

PATHOLOGICAL OBSERVATIONAL STUDY OF SALIVARY GLAND NEOPLASMS IN A TERTIARY CARE HOSPITAL WITH SPECIAL REFERENCE TO PERIODIC ACID SCHIFF AND ALCIAN BLUE STAINS

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**Abstract**

Background: Salivary gland tumours show diverse morphological patterns between various tumor types and on rare occasions within the same tumours, thus causing interpretation challenges among the pathologists. **Materials and Methods:** This was a one-year observational study of salivary gland neoplasm's with special references to Periodic Acid Schiff (PAS) and Alcian Blue (AB) stain. Thirty-two cases were diagnosed with salivary gland neoplasm. This prospective observational study was done in Department of Pathology in a Tertiary care Hospital. A total of 32 patients were included in which 22 of them were male and rest 10 were female. **Result:** Total of 32 were diagnosed with salivary gland tumor in out of 23 were cases of Benign tumors and remaining 9 were cases of Malignant tumors. Parotid gland 19 (59.37%) was the commonest site involved followed by submandibular gland 9(28.12%) minor salivary glands 4 (12.50%). Thus, submandibular gland minor salivary gland was more likely to be involved than the minor salivary gland in our study. **Discussion:** Parotid was commonest site was seen in present study and correlated with other studies. Parotid gland is the most frequently involved gland by salivary gland tumors. Most commonly presented in middle to older age group. Second most common site in our study is Sub mandibular followed by minor salivary gland? **Conclusion:** Salivary gland tumours' both benign and malignant are more frequent in males. A detailed clinical, histopathological and histochemical analysis in salivary gland tumors will provide a better insight to the pathophysiology of the disease, tumor differentiation and prognostic implications. Thus, emphasizing on a more 'pleomorphic' approach to PAs.

INTRODUCTION

Salivary gland tumors show diverse morphological patterns between various tumor types and on rare occasions within the same tumor, thus causing interpretation challenges among the pathologists.^[1] Studies from different countries have shown geographic variations in the relative incidence and clinico- pathological discrepancies between benign and malignant salivary gland tumours.^[2] The annual incidence worldwideranges from 0.4 to 13.5 cases per 1,00,000 people. Majority of salivary gland tumors show a female preponderance.^[3] Although the aetiology of salivary gland tumors is unknown, literature reports certain risk factors implicated in causation of these tumors. Among the viruses, Epstein-Barr virus, polyoma virus, cytomegalovirus, human immunodeficiency virus and human papilloma virus types 16, 18 are

likely to cause salivary gland neoplasms. Patients exposed to head and neck radiation for treatment showed increased incidence. Certain occupations such as asbestos mining, manufacturing of rubber products and plumbing (exposure to metals) and working in the automobile are other risk factors.^[4,5] Clinically, salivary gland tumours present as a slow growing mass, however, rapid growth, pain, nerve involvement and fixation to skin or underlying muscles indicate malignancy.^[7] Benign tumours commonly occur in the age group of 30-70years whereas peak incidence of malignant tumors is in the 6th to 7th decades. Malignant tumors are more frequent in women than men.^[8] Among the pediatric age group, 35% of salivary gland tumors are considered malignant.^[2] Majority of Epithelial tumours are benign (75%) among which pleomorphic adenoma is the most common tumor and constitutes about 65% of all salivary gland

neoplasms. Among the malignant neoplasms, mucoepidermoid carcinoma is the commonest. Of the major salivary glands, parotid gland is frequently involved.^[4,5]

The distribution of salivary neoplasms between sites has followed a rule of 100:10:10:1 ratio for parotid, submandibular, minor salivary glands and sublingual tumors, respectively.^[9]

MATERIALS AND METHODS

This was a one-year observational study of salivary gland neoplasm's with special references to Periodic Acid Schiff (PAS) and Alcian Blue (AB) stain. Thirty-two cases were diagnosed with salivary gland neoplasm. This prospective observational study was done in Department of Pathology in a Tertiary care Hospital. The study duration was 12 months from November 2018 to October 2019. A total of 32 patients were included. All the epithelial salivary gland tumors were classified according to WHO (2005) histological classification. Ethical committee clearance was obtained before the study.

Inclusion Criteria

All 32 patients of salivary gland neoplasm with benign and malignant neoplasm.

Exclusion Criteria

Inflammatory, non- neoplastic and non-epithelial tumours' of salivary gland were excluded.

Salivary gland specimens were obtained after written and informed consent. Details of clinical data and investigations performed collected for available cases from histopathological requisition

form. All specimens were fixed in 8% Formalin for a period of 11 – 13 hours. Specimens gross and cut surface examined for size, shape, colour, circumscribed or infiltrative borders and presence of cystic changes were noted. The sections were taken from the lesion, its margins, surrounding tissues.

Tissues were processed, embedded in paraffin and sections were cut at 3-5 microns thick and were stained with Haematoxylin and Eosin. Selected cases special stains like Periodic acid Schiff (PAS), Alcian blue (AB).

RESULTS

All 32 patients of salivary gland neoplasm with benign and malignant neoplasm. Out of 32 patients in which 10 were female and 22 were male. Age of the patients vary from 13 to 76 year. A total of 32 salivary gland tumors during study period out of which 23(71.87%) were benign tumours and 9(28.12%) were malignant tumors.

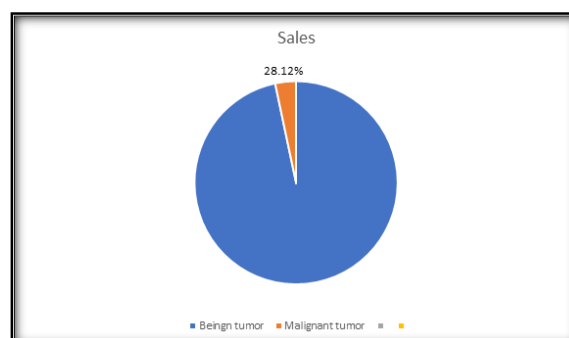


Figure 1: Showing site distribution of benign and malignant salivary gland lesion

Table 1: Number, percentage and Sex distribution of salivary gland tumors

S no.	Tumors	No.	%	Male	Female	Total
1.	Benign Tumors					
	Pleomorphic adenoma	10	31.25	7	3	9
	Warthin's tumor	6	18.75	4	2	8
	Oncocytoma	7	21.87	5	2	6
	Total	23	71.87	16	7	23
2.	TumorsMalignant					
	Acinic cell carcinoma	0	0	0	0	0
	Mucoepidermoid carcinoma	2	06.25	2	0	2
	Adenoid cystic carcinoma	3	09.37	1	2	3
	Clear cell carcinoma,NOS	1	03.12	1	0	1
	Salivary duct carcinoma	2	06.25	1	1	2
	Adenocarcinoma,NOS	0	0	0	0	0
	Squamous cell carcinoma arising in Warthin's tumour	1	03.12	1	0	1
	Total	9	28.11	6	3	9
	Grand Total	32	100	22	10	32

The parotid gland with 19 (59.37%) cases was the commonest site for various tumors, notable exception being adenoid cystic carcinoma, adenocarcinoma NOS, clear cell carcinoma NOS, which showed predilection for the minor salivary glands 4 (12.5%). Nine cases (28.12%) involved the submandibular gland. However, no case was seen in the sublingual gland. Histology of Pleomorphic adenoma revealed classical histological features. One case showed squamous metaplasia of the ducts with epithelial pearl formation and one case showed adenoid cystic – like areas. Six cases of warthin tumor accounted for 18.75 % of total, occurring in an age range of 48 and 60 years, frequent in males. In the present study, seven case of oncocytoma was found in a 40-year female patient involving the parotid gland and showed typical histological features.

There is no any case of Acinic cell carcinoma and Adenoid cystic carcinoma. Histological features (Table 1). There was a higher incidence of low grade tumors and only one tumor with intermediate grade showed lymph node metastasis. Two cases of Mucoepidermoid carcinoma accounted 06.25% of total, with an age range of 51-56 years and involving mainly males and

minor salivary glands. Microscopically, cribriform arrangement was the predominant pattern observed and two cases showed perineural invasion and one case showed muscle invasion.

[Table 2] displays various morphological types of benign tumors. Pleomorphic adenoma 10(43.47%) was the commonest histologic type followed by Warthin's tumor 6(26.08%).

Table 2: Distribution of benign tumors in salivary glands

Benign Tumors	No. of Cases	Parotid	Parotid Percentage	Sub-mandibular	Sub-mandibular Percentage	Minor Salivary Gland	Minor Percentage
Pleomorphic adenoma	10	8	34.78	2	08.69	0	0
Warthin's tumor	6	5	21.73	0	0	1	04.34
Oncocytoma	7	4	17.39	2	08.69	1	04.34
Total	23	17	73.60	4	17.38	2	8.68

[Table-2]. The commonest malignant tumor of parotid is Mucoepidermoid carcinoma and Adenoid cystic carcinoma (11.11%) and submandibular gland Adenoid cystic carcinoma (22.22%), whereas of the minor salivary gland (22.22%) was Adenoid cystic carcinoma.

Table 3: Distribution of malignant tumors in salivary glands

Malignant Tumors	No. of Cases	Parotid	Parotid Percentage	Sub-mandibular	Sub-mandibular Percentage	Minor Salivary Gland	Minor Percentage
Acinic cell carcinoma	0	0	0	0	0	0	0
Mucoepidermoid carcinoma	2	1	11.11	1	11.11	0	0
Adenoid cystic carcinoma	3	1	11.11	2	22.22	0	0
Clear cell carcinoma, NOS	2	0	0	0	0	2	22.22
Salivary duct carcinoma	1	0	0	1	11.11	0	0
Adenocarcinoma, NOS	0	0	0	0	0	0	0
Squamous cell carcinoma arising in Warthin's tumor	1	0	0	1	11.11	0	0
Total	9	2	22.22	5	55.55	2	22.22

Total five cases of sub-mandibular is obtained i.e. 55.55% in which most common case of Adenoid cystic carcinoma accounted for 22.22% of total occurring in an age range of 51-59 years. One case of Mucoepidermoid carcinoma and Adenoid cystic carcinoma accounted for 11.11% of total, occurring in an age range of 48-56 years with a male predominance. Sub-mandibular was the commonest site followed by parotid and minor salivary glands and showed typical histological features (Table 3). There was a higher incidence of low grade tumors and only one tumor with intermediate grade showed lymph node metastasis.

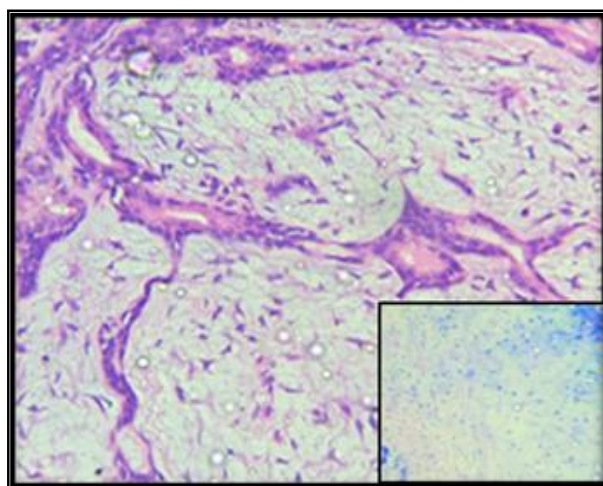


Figure 1: Microphotograph of PA showing ductal structures surrounded by chondromyxoid stroma with, inset Alcian blue stain shows positive chondromyxoid areas (H&E, 40x and AB, 40x).

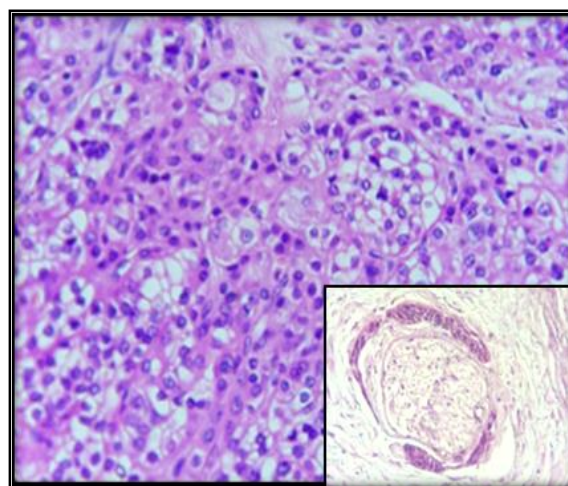


Figure 2: Mucoepidermoid Carcinoma: Microphotograph showing three types of cells: squamous cells, intermediate cells and mucin producing cells. Inset perineural invasion (H&E, 40x).

DISCUSSION

During this observational study some of important points were noticed which is quite similar to other study done. Present study frequency of occurrence of benign tumors were more common when

compared to malignant tumours.^[1,4] This result is quite similar to this observational study in out of total thirty two patients twenty were diagnosed with Benign tumour accounted for 71.87% together with nine patients were diagnosed with Malignant tumour accounted for 28.12% except in observations made

by Nagarkar et al and Tilakaratne WM et al.^[2,11] wherein malignant tumors were predominant. The mean age of benign tumor was 46yrs and for malignant tumor was 51yrs . In this study there was male preponderance in both benign and malignant tumors, similar to other studies.

The mean age observed was 40 years with an age range of 10 to 79 years. Benign tumors were common in age group of 40 to 49 years and the peak age incidence for malignant tumors was 50 to 59 years; which is in accordance with the observation by Chatterjee et al.^[1] wherein benign tumors occurred at a lower age group as compared to malignant tumors. A male preponderance (22 cases among 32) with a sex ratio of 1:2.3 was noted. Our findings were different to the previous studies.^[15]

A male predominance was observed among benign tumors which does not supporting the findings of previous studies.^[9,15] However; other authors.^[16,17] have observed a male predominance. A male predominance was seen in malignant tumours which can be easily supported by the findings of previous study. Parotid gland 19 (59.37%) was the commonest site involved followed by submandibular gland 9 (39.13%) minor salivary glands 4 (12.50 %). Thus, submandibular gland minor salivary gland was more

likely to be involved than the minor salivary gland in our study which seems to be quite different than the previous studies.^[1,12,11] However, in other series.^[14,15] the sequence of involvement of the tumors was parotid gland followed by submandibular gland and minor salivary glands through which this study can be easily justified.

Pleomorphic adenoma was the most common tumor accounting for 8 (34.78%) of benign tumors and 10 (31.25%) of all tumors. This is similar to the results of other studies.^[5,12,3,8] The peak age incidence of pleomorphic adenoma was 41-52 years with a male preponderance. These findings are different to De Oliveria FP et al.^[10] Sevencase of oncocytoma was recorded, accounting for 21.87% of all salivary gland tumors this percentage of oncocytoma were extremely different from other studies. In other studies, Tilakaratne WM et al, Subhashraj K et al, Ito et al.^[11,12] studies found oncocytoma accounting for less than 1% of all salivary gland tumors. However, in Vuhahula E A.M.^[9] series, it accounted for 2.6%. Parotid gland was the commonest site involved which was similar to that observed by Vuhahula E A.M.^[9] and Subhashraj K et al.^[12] The present study showed an higher incidence of Pleomorphic adenoma which was similar to the findings of Ito et al.^[12] Other studies.^[1,9,12,10] showed a lower incidence as compared to the present study. The peak incidence was 49-60 years which was similar to study of De Oliveria FP et al.^[12] One case of salivary duct carcinoma was seen in a 54-year male, comprising 03.12% of all tumours involving the parotid gland, which correlates with the study of Gonzalvez-Alva P et al.^[12]

Zero case was observed in Acinic cell carcinoma and Adenocarcinoma, NOS.

CONCLUSION

The PAS stain showed a strong reaction with the mucinous material in the tubules and myxoid areas. Alcian blue stain was positive for chondromyxoid areas in Pleomorphic Adenoma. Salivary gland tumours' both benign and malignant are more frequent in males. Pleomorphic adenoma is most common salivary gland tumor and Oncocytoma is the most common malignant salivary gland tumor. Very less literature is available on the mucin histochemistry of salivary gland tumors. Mucin stain acts as an adjunct to the routine H and E staining. Histopathological examination is gold standard method for diagnosis, predicting prognosis of malignant neoplasm's of salivary gland. A detailed clinical, histopathological and histochemical analysis in salivary gland tumors will provide a better insight to the pathophysiology of the disease, tumor differentiation and prognostic implications. Thus, emphasizing on a more 'pleomorphic' approach to PAs.

REFERENCES

1. Chatterjee T, Panda PK. A pathological study of benign and malignant tumours of salivary glands. *Med J Armed Forces India*. 2000 Oct; 56 (4): 282-286. doi: 10.1016/S0377-1237(17)30208-3. Epub 2017 Jun 12.
2. Nagarkar NM, Bansal S, Dass A, et al. Salivary gland tumors - our experience. *Indian J Otolaryngol Head Neck Surg*. 2004 Jan;56 (1):31-4. doi: 10.1007/BF 02968769.
3. Barnes LB, Eveson JW, Reichart P, Sidransky D. editors. *Pathology and Genetics of Head and Neck Tumors*. Lyon: IARC Press; 2005.
4. Hashemi P, Zarei MR, Chamani G, Rad M. Malignant salivary glands tumors in Kerman province: a retrospective study. *Dent Res J* 2007; 4: 4-10.
5. Hamada T, Matsukita S, Goto M, Kitajima S, Batra SK, Irimura T, et al. Mucin expression in pleomorphic adenoma of salivary gland: A potential role for MUC1 as a marker to predict recurrence. *J Clin Pathol* 2004;57:813-21.
6. Da-Quan M, Guang-Yan Y. Tumors of the minor salivary glands: A clinicopathological study of 243 cases. *Acta Otolaryngol* 1987;103:325-31.
7. Jaafari-Ashkavandi Z, Ashraf MJ, Moshaverinia M. et al. Salivary gland tumors: a clinicopathologic study of 366 cases in southern Iran. *Asian Pac J Cancer Prev*. 2013;14(1):27-30.
8. Ahmed S, Lateef M, Ahmad R. Clinicopathological study of Primary Salivary-gland tumors in Kashmir. *JK Pract* 2002; 9 (4): 231-3.
9. Vuhahula E A.M. Salivary Gland tumors in Uganda: Clinical Pathological study. *African Health Sciences* 2004; 4 (1): 15-23.
10. Shetty Archana, V Geethamani. Spectrum of Major Salivary Gland Tumours: Clinicopathologic Study. *Sch. J. App. Med. Sci.* 2014; 2(3):1088-90.
11. Tilakaratne WM, Jayasooriya PR, Tennakoon TM, et al. Epithelial salivary tumors in Sri Lanka: a retrospective study of 713 cases. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod*. 2009 Jul; 108 (1): 90-8. doi: 10.1016/j.tripleo.2009.01.026. Epub 2009 Apr 29.
12. Subhashraj K. Salivary gland tumors: a single institution experience in India. *Br J Oral Maxillofac Surg*. 2008 Dec; 46 (8):635-8. doi: 10.1016/j.bjoms. 2008. 03.020. Epub 2008 Jul 11.

13. Ito FA, Ito K, Vargas PA, et al. Salivary gland tumors in a Brazilian population: a retrospective study of 496 cases. *Int J Oral Maxillofac Surg.* 2005 Jul;34 (5): 533-6.
14. Nepal A, Chettri ST, Joshi RR, et al. Primary salivary gland tumors in eastern Nepal tertiary care hospital. *J Nepal Health Res Counc.* 2010Apr; 8 (1): 31-4.
15. Kalburge JV, Kalburge V, Latti B, Kini Y. Salivary Gland Tumors: Clinicopathologic analysis of 73 cases. *J Cranio Max Dis* 2014; 3(2):111-5.
16. Shrestha S, Pandey G, Pun C.B, Bhatta R, Shahi R. Histopathological Pattern of Salivary Gland Tumors. *Journal of Pathology of Nepal* 2020; 04(7): 520 –24.
17. Laishram RS, Kumar KA, Pukhrabam GD, et al. Pattern of salivary gland tumors in Manipur, India: A 10 year study. *South Asian J Cancer.* 2021 Oct;2(4): 250-3. doi: 10.4103/2278-330X.119886.
18. C. M. Dharaiya, M. B Patel. Comparative study of Cytodiagnosis of salivary gland neoplasm with Histopathology. *Trop J Path Micro* 2022;4(1):88-92.