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Original Article

Pathology Section

Diagnosis of Salivary Gland Lesions By Fine Needle Aspiration Cytology and Its Histopathological Correlation in A Tertiary Care Center of Southern India

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ABSTRACT

Background: Salivary glands may enlarge either due to inflammation or neoplastic conditions and the diagnosis is possible by fine needle aspiration cytology (FNAC).

Aim: The present study was undertaken to determine utility of FNAC in the diagnosis of salivary gland lesions.

Materials and Methods: In this retrospective study, a total of 186 FNACs of salivary gland lesions were retrieved and evaluated. Of these, 146 cases had follow-up histopathological diagnosis. FNAC diagnoses were compared to histopathological diagnoses.

Results: The parotid glands were more commonly involved than others. Among the various diagnostic categories used in FNAC reports, Non neoplastic category was seen in 24 (16.4%), benign category in 86 (58.9%) and malignant category in 30 (20.6%) and unsatisfactory category in 6 (4.1%) of 146 cases. The overall sensitivity, specificity, accuracy, positive predictive value and negative predictive value of FNAC in the diagnosis of salivary gland lesions were 86.6%, 94.6%, 93.6%, 88.3%, and 94.6% respectively.

Conclusion: The present study concluded that FNAC in the diagnosis of salivary gland lesions is highly sensitive, specific and accurate method. Hence, FNAC is a useful, quick and reliable diagnostic tool. It also appears to be a safe, cost effective and minimally invasive procedure, which provides information for management of salivary gland lesions.

Keywords: Diagnostic tool, Diagnostic utility, Non neoplastic

INTRODUCTION

Fine needle aspiration cytology (FNAC) has been widely used as a diagnostic tool for the management of various head and neck lesions [1]. It is a minimally invasive, easy to perform technique. The smear evaluation is immediate and the procedure can be repeated many times to obtain more tissues for diagnosis or special investigations [2,3]. Salivary gland neoplasms comprise of 3%-10% of head and neck tumours [4]. Since FNAC can differentiate inflammatory lesions from neoplastic conditions, lymphomas from epithelial malignancies and primary tumours from secondary tumours [5], patients with salivary gland lesions should undergo FNAC for triaging and thereby patients can be planned for next step of therapeutic approach. The present study was undertaken to determine utility of FNAC in the diagnosis of salivary gland lesions.

MATERIALS AND METHODS

This retrospective study was carried out to review the salivary gland lesions over a period of 3 years between March 2012 and February 2015 at our institution after obtaining approval from institutional ethical committee. Details of age, gender and other relevant clinical informations were collected from medical records. May-Grunwald Giemsa (MGG) and Haematoxylin and Eosin (H&E) stained FNAC slides of 182 cases were retrieved and evaluated. Slides stained with Periodic Acid-Schiff (PAS) was also retrieved and studied for the demonstration of mucus producing tumours. MGG stain was done to identify chondromyxoid matrix/metachromatic stroma/basement membrane materials as it is seen in tumours such as pleomorphic adenoma, basal cell adenoma, adenoid cystic carcinoma, polymorphous low grade adenocarcinoma and epithelial-myoepithelial carcinoma. Cytological diagnoses were classified into four categories- non-neoplastic, benign, malignant and unsatisfactory. Chronic sialadenitis and lymphoepithelial cyst were considered as non-neoplastic category, pleomorphic adenoma (PA), basal cell adenoma (BCA) and Warthin's tumour (WT) were considered as benign category, mucoepidermoid carcinoma (MEC), acinic cell carcinoma (ACC), adenoid cystic carcinoma, carcinoma ex pleomorphic adenoma, epithelial-myoepithelial carcinoma and squamous cell carcinoma were considered as malignant category. Among 182 FNAC cases, 146 patients had histologic follow up. Histopathological results were categorized as negative and positive test results. Non-neoplastic and benign lesions were considered as negative test result and malignant lesions were considered positive test result. On correlation of FNAC diagnoses with histopathological diagnoses, the sensitivity, specificity, accuracy, false positive rate, false negative rate, positive predictive value and negative predictive value of FNAC were calculated. The unsatisfactory results were excluded from the analysis. The results were tabulated and statistical analyses were done with the IBM SPSS statistical software, version 20.

RESULTS

Age and gender distribution

The mean age of the patients with non-neoplastic lesions was 29 years with a range from 18 to 75 years, benign lesions was 32 years with a range from 22 to 78 years and malignant lesions was 53 years with a range from 39 to 80 years among 182 cases. There were 112 males and 70 females with overall male to female ratio of 1.6:1.

FNAC diagnosis

The parotid gland (138/182, 75.8%) was the most commonly involved salivary gland followed by the submandibular gland (40/182, 22%) and minor salivary glands (4/182, 2.2%). Among 182 FNAC cases, non-neoplastic category consisted of 29 (15.9%), benign category consisted of 117 (64.3%) malignant category consisted of 30 (16.5%) and unsatisfactory category consisted of 6 (3.3%) cases as shown in [Table/Fig-1]. Most commonly diagnosed non-

Diagnostic category	Number of cases (%)
Non-neoplastic	29 (15.9%)
Chronic sialadenitis	24 (13.2%)
Lymphoepithelial cyst	5 (2.7%)
Benign	117 (64.3%)
Pleomorphic adenoma	91 (50%)
Basal cell adenoma	4 (2.2%)
Warthin's tumour	22 (12.1%)
Malignant	30 (16.5%)
Mucoepidermoid carcinoma	9 (5%)
Acinic cell carcinoma	7 (3.8%)
Adenoid cystic carcinoma	6 (3.3%)
Carcinoma ex pleomorphic adenoma	5 (2.8%)
Epithelial-myoepithelial carcinoma	1 (0.5%)
Squamous cell carcinoma	2 (1.1%)
unsatisfactory	6 (3.3%)

[Table/Fig-1]: Distribution of FNAC diagnoses in salivary gland lesions (n=182)

neoplastic, benign and malignant condition in our study by FNAC were 24 (13.2%) cases of sialadenitis, 91 (50%) cases of PA and 9 (5%) cases of MEC respectively.

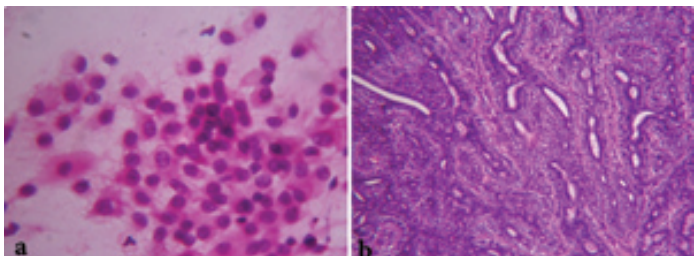
Correlation of FNAC and histopathological diagnoses

Among 182 cases of FNAC, 146 patients underwent surgical intervention and corresponding final histopathological diagnoses were available [Table/Fig-2]. The correlation of 146 FNAC diagnoses with corresponding histopathological diagnoses was done.

In 24 cases of non-neoplastic lesions by FNAC, 21 cases correlated with final histopathological diagnoses. Out of 3 cases reported as sialadenitis on FNAC, 1 case turned out to be MEC and 2 cases, WT. In 86 cases of benign tumours, 75 were consistent with corresponding histopathological diagnoses. PA was confirmed in 55 cases out of 62 FNAC diagnoses. In 7 cases, cytological diagnosis of PA, 1 case turn to MEC, 3 cases as adenoid cystic carcinoma and 3 cases as BCA. Four cases interpretated as BCA on FNAC, 2 cases were correctly cyto-typed [Table/Fig-3a,b] and 2 cases turn to PA. WT was consistent in 18 of 20 cases with one case of sialadenitis and another case of low grade MEC on histopathological examination.

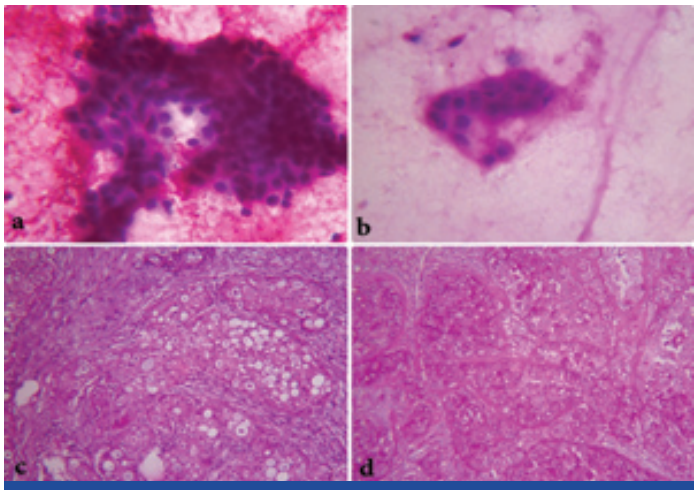
Histopathological diagnoses	Number of cases (%)
Normal salivary gland	1 (0.7%)
Non-neoplastic	25 (17.1%)
Chronic sialadenitis	18 (12.3%)
Kuttner's tumour	1 (0.7%)
Lymphoepithelial cyst	6 (4.1%)
Benign	88 (60.2%)
Pleomorphic adenoma	58 (39.7%)
Basal cell adenoma	6 (4.1%)
Warthin's tumour	24 (16.4%)
Malignant	32 (22%)
Mucoepidermoid carcinoma	11 (7.5%)
Acinic cell carcinoma	6 (4.1%)
Adenoid cystic carcinoma	8 (5.5%)
Carcinoma ex pleomorphic adenoma	4 (2.8%)
Epithelial-myoepithelial carcinoma	1 (0.7%)
Squamous cell carcinoma	2 (1.4%)
Total	146 (100%)

[Table/Fig-2]: Histopathological diagnoses of salivary gland lesions (n=146)

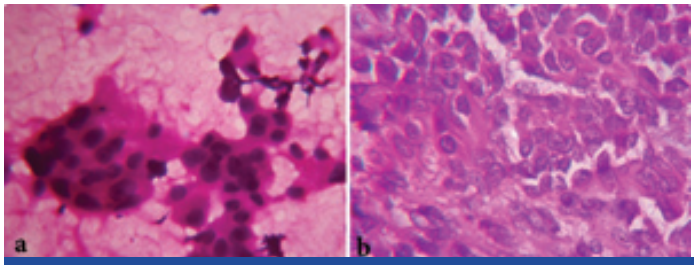


[Table/Fig-3a,b]: Basal cell adenoma. (a) Moderately cellular smear showing singly scattered and cluster of basaloid cells with many naked nuclei (H and E x100). (b) Photomicrograph showing tumour composed of cords and nests of neoplastic epithelial cells with peripheral palisading and scanty stroma (H & E x100)

In malignant group, 25/30 cases of cytological diagnosis was comparable with histopathological diagnosis. PAS stain was done in both cytology and histopathology slides to demonstrate mucus cells in MEC [Table/Fig-4a,b]. Five cases diagnosed as malignancy on FNAC, 3 cases turns to benign [Table/Fig-5a,b], 2 cases as non-neoplastic lesions and 1 case was diagnosed as normal salivary gland on final histopathological interpretation [Table/Fig-6].



[Table/Fig-4a-d]: Mucoepidermoid carcinoma. (a) Cytosmear showing a cluster of tumour cells with mucin secreting cells (MGG x100). (b) Cytosmear revealing tumour cells and PAS positive mucus cells (PAS x100). (c) Photomicrograph exhibiting nests of malignant squamous epithelial cells and many cystic spaces filled with mucin (MGG x100). (d) Photomicrograph showing malignant squamous cells and PAS positive mucus cells (PAS x100)



[Table/Fig-5a,b]: False positive case-Carcinoma ex pleomorphic adenoma turned out to be Pleomorphic adenoma. (a) Cytosmear showing a cluster of tumour cells having moderate cytoplasm and markedly pleomorphic hyperchromatic nuclei with few benign appearing cells suggestive of Carcinoma ex pleomorphic adenoma (MGG x400). (b) Follow-up histopathology section revealing nuclear atypia in the ductal epithelial cells were mistaken as malignancy on FNAC (H & E x400)

The sensitivity, specificity and accuracy for diagnosing non-neoplastic salivary gland lesions were 84%, 97.4% and 95%, respectively. The sensitivity, specificity and accuracy for diagnosing benign lesions were 95.2%, 91.07% and 93.6% respectively. The sensitivity, specificity and accuracy value for diagnosing malignant lesions were 80.7%, 95.4% and 92.1% respectively. The overall sensitivity, specificity, accuracy, positive predictive value and negative predictive value for diagnosing salivary gland lesions were 86.6%, 94.6%, 93.6%, 88.3%, and 94.6% respectively. False positive and false negative rate were 4.6% and 19.3% respectively.

DISCUSSION

The aim of FNAC is to determine if a mass is inflammatory, reactive, benign, or malignant and if possible, to render a specific diagnosis which helps the clinicians in the patient management. On comparison with other diagnostic methods, FNAC has many advantages [6] viz inexpensive, fast, efficient, safe and well tolerated [4].

Non-neoplastic lesions

In our study, the rate of non-neoplastic lesion was 15.9% (29/182) cases in contrast to those of other studies, ranging from 20% to 72.9% [7-10]. The most common non-neoplastic lesion was chronic sialadenitis followed by lymphoepithelial cyst. Two cases of WT

FNAC Diagnoses	Number of cases	Final histopathological diagnoses	Number of cases
Non-neoplastic	24		
Chronic sialadenitis	19	Chronic sialadenitis	16
		Warthin's tumour	2
		Low grade mucoepidermoid carcinoma	1
Lymphoepithelial cyst	5	Lymphoepithelial cyst	5
Benign	86		
Pleomorphic adenoma	62	Pleomorphic adenoma	55
		Basal cell adenoma	3
		Mucoepidermoid carcinoma	1
		Adenoid cystic carcinoma	3
Basal cell adenoma	4	Basal cell adenoma	2
		Pleomorphic adenoma	2
Warthin's tumour	20	Warthin's tumour	18
		Chronic sialadenitis	1
		Low grade mucoepidermoid carcinoma	1
Malignant	30		
Mucoepidermoid carcinoma	9	Mucoepidermoid carcinoma	7
		Chronic sialadenitis	1
		Kuttner's tumour	1
Acinic cell carcinoma	7	Acinic cell carcinoma	6
		Normal salivary gland	1
Adenoid cystic carcinoma	6	Adenoid cystic carcinoma	5
		Basal cell adenoma	1
Carcinoma ex pleomorphic adenoma	5	Carcinoma ex pleomorphic adenoma	4
		Pleomorphic Adenoma	1
Epithelial-myoepithelial carcinoma	1	Epithelial-myoepithelial carcinoma	1
Squamous cell carcinoma	2	Squamous cell carcinoma	2
Unsatisfactory	6	Lymphoepithelial cyst	1
		Warthin's tumour	5
Total	146	Total	146

[Table/Fig-6]: Correlation of FNAC and histopathological diagnoses of salivary gland lesions (n=146).

and one case of MEC were misdiagnosed as chronic sialadenitis. This could be due sampling errors and misinterpretation. Sufficient aspiration of material, correct interpretation and correlation of clinical findings could have been eliminated the wrong diagnosis.

Benign lesions

In this study, benign neoplasm consisted of 117 (64.3%) cases which were comparable with most of the published studies ranging from 49% to 83% [7,11-14] and higher than Omhare et al., [15] study (31.5%, 39/124). PA (50%) was most common benign neoplasm found in our study followed by WT (12.1%), similar to previously published studies [8,10,12,16-18]. Three cases each of BCA and adenoid cystic carcinoma, one case of MEC were misdiagnosed as PA on FNAC. BCA is one of the differential diagnoses which can easily misinterpretated as PA. Nonfibrillary chondromyxoid matrix, peripheral palisading and more number of naked nuclei favour the BCA [19]. In our case, we observed high cellularity, cells with slight spindling and clump of basement membrane materials; hence diagnosis of PA was made. In three cases, there was mild nuclear atypia along with hyaline globules and few myoepithelial cells. The diagnosis of PA over adenoid cystic carcinoma was made as the nuclear atypia was only noted. Failure to make MEC could be due to mucous in the background being mistaken as chondromyxoid matrix which leads the diagnosis of PA. Two cases of PA were diagnosed as BCA on FNAC. This could be due to matrix poor tumour cells and scanty myoepithelial cells [19]. One case of chronic sialadenitis was misdiagnosed as WT. Chronic inflammatory and duct lesions can accumulate fluid, show oncocytic metaplasia and contain plenty of lymphocytes which can easily confused with WT [19]; this could be the explanation for wrong diagnosis of chronic sialadenitis. Foamy histiocytes are close mimickers of mucous secreting cells and cells with true or mature squamous cell differentiation are almost never seen in low grade MEC [19]. In our case we found foamy histiocytes, no squamous cells and bland cytological features; hence we missed the low grade MEC. The rate of agreement of WT between FNAC and histopathology in this study was 90% (18/20) which was almost similar to Diaz et al., (100%) and Flezar et al., (95%) studies [4,20].

Malignant lesions

Various authors have reported the incidence of malignant tumours, ranged from 15% to 32% [8,9]. The incidence of malignant tumours in the present study was 16.5% (30/182) almost comparable to Omhare et al., [15] study (15.32%, 19/124) and higher than Nguansangiam et al., [7] study (8.3%, 11/133). In this study, MEC was most common malignant tumour (30%, 9/30) similar to Omhare et al., [15] study (42.1%, 8/19) followed by ACC. Whereas, Diaz et al., [4] have observed, adenoid cystic carcinoma was most common malignant tumour (27.3%, 6/22) followed by poorly differentiated carcinoma, carcinoma with squamous differentiation and adenocarcinoma.

One case of chronic sialadenitis and another case of Kuttner's tumour were misdiagnosed as MEC on FNAC. Ductal epithelium can show metaplastic changes such as squamous, mucous and oncocytic metaplasia due to long standing chronic inflammation and sialolithiasis and it can also cause mucous accumulation which lead to cystic dilatation, acinar cell atrophy and fibrosis. Metaplastic squamous cells and mucous cells may be mistaken for intermediate or squamous cells and mucous producing cells of MEC [19]; this could be the reason for wrong diagnosis of MEC. The present study diagnosed 6 cases of ACC and one case of normal salivary gland on histopathological examination among 7 cases of ACC diagnosed by FNAC. The important and major differential diagnosis of ACC is normal salivary gland acinar cells [21]. In our study, patient diagnosed as ACC which was turned out to be normal salivary gland, presented with minimal enlargement of parotid gland in a short duration. The FNAC slides of ACC showed moderately cellular smears with clusters and few singly scattered acinar cells having clear to vacuolated cytoplasm and eccentric nuclei; presence of singly scattered acinar cell made us to diagnose ACC. All the 6 cases of adenoid cystic carcinoma diagnosed by FNAC were confirmed except one case which was turned out to be BCA on histopathological examination. We have made diagnosis of adenoid cystic carcinoma on FNAC by presence of hyaline globules and cylinders of metachromatic stroma with few clusters of small, uniform cells. However these hyaline globules and cylinders of metachromatic stroma are not specific or unique to adenoid cystic carcinoma. It can be seen in BCA and other salivary gland tumours [22]. Presence of hyaline globules and cylinders led to diagnose adenoid cystic carcinoma over BCA. The rate of agreement between FNAC and histopathological diagnosis of adenoid cystic carcinoma was 83.3% which is almost similar to the results of Diaz et al., study [4], who achieved 100% accuracy in 4 cases. One case of PA was diagnosed as carcinoma ex PA on FNAC; however other 4 cases of carcinoma ex PA were cytotyped correctly with an agreement of 80%. The aspirate, which was diagnosed as carcinoma ex PA on FNAC showed mild to moderate atypia of the tumour cells with scanty myoepithelial cells and the patient had history of sudden increase in size. This led to us to give erroneous diagnosis. Epithelial-myoepithelial carcinoma (1/1) and squamous cell carcinoma (2/2) were correctly cytotyped with an agreement of 100%.

The false positive rate in our study was 4.6% similar to those of other studies, ranged from 0% to 4.7% [7,15,17]. The false negative rate in this study was 19.3% comparable to those of other studies, ranged from 2.2% to 24.5% [7,15,17]. The unsatisfactory specimens in the present study were 3.3% in contrast to studies done by Nguansangiam et al., [7] and Diaz et al., [4] (5.2% and 7% respectively).

In our study, the overall sensitivity, specificity, accuracy, positive predictive value and negative predictive value of FNAC in the

diagnosis of salivary gland lesions were 86.6%, 94.6%, 93.6%, 88.3%, and 94.6% respectively which was comparable with previously published studies [4,7,9,11,12,15,17,18]. The comparison of sensitivity, specificity, accuracy, positive predictive and negative predictive value of various previous studies with present study is shown in [Table/Fig-7].

Study	Number of patients	Sensitivity (%)	Specificity(%)	Accuracy(%)	PPV (%)	NPV (%)
Stow et al., [23]	104	86.9	92.3	92.3	96.8	86.6
Postema et al., [24]	380	88	99	96	95	97
Stramandinoli et al., [25]	79	68.2	87.7	82.3	68.2	87.7
Nguansangiam et al., [7]	133	81.3	99.1	97	92.9	97.5
Omhare et al.,[15]	86	88.2	97.1	95.3	88.2	97.1
Diaz et al., [4]	182	94	100	99	100	100
Present study	146	86.6	94.6	93.6	88.3	94.6

[Table/Fig-7]: Comparison of results of present study with previous studies
PPV: Positive predictive value, NPV: Negative predictive value

CONCLUSION

The present study concluded that FNAC in the diagnosis of salivary gland lesions is highly sensitive, specific and accurate method. Hence, FNAC is a useful, quick and reliable diagnostic tool. It also appears to be a safe, cost effective and minimally invasive procedure, which provides information for management of salivary gland lesions.

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