Section: Pathology

Original Research Article

CORRELATION OF CYTOMORPHOLOGICAL PATTERNS WITH HISTOPATHOLOGICAL SUBTYPES IN SALIVARY GLAND NEOPLASMS

Amandeep Kaur[1], Aakanksha Paniya[2], Mansi Khamesra[3]

[1,3]Assistant Professor, Department of Pathology, Pacific Medical College and Hospital, Udaipur, Rajasthan, India.
[2]Assistant Professor, Department of Pathology, Geetanjali Medical College and Hospital, Udaipur, Rajasthan, India.

Received : 07/06/2025
Received in revised form : 12/08/2025
Accepted : 31/08/2025

Keywords:
Salivary Gland Neoplasms, Fine Needle Aspiration Cytology, Histopathology, Diagnostic Accuracy, Cytomorphology, Milan System.

Corresponding Author:
Dr. Mansi Khamesra,
Email: mansikhamesra12@gmail.com

DOI: 10.47009/jamp.2025.7.5.9

Source of Support: Nil,
Conflict of Interest: None declared

Int J Acad Med Pharm
2025; 7 (5); 38-42

ABSTRACT

Background: Salivary gland neoplasms encompass a wide spectrum of benign and malignant tumors with significant morphological diversity. Fine Needle Aspiration Cytology (FNAC) is a crucial first-line investigation for the pre-operative assessment of salivary gland lesions, guiding patient management and surgical planning. However, its diagnostic accuracy can be challenged by overlapping cytomorphological features among different tumor subtypes. **Materials and Methods:** A retrospective study was conducted on 150 patients who underwent FNAC followed by surgical excision of salivary gland tumors. Cytology slides were reviewed and categorized based on established morphological criteria. These cytological diagnoses were then compared with the definitive histopathological reports, which served as the gold standard. Statistical analysis was performed using the Chi-square test to determine the correlation, and diagnostic accuracy metrics were calculated. **Result:** The study included 150 cases with a mean age of 48.5 ± 15.2 years and a female-to-male ratio of 1.14:1. The parotid gland was the most common site (82.0%). Histopathology confirmed 115 (76.7%) benign and 35 (23.3%) malignant neoplasms. The overall diagnostic accuracy of FNAC was 92.7%. For distinguishing benign from malignant lesions, FNAC demonstrated a sensitivity of 82.9%, a specificity of 95.7%, a positive predictive value (PPV) of 85.3%, and a negative predictive value (NPV) of 94.8%. The correlation between cytological and histopathological diagnoses was statistically significant (χ^2 = 225.4, $p < 0.001$). The most common discordant diagnoses occurred within basaloid neoplasms and cystic lesions, where pleomorphic adenoma was occasionally misdiagnosed as basal cell adenoma, and low-grade mucoepidermoid carcinoma was misinterpreted as a benign cystic lesion. **Conclusion:** FNAC is a highly reliable and accurate diagnostic tool for the initial evaluation of salivary gland neoplasms. It provides crucial pre-operative information with high specificity and NPV. Nevertheless, diagnostic pitfalls exist, particularly in tumors with overlapping basaloid or cystic features, underscoring the indispensable role of histopathology for definitive classification and grading.

INTRODUCTION

Salivary gland neoplasms are a heterogeneous group of tumors characterized by complex clinicopathological and morphological features. They account for approximately 3–6% of all head and neck tumors, with the majority arising in the parotid gland.[1] The histological diversity of these neoplasms, ranging from common benign entities like pleomorphic adenoma (PA) to a wide array of malignant carcinomas, presents a significant diagnostic challenge.[2] Accurate pre-operative diagnosis is paramount as it directly influences the extent of surgery, the need for neck dissection, and potential adjuvant therapies.[3]

Fine Needle Aspiration Cytology (FNAC) has long been established as the primary diagnostic modality for palpable salivary gland masses. It is a minimally invasive, cost-effective, and safe procedure that provides valuable information to clinicians, helping to differentiate neoplastic from non-neoplastic processes and, crucially, benign from malignant tumors.[4,5] The information gleaned from FNAC allows for appropriate patient counseling and surgical planning, potentially avoiding unnecessary surgery

ISSN (O): 2687-5365; ISSN (P): 2753-6556

for non-neoplastic conditions or enabling more radical procedures when malignancy is suspected.[6]

Despite its widespread use, the diagnostic utility of FNAC in salivary gland pathology is not without limitations. The cytological interpretation can be confounded by significant morphological overlap between various tumor types. For instance, the spectrum of basaloid neoplasms, which includes benign entities like basal cell adenoma and malignant ones like adenoid cystic carcinoma, often shares similar cytological features, making a definitive distinction challenging on smears alone.[7] Similarly, cystic changes, which can be seen in benign lesions like Warthin's tumor and malignant tumors like low-grade mucoepidermoid carcinoma, can lead to paucicellular aspirates and diagnostic ambiguity.[8]

To standardize reporting and improve communication between cytopathologists and clinicians, the Milan System for Reporting Salivary Gland Cytopathology (MSRSGC) was introduced. This system stratifies diagnoses into distinct categories, each with an associated risk of malignancy, thereby guiding clinical management [9, 10]. Recent studies utilizing the MSRSGC have reaffirmed the high overall accuracy of FNAC but have also continued to highlight specific areas of diagnostic difficulty.[11] Continuous evaluation of institutional FNAC performance against the gold standard of histopathology is essential for quality assurance and for identifying recurring diagnostic pitfalls. While numerous studies have addressed this correlation, there is a persistent need to analyze these patterns within specific patient populations and institutional practices to refine diagnostic criteria and enhance pathologist expertise.

This study was therefore undertaken to systematically correlate the cytomorphological patterns observed on FNAC with the final histopathological subtypes of salivary gland neoplasms at our institution. The primary aim was to evaluate the overall diagnostic accuracy, sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) of FNAC in this context. A secondary aim was to identify and analyze the specific patterns of concordant and discordant cases to better understand common diagnostic challenges.

MATERIALS AND METHODS

Study Design and Population: This was a single-center, retrospective, cross-sectional study conducted at the Department of Pathology. We reviewed the archival records from January 1, 2018, to December 31, 2022. All patients who presented with a salivary gland swelling, underwent FNAC, and subsequently had surgical excision with a definitive histopathological diagnosis were included.

Inclusion and Exclusion Criteria: Inclusion criteria were: (1) all cases with a definitive cytological diagnosis of a salivary gland neoplasm, and (2) corresponding surgical resection specimen available for histopathological review. Exclusion criteria were: (1) FNAC smears reported as inadequate for evaluation (Category I of the MSRSGC), (2) cases diagnosed as non-neoplastic or inflammatory lesions, (3) cases without subsequent histopathological follow-up, and (4) cases with incomplete clinical or pathological data.

Sample Size: A total of 188 cases with both FNAC and histopathology were initially screened. After applying the exclusion criteria, 150 cases were found suitable for inclusion in the study.

Procedure

- **FNAC Procedure and Smear Preparation:** FNAC was performed by trained pathologists using a 22- or 23-gauge needle attached to a 10 mL syringe, often with a syringe holder. Multiple passes were made to ensure adequate sampling. The aspirated material was expelled onto clean glass slides. Both air-dried smears stained with May-Grünwald-Giemsa (MGG) and wet-fixed (95% ethanol) smears stained with Papanicolaou (Pap) stain were prepared for each case.
- **Cytomorphological Evaluation:** All FNAC slides were retrieved and retrospectively reviewed by two independent pathologists who were blinded to the original reports and final histopathological diagnoses. A consensus was reached in cases of disagreement. The cytological features evaluated included overall cellularity, architectural patterns (e.g., acinar, sheets, papillary, cribriform), cell morphology (e.g., basaloid, oncocytic, plasmacytoid, squamoid, clear), stromal components (e.g., chondromyxoid, fibrillary, hyaline globules), and background elements (e.g., mucin, colloid, lymphoid infiltrate). Diagnoses were categorized into specific neoplastic entities where possible.
- **Histopathological Evaluation:** The corresponding surgically excised specimens had been fixed in 10% neutral buffered formalin, processed routinely, embedded in paraffin, and sectioned at 4-5 μm thickness. Sections were stained with Hematoxylin and Eosin (H&E). The histopathological diagnoses were considered the gold standard for this study and were based on the criteria outlined in the 2017 World Health Organization (WHO) Classification of Head and Neck Tumours.

Statistical Analysis: Data were entered and analyzed using SPSS Statistics for Windows, Version 25.0 (IBM Corp., Armonk, NY). Descriptive statistics, including mean, standard deviation (SD), frequencies, and percentages, were used to summarize demographic and clinicopathological data. The correlation between the cytological diagnosis and the final histopathological diagnosis was assessed using the Chi-square (χ^2) test. Diagnostic accuracy, sensitivity, specificity, PPV, and NPV were calculated for the differentiation of

ISSN (O): 2687-5365; ISSN (P): 2753-6556

benign and malignant lesions using standard formulas. A p-value of < 0.05 was considered statistically significant.

RESULTS

Patient Demographics and Tumor Characteristics: A total of 150 cases met the inclusion criteria. The age of the patients ranged from 16 to 78 years, with a mean age of 48.5 ± 15.2 years. There was a slight female preponderance, with 80 (53.3%) females and 70 (46.7%) males (ratio 1.14:1). The most commonly affected salivary gland was the parotid gland (123 cases, 82.0%), followed by the submandibular gland (22 cases, 14.7%) and minor salivary glands (5 cases, 3.3%).

Histopathological Findings: out of the 150 neoplasms, 115 (76.7%) were benign and 35 (23.3%) were malignant on final histopathology. Pleomorphic adenoma was the most frequent benign tumor (n=75, 50.0% of total cases), while mucoepidermoid carcinoma was the most common malignancy (n=14, 9.3% of total cases). The distribution of all histologically confirmed neoplasms is detailed in Table 1.

Table 1: Distribution of Salivary Gland Neoplasms on Histopathology (n=150)

Histopathological Diagnosis	Frequency (n)	Percentage (%)
Benign Neoplasms (n=115)		
Pleomorphic Adenoma	75	50.0
Warthin's Tumor	28	18.7
Basal Cell Adenoma	8	5.3
Oncocytoma	4	2.7
Malignant Neoplasms (n=35)		
Mucoepidermoid Carcinoma	14	9.3
Adenoid Cystic Carcinoma	9	6.0
Acinic Cell Carcinoma	5	3.3
Carcinoma ex Pleomorphic Adenoma	3	2.0
Salivary Duct Carcinoma	2	1.3
Adenocarcinoma, NOS	2	1.3
Total	**150**	**100.0**

Correlation between Cytology and Histopathology: The overall cyto-histological concordance rate for specific tumor types was 86.0% (129/150 cases). FNAC correctly identified 110 of 115 benign lesions and 29 of 35 malignant lesions. A strong statistically significant correlation was found between the cytological and histopathological diagnoses when categorized as benign or malignant ($\chi^2 = 225.4$, $p < 0.001$). The detailed correlation, including concordant and discordant cases, is presented in Table 2.

Table 2: Correlation Matrix of Cytological and Histopathological Diagnoses

Cytological Diagnosis	Pleomorphic Adenoma (H)	Warthin's Tumor (H)	Other Benign (H)	Mucoepidermoid Ca (H)	Adenoid Cystic Ca (H)	Other Malignant (H)	Total
Pleomorphic Adenoma (C)	**70**	0	2 (BCA)	1 (LG)	2 (ACC)	1 (Ca ex PA)	76
Warthin's Tumor (C)	0	**27**	1 (Oncocytoma)	1 (LG)	0	0	29
Other Benign (C)	3 (PA)	1 (WT)	**9** (BCA, Onco)	0	0	0	13
Mucoepidermoid Carcinoma (C)	0	0	0	**11**	0	0	11
Adenoid Cystic Carcinoma (C)	2 (PA)	0	0	0	**7**	0	9
Other Malignant (C)	0	0	0	1 (MEC)	0	**11**	12
Total	**75**	**28**	**12**	**14**	**9**	**12**	**150**

Discordant cases included 5 false negatives (malignant tumors misdiagnosed as benign) and 5 false positives (benign tumors misdiagnosed as malignant). The most notable false negatives were two cases of low-grade mucoepidermoid carcinoma diagnosed as Warthin's tumor and a benign cyst, respectively, due to prominent cystic change and scant cellularity. False positives primarily involved basaloid neoplasms, where two cases of pleomorphic adenoma with high cellularity were misdiagnosed as adenoid cystic carcinoma.

Diagnostic Accuracy of FNAC: The performance of FNAC in differentiating benign from malignant neoplasms was evaluated. The overall diagnostic accuracy was 92.7%. The detailed metrics are shown in Table 3.

ISSN (O): 2687-5365; ISSN (P): 2753-6556

Table 3: Diagnostic Performance of FNAC for Malignancy (n=150)

Parameter	Value (%)	95% Confidence Interval
Sensitivity	82.9%	67.4% – 93.4%
Specificity	95.7%	90.3% – 98.6%
Positive Predictive Value (PPV)	85.3%	69.9% – 94.7%
Negative Predictive Value (NPV)	94.8%	89.2% – 98.0%
Diagnostic Accuracy	92.7%	87.2% – 96.3%

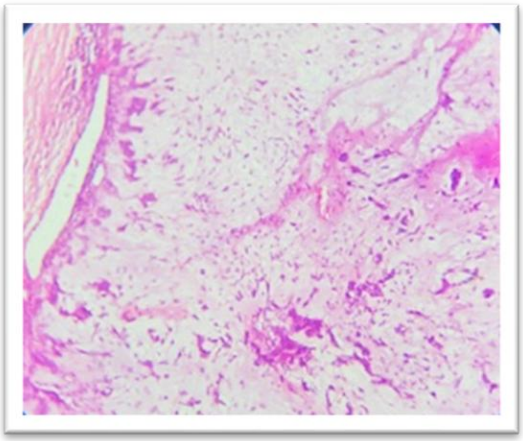

Figure1 a– Photomicrograph of Pleomorphic adenoma showing epithelial and stromal components. (H and E stain, 100X)

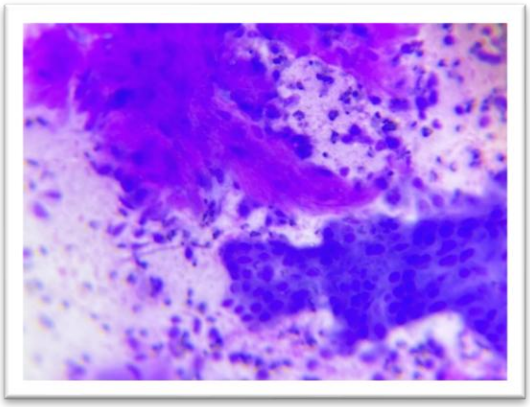

Figure1 b – Photomicrograph of Pleomorphic adenoma showing ductal cells, myoepithelial cells and extra cellular matrix. (MGG stain, 400X)

DISCUSSION

The pre-operative diagnosis of salivary gland neoplasms is critical for appropriate surgical management, and FNAC serves as the cornerstone of this initial assessment. This study confirms the high utility of FNAC, demonstrating an overall diagnostic accuracy of 92.7% in distinguishing benign from malignant lesions, which is consistent with the range of 85–96% reported in extensive reviews and meta-analyses.[8,12] Our calculated sensitivity of 82.9% and specificity of 95.7% also align with published data, underscoring the procedure's reliability in a routine clinical setting.[4,13]

The strength of FNAC lies in its high specificity and high negative predictive value (NPV). In our cohort, the specificity was 95.7% and the NPV was 94.8%. This implies that a benign diagnosis on FNAC is highly reliable, allowing clinicians to confidently plan for more conservative surgical approaches, such as superficial parotidectomy or simple enucleation, thereby minimizing patient morbidity.[5] The most common benign neoplasms, pleomorphic adenoma and Warthin's tumor, were correctly identified in a high proportion of cases (93.3% and 96.4%, respectively). The classic cytomorphological features—a combination of epithelial/myoepithelial cells in a chondromyxoid stroma for PA, and a dual population of oncocytes and lymphocytes for Warthin's tumor—are sufficiently distinct to allow for an accurate diagnosis.[10,14]

Despite its high accuracy, this study highlights several well-documented diagnostic pitfalls that challenge cytopathologists. The primary source of diagnostic error in our series, as in others, was the significant morphologic overlap among certain tumor types.[7] The most problematic group was basaloid neoplasms. We observed two cases of pleomorphic adenoma misdiagnosed as adenoid cystic carcinoma (ACC) and two cases of ACC misdiagnosed as PA. This confusion arises because both tumors can present with cohesive clusters of small, uniform basaloid cells. The key distinguishing feature of ACC is the presence of acellular hyaline globules, but these can be mimicked by the myxoid stroma of PA, particularly in cellular variants of PA. Conversely, some ACCs may lack the classic cribriform pattern on cytology.[13] This overlap underscores the importance of careful examination for subtle clues and often necessitates a diagnosis of "Neoplasm of Uncertain Malignant Potential" (SUMP) or "Suspicious for Malignancy" as per the MSRSGC guidelines.[9]

Another significant challenge is posed by cystic salivary gland lesions. In our study, two cases of low-grade mucoepidermoid carcinoma (MEC) were misdiagnosed as benign (one as a Warthin's tumor and another as a simple cyst). Cystic degeneration is common in low-grade MEC, and aspiration may yield only watery fluid with scant cellularity, composed predominantly of muciphages and a few bland epithelial cells, leading to a false-negative diagnosis.[11,15] The presence of any atypical or mucin-producing cells in a cystic aspirate should raise suspicion for MEC and prompt a recommendation for complete excision.

The limitations of this study include its retrospective nature, which may introduce selection bias, as only patients who underwent surgery were included. This inherently excludes cases managed non-surgically and cases with inadequate aspirates, which may artificially inflate the reported diagnostic accuracy.

ISSN (O): 2687-5365; ISSN (P): 2753-6556

Furthermore, as a single-institution study, the findings may not be generalizable to all practice settings. Ancillary techniques such as immunocytochemistry or molecular analysis were not routinely performed on the cytology specimens in this cohort but have been shown in other studies to be valuable in resolving diagnostically ambiguous cases, particularly in distinguishing between basaloid tumors or confirming the myoepithelial differentiation in PA.[7]

CONCLUSION

This study validates that Fine Needle Aspiration Cytology is a highly accurate and reliable tool for the pre-operative evaluation of salivary gland neoplasms, with high specificity and negative predictive value for ruling out malignancy. It effectively guides clinical decision-making and helps tailor surgical approaches. However, practitioners must remain vigilant of inherent diagnostic limitations, especially in the interpretation of basaloid and cystic neoplasms, where cytomorphological features can overlap significantly between benign and malignant entities. A multidisciplinary approach, combining clinical findings, radiological imaging, and cytological interpretation, is essential for optimal patient care. Histopathological examination of the excised specimen remains the undisputed gold standard for definitive diagnosis, classification, and grading of these complex tumors.

REFERENCES

1. Speight PM, Barrett AW. Salivary gland tumours. Oral Dis. 2002;8(5):229-40. DOI: 10.1034/j.1601-0825.2002.02870.x.
2. Ellis GL, Auclair PL. Tumors of the Salivary Glands. AFIP Atlas of Tumor Pathology, 4th series, fascicle 9. Washington, DC: American Registry of Pathology; 2008.
3. Guzzo M, Locati LD, Prott FJ, Gatta G, McGurk M, Licitra L. Major and minor salivary gland tumors. Crit Rev Oncol Hematol. 2010;74(2):134-48. DOI: 10.1016/j.critrevonc.2009.10.004.
4. Schmidt RL, Hall BJ, Wilson AR, Layfield LJ. A systematic review and meta-analysis of the diagnostic accuracy of fine-needle aspiration for salivary gland lesions. Am J Clin Pathol. 2011;136(1):45-59. DOI: 10.1309/AJCPRT8Y31MASGDF.
5. Al-Abbadi MA, Al-awad Sama. Salivary Gland FNAs: A Journey of Challenges, Rewards, and Checkpoints. A Personal Review. Acta Cytol. 2017;61(4-5):333-344. DOI: 10.1159/000479007.
6. Stow N, Veale D, Titus J, diaspora C, mcgurk M. The role of fine needle aspiration cytology in the management of parotid tumours. Ann R Coll Surg Engl. 2009;91(7):577-81. DOI: 10.1308/003588409X464732.
7. Griffith CC, Schmitt AC, Little JA, Magliocca KR. The Milan System for Reporting Salivary Gland Cytopathology: A 4-year institutional experience with a focus on risk stratification of the Salivary Gland Neoplasm of Uncertain Malignant Potential (SUMP) category. Cancer Cytopathol. 2019;127(11):707-716. DOI: 10.1002/cncy.22177.
8. Krane JF, Faquin WC. The Milan System for Reporting Salivary Gland Cytopathology. Cham, Switzerland: Springer; 2018.
9. Faquin WC, Rossi ED, Baloch Z, Barkan GA, Foschini MP, Kurtycz DF, et al. The Milan System for Reporting Salivary Gland Cytopathology. J Am Soc Cytopathol. 2016;5(3):185. DOI: 10.1016/j.jasc.2016.05.003.
10. Rossi ED, Faquin WC, Baloch Z, Barkan GA, Foschini MP, Pusztaszeri M, et al. The Milan System for Reporting Salivary Gland Cytopathology: Analysis and suggestions of initial survey. Cancer Cytopathol. 2017;125(11):757-766. DOI: 10.1002/cncy.21911.
11. Layfield LJ, Gopez EV. Cystic lesions of the salivary glands: cytologic and clinical features. Diagn Cytopathol. 2002;27(4):197-204. DOI: 10.1002/dc.10163.
12. Wei S, LiVolsi VA, Montone KT, Morrissette JJD, Baloch ZW. The diagnostic utility of fine-needle aspiration in the evaluation of salivary gland lesions. Cancer Cytopathol. 2013;121(11):643-51. DOI: 10.1002/cncy.21323.
13. Klijanienko J, Vielh P. Fine-needle sampling of salivary gland lesions. I. Cytology and histology correlation of 412 cases of pleomorphic adenoma. Diagn Cytopathol. 1996;14(3):195-200. PMID: 8724128.
14. Rohilla M, Singh P, Rajwanshi A, Gupta N, Srinivasan R, Gautam U. Three-year cytohistological correlation of salivary gland neoplasms with the application of the Milan system. Cytopathology. 2019;30(1):65-71. DOI: 10.1111/cyt.12643.
15. Bongiovanni M, Di LMA, Taffon C, Fadda G. The role of immunocytochemistry in salivary gland fine-needle aspiration. Cytopathology. 2012;23(5):287-95. DOI: 10.1111/j.1365-2303.2012.00998.x.

ISSN (O): 2687-5365; ISSN (P): 2753-6556