Section: Neurosurgery

Original Research Article

OUR INSTITUTIONAL EXPERIENCE IS USE OF "SQUASH" CYTOLOGY AS AN INTRAOPERATIVE GUIDE IN NEUROSURGICAL PRACTICE

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Abstract

Background: The accurate pathological diagnosis during neurosurgical procedures is imperative. As neurosurgery continues to expand into developing countries, it has been noted that many newly established centers in these regions lack the capacity for frozen section analysis initially. This has led to an increase in the reliance on intraoperative cytological diagnosis via the "squash" technique. Therefore, it is necessary to reevaluate the effectiveness of "squash" cytology for prompt diagnosis and its potential application in stereotactic biopsy procedures. The objective of this study was to determine the efficacy of using squash cytology for prompt diagnosis during intraoperative consultation in neurosurgical biopsy procedures. **Materials and Methods:** For this study, data was collected from 433 consecutive biopsy cases of central nervous system lesions that were submitted for intraoperative consultation. The biopsy samples were obtained in isotonic saline solution and processed immediately to prepare smears. The smears were then stained using a rapid Hematoxyline and Eosin staining method. The cytomorphological characteristics of the samples were compared with the histopathological findings obtained from paraffin section analysis. **Result:** In 78% of cases, the intraoperative cytological diagnosis was found to be consistent with the final diagnosis. The highest accuracy of intraoperative diagnosis was observed in cases of glioblastoma, meningioma, metastasis, and medulloblastoma. However, a marked decrease in diagnostic accuracy was noted in cases of non-neoplastic lesions and ependymoma. In addition to providing a diagnosis and grading, the smear cytology results also assisted in the identification of well-defined tumors and in guiding their surgical resection. **Conclusion:** Squash cytology has been determined to be a relatively accurate, simple, reliable, safe, and cost-effective tool for the rapid diagnosis of central nervous system lesions during surgery. Its utilization can aid in the targeting and resection of lesions, making it a valuable tool in the context of rural neurosurgical practice.

INTRODUCTION

Intraoperative pathological diagnosis plays a critical role in neurosurgery. As the field expands to new centers in developing countries, it has been noted that these centers often initially have the capacity for routine histopathological examination (HPE) and cytology, but it takes some time before facilities for frozen section analysis are established. This has resulted in an increase in the utilization of intraoperative cytological diagnosis using the "squash"technique.[1] Thus, there is a need to reevaluate the utility of "squash" cytology for rapid diagnosis, particularly in the context of stereotactic biopsies, where there may be limited tissue available for intraoperative diagnosis.[2]

The technique of intraoperative cytology preparation was first introduced in the early 1930sby Eisenhardt,[3] and Cushing and was further developed and documented by Russell et al. and Sir Hugh Cairns in the 1930s. The technique has gained renewed importance with the advent of CT and

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MRI-guided stereotactic biopsies. In neurosurgical practice, a prompt pathological diagnosis of a space-occupying lesion of the nervous system enables the surgeon to plan and modify the extent of surgery as necessary.

Intraoperative consultations regarding the pathology of brain lesions are frequently requested for a number of reasons, including:

1. Confirming the identification of a lesion.
2. Differentiating neoplastic from reactive lesions.
3. Differentiating metastatic from primary neoplasms.
4. Estimating the degree of malignancy.
5. Determining tumor margins.
6. Obtaining tissue for culture or other special procedures, such as immunohistochemistry.

The intraoperative cytology preparation was first introduced by Eisenhardt and Cushing in early 1930 and by Badt in 1937. This technique was further championed and documented by Russell et al., in 1937. The present technique was introduced by her along with Sir Hugh Cairns in 1930s.

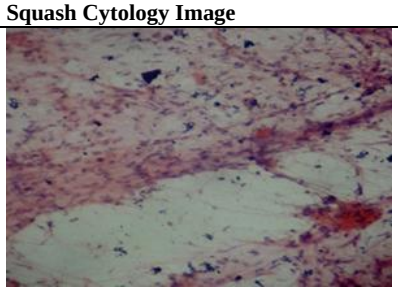
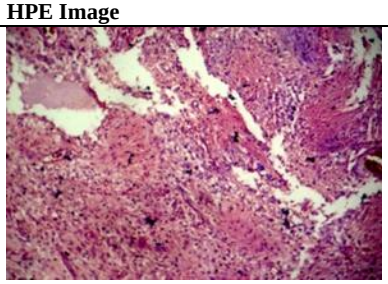
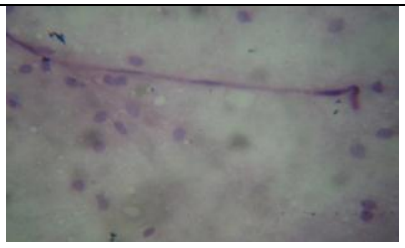
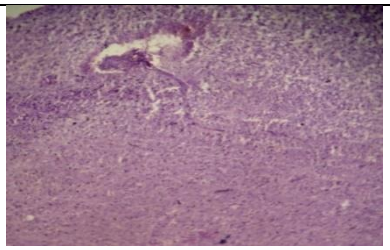
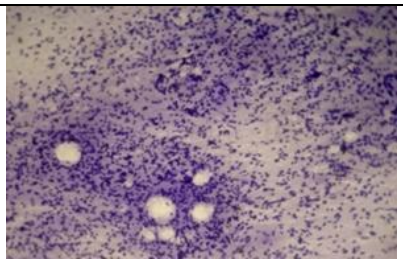
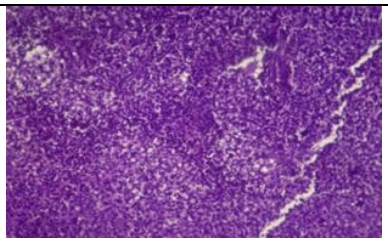
MATERIALS AND METHODS

The materials for this study were collected from 433 consecutive biopsies of central nervous system (CNS) lesions that were evaluated through intraoperative smear cytology. The biopsy specimens were collected during surgery and were immediately transported to the neuropathology laboratory in isotonic saline solution for processing. The smear preparation involved taking 1-2 millimeters of the biopsy material with a scalpel blade, placing it on a slide, and crushing it with another slide, applying just enough pressure to spread the tissue into a thin film. The smear was then fixed in 95% alcohol and stained with Hematoxylin and Eosin (H&E). The duration of the smear technique was only 20 minutes.

In addition, paraffin sections were prepared from the resected tissue and stained with H&E. Relevant clinical and radiological information was recorded. The smear cytology diagnosis was compared with the histopathological findings. The tumors were classified according to the World Health Organization (WHO) classification of CNS neoplasms. The observations were analyzed using statistical methods.

RESULTS

Of the 433 cases included in the study, the intraoperative cytological diagnosis agreed with the final diagnosis in 339 (78%) cases, but did not agree in 94 (22%) cases.

S.No	Remarks	Squash Cytology Image	HPE Image
1	Pilocytic astrocytoma cytology: In cases of pilocytic astrocytoma, the cytology was often difficult to spread due to thick clusters of spindled and stellate cells with bipolar fibrillary processes. Occasionally, Rosenthal fibers and eosinophilic granular bodies were seen.		
2	Diffuse astrocytoma cytology: In cases of diffuse astrocytoma, which contributed 21.7% of the cases, the smears spread easily and were composed of monomorphic neoplastic astrocytes around blood vessels with a fibrillated background.		
3	Anaplastic astrocytoma cytology: In cases of anaplastic astrocytoma, which contributed 5% of the cases, the smears spread evenly and were highly cellular. The cells aggregated around blood vessels and had pleomorphic nuclei with accentuated angulations of the nuclear outline and loose chromatin.		

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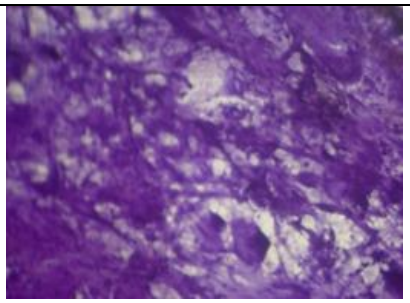
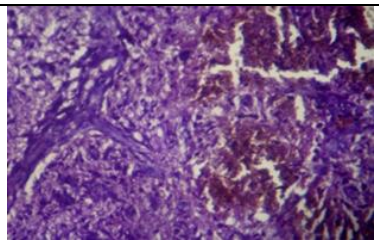
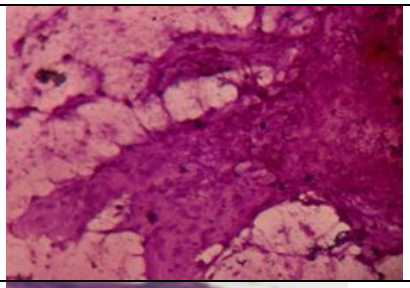
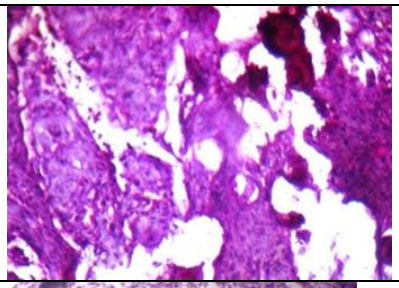
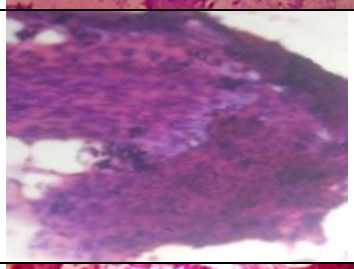
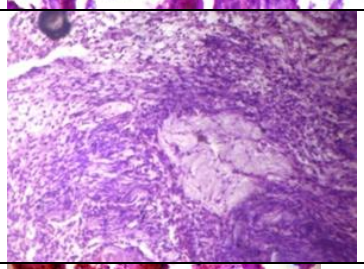
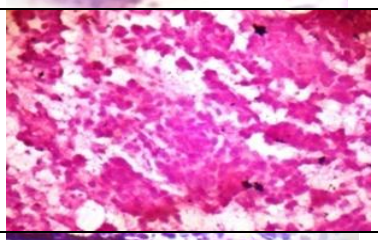
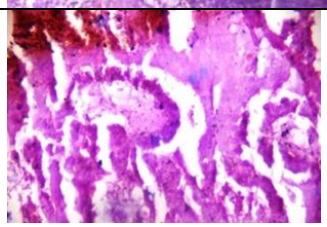
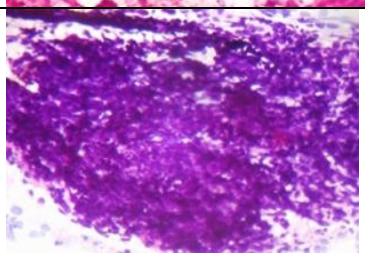
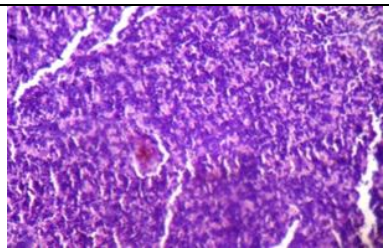
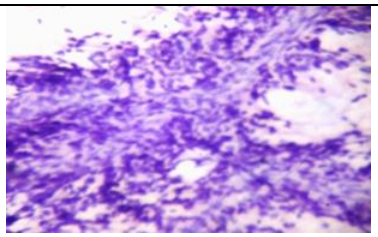
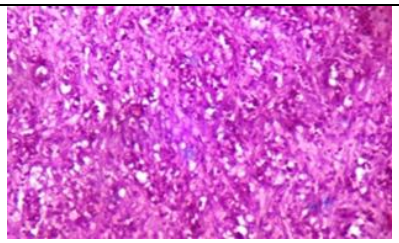
4	Gliobastoma multiforme grade IV cytology: In cases of grade-IV glioblastoma multiforme, which contributed 15.4% of the cases, the smears spread evenly and were highly cellular with mild to moderate nuclear pleomorphism. The cells were spindle-shaped, contained gemistocytes and bizarre tumor giant cells, and had a fibrillated background with necrotic debris. Significant endothelial proliferation with necrosis was seen as granular eosinophilic debris.		
5	Meningioma cytology: In cases of meningioma, which accounted for 6.23% of the cases, the smears did not spread well and were composed of rubbery, uneven, tightly cohesive clusters, whorls, and monolayered sheet waves. The nuclei had pseudo inclusion, stippled chromatin, and psammoma bodies.		
6	Schwanoma cytology: In cases of schwannoma, which were difficult to spread due to their cohesive nature, the smears had dimorphic features and were composed of spindle-shaped cells with elongated, wavy nuclei, with or without palisading.		
7	Secondaries cytology: In cases of secondary tumors, the smears were easily spread and contained pleomorphic cells with a high nucleo-cytoplasmic ratio and mitotic activity in cohesive clusters.		
8	Medulloblastoma cytology: In cases of medulloblastoma, which contributed 4.8% of the cases, the smears were highly cellular and spread easily. The cells were seen in discohesive sheets composed of small cells devoid of identifiable cytoplasm, round hyperchromatic nuclei with abundant mitosis, and individual cell necrosis.		
9	Ependymoma cytology: Ependymoma, present in 3.7% of the samples, exhibit smears that are highly cellular and unevenly spread, with fibrillated and well-vascularized characteristics. The nuclei may display clefting, while the tumor cells are observed to adhere to blood vessels, with occasional perivascular rosettes present.		

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In our series, the most common tumors were Grade-II and Grade-IV astrocytoma, which constituted 21.7% and 15.4% of the samples, respectively.

HPE Diagnosis	Number	%	CytologyCorrelating with HPE	% of correlation with HPE
Grade-I astrocytoma	30	6.92	23	77
Grade-II astrocytoma	94	21.70	78	83
Grade-IIIastrocytoma	22	5.08	17	77
Grade-IVastrocytoma	67	15.47	60	90
Medulloblastoma	21	4.84	17	81
Ependymoma	16	3.69	9	56
Meningioma	27	6.23	20	74
Secondaries	30	6.92	26	87
Schwannoma	26	6.00	19	73
Tuerculoma	21	4.84	12	57
Pituitary adenoma	20	4.61	15	75
Hemangioblastoma	12	2.77	9	75
Craniopharyngioma	16	3.69	12	75
PNET	8	1.84	6	75
Abscess	9	2.07	6	67
Neurofibroma	10	2.30	7	70
Epidermoid cyst	4	0.92	3	75
Total	433	100.00	339	

Previous studies have reported diagnostic accuracy in intraoperative diagnoses ranging from 80% to 97.3%.

S No	Studies	Accuracy rates (%)	Number of cases
01	Roessler et al[4]	95	4,172
02	Torres et al[5]	97.3	650
03	Ghoshal et al[6]	93	306
04	Shukla et al[7]	87.76	278
05	Shah et al[8]	89.7	183
06	Iqbal et al[9]	95.36	151
07	Kamechian et al[10]	84	139
08	Mitra et al[11]	88.5	114
09	Kiniet al[12]	86	100
10	Nigam et al[13]	89.3	75
11	Vermaet al[14]	88.9	63
12	Arpita jindal et al[15]	94	

DISCUSSION

Among the different types of tumors, the most accurate intraoperative diagnoses were obtained in cases of glioblastoma, metastasis, meningioma, and medulloblastoma. However, a significant reduction in diagnostic accuracy was observed in cases of non-neoplastic lesions, such as tuberculoma, abscess, and ependyoma.

The diagnostic accuracy in our study was determined to be 78% when compared with histopathology results. Among the different types of tumors, the highest accuracy of intraoperative diagnoses was observed in cases of glioblastoma, meningioma, metastasis, and medulloblastoma. However, a significant reduction in accuracy was noted in cases of non-neoplastic lesions, including ependymoma. The presence of extensive necrosis can pose a challenge in interpretation, as was the case with tuberculoma, due to the firm nature of the lesion, making it difficult to obtain a usable specimen.

The limitations of the smear technique include lesions that are too firm to obtain a specimen, as well as the interpreter's level of experience and ability to accurately identify the tissue. Other factors that may affect the accuracy of the diagnostic results include inadequate tissue samples, unrepresentative specimens, faulty staining techniques, and inadequate fixation.

Powell et alsuggest that squash cytology prepared with a sufficient amount of tissue provides a rich source of both cytological and histological details.[16] This technique allows for the clear differentiation of cell types, including neurons, oligodendroglial cells, and astrocytes, based on their distinct nuclear and cytoplasmic features.[8,17-20]

The ideal technique for intraoperative pathological diagnosis should be characterized by high accuracy, rapidity, and the preservation of tissue for paraffin section analysis. While frozen sections provide the best architectural details,[17,18]the inherent softness and high water content of nervous tissue can result in poor-quality sections. As a result, alternative techniques, such as squash smears and touch imprint cytology, which are technically simpler and more cost-effective, are often used.[19,20]These techniques are especially advantageous as they allow for the use of a small amount of tissue, which is critical in surgical procedures involving intracranial lesions localized in functionally important areas of the brain and the preservation of enough tissue for paraffin section analysis.

Several studies have compared the results of squash cytology with frozen sections and found the accuracy of cytology to be comparable to that of

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frozen sections.[21]Moreover, the elimination of freezing artifacts4 and reduced cryostat contamination from potentially infected tissue make squash cytology a preferable alternative to frozen sections.[22-24]

Squash smears are limited by the inability to control thickness, the potential for crushing artifacts, and difficulties with smearing. Folkerth has also noted the aforementioned advantages and disadvantages of squash smear cytology.[3,4]

In the study, soft lesions such as astrocytomas, oligodendrogliomas, and pituitary adenomas smeared well, however, lesions with increased fibrous component, calcification, or firm consistency such as meningiomas, schwannomas, and tuberculomas resulted in smears of poor quality. This is as noted by Folkerth,[4] and Asha et al.[25] According to Nigam et al,[10,13] a well-prepared squash smear may reduce the need for a frozen section or at least provide additional information for its interpretation. Some studies suggest that cytological preparation offers better morphological information than frozen section even with minimal biopsy material.[22,26,27]

However, in our study, a significant reduction in diagnostic accuracy was observed in cases of non-neoplastic lesions including abscess, tuberculoma, neurofibroma, and also lesions like ependymoma. There have been several previous studies documenting improper grading of central nervous system tumors in cytological preparations.

CONCLUSION

The results of our study indicate that the overall diagnostic accuracy of squash cytology was 78%. While it is a reliable method, it should not be considered a substitute for histopathology in the diagnosis of brain tumors. Rather, it should be considered a valuable supplement.

Squash cytology is a simple, efficient, reliable, and cost-effective diagnostic tool for rapidly identifying central nervous system (CNS) lesions during surgical procedures. It facilitates the precise targeting and removal of lesions, making it a useful addition to the arsenal of tools used in rural neurosurgical practices.

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