DIAGNOSTIC PRECISION OF FNAC FOR SALIVARY GLAND LESIONS

Shabana Naz[1], AbdurRehman[2], Shagufta Naeem[1], Farhad Ali[3], Khurshid Ali[4], Sidra Maqbool[5], Hamza Javed[6]

[1]Department of Histopathology, Ayub Medical College, Abbottabad

[2]Department of Oral Pathology, Dental Section, Ayub Medical College, Abbottabad

[3]Department of R&D, Khyber College of Dentistry, Peshawar

[4]Department of Oral Pathology, Khyber College of Dentistry, Peshawar

[5]Department of Pathology, Ayub Medical College, Abbottabad

[6]Final year MBBS student, Ayub Medical College, Abbottabad

ABSTRACT

Objective: *The objective of this study was to evaluate the validity of FNAC as a diagnostic investigation for salivary gland lesions (SGL) and to discuss the spectrum of SGL in our setup.*

Materials and Methods: *In the Pathology department of Ayub medical college and Advance laboratory Abbottabad, 82 salivary gland FNAC cases from Jan 2018 to June 2020 were studied to identify their cytological characteristic and correlated with histopathological findings.*

Results: *In this study, 82 patients with salivary gland swellings were included. The age range from 24-73 years with a mean age of 47 SD13yrs, with male to female ratio of 1.8:1. Among 82 cases non-neoplastic lesions constituted about 21 cases, neoplastic cases were 61. Out of which 52 cases were of pleomorphic adenoma,2 cases of within's tumor, 3 cases were of adenoid cystic carcinoma, 2 mucoepidermoid carcinomas low grade, one each acinic cell carcinoma and follicular lymphoma. Histopathological correlation was available in all the neoplastic cases.*

Conclusion: *FNAC is an accurate and effective modality in terms of sensitivity, specificity, and accuracy for diagnosing the SGL. Owing to its safety, affordability, and quick results, it should be considered as an investigation of the first choice for SGL before devising a definitive management plan. On the other hand, due to the overlap between the cytological features of certain salivary gland tumors, tissue biopsy for histopathology examination is the gold standard.*

Keywords: *FNAC, Salivary gland lesions, pitfalls, Diagnostic Accuracy, pleomorphic adenoma, adenoid cystic carcinoma.*

INTRODUCTION

The swelling of the salivary glands usually is a diagnostic challenge for the cytologist with regards to its site of origin and nature. Primary Salivary Gland tumors include a heterogeneous set of 45 morphologic types as described by the World Health Organization (WHO).[1] In the 4th edition of the WHO Classification of Head and Neck Tumors, many entities were compressed into their broader categories due to their clinical and cytomorphological likeness or put into a different group providing pathologists flexibility in recognition of the SGL.[2,3] Generally, Salivary Gland Lesions (SGL) are classified into non-neoplastic/ inflammatory or neoplastic groups, with benign and malignant categories in the neoplastic group.[4] Salivary Gland tumors comprise 2 – 6.5% of all head and neck neoplasms.[5-8] Population-based surveys report that 8-9 malignant SGL can be expected in a population of one million per

Correspondence:
Dr. Shagufta Naeem
Associate Professor, Department of Histopathology,
Ayub Medical College, Abbottabad
Email: drshaguftanaeem@yahoo.com
Contact: +923348952197

annum.[9] Malignant SGL accounts for >0.5% of all malignancies.[9,10] Age incidence of SGL varies from children to over 80 years of age with the mean age of incidence in the 5th decade of life.[6]

Fine Needle Aspiration Cytology (FNAC) is a diagnostic test based on the morphological findings of individual cells, clumps of cells, and micro shreds of tissue, aspirated using a 23 G needle. FNAC is a fairly simple, rapid, and repeatable technique.[7,11] Being minimally invasive, it leaves no scar and there is no risk of seeding of the tumor along the needle tract, thus it is safe enough to be performed as a routine outpatient procedure.[9] Cytology is included in the triage of imaging and clinical exam in the management of SGL.[12] FNAC plays a role not only in the confirmation of the origin of the lesion but also in the initial diagnosis of the nature of the lesion before approaching the definite management i.e. radical surgery for malignant lesions versus a conservative approach towards the removal of only the benign mass.[13,14]

Analysis of Layfield et al on the cost efficiency of FNAC in the diagnostic workup of SGL concluded at FNAC reduced the number of unnecessary surgical procedures and overall morbidity along with a 33% reduction in the financial burden.[15,16] The trend of using FNAC in the diagnostic workup of SGL is due to the high accuracy (approaching 100%) of FNAC in differentiating Inflammatory from neoplastic lesions.[5,14] FNAC can accurately distinguish between benign and malignant lesions in about 81 – 100% cases but the diagnostic precision to subtype neoplasms varies between 48 – 94%.[17,18]

This study aims to evaluate the validity of FNAC as a diagnostic investigation for salivary gland lesions and to discuss the spectrum of salivary gland lesions in our setup.

MATERIALS AND METHODS

This cross-sectional descriptive study was conducted in the department of pathology of Ayub Medical College and Advanced Lab Abbottabad. Data of patients who underwent FNAC from Jan 2018 till June 2020 were collected and analyzed. A total of 82 cases of FNAC of the salivary glands were included via a non-probability consecutive sampling technique, in this study. Informed consent was taken for the procedure before recording the relevant details. Ultrasound of the swelling was done before FNAC. The general contraindications for performing FNA like vascular lesions, bleeding diathesis, or patients on anticoagulant therapy were noted before the procedure. FNAC was done after taking aseptic precautions. Using a 5cc BD disposable syringe and 23 G needle, aspiration of material from the salivary gland lesions was obtained. The aspirate was immediately spread onto clean glass slides. Two slides were fixed in 95% alcohol and stained with Haematoxylin and Eosin (H&E), and four slides were air-dried and fixed with diff quick stain.

Subsequent Histopathology examination was done only on the surgically excised neoplastic lesions. Specimens were brought to the pathology laboratory, fixed in 10% buffered formalin. Specimens were grossly examined for the size of lesion, consistency, and presence of cystic changes or necrosis. Suspicious areas were carefully sampled and processed using an automatic tissue processor. H&E stained slides were prepared from the paraffin tissue blocks for light microscopy.

The diagnosis of non-neoplastic/ inflammatory salivary gland lesions was made according to conventional criteria. Histopathological diagnoses of the lesions were done following the WHO classification criteria of salivary gland lesions. The cytological diagnoses were compared to the Histopathology examination reports.

All patients presenting with clinically palpable salivary gland swelling who underwent FNAC were included in the study. Data was entered and analyzed using statistical software SPSS version 16.0 Quantitative continuous data i.e. age was expressed as mean and SD while Categorical data i.e. gender, categories of salivary gland lesions, etc were expressed as percentages and frequencies. The validity of the test was performed taking histopathology reports of the excised salivary gland specimen as the gold standard. Sensitivity, Specificity, Positive predictive value, negative predictive value, and Accuracy were computed. Post-stratification, chi-square test of significance was used to see any association between benign and malignant SGL and FNAC based diagnoses. A P-value of <0.05 was considered statistically significant. Data is presented in the pie diagram, bar diagrams, and tables.

RESULTS

82 cases of Salivary Gland Lesions were included in this study. The age of the patient ranged from 24 to 73 years with a mean age of 47±13 years. The commonest age group was from the fourth and fifth decade of life for the neoplastic lesions while it was the third and fourth decade for the non-neoplastic lesions.

This horizontal bar chart shows the gender distribution of the cases included in this study. As evident, 64.6% (53/ 82) were Male while 35.4% (29/ 82) were Female patients with a Male Female ratio of 1.8:1. This doughnut diagram is showing the gland-wise distribution of the SGL.

As shown in the above figure, 63.4% (53/ 82) cases included in this study were affecting the parotid gland while the rest were in the sub-mandibular gland 26.83% (22/82) and sub-mental Salivary gland 8.54% (7/82). This pie chart depicts the frequency of different categories of salivary gland lesions.

As shown in this pie diagram, 25.61% (21/82) lesions were Non-neoplastic only on FNAC. 65.85% (54/82) were diagnosed as Benign Tumors while 8.54% (7/82) cases were diagnosed as Malignant Tumors both on FNAC and subsequently on Histopathologic examination.

This table is showing the different types of lesions from each category that are included in this study along with its frequency, percent of individual lesion out of the total lesions from all categories, and percent of each lesion in its category.

As shown in the above table, among the Non-neoplastic category, chronic Sialadenitis was the commonest encountered lesion making up 42.86% (9/21) of the total. Sialadenosis was found 23.81% (5/21) times as well as cystic lesions. 3/5 of the cysts were mucocele while 2/5were inflammatory cysts. Enlarged intra-parotid lymph nodes were studied by FNAC in 9.52% (2/21) cases which showed reactive inflammatory changes. Histopathologic examination was not done in any of these cases.

Histopathology was done on all the neoplastic cases as these lesions were surgically excised.

Among the Benign lesions category, which is making major bulk of our study population, Pleomorphic Adenoma has seen in 96.3% (52/ 82) cases. The other benign lesion that we encountered was Warthin's Tumor. It was seen in 3.7% (2/82) cases.

In the Malignant Neoplasm category, we found Adenoid Cystic Carcinoma to be most prevalent in our settings. It was seen in 42.86% (3/7) cases. Following it in the decreasing order of occurrence was Mucoepidermoid Carcinoma, seen in 28.58% (2/7) cases. Both Acinic Cell Carcinoma and Follicular lymphoma were observed in 14.28% (1/7) both on FNA cytology as well as Histopathology.

This table is showing the frequency of FNAC based diagnoses of the different categories of the neoplastic lesions along with their Histopathology correlation and the false positive as well as the false-negative cases.

As shown in the above table, the histopathology correlation is available in all the 61 neoplastic cases. Pleomorphic Adenoma was diagnosed on FNAC in 52 cases. Histopathology confirmed 51 cases while one case was a false negative diagnosis of Pleomorphic Adenoma on FNAC which was proved to be Adenoid Cystic Carcinoma on subsequent Histopathology.

Three cases of Adenoid Cystic Carcinoma were diagnosed on FNAC where two cases were coherent with the Histopathology examination while one case was false negatively diagnosed on FNAC to be Adenoid Cystic Carcinoma which was a Pleomorphic Adenoma on Histopathology. FNAC based diagnoses of all the other lesions were in perfect coherence with their Histopathology examination.

This Bar chart shows the FNAC based diagnoses of the different categories of the salivary gland lesions.

As evident from the above bar chart, FNAC correctly diagnosed 98.15% (53/ 54) benign cases and 85.7% (6/7) malignant cases. The rate of False Positive as well as False Negative diagnosis on the FNAC was calculated to be 1.64% in diagnosing the Benign and Malignant Lesions of the salivary glands.

As shown in the above table, 53 of the FNAC based negative for malignancy cases were coherent with the Histopathology examination i.e., True Negative cases and so are the 6 positive for malignancy cases confirmed by histopathology i.e., True Positive Cases. One benign case was false positively diagnosed to be malignant and also one malignant

case was false negatively diagnosed to be benign on the FNAC. The Validity Parameters for the ability of FNAC to diagnose benign from malignant SGL was calculated from the above 2×2 contingency table using the following formulae:

Accuracy = (TP+ TN) × 100/(TP + TN + FP + FN). Sensitivity = (TP × 100) / (TP + FN).

Specificity = (TN × 100) / (TN + FP).

Positive predictive value (PPV) = (TP × 100) / (TP + FP).

Negative predictive value (NPV) = (TN× 100) / (TN + FN).

The calculated values for the Validity parameters are given in the table below.

Out of the 61 confirmed cases on histopathology, 6 cases were positive for malignancy on FNAC i.e., TP yielding of the sensitivity of 85.7% of FNAC while 1 case was FN. Of the 54 confirmed negative for malignancy cases on histopathology, 53 cases were True Negative on FNAC yielding specificity of 98.15% of FNAC. Of the 7 positive cases on FNAC, 6 were true positive that revealed positive predictive value 85.7% of FNAC in the diagnosis of Malignancy. Of the 54 cases that were negative on FNAC for Malignancy, 53 cases were true negative that revealed a negative predictive value of 98.15% for FNAC. The overall accuracy of FNAC in the diagnosis of Malignancy from benign lesions was 96.72%.

Chi-square test of association was applied to see any correlation between FNAC ability to diagnose malignant from benign lesions as compared to the gold standard which is Histopathology examination and calculate a p-value of 0.001. This p=0.001 shows a strong association between FNAC based diagnoses of the benign or neoplastic lesion and the actual presence of such a lesion.

Table: 1 Diagnostic Categories & Types of Salivary Gland Lesions

Diagnostic category	Lesion	F	Percent in its category	%age of Total
Non-neoplastic/ Inflammatory Lesions 21/ 82 (25.61%)	Chronic Sialadenitis	9	42.86	11.0
	Intraparotid Lymph Node	2	9.52	2.4
	Sialadenosis	5	23.81	6.1
	Benign Cysts	5	23.81	6.1
Benign Lesions 54/ 82 (65.85%)	Pleomorphic Adenoma	52	96.3	63.4
	Warthin's Tumor	2	3.7	2.4
Malignant Lesions 07/ 82 (8.54%)	Adenoid Cystic Carcinoma	3	42.86	3.7
	Mucoepidermoid Carcinoma	2	28.58	2.4
	Acinic Cell Carcinoma	1	14.28	1.2
	Follicular lymphoma	1	14.28	1.2
	Total	82	-	100.0

Table: 2 FNAC Based Diagnoses with Histopathology Correlation

Type of Lesion	FNAC Based Diagnoses	Histopathology Consistent with FNAC	Histopathology Not consistent with FNAC	False-positive	False-negative
Pleomorphic Adenoma	52	51	01	1 (Adenoid cystic Ca)	-
Warthin's Tumor	02	02	-	-	-
Mucoepidermoid Ca	02	02	-	-	-
Acinic Cell Ca	01	01	-	-	-
Adenoid Cystic Ca	03	02	01	-	1 (Pleomorphic adenoma)
Follicular lymphoma	01	01	-	-	-
Total	61	59	02	1	1

Table: 3 2×2 Comparison Table of FNAC with Histopathology

FNAC diagnosed cases	Histopathology reported confirmed cases			
		Benign	Malignant	Total
	Benign	TN= 53	FN=1	54
	Malignant	FP=1	TP=6	7
	Total	54	7	61

TN: True Negative FN: False Negative

FP: False Positive TP: True Positive

Table: 4 Validity Parameters for FNAC

Sensitivity	85.7%
Specificity	98.15%
PPV	85.7%
NPV	98.15%
Accuracy	96.72%

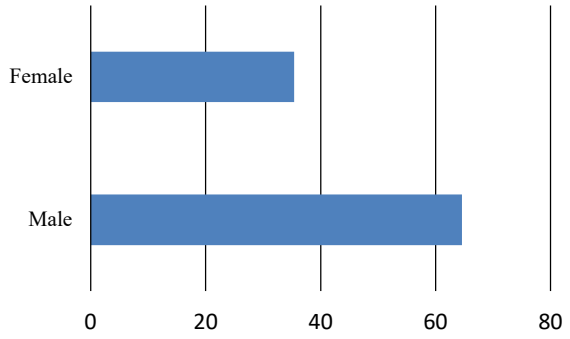

Fig 1: Distribution of Gender

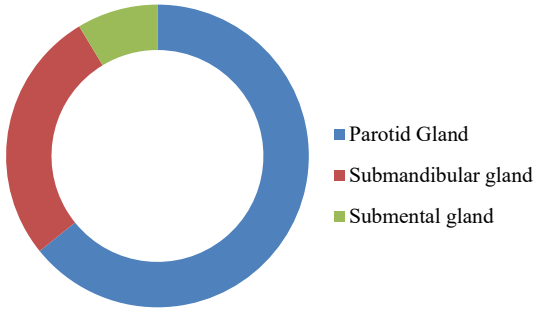

Fig 2: Gland-Wise Distribution of Salivary Gland Lesions

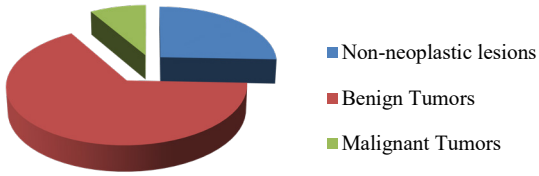

Fig 3: Frequency of Salivary Gland Lesions

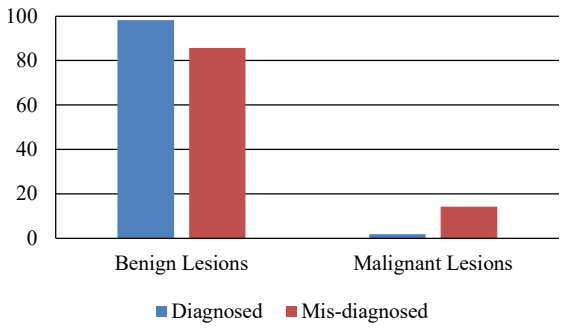

Fig 4: FNAC Based Diagnoses of Neoplastic Lesions

DISCUSSION

A total of 82 cases of SGL were included in this study. The patient's age ranged from 24 to 73 years. The mean age at presentation for FNAC was calculated to be 47±13 years. The commonest age group was from the fourth and fifth decade of life for the neoplastic lesions while it was the third and fourth decade for the non-neoplastic lesions. The mean age at presentation with neoplastic SGL has been reported to be the fifth decade.[5,6,9,14]

The male gender has been reported to be more often affected by the SGL than the female gender.[6,11] In this study, 64.6% (53/ 82) of the patient were Male while 35.4% (29/ 82) were Female patients with a Male to Female ratio of 1.8:1. Similar results for gender distribution have been published by Arul P et al and Gupta R et al, both from India.[9,19]

The majority of the cases of SGL have been reported to occur in the Parotid Gland. The occurrence of SGL in the parotid gland has been reported from

41.9% by Gupta R et al to 76.2% by Ameli F et al.[5,19] In this study, SGL was present in the parotid gland in 63.4% (53/ 82) cases while the rest were in the submandibular gland 26.83% (22/82) and submental Salivary gland 8.54% (7/82). Distribution of SGL in the different glands in our setting was found to be the same as those reported from Puducherry, Ambala, and Punjab, the three different regions of India.[4,9,11] This similarity seems to be due to the same geographical region and living standards of the people of the two neighboring countries.

Salivary gland masses have a variety of causes including Non-neoplastic/ inflammatory conditions and neoplastic lesions including benign and malignant tumors. In our study, Non-neoplastic lesions were encountered in 25.61% of cases whereas the major bulk of the study population was diagnosed to have benign lesions (65.85%). Only 8.54% of cases were diagnosed with malignant lesions. A similar distribution pattern for the different categories of SGL has been reported by Ameli F et al from Malaysia, Kakoty S et al and Shelly S, both from India.[5,9,11]

Among the Non-neoplastic/Inflammatory lesions, chronic Sialadenitis was the commonest lesion being diagnosed in 11% cases, followed by sialadenosis and cysts diagnosed in 6.1% cases. By far, the most common non-neoplastic lesion affecting the Salivary glands was reported to be chronic sialadenitis.[7,14,20] The ability of FNAC to diagnose Non-neoplastic SGL is approaching 100%.[10,17] Therefore, the non-neoplastic lesions were not excised in any of the cases.

Clinically salivary gland tumors present as a slowly growing mass which is usually asymptomatic except in the case of adenoid cystic carcinoma due to its perineural invasive propensity or any other tumor that causes entrapment of the facial nerve.[5]

The category of benign lesions is making the bulk of this study population but only two types of benign lesions have been encountered. Pleomorphic Adenoma being the most prevalent of all SGL lesions was diagnosed in 63.4% of patients. Pleomorphic adenoma is a biphasic tumor cytologically revealing a combination of bland epithelial cells forming sheets and groups along with fragments of fibromyxoid stroma.[5] The diagnostic problem arises when the stromal component is less as in cellular PA. Also, the pleomorphic adenoma may contain areas resembling adenoid cystic carcinoma, giving a false-positive result for malignancy. Adenoid cystic carcinoma is famous for neural invasion so it is often associated with severe pain a feature that guides in diagnosis.[17] Warthin's Tumor (also called papillary cystadenoma lymphomatosum) was the other benign lesion diagnosed in 2.4% of patients. It is believed to be common in smokers and male patients. The cytological smears showed aggregates of oncocytic cells along with lymphocytes. If the lymphoid element is not sampled it can be wrongly diagnosed as oncocytoma.[18]

Mairemban P from London and Tess PJ from India also reported PA to be the most common benign lesions in their study populations.[6,1] A total of 52 cases were diagnosed for PA on FNAC. Subsequent Histopathology examination confirmed 51 cases. The one case which was false negatively diagnosed as PA on FNAC was confirmed to be Adenoid Cystic Carcinoma. Both cases of Warthin's tumor were confirmed by histopathology to have been accurately diagnosed by FNAC.

The malignant cases in this study made up 8.54% of the total cases. This was below the average expected rate of the malignant lesions in any population i.e., 15-30%.[5] This might be due to better access to people of this setting to tertiary care hospitals as there are four teaching hospitals in this location. Prompt diagnosis and excision of benign lesions can substantially reduce the risk of malignant transformation, which is a common phenomenon in most of the salivary gland neoplasm i.e., PA to carcinoma ex pleomorphic adenoma, etc.[5] Adenoid cystic carcinoma was the most prevalent malignant neoplasm in our settings making up 3.7% of the cases followed by Mucoepidermid carcinoma which was diagnosed in 2.4% cases. Acinic Cell Carcinoma and Follicular Lymphoma were diagnosed each in 1.2% cases. Similar results for the prevalence of the malignant neoplasms have been documented by Arul p et al, Gupta R et al and Sandhu VK et al.[4,19,20] All the FNAC diagnoses were in coherence with the Histopathology examination except one case of Adenoid Cystic Carcinoma which was false positively diagnosed by FNAC that turned out to be pleomorphic adenoma on histopathology examination. The distinction between pleomorphic adenoma and adenoid cystic carcinoma on FNAC may be difficult on account of several features-myxoid, acellular material may be found in both

and hyaline globules characteristic of adenoid cystic carcinoma may also be seen in pleomorphic adenoma.[17,18] Cystic change occurring in salivary gland tumors can cause diagnostic confusion as it could be due to the cystic nature of the tumor or necrosis.[18] A cytologic diagnosis of mucoepidermoid carcinoma requires a background of mucous and debris and a variable population of cells.[13,17]

Out of the 61 FNAC cases which were followed by Histopathology examination, 59 were confirmed to be correctly diagnosed by FNAC. Thus, the concordance between FNAC and Histopathology in this study was calculated to be 96.72%. Rossi ED et al from Italy, Rome, in their retrospective analysis of 1729 salivary gland FNAC, also reported the concordance between FNAC and Histopathology to be 92.2%.[16] In this study, one case of adenoid cystic carcinoma was false negatively diagnosed to be PA, giving the percentage of false-negative diagnosis of 1.64%. Also, one case of PA was false positively diagnosed on FNAC to be adenoid cystic carcinoma, giving the percentage of false-positive diagnosis of 1.64%. Rohilla M et al, in their retrospective evaluation of 631 salivary gland aspirates, reported the percentage of FP and FN diagnoses by FNA of 1.1% and 7.4% respectively.[21]

A study addressing the diagnostic problems in the SGL by FNAC reasoned that it is due to the marvelous diversity and cytomorphological overlap that exists between the SGL. A[13] Inadequate sampling and low cellularity of the FNA are other common causes that at times make a diagnosis on FNAC quite challenging even for an experienced cytopathologist.[2,18,21] These problems can be countered by proper lesion sampling, adequate cellularity of the smear, use of both air-dried and 95% alcohol fixed slides, and an experienced cytopathologist.[4,9] A study indicated that the more experienced and skilled the cytopathologist, the greater will be the diagnostic accuracy of FNAC.[5,13]

Wei S et al, in their review of 29 articles concluded that FNAC has sensitivity and specificity of 95% and 93% respectively to rule out the presence of benign neoplasm and sensitivity and specificity of 87% and 85% respectively to detect malignancy. FNAC can predict the presence of a tumor with more than 90% accuracy.[10]

This table given below shows the comparison of the results of this study with others around the globe.

No	Year		Author	Sensitivity	Specificity	Accuracy	PPV	NPV
1	2015	Malaysia	Ameli F	80	98.8	95.87	92.3	96.4
2	2015	India	Arul P	86.6	94.6	93.3	88.3	94.6
3	2015	Bangladesh / Pakistan	Naz S	77.7	86.3	83.3	70	90.4
4	2016	Rome	Rossi ED	69.1	98.9	92.2	91.8	94.1
5	2017	India	Kakoty S	90.91	96.42	94.87	90.91	96.42
6	2017	India	Rohilla M	79.4	98.3	91.4	96.4	89.2
7	2018	India	Shelly S	89.5	100	85	100	50.0
This study	2020	Pakistan	Naz S	85.7%	98.15%	96.72%	85.7%	98.15%

As evident from the above table, our results are comparable to other studies conducted by different researchers in the world.

CONCLUSION

FNAC is an accurate and effective modality in terms of sensitivity, specificity, and accuracy for diagnosing the SGL. Owing to its safety, affordability, and quick results, it should be considered as an investigation of the first choice for SGL before devising a definitive management plan. On the other hand, due to the overlap between the cytological features of certain salivary gland tumors, tissue biopsy for histopathology examination is the gold standard.

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