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## Cytological Study of Salivary Gland Lesions along with Histopathological Correlation in a Tertiary Care Centre

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### ABSTRACT

**Background and aim:** This study aimed to analyze the relative frequency and distribution of different salivary gland lesions on Fine needle aspiration cytology, study histopathological correlation, and evaluate the accuracy and efficacy of this procedure in diagnosing the lesions.

**Material and methods:** A five-year study was performed on salivary gland swellings in a tertiary health care centre. Fine needle aspiration cytology was done, and smears were stained with Leishman, Haematoxylin, and eosin stains. Histopathology was done wherever possible. Cytological and histopathological correlation was done and the results were statistically analyzed.

**Results:** One hundred cases were studied. Lesions were categorized into non-neoplastic and neoplastic lesions. Chronic sialadenitis was the most common non-neoplastic lesion, and Pleomorphic Adenoma was the commonest benign lesion. Malignant lesions reported were Adenoid Cystic carcinoma, Mucoepidermoid carcinoma, Carcinoma ex pleomorphic adenoma, and three cases were interpreted as positive for malignancy. 38 cases were available for histopathology, out of which 34 showed a positive correlation. The diagnostic accuracy, sensitivity, specificity, positive predictive value, and negative predictive value of the present study are 94.74%, 75%, 100%, 100%, and 100%, respectively.

**Conclusions:** Fine needle aspiration cytology is a rapid, cost-effective, and safe investigation for the primary categorization of salivary gland lesions into inflammatory, benign and malignant lesions. The high diagnostic accuracy and low false positive and false negative diagnosis suggest that it can be used as a first-line diagnostic procedure to evaluate patients with various salivary gland lesions. However, histopathological diagnosis remains the gold standard.

### 1. Introduction

Fine needle aspiration cytology (FNAC) is an essential and integral component of cytology that can be done in any swelling or organ of the body with minimal or no complications at all. The procedure is simple, requires very little time, and can deliver the report within a few hours. It is cost-effective, rapid, and with high sensitivity and specificity. Fine needle aspiration cytology is useful for diagnosing salivary gland lesions. It is preferred over incisional biopsy as the latter can lead to fistula formation, infection in the plane of surgery, and facial nerve palsy. It also helps differentiate a neoplastic from a non-neoplastic lesion. Conservative treatment may be helpful in the case of non-neoplastic inflammatory lesions.<sup>[1]</sup> Diagnostic methods like sialography, ultrasonography, and Computed Tomography scan provide some diagnosis but often cannot differentiate the nature and type of salivary gland lesion. In such cases, FNAC can diagnose the lesions with high accuracy.<sup>[2]</sup> Benign salivary gland tumours are usually seen in the age group of 30 to 70 years and are most frequent in

parotid than in the Submandibular gland. Malignant tumours are more frequent in women as compared to men. The non-neoplastic lesions comprise acute sialadenitis, chronic sialadenitis, retention cysts, granulomatous sialadenitis, and non-specific reactive changes. The neoplastic lesions include benign lesions to high-grade malignancies. The benign lesions frequently seen are pleomorphic adenoma within's tumour, myoepithelioma, and basal cell adenoma. The commonest lesion is pleomorphic adenoma. Malignant cases include adenoid cystic carcinoma, mucoepidermoid carcinoma, acinic cell tumour, adenocarcinoma, and squamous cell carcinoma.<sup>[3]</sup> In this study, most of the lesions were diagnosed by FNAC and categorized into non-neoplastic and neoplastic lesions. Histopathology (HPE) was done wherever possible, and a correlation of cytological and histopathological findings was made to assess the diagnostic accuracy and efficacy of FNAC.<sup>[3]</sup>

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**2. Material and methods**

One hundred cases were studied, and the cases presented with palpable lesions. FNAC was advised in all the cases. After obtaining consent, complete history taking and local examination were performed. Relevant laboratory and radiological investigations were recorded. The procedures followed were in accordance with the ethical standards of the responsible committee on human experimentation (institutional and regional). FNAC was performed on the same lesion using a 10ml syringe with a 22 gauge needle. Smears were made, half of which were air dried and stained with Leishman stain. The remaining were fixed in 95% alcohol, stained with haematoxylin and eosin (H&E) stain and Papanicolaou stain. Smears were analyzed, observations were recorded, and the cytological diagnosis was given. The lesions were placed in two broad groups- Non-neoplastic and Neoplastic. Neoplastic lesions were further divided into benign and malignant neoplasms. Out of the 100 cases, 38

specimens were available for histopathology. The specimens were fixed in 10% formalin and processed routinely. The slides were stained with haematoxylin and eosin stain. Special stains and Immunohistochemistry were used wherever necessary. The results were analyzed statistically by sensitivity, specificity, and positive and negative predictive values. The diagnostic accuracy of FNAC was determined.

**3. Results**

One hundred cases were studied, out of which 55 were male, and 45 were female (male to female ratio was 1.2:1). There was a slight male preponderance. Maximum patients were in the third and fourth decades of life. Among females, the majority of cases were in the fourth and fifth decades, whereas in males majority of cases were in the first two decades (Table 1).

**Table 1. Age and sex distribution of salivary gland lesions (n=100).**

Age Group (in years)	Sex		Grand Total
	Female	Male	
< or=20	2	14	16
21-30	8	12	20
31-40	13	7	20
41-50	13	5	18
51-60	6	11	17
>60	3	6	9
Grand total	45	55	100

Maximum cases were found to involve the parotid gland (73 cases). Twenty-three cases were seen in the submandibular gland. Other locations

included only 4 cases, in which palate was the common location (Table 2).

**Table 2. Distribution of location-wise salivary gland lesions.**

No	Location	No. of Cases	% of Cases
1	Parotid	73	73%
2	Submandibular	23	23%
3	Others	4	4%
Total		100	100%

Cytologically, most cases were benign neoplasms (55%), followed by non-neoplastic lesions (39%). Very few cases were malignant neoplasms (6%). Among the benign neoplasms, most cases were of Pleomorphic Adenoma (43 cases). Chronic Sialadenitis was predominant among non-

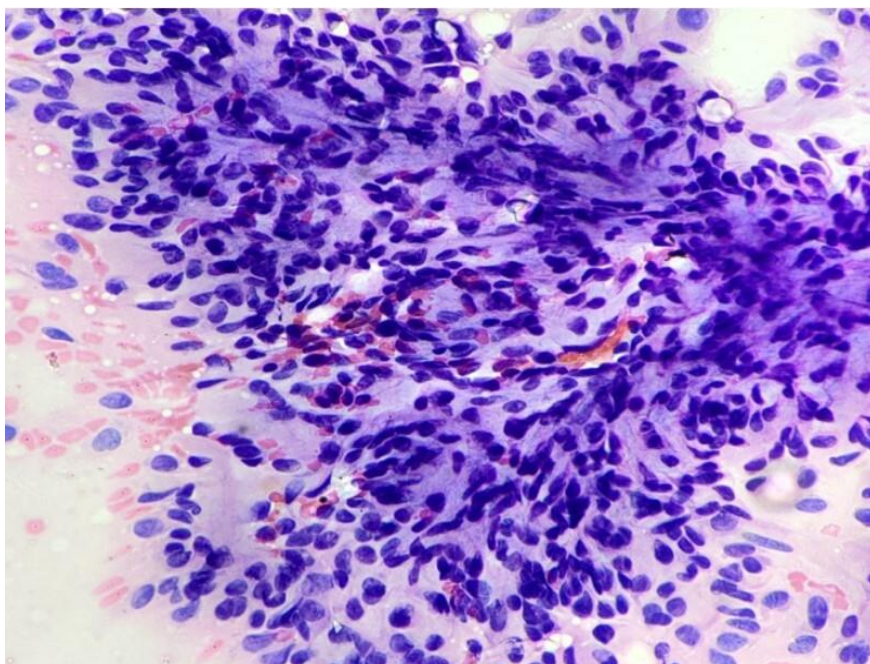
neoplastic lesions (13 cases). Among the malignant neoplasms was one case of Adenoid cystic carcinoma, Mucoepidermoid Carcinoma, and Carcinoma Ex Pleomorphic Adenoma. Three cases were reported as positive for malignancy, but a definite diagnosis was not possible on FNAC (Table 3).

**Table 3. The cytological spectrum of salivary gland lesions (n=100).**

Salivary Gland Lesions	No. of Cases	% of Cases
Non-neoplastic lesions	----	39%
Submandibular abscess	3	----
Parotid abscess	6	----
Cystic lesion	9	----
Acute sialadenitis	8	----
Chronic sialadenitis	13	----
Neoplastic lesions	----	----
<b>1. Benign tumors</b>	----	55%
Pleomorphic adenoma	43	----
Basal Cell adenoma	3	----
Warthin's Tumor	3	----
Benign Lymphoepithelial lesion/Mickulicz	1	----
Other benign lesions	5	----
<b>2. Malignant tumors</b>	----	6%
Adenoid cystic carcinoma	1	----
Mucoepidermoid carcinoma	1	----
Carcinoma ex pleomorphic adenoma	1	----
Positive for malignancy	3	----
Total	100	100%

Forty-three cases of Pleomorphic Adenoma (PA) were reported, which was the commonest lesion. Twenty were male, and 23 were female (male-to-female ratio-1:1.15). The cytology smears were cellular and showed clusters and sheets of benign epithelial cells that were round to oval. Myoepithelial cells appeared plasmacytoid; few were spindle-shaped in the chondromyxoid

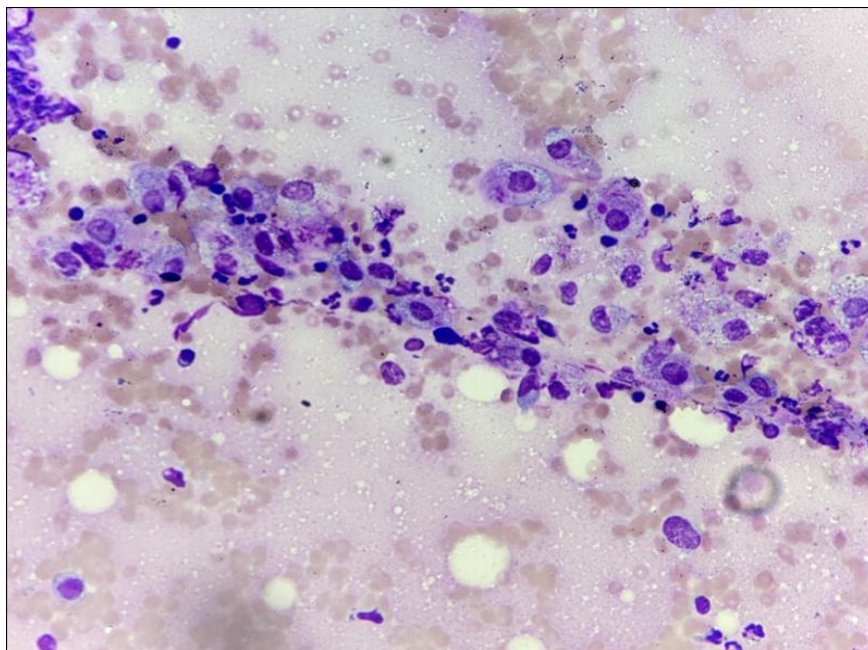
stroma. Myoepithelial cells showed polygonal, epitheloid, or clear appearance in a few cases. In other cases, the epithelial component formed the main tumour mass, whereas some myxoid stromal component was abundant, and the cellular component was scanty (Fig. 1).



**Fig. 1. Cytology image of Pleomorphic Adenoma showing round to oval epithelial cells and spindle-shaped myoepithelial cells in chondromyxoid stroma (H&E, 400X).**

Of the 43 cases, 33 were found in the parotid gland. Eight were seen in the submandibular gland and 2 in minor salivary glands. Smears of chronic sialadenitis were cellular and showed inflammatory cells comprising

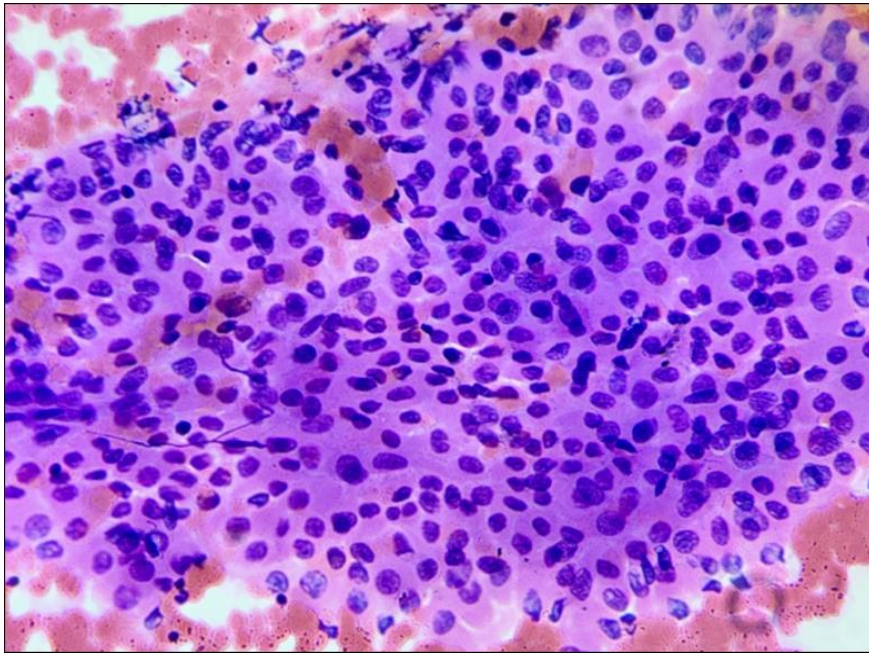
polymorphs, plasma cells, and lymphocytes with plump epithelial and myoepithelial cells against a hemorrhagic and fibromyxoid background (Fig. 2).



**Fig. 2. FNAC of Chronic Sialadenitis showing inflammatory cells with plump epithelial and myoepithelial cells against a hemorrhagic and fibromyxoid stroma (H&E, 400X).**

Warthin's tumour showed sheets of oncocytic cells that were uniform with regular round nuclei and abundant cytoplasm. Thin cystic material to

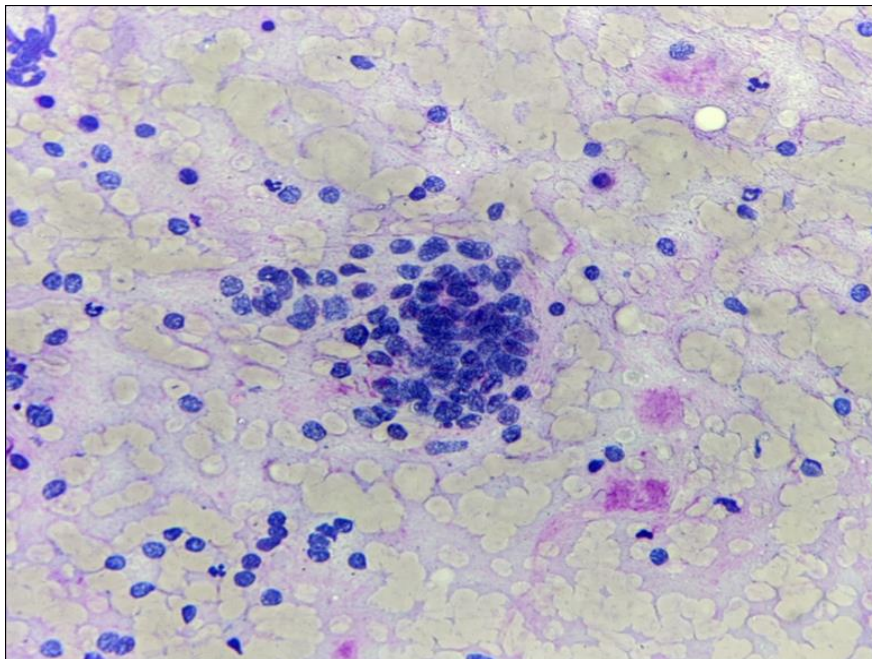
granular amorphous substance and plenty of lymphocytes were seen in the background (Fig. 3).



**Fig. 3. Warthin's Tumour Cytology showing sheets of oncocytic cells with regular round nuclei. (H&E, 400X).**

Smears of Adenoid cystic carcinoma were highly cellular and showed numerous scattered and loose clusters of acinar cells. Cells were large but morphologically resembled normal acinar cells and showed abundant

granular cytoplasm. Nuclei were round, monomorphic, had granular chromatin, and a few showed inconspicuous nucleoli (Fig. 4).



**Fig. 4. Adenoid Cystic Carcinoma cytology showing a cluster of cells with round monomorphic nuclei, granular chromatin, and inconspicuous nucleoli. (H&E,400X).**

Thirty-eight cases were available for the histopathological examination, out of which 34 cases showed a positive correlation (Table 4).

**Table 4. Histopathologic diagnosis of salivary gland lesions(n=38).**

	Histopathologic Diagnosis	No. of Cases	% of Cases
1	<b>Non-neoplastic lesions</b>	-----	18.42%
	Submandibular abscess	2	-----
	Parotid abscess	1	-----
	Acute sialadenitis	1	-----
	Chronic sialadenitis	2	-----
	Lymphoepithelial Cyst	1	-----
2	<b>Benign neoplastic lesions</b>	-----	60.53%
	Pleomorphic adenoma	19	-----
	Basal cell adenoma	1	-----
	Warthin's tumor	2	-----
	Lymphoepithelial sialadenitis/ Mickulicz syndrome	1	-----
3	<b>Malignant neoplastic lesions</b>	-----	21.05%
	Adenoid cystic carcinoma	2	-----
	Mucoepidermoid carcinoma	2	-----
	Carcinoma ex pleomorphic adenoma	2	-----
	Myoepithelial carcinoma	1	-----
	Metastatic deposits of squamous cell carcinoma	1	-----
	<b>Total</b>	38	100%

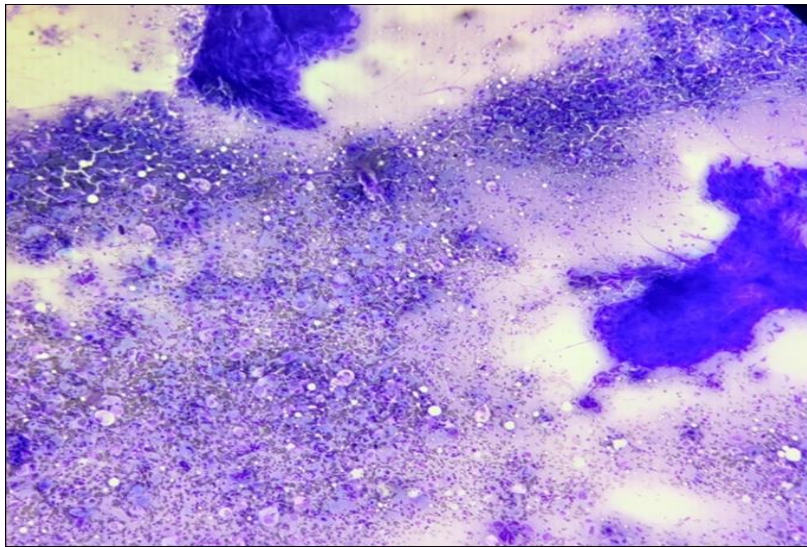
Only 4 cases showed cyto-histological discordance (Table 5). In our study, the diagnostic accuracy, sensitivity, specificity, positive predictive

value, and negative predictive value of overall lesions were 94.74%, 75%, 100%, 100%, and 100%, respectively.

**Table 5. Correlation of cytodiagnosis with histodiagnosis.**

No	FNAC Diagnosis	No. of Cases	Histopathology Diagnosis	No. of Cases	Positive Correlation	Negative Correlation
1	Acute sialadenitis	1	Acute sialadenitis	1	1	-----
2	Chronic sialadenitis	2	Chronic sialadenitis	1	1	-----
	-----	-----	Adenoid cystic carcinoma	1	-----	1
3	Submandibular abscess	2	Submandibular abscess	2	2	-----
4	Parotid abscess	2	Parotid abscess	1	1	-----
	-----	-----	Pleomorphic adenoma	1	-----	1
5	Benign cystic lesion	3	Lymphoepithelial cyst of parotid	1	1	-----
	-----	-----	Chronic sialadenitis	1	-----	1
	-----	-----	Mucoepidermoid carcinoma	1	-----	1

6	Pleomorphic adenoma	18	Pleomorphic adenoma	18	18	-----
7	Basal Cell adenoma	1	Basal cell adenoma	1	1	-----
8	Warthin's tumor	1	Warthin's tumor	1	1	-----
9	Mickuliczdiasease	1	Lymphoepithelial sialadenitis	1	1	-----
10	Benign parotid lesion	1	Warthin's tumor	1	1	-----
11	Adenoid cystic carcinoma	1	Adenoid cystic carcinoma	1	1	-----
12	Mucoepidermoid carcinoma	1	Mucoepidermoid carcinoma	1	1	-----
13	Carcinoma ex pleomorphic adenoma	1	Carcinoma ex pleomorphic adenoma	1	1	-----
14	Positive for malignancy	3	Metastasis of squamous cell carcinoma	1	1	-----
	-----	-----	Carcinoma ex pleomorphic adenoma	1	1	-----
	-----	-----	Myoepithelial carcinoma	1	1	-----



**Fig. 5. Mucoepidermoid Carcinoma- Cytology smears showing atypical epithelial cells (squamous) arranged in clusters and sheets against a dirty background of mucus and debris (Leishman, 100X).**

#### 4. Discussion

This study showed slight male preponderance, with a male-to-female ratio of 1.2:1. Maximum incidence was seen in the third and fourth decades of life. These findings were in accordance with other studies.<sup>[2-5]</sup> However, few studies showed a female preponderance in the distribution.<sup>[4, 5]</sup> The cytologic examination revealed a higher incidence of neoplastic lesions (61 cases) compared to non-neoplastic lesions (39 cases). Among the non-neoplastic lesions, inflammatory lesions were predominant. Chronic sialadenitis was the most common entity. Similar findings were seen in another study.<sup>[2]</sup> Other lesions were submandibular and parotid abscesses and cystic lesions. Benign neoplasms outnumbered malignant neoplasms. Among the benign neoplasms, Pleomorphic Adenoma was the most common lesion (78.18%). Other benign lesions included Basal cell Adenoma, benign lymphoepithelial lesion, and warthin's tumour, and 5 cases were diagnosed as

benign salivary gland lesions. It was in accordance with other studies.<sup>[6, 7]</sup> Among the malignant lesions, there was one case each of Adenoid Cystic Carcinoma, Mucoepidermoid Carcinoma, and Carcinoma Ex Pleomorphic Adenoma. Three cases were interpreted as positive for malignancy. Similar distribution and incidence of lesions on FNAC were also observed in other studies.<sup>[5, 7, 8]</sup> There were 39 non-neoplastic lesions on cytology, of which only ten cases underwent HPE. One case of acute sialadenitis, one case of chronic sialadenitis, two submandibular abscesses, one parotid abscess, and one cystic lesion were confirmed on HPE. A case of Mikulicz's disease was diagnosed based on cytological features showing lymphoepithelial lesions and correlating clinical features. So diagnostic accuracy for overall non-neoplastic lesions was 60% in our study. However, other studies showed accuracy up to 75-100%.<sup>[2, 3]</sup>

In the present study, 43 cases were diagnosed cytologically as Pleomorphic Adenoma. The male-to-female ratio was 1:1.15. Eighteen cases were available for histopathologic correlation. All the cases were correctly diagnosed on HPE, thus giving the diagnostic accuracy of 100% in diagnosing Pleomorphic Adenoma on FNAC. It was an excellent finding in our study as various other studies did not show accuracy to this extent.<sup>[2,3]</sup> Three cases of basal cell adenoma were diagnosed on FNAC, out of which one case underwent histopathological examination. The HPE diagnosis was consistent with the cytological diagnosis, thereby making the accuracy of FNAC in diagnosing Basal Cell Adenoma 100%. However, in a few other studies, the cytological diagnosis of Basal cell Adenoma was not this accurate. A case of Basal cell adenoma on FNAC turned out to be Basal cell carcinoma on HPE.<sup>[2]</sup> In another study, one case turned out to be pleomorphic adenoma after HPE. The high cellularity of the PA, along with scanty stromal material, led to misinterpretation.<sup>[9]</sup> There were five benign lesions where a definite cytological diagnosis could not be done. One such case was available for histopathological examination. This case was diagnosed as Warthin's Tumour on HPE. On cytology, smears were paucicellular with few glandular epithelial cells in small groups and clusters and had eosinophilic cytoplasm. On HPE, the tumour showed a double layer of epithelial cells with the upper layer of palisading columnar cells with abundant finely granular eosinophilic cytoplasm, while the lower layers were composed of cuboidal to polygonal cells. Polypoid projections of lymphoepithelial components narrowed cystic spaces. Three cases interpreted as positive for malignancy on cytology were finally diagnosed as myoepithelial carcinoma, metastatic deposits of squamous cell carcinoma, and carcinoma ex pleomorphic adenoma on HPE.

#### Discordant cases

A case reported as a Parotid abscess on cytology turned out to be Pleomorphic Adenoma on HPE. Cytology smear showed necrotic material and degenerated cells. The most aspirate was not appropriately obtained, which might have led to an incorrect diagnosis. Another case of adenoid cystic carcinoma was interpreted as chronic sialadenitis on cytology. HPE showed tumour cells composed of small cells having dark, compact nuclei and scant cytoplasm. These cells could have been incorrectly interpreted as lymphocytes, leading to a false diagnosis on cytology. Two benign cystic lesions were reported as chronic sialadenitis and mucoepidermoid carcinoma (MEC) on HPE. Aspirate must have been obtained from the mucus-secreting cells of mucoepidermoid carcinoma, which led to a diagnosis of cystic lesion on cytology. Cystic mucus-containing lesions often cause a diagnostic dilemma in cytology as they show similar features in cases of Warthin's Tumour, sialadenitis, and low-grade MEC.<sup>[10-12]</sup> All the malignant lesions on cytology were confirmed on histopathology. Thus the accuracy of diagnosing malignancies on FNAC in our study was 100%. Similar results were seen in other studies also.<sup>[3,7,13,14]</sup> In accordance with other studies, we can say that cytology of Salivary gland lesions can help surgeons counsel and further manage the patients. In order to reach a final diagnosis, a team discussion between the pathologist, radiologist, and surgeon is essential.<sup>[15]</sup> The Milan System for Reporting Salivary Gland Cytopathology has also been introduced to ensure universal uniformity in the reporting of salivary gland lesions on cytology in order to improve clinicopathological communication for better patient management.<sup>[16]</sup>

#### 5. Conclusion

FNAC is a rapid, effective, and safe mode of investigation for the primary

categorization of salivary gland lesions into inflammatory, benign and malignant lesions. Multiple aspirations should be done in cases of cystic lesions, lesions with cystic and solid areas, and large swellings to avoid false diagnoses. Pre-operative diagnosis plays a crucial role in avoiding unnecessary surgeries in inflammatory conditions as they can be treated medically. Communication and co-operation between clinician and cytopathologist should be done in doubtful cases. Before FNAC, Ultrasound findings should be available to improve the accuracy rate. This study's high diagnostic accuracy and low false positive and false negative diagnosis suggest that FNAC can be used as a first-line diagnostic procedure to evaluate patients with various salivary gland lesions. However, Histopathological diagnosis remains the gold standard due to the heterogeneous nature of many benign and malignant tumours, many of which show overlapping cytological features, thus making diagnosis difficult at times.

#### Conflict of Interest

The authors declared that there is no conflict of interest.

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