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Role of Squash Cytology in Intraoperative Diagnosis of Meningioma

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ABSTRACT

Introduction: Primary Central Nervous System (CNS) tumours constitute less than 2% of overall cancers in adults and are the second most frequently encountered tumours in children. Meningiomas form 24-30% of primary intracranial tumours. Most intrinsic brain tumours are soft and gelatinous in consistency, smear preparation can readily made which gives excellent cytological details when compared to frozen section as the latter produces ice crystal artifacts.

Aim: To assess the diagnostic utility of squash cytological evaluation of meningiomas and its comparison with final histopathological diagnosis.

Materials and Methods: The cross-sectional study was done at Thanjavur Medical College, Thanjavur, Tamil Nadu, India for period of three years from January 2015 to December 2017. Total of 54 clinically diagnosed and radiologically suspected case of meningiomas were selected. Smears were prepared from the biopsy samples sent in normal saline and stained by Haematoxylin and Eosin (H&E) method. The cytological features were noted and matched with biopsy findings. Descriptive statistics were used to analyse the results.

Results: Total of 54 squash smears with male to female ratio was 1:1.5. Maximum number of cases were seen between 41-50 years followed by 51-60 years. Complete concordance was obtained in 51 cases (94.44%) and partial concordance was noted in a case due to underestimation of malignancy grade in squash cytology. Out of 54 cases, two cases were found to be discordant with final histopathological diagnosis.

Conclusion: Intraoperative squash cytology is easy, rapid, reliable and cost-effective technique for neurosurgical consultation with fairly high accuracy in diagnosing meningiomas.

Keywords: Central nervous system tumours, Cytological-histological comparison, Cytomorphological features, Squash smear cytology

INTRODUCTION

Primary Central Nervous System (CNS) tumours constitute less than 2% of overall cancers in adults and are the second most frequently encountered tumours in children [1]. It is estimated that annual incidence of CNS tumours ranges from 10-17 per 1,00,000 persons for intra-cranial tumours and 1-2 per 1,00,000 persons for intraspinal tumours [1]. Meningiomas form 24-30% of primary intracranial tumours [2].

Most intrinsic brain tumours are soft and gelatinous in consistency, smear preparation can readily made which gives excellent cytological details when compared to frozen section as the latter produces ice crystal artifacts [3]. Intraoperative squash cytology preparation was first introduced by Eisenhardt and Cushing in early 1930 and by Badt in 1937 [4-7]. This technique was furthered and documented by Russell et al., in 1937 [5]. Recently, the role of intraoperative smear preparation technique has gained importance because of advent of Computed Tomography (CT) and Magnetic Resonance Imaging (MRI) guided stereotactic biopsies [5,6].

The advantages of squash smear technology are: (1) simple and rapid technique; (2) no technical expertise is required for preparation of smear; (3) provides both cytological and architectural features of CNS tumours; (4) background matrix and necrosis are easy to appreciate; (5) rapid diagnosis aids the neurosurgeon to plan the extent of surgery [8].

Objectives

- To assess the utility of intraoperative squash smear cytology in diagnosing meningioma and its limitations.
- To study about cytomorphological features of different types of meningioma in squash preparation.

MATERIALS AND METHODS

This cross-sectional study was done in Thanjavur Medical College and Hospital, Thanjavur, Tamil Nadu, India, for a period of three years from January 2015 to December 2017. The study was conducted after obtaining approval from Institutional Ethical Committee, Thanjavur (IEC NO. 117/dated/10.04.2015). A total of 54 clinically diagnosed and radiologically suspected cases of meningiomas were included and other CNS tumours were excluded from the study.

Study Procedure

For all the 54 cases, unfixed fresh biopsy material sent in normal saline from operation theatre was obtained. Initially, gross examination of specimen was done to evaluate the nature of spread. Care was taken not to allow the tissue to dry out. To prepare squash smears, small pin head sized or 2-3 mm tissue was taken and placed on one end of the slide. Flat surface of another slide placed at right angle on the top of the specimen and advanced with uniform motion without exerting undue pressure on the tissue. Minimum of 4-6 smears were made depending on the amount and sample of the tissue received. All the smears were fixed in 99.9% isopropyl alcohol and stained with Harris haematoxylin and eosin stain and examined.

Remaining tissue was fixed in 10% neutral buffered formalin for histopathological examination. Intraoperative squash smear cytological diagnosis was then compared with histopathological findings. Histopathological diagnosis was considered as final gold standard diagnosis to estimate the accuracy of squash smear cytology. The diagnosis was made, based on World Health Organisation (WHO) 2007 classification of CNS neoplasm and graded accordingly [2]. In all cases, relevant clinical data and radiological findings were obtained.

Cytology results were classified into the following categories [9]:

Complete agreement- Cases with same diagnosis and grade on cytology and histopathology.

Partial agreement- Cases with same diagnosis, but the grade was misdiagnosed on cytology.

Disagreement- Cases where there is difference in the cell of origin.

The overall accuracy rate was calculated by including the cases which show complete concordance with final histopathological diagnosis.

STATISTICAL ANALYSIS

The data obtained were analysed using Statistical Package for the Social Sciences (SPSS) and then the parameters were evaluated and expressed in percentage.

RESULTS

A total of 54 squash smears comprising 38 females and 26 males were included in the study. Male to female ratio was 1:1.5 irrespective of age group with female predilection. The age group ranged from 21 years to 80 years. Maximum number of cases was seen between 41-50 years followed by 51-60 years comprising 21 cases and 14 cases respectively [Table/Fig-1].

Age group (years)	No. of cases	Percentage (%)
21-30	3	5.6%
31-40	12	22.2%
41-50	21	38.9%
51-60	14	25.9%
61-70	3	5.6%
71-80	1	1.8%

[Table/Fig-1]: Age distribution of patients with meningioma.

Meningiomas were encountered in the following sites like parasagittal region, sphenoid wing region, olfactory groove adjacent to cribriform plate, the tentorium, falx cerebri, free surfaces of cerebral convexities of frontal, temporal, parietal region followed by Cerebellopontine (CP) angle/posterior fossa location Convexities of cerebral hemispheres followed by Cerebellopontine angle. Most common site of presentation for meningiomas in the present study was angle/posterior fossa location [Table/Fig-2].

Site of lesion	Number of cases
Cerebral convexities of frontal, temporal and parietal region	21
Cerebellopontine angle/posterior fossa location	9
Sphenoid wing	8
Olfactory groove	7
Parasagittal region	4
Falx cerebri	3
Tentorium	2

[Table/Fig-2]: Site of lesion in cases.

Out of 54 cases, 51 cases showed complete concordance with final histopathological findings and partial concordance was obtained in a case due to underestimation of grading of malignancy in squash cytology. Two cases were found to be discordant with final histopathological diagnosis. The overall diagnostic accuracy of squash cytology in diagnosing meningioma was 94.44% [Table/Fig-3,4].

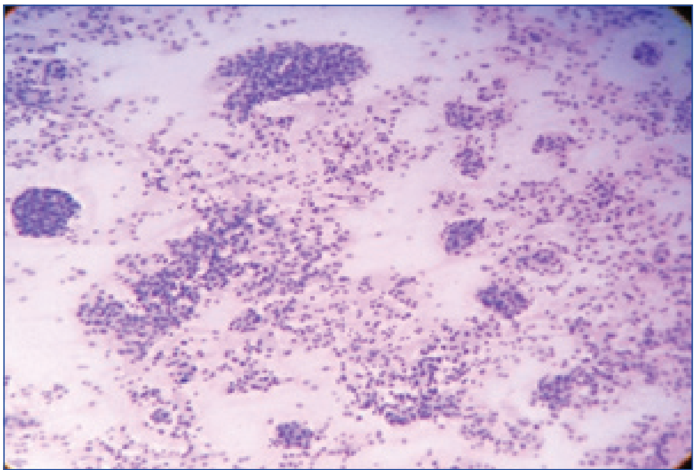
Distribution of cases	Number of cases
Complete agreement	51
Partial agreement	1
Disagreement	2
Total no. of cases	54
Accuracy rate	94.44%

[Table/Fig-3]: Overall accuracy rate.

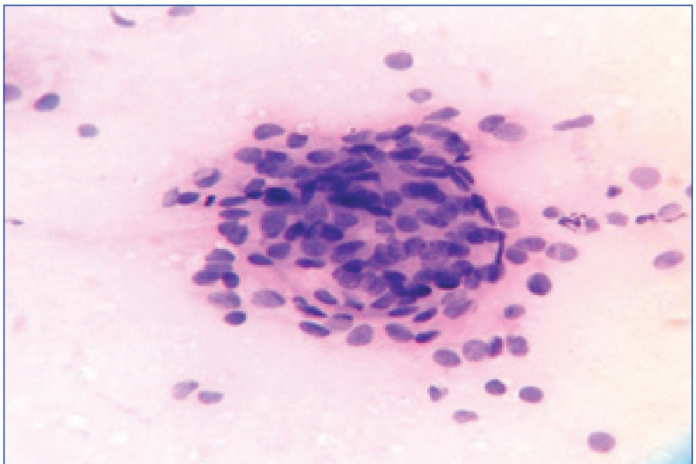
Cytological diagnosis	Histopathological diagnosis	Number of cases	Limitations encountered
Cases with partial agreement (Grading error)			
Meningioma-grade I	Anaplastic meningioma	1	Sampling error (Non representative sampling)
Disagreement cases (Cell type error)			
Fibrous meningioma	Schwannoma	1	Spindle cell nature of both
Anaplastic meningioma	Glioblastoma multiforme	1	Loss of architecture and extensive areas of necrosis (Non representative sampling)

[Table/Fig-4]: Partial agreement and disagreement cases.

Total of 44 cases of meningothelial and transitional meningioma showed plump to ovoid meningothelial cells arranged in syncytial pattern with characteristic whorled configuration and were unrelated to blood vessels as shown in [Table/Fig-5,6]. Occasional concentric calcification termed as psammoma bodies which appear as round dark blue formations was noted in few cases as shown in [Table/Fig-7]. Histopathological examination revealed lobules of meningothelial cells with indistinct cell borders having syncytial cytoplasm as shown in [Table/Fig-8]. Nuclei are pale round to oval with marginated chromatin.



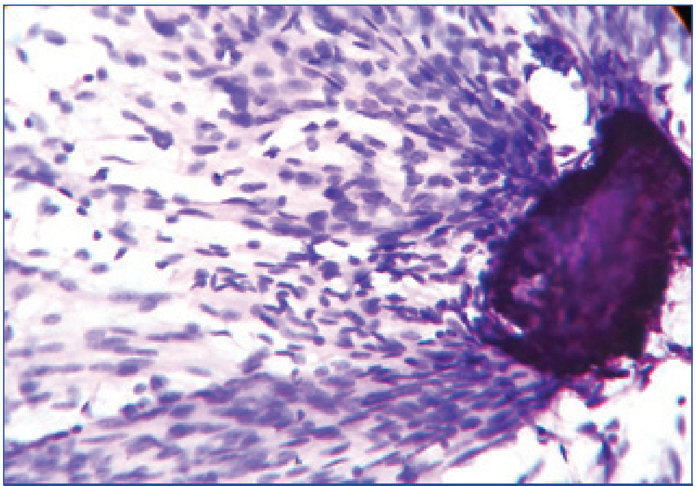
[Table/Fig-5]: Meningioma- Squash smear shows whorls and syncytial arrangement of meningothelial cells (H&E, x100).



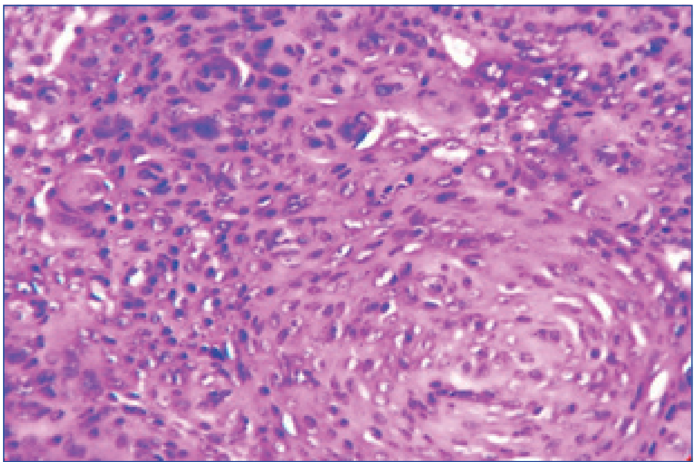
[Table/Fig-6]: Meningioma- Squash smear shows whorling configuration of meningothelial cells (H&E, x400).

Four cases of psammomatous meningioma showed numerous psammoma bodies occupying more than half of the slide with very few tumour cells as shown in [Table/Fig-9].

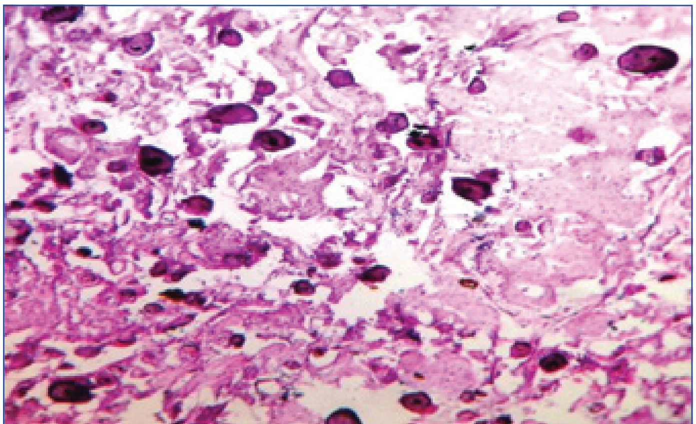
Two cases of angiomatoid and a case of metaplastic meningioma were really very difficult to make smear and it showed occasional clusters of meningothelial cells in syncytial pattern, reported as grade I meningioma in smear cytology. Nuclei in maximum cases



[Table/Fig-7]: Meningioma- Squash smear shows meningothelial cells with concentric calcification (Psammoma bodies) (H&E, x400).



[Table/Fig-8]: Meningioma- Section shows meningothelial cells with syncytial cytoplasm in whorling configuration (H&E, x400).

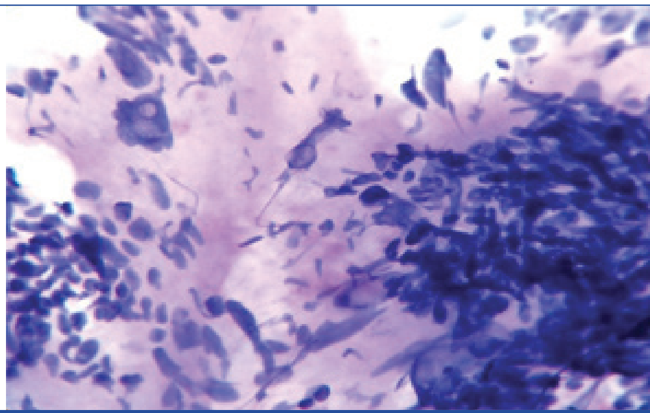


[Table/Fig-9]: Psammomatous meningioma- Section shows numerous psammoma bodies occupying more than half of the slide (H&E, x100).

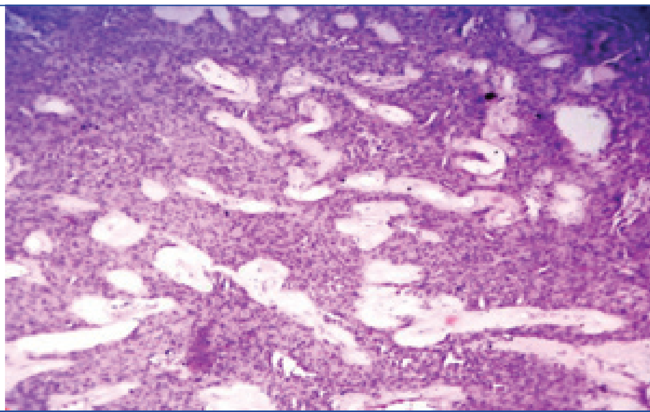
were round to ovoid with delicate nuclear chromatin and scant cytoplasm. Intranuclear inclusions were noted in few cases in the present study [Table/Fig-10].

Histopathological examination of angiomatoid meningioma revealed numerous small to large vascular spaces lined by flattened endothelial cells with thickened hyalinised vessel walls, and surrounding nests of meningothelial cells having syncytial cytoplasm as shown in [Table/Fig-11]. Metaplastic meningioma showed syncytial nests of meningothelial cells admixed with lobules of mature adipocytes with eccentrically placed nuclei as shown in [Table/Fig-12].

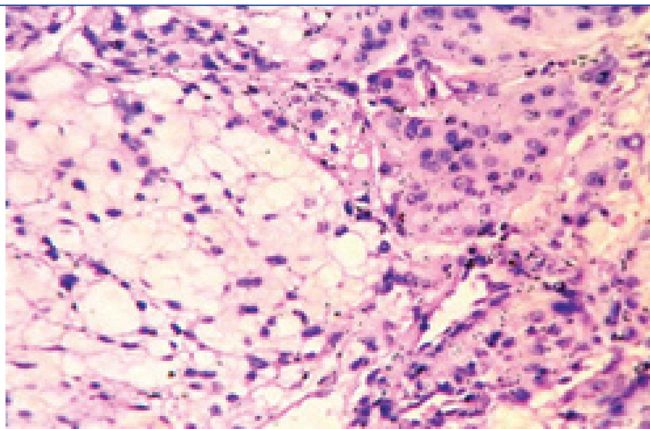
A case of schwannoma in posterior fossa location was reported as fibrous meningioma, which lacks characteristic whorls in squash smear cytology [Table/Fig-13]. As a result of non representative sampling, a case of anaplastic meningioma downgraded as grade I



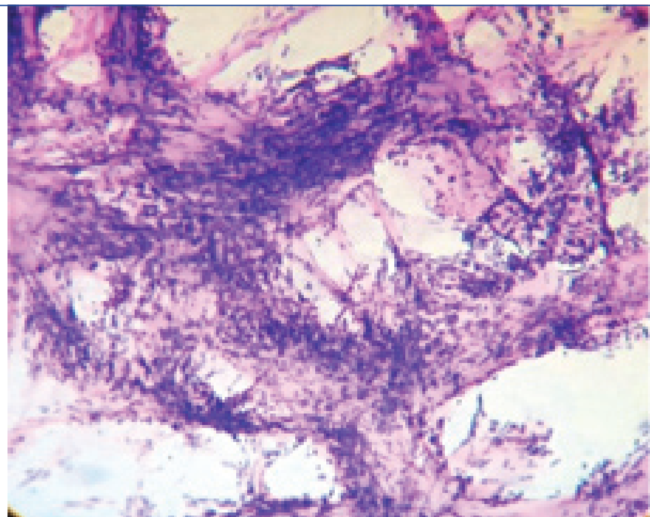
[Table/Fig-10]: Meningioma- Squash smear shows intranuclear inclusion in a case of transitional meninigioma (H&E, x400).



[Table/Fig-11]: Angiomatous meningioma-Section shows numerous thick walled hyalinised blood vessels surrounded by meningothelial cells (H&E, x100).



[Table/Fig-12]: Metaplastic meningioma-Section shows meningothelial cells with syncytial cytoplasm and lobules of mature adipocytes (H&E, x400).



[Table/Fig-13]: Squash smear of schwannoma diagnosed as fibrous meningioma due to the spindle cell nature of both (H&E, x400).

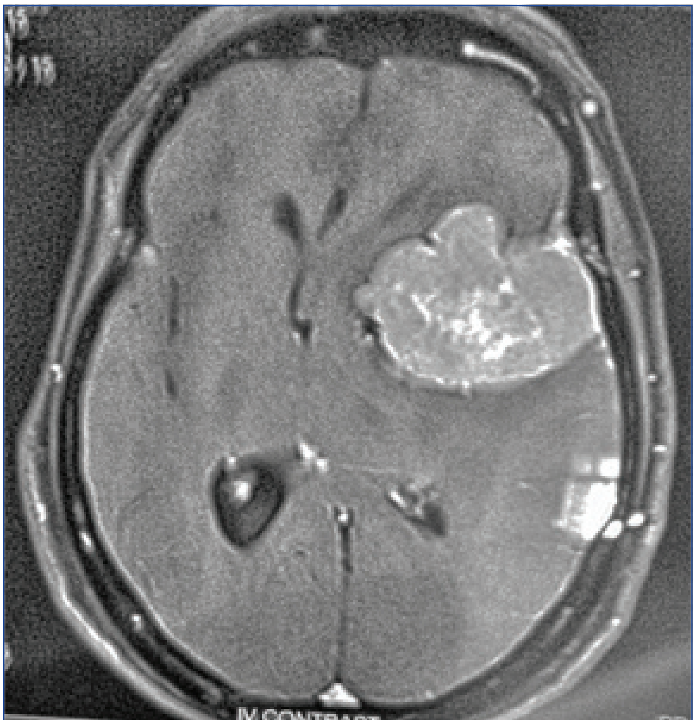
meningioma, and a case of anaplastic meningioma on squash cytology turned out to be glioblastoma (WHO Grade IV) on HPE.

DISCUSSION

Meningiomas are benign slow growing neoplasm arising from meningothelial cells of arachnoid layer [10]. It is the most frequently reported brain tumour accounting for 36% of all brain tumours [10]. About 20-25% and 1-6% of meningiomas are WHO grade II and III, respectively [10]. High water and fat content and innate fragility of brain tissue may give rise to freezing artifacts on frozen section studies [13]. Neurosurgeons often depend upon rapid intraoperative diagnosis for immediate surgical management in Central Nervous System (CNS) lesions [6]. Squash smear technique is frequently offered by neurosurgeons for rapid and reliable diagnosis and also helps to plan the extent of surgery intraoperatively and modify the treatment accordingly [6].

The current study was to assess the diagnostic accuracy of intraoperative squash smear cytology of meningiomas and to compare it with the final histopathological diagnosis. The overall diagnostic accuracy in the present study was 94.44%. Following studies have also reported higher degree of accuracy rate in diagnosing meningiomas on squash smear cytology Karanjekar SR and Parate SN, Gopal R and Lalitha, Rao S et al., Dumitrescu G et al., Shukla K et al., Roessler K et al., and Acharya S and Azad S, [11-17].

Clinically, most of the patients presented with headache, seizures and other pressure symptoms due to compression of underlying brain parenchyma. Radiological finding in most of the cases were, uniformly contrast enhancing circumscribed dural mass with well-defined brain tumour interface [Table/Fig-14]. Higher diagnostic accuracy rate in squash smear cytology is achieved by correlating cytological features with clinical and radiological findings [16].



[Table/Fig-14]: Computed tomography picture of sphenoid wing meningioma.

Out of 54 cases, 33 cases were reported as meningothelial meningioma, 11 cases of transitional meningioma, followed by four cases of psammomatous meningioma, two cases of angiomatous meningioma and one case of metaplastic meningioma. The tumours were classified and graded according to the World Health Organisation (WHO) classification of CNS neoplasms 2007. Characteristic type of each meningioma was finally confirmed in histopathological diagnosis mainly for transitional, metaplastic and angiomatous meningioma. Jha B et al., also stated that, it was not possible to provide clear cut differentiation between types of meningioma on cytology as in the present study [3].

Most of the cases of meningothelial and transitional meningioma showed the characteristic whorling pattern of meningothelial cells as stated by Karanjekar SR and Parate SN [11].

Krishna Prasad HV et al., stated that psammomatous meningiomas are invariably occurs in spinal canal, but in the present study most of the cases located in the supratentorial region as extradural space occupying lesion [4]. Basically, meningiomas are easy to spread but in psammomatous meningioma and transitional type with high fibrous component were difficult to make a uniform smear [2,18].

Exact subtyping of angiomatoid and metaplastic meningioma was found to be difficult due to loss of specific features in the tissue submitted for squash preparation [17]. Each meningioma shows variable cellularity found to be high in meningothelial and transitional meningioma, low in fibrous meningioma [4]. Metaplastic changes include xanthomatous, cartilaginous, osseous, myxoid changes [19,20]. Foci of meningothelial cells give a clue to the diagnosis.

Schwannoma and meningioma, particularly the fibrous type resist smearing [19-26]. Features such as whorling, psammoma bodies and plump to ovoid cells with syncytial cytoplasm are more in favour of meningioma.

Possible reasons for the discordance in a case of anaplastic meningioma diagnosed as glioblastoma were, one of the reasons was the exact tumour's anatomic relationship with dura and brain was obscured on imaging. Another reason was found to be sampling error as the smear shows only varying amount of oval to spindle shaped cells with necrotic debris in the background. Hence, in a radiologically high grade tumour it is wise to get more representative sample, if needed multiple samples from various sites before coming to a definitive diagnosis. This needs proper communication with the neurosurgeon.

Due to the non representative sampling, a case of atypical meningioma was diagnosed as grade I meningioma in squash smear cytology. This situation signifies the importance of radiological finding and representative sampling in diagnosing CNS tumours using smear cytology [10,15,24].

Limitation(s)

Major limitation encountered was non representative sampling, in which necrotic areas misleads the final diagnosis in the present study. This could overcome by representative sampling.

CONCLUSION(S)

This study shows a higher degree of cytological-histological matching in diagnosing meningiomas. It needs adequate clinical history, neuroimaging details and intraoperative impression of neurosurgeon in aiding the pathologists to improve the diagnostic accuracy. Squash smear cytology is fairly accurate, relatively safe, rapid, and simple tool to diagnose meningioma intraoperatively and helps the neurosurgeon to plan about the extent of surgery.

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