield of intraoperative squash smear technique in the rapid diagnosis of CNS lesions. *Bombay Hospital Journal*, vol 52, No. 2, 2010.
 - 17) K Sanjeev et al. Intraoperative squash cytology of central nervous system and spinal cord lesions with histological correlation. *Annals of Pathology and Laboratory Medicine*, Vol. 03, No. 02, April - June 2016
 - 18) Indrani Krishnappa et al. Diagnostic utility of intraoperative cytology in CNS lesions. *International Journal of Biomedical Research* 2016; 7(10): 726-732
 - 19) Goel D, Sundaram C, Paul TR et al. Intraoperative cytology (squash smear) in neurosurgical practice pitfalls in diagnosis experience based on 3057 samples from single institution. *Cytopathology Official Journal of the British Society for Clinical Cytology*. 2007;18(5):300-8
 - 20) N Krishnani et al. Intraoperative squash cytology :accuracy and impact on immediate surgical management of central nervous system tumours. ©2011, Blackwell Publishing, Ltd *Cytopathology* 2012, 23, 308- 314.
 - 21) Shukla K, Parikh B, Shukla tumors in crush preparation. *Indian J Pathol Microbiol J*, Trivedi P, Shah B. Accuracy of cytologic diagnosis of central nervous system tumors in crush preparation. *Indian J Pathol Microbiol* 2006; 49(4):483-486.
 - 22) Padma M, Jigiyabai, Prasad babu. Evaluation of imprint cytology in surgical specimens. *GMC, Path- library* 2005; 82-86.
 - 23) Brommeland T, Lindal S, Straume B, Dahl IL, Henning R. Does imprint cytology of brain tumors improve intraoperative diagnosis? *Acta Neurol Scand* 2003; 108(8):153-156.
 - 24) Martinez AJ, Pollack I, Hall WA, Lunsford LD. Touch preparations in the rapid intraoperative diagnosis of central nervous system lesions. A comparison with frozen sections and paraffin- embedded sections. *Mod. Pathol.* 1988; 1(5):378-84.
 - 25) Srinivas N S Nori et al. A study to determine the accuracy of cytological diagnosis of CNS lesions by squash smears and correlation with histopathology *Med Pulse International Journal of Pathology*, 2017; 4(1): 21-28
 - 26) S Acharya et al. Squash Smear Cytology, CNS Lesions – Strengths and Limitations. *National Journal of Laboratory Medicine*. 2016 Jul, Vol-5(3): PO01-PO07.
 - 27) Kobayashi S. Meningiomas, neurilemmoma and astrocytoma specimens obtained with the squash method for cytodiagnosis. A cytological and immunohistochemical study. *Acta Cytol* 1993;37:913-22.
 - 28) Shalini Rao et al. Challenges in neurosurgical intraoperative consultation. *Neurology India / Jul-Aug 2009/ vol 57/ issue 4.*
 - 29) S Patil et al. Utility of squash smear cytology in intraoperative diagnosis of central nervous system tumors. *Journal of Cytology / October 2016 / Volume 33 / Issue 4.*
 - 30) Marshall LF, Adams H, Doyle D, Graham DI. The histological accuracy of the smear technique for neurosurgical biopsies. *J Neurosurg* 1973;39:82-8.
 - 31) V Narang et al. Intraoperative diagnosis of central nervous system lesions: comparison of squash smear, touch imprint and frozen section. *Journal of Cytology / July 2015 / volume 32 / Issue 3.*