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Original Research Paper

Pathology

IMMUNOHISTOCHEMICAL EXPRESSION OF P16 AND P53 AS MARKERS OF HPV ASSOCIATION AND MALIGNANT TRANSFORMATION IN SCHNEIDERIAN PAPILLOMAS: A CROSS-SECTIONAL STUDY FROM A TERTIARY CARE CENTER

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ABSTRACT

Background: Schneiderian papillomas (SPs) are benign sinonasal tumors with potential for recurrence and malignant transformation to squamous cell carcinoma (SCC). The roles of p16, p53 and Human Papillomavirus (HPV) remain controversial. **Aim:** To evaluate p16 and p53 expression as markers of HPV association and malignant transformation in SPs. **Materials and Methods:** Thirty histologically confirmed SPs were studied over two years. Cases were reviewed, classified, and immunohistochemistry done for p16 and p53; HPV status was evaluated. **Results:** Among 30 cases, 12 (40%) were SPs without dysplasia, 9 (30%) showed dysplasia, and 9 (30%) were SCC. Benign SPs (73.3%) showed strong p16 expression, SCC cases showed absent/non-diffuse staining. Strong p53 expression was significantly associated with SCC and dysplasia. Only 1 case (3.3%) was HPV positive. **Conclusion:** p53 overexpression correlates with malignant transformation, p16 is an unreliable indicator of HPV association and HPV has limited role in SP pathogenesis in this population.

KEYWORDS : Schneiderian Papilloma, p16, p53, HPV, Malignant Transformation

INTRODUCTION

Schneiderian papillomas are uncommon benign epithelial neoplasms arising from the Schneiderian membrane lining the nasal cavity and paranasal sinuses. They account for less than 5% of all sinonasal tract irs and are classified into inverted, exophytic, and oncocytic types (1). Among these, inverted papilloma is the most common subtype and demonstrates a male predominance with peak incidence in the fifth to eighth decades of life (1,2). Although histologically benign, Schneiderian papillomas exhibit locally aggressive behavior, high recurrence rates, and a well-documented potential for malignant transformation, reported in approximately 10–15% of cases (2,3).

The etiopathogenesis of Schneiderian papillomas remains multifactorial. Human papillomavirus (HPV), particularly low-risk types 6 and 11 and occasionally high-risk type 16, has been detected in a subset of cases using molecular techniques (4,5). However, the reported prevalence varies widely across studies and geographic populations. In HPV-driven tumors, viral E7 protein inactivates the retinoblastoma (Rb) pathway, leading to overexpression of p16, which is therefore considered a surrogate marker of transcriptionally active HPV infection in oropharyngeal carcinomas (6). However, p16 overexpression may occur independently of HPV, limiting its specificity in certain head and neck sites (6,7).

The TP53 gene encodes p53, a critical tumor suppressor protein involved in cell cycle regulation and apoptosis. TP53 mutations are among the most common genetic alterations in head and neck squamous cell carcinomas and are associated with tumor progression and poor prognosis (7). Immunohistochemical overexpression of p53 often reflects underlying TP53 mutation due to prolonged protein half-life (7,8). Several studies have investigated p53 and p16 expression patterns in Schneiderian papillomas to clarify their role in malignant transformation (3,5,8).

Given the inconsistent evidence regarding HPV involvement and the predictive value of p16 and p53, this study aimed to evaluate their expression in Schneiderian papillomas and assess their association with dysplasia and malignant transformation in a tertiary care population.

MATERIALS AND METHODS

Study Design

This study was designed as a cross-sectional observational study to evaluate the immunohistochemical expression of p16 and p53 in Schneiderian papillomas and to assess their association with dysplasia and malignant transformation.

Study Setting and Duration

The study was conducted in the Department of Pathology, Osmania Medical College, Hyderabad, in collaboration with the Government ENT Hospital, Koti, Hyderabad. The study was carried out over a period of two years from the date of institutional approval (ECR/300/Inst/AP/2013/RR-19).

Study Population and Sample Size

The study population consisted of patients diagnosed with Schneiderian papillomas who underwent surgical excision or biopsy of sinonasal lesions at the Government ENT Hospital, Koti, Hyderabad. A total of 30 cases of histopathologically confirmed Schneiderian papillomas were included in the study. The cases comprised Schneiderian papillomas with and without dysplasia, as well as those associated with malignant transformation.

Inclusion Criteria

Patients diagnosed histopathologically with Schneiderian papillomas were included in the study. Cases of Schneiderian papillomas showing epithelial dysplasia and those associated with malignant transformation were also included.

Exclusion Criteria

Patients who had received prior radiotherapy or chemotherapy were excluded from the study as these treatment modalities may alter the histological and immunohistochemical characteristics of the tissue. Cases with inadequate tissue representation in slides or paraffin blocks were excluded. Specimens showing autolysis or poor preservation were also excluded. Infective and inflammatory lesions of sinonasal polyps and other primary sinonasal malignancies not arising from Schneiderian papillomas were excluded from the study.

Specimen Collection and Processing

Biopsy and resection specimens of sinonasal papillomas

arising from the nasal cavity and paranasal sinuses were collected and fixed in 10% neutral buffered formalin to preserve tissue morphology and antigenicity. Following adequate fixation, the specimens were processed through routine histopathological procedures including dehydration, clearing, and paraffin embedding. Formalin-fixed paraffin-embedded tissue blocks were prepared and serial sections of approximately 3–5 μm thickness were cut using a microtome and mounted on glass slides for histopathological and immunohistochemical analysis.

Histopathological Examination

Routine histological examination was performed using hematoxylin and eosin staining. The stained sections were examined under a light microscope to confirm the diagnosis of Schneiderian papilloma and to evaluate histopathological features such as the type of papilloma, presence or absence of epithelial dysplasia, and evidence of malignant transformation. Additional features including stromal characteristics and inflammatory cell infiltrates were also assessed.

Immunohistochemical Analysis

Immunohistochemical staining was performed on representative tissue sections using antibodies against p16 and p53 proteins. The paraffin sections were first deparaffinized in xylene and rehydrated through graded alcohols to water. Antigen retrieval was carried out using appropriate buffer solutions to enhance antigen exposure. Endogenous peroxidase activity was blocked prior to incubation with primary antibodies against p16 and p53. The sections were then incubated with secondary antibodies and visualized using a chromogenic detection system. Finally, the slides were counterstained with hematoxylin and mounted for microscopic evaluation.

Evaluation of Immunohistochemical Staining

The immunohistochemically stained slides were examined under light microscopy. The expression of p16 and p53 was assessed based on staining intensity, percentage of positively stained cells, and distribution of staining within the epithelial layers. p16 expression was interpreted as diffuse strong nuclear and cytoplasmic staining in tumor cells, whereas p53 expression was evaluated based on nuclear staining patterns and the proportion of positively stained cells. These findings were correlated with histopathological features including dysplasia and malignant transformation.

HPV Assessment

Paraffin-embedded biopsy specimens were deparaffinized with xylene, centrifuged, and rehydrated through graded ethanol washes before air-drying. The tissue pellet was resuspended in lysis buffer containing proteinase K and incubated at 56 °C until complete digestion. DNA was extracted using a commercial nucleic acid extraction kit following the manufacturer's protocol and eluted for analysis. The extracted DNA served as a template for real-time PCR targeting HPV 16 and 18. The reaction mixture included specific ready mixes, internal control, and appropriate positive and negative controls. Amplification was carried out under standardized cycling conditions with sequential denaturation, annealing, and extension steps. Fluorophores were used for detection of HPV types and internal control signals, and analysis was performed using a real-time PCR system.

Statistical Analysis

The collected data were compiled and analyzed using appropriate statistical methods. Descriptive statistics were used to summarize the clinicopathological characteristics of the study population. Associations between immunohistochemical expression of p16 and p53 and histopathological findings such as dysplasia and malignant transformation were analyzed using appropriate tests of statistical significance. A p-value of less than 0.05 was considered statistically significant.

RESULTS

Demographic Characteristics

A total of 30 histopathologically confirmed cases of Schneiderian papilloma were included in the study. The age of the patients ranged from 26 to 90 years, with the majority of cases occurring in the fifth to seventh decades of life, indicating that Schneiderian papillomas predominantly affect middle-aged and elderly individuals. A marked male predominance was observed in the study population. Out of the 30 cases, 24 patients (80%) were male and 6 patients (20%) were female, resulting in a male-to-female ratio of 4:1 (Figure 1).

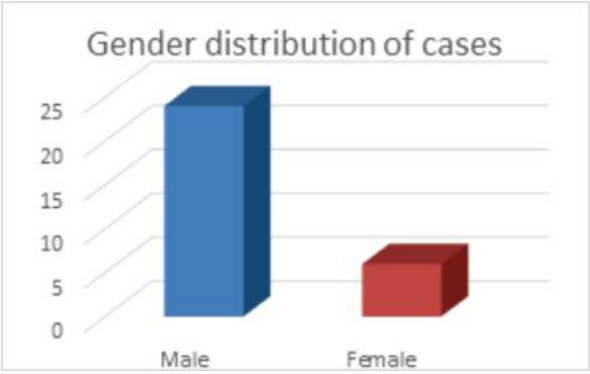

Figure 1. Bar Graph Showing Male vs Female Distribution Among Cases

Clinical Presentation

The most common presenting symptom among patients was nasal obstruction, which was reported in the majority of cases. Other associated symptoms included nasal discharge, epistaxis, and facial discomfort, although these were less frequently observed.

Histopathological Findings

Histopathological examination of hematoxylin and eosin (H&E) stained sections revealed the characteristic features of Schneiderian papillomas, including papillary epithelial proliferation, thickened squamous or transitional epithelium, and fibrovascular stromal cores. Based on histopathological evaluation, among the 30 cases studied, 12 cases (40%) showed benign Schneiderian papillomas without dysplasia, 9 cases (30%) demonstrated varying degrees of epithelial dysplasia, 9 cases (30%) were associated with squamous cell carcinoma (Table 1).

Table 1. Histopathological Distribution of Cases

Histopathological Category	Number of Cases	Percentage
Schneiderian papilloma without dysplasia	12	40%
Schneiderian papilloma with dysplasia	9	30%
Schneiderian papilloma with SCC	9	30%
Total	30	100%

Immunohistochemical Expression of p16

Immunohistochemical analysis demonstrated strong p16 expression in the majority of benign Schneiderian papillomas (Figure 2). Out of the total 30 cases, 22 cases (73.3%) showed strong p16 positivity, 5 cases (16.7%) showed moderate staining, 3 cases (10%) demonstrated weak or negative staining. The staining pattern was predominantly diffuse nuclear and cytoplasmic staining within the epithelial cells. However, diffuse strong p16 expression did not correlate with HPV positivity in most cases.

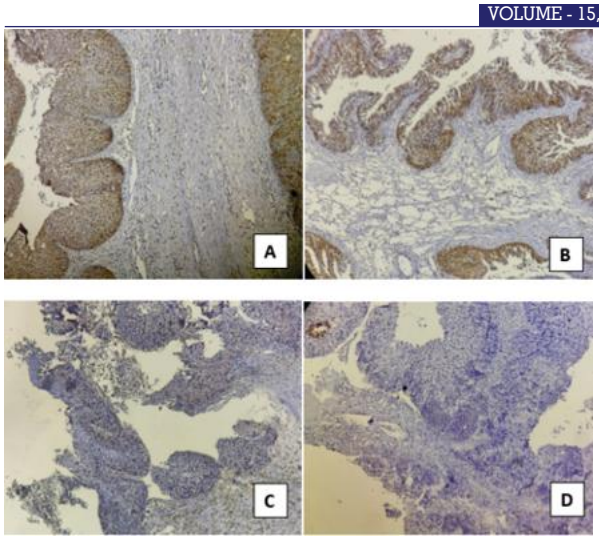

Figure 2. A - Immunohistochemical Staining Showing Strong Diffuse p16 Expression in Benign Schneiderian Papilloma. B - p16 Immunostaining Showing Moderate Expression in Dysplastic Schneiderian Papilloma. C - p16 Expression in Severe Dysplasia within Schneiderian Papilloma. D - p16 Expression is Absent in Squamous Cell Carcinoma Arising in Schneiderian Papilloma.

Immunohistochemical Expression of p53

p53 expression demonstrated a distinct pattern compared to p16. Strong p53 overexpression was observed predominantly in dysplastic lesions and in cases associated with squamous cell carcinoma. Among the 30 cases, benign papillomas without dysplasia showed minimal or low p53 expression, dysplastic papillomas demonstrated moderate to strong p53 expression, and all cases associated with squamous cell carcinoma showed strong p53 overexpression.

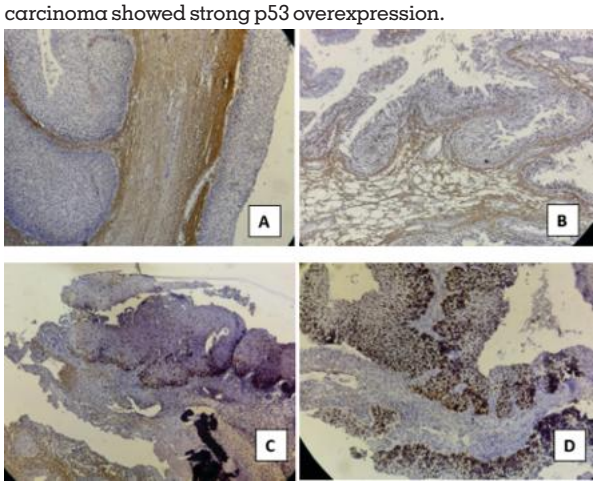

Figure 3. A - Immunohistochemical Staining Showing Low p53 Expression in Benign Schneiderian Papilloma. B - p53 Immunostaining Showing Moderate Expression in Dysplastic Schneiderian Papilloma. C - p53 Overexpression in Severely Dysplastic Schneiderian Papilloma. D - p53 Overexpression in Malignant Transformation.

Correlation Between Histopathology and Immunohistochemistry

Correlation analysis demonstrated that p16 expression was common in benign papillomas but did not correlate with HPV positivity. In contrast, p53 overexpression showed a strong association with dysplasia and malignant transformation. These findings suggest that p53 may serve as a useful biomarker for identifying dysplastic and malignant progression in Schneiderian papillomas, whereas p16 expression alone may not reliably indicate HPV involvement in these lesions.

HPV Detection using Real Time PCR

HPV testing revealed that only one case (3.3%) was positive for HPV, while 29 cases (96.7%) were negative. The HPV-positive case corresponded to a moderately differentiated squamous cell carcinoma arising in a Schneiderian papilloma in an elderly patient.

DISCUSSION

The present study provides a comprehensive clinicopathological and immunohistochemical analysis of 30 Schneiderian papilloma cases, with emphasis on p16 and p53 expression patterns and their correlation with histopathological progression and HPV status.

The demographic characteristics in our study align with established epidemiological data. Patient age ranged from 26 to 90 years, with peak incidence in the fifth to seventh decades, consistent with large series reporting mean ages of 50-60 years [17]. A marked male predominance was observed (80% male; male:female ratio 4:1), corroborating literature reports of male-to-female ratios ranging from 3:1 to 5:1 [18,19]. Smoking, more prevalent in males, may contribute to this disparity [20]. Nasal obstruction was the predominant presenting symptom, consistent with the typical clinical manifestation of these tumors, frequently accompanied by epistaxis, rhinorrhea, and facial discomfort [21].

Schneiderian papillomas (SPs) are unique benign epithelial neoplasms of ectodermal origin arising from the sinonasal mucosa. Despite their benign histology, their biological behavior is complex due to recurrence, local aggressiveness, and malignant transformation potential. In the present study, histopathological evaluation revealed 12 cases (40%) of benign Schneiderian papilloma without dysplasia, 9 cases (30%) with varying degrees of epithelial dysplasia, and 9 cases (30%) associated with squamous cell carcinoma. This proportion of SCC is higher than that reported in many published series and may reflect referral bias associated with a tertiary care centre, as well as the relatively small sample size of the present study.

Immunohistochemical analysis demonstrated strong p16 expression in 22 cases (73.3%), with moderate staining in 5 cases (16.7%), and weak/negative staining in 3 cases (10%). However, this robust p16 expression did not correlate with HPV positivity, as only one case (3.3%) demonstrated detectable HPV DNA by real-time PCR. This dissociation between p16 overexpression and HPV infection represents a critical finding.

The role of HPV in SP remains controversial. This low prevalence aligns with several studies reporting inconsistent HPV detection rates in sinonasal papillomas (4,5,10). Syrjänen (10) reported that while low-risk HPV types (6 and 11) are frequently detected in exophytic papillomas, inverted papillomas show wide variability in HPV prevalence across geographic regions. Similarly, Bishop et al. (11) demonstrated that HPV-driven oncogenesis is significantly less common in sinonasal carcinomas compared to oropharyngeal carcinomas. Recent meta-analyses have highlighted substantial heterogeneity in HPV detection rates in inverted papillomas, ranging from 0% to over 60%, largely depending on detection methodology (PCR vs in situ hybridization) and population studied (12). The extremely low HPV positivity in our study suggests that HPV-independent pathways may predominate in this regional population. Interestingly, although strong p16 expression was observed in 73.3% of cases, it did not correlate with HPV positivity. This finding supports growing evidence that p16 overexpression in sinonasal lesions may occur independently of transcriptionally active HPV infection (5,8,11). Lewis et al. (13) emphasized that p16 is a reliable surrogate marker only in oropharyngeal squamous cell carcinoma, but not universally

applicable to all head and neck subsites. Therefore, extrapolating the p16-HPV paradigm from oropharynx to sinonasal tract may be misleading.

In contrast to p16, p53 expression demonstrated a clear association with dysplasia and carcinoma in our study. All cases of malignant transformation showed strong p53 overexpression. This observation is consistent with the known role of TP53 mutations in head and neck carcinogenesis (7,14). Nudell et al. (3) reported frequent p53 overexpression in carcinoma ex-Schneiderian papilloma and suggested that TP53 pathway disruption represents a critical step in malignant progression. Altavilla et al. (4) also demonstrated increased p53 expression in dysplastic and malignant areas of inverted papillomas compared to benign regions. Similarly, Lin et al. (8) found that p53 staining intensity increased progressively from benign papilloma to carcinoma. From a molecular standpoint, TP53 mutations lead to accumulation of dysfunctional p53 protein due to increased half-life, allowing detection by immunohistochemistry (7). Recent genomic studies have further confirmed that TP53 mutations are common in sinonasal squamous cell carcinoma, particularly in HPV-negative tumors (15). Udager et al. (15) demonstrated that HPV-negative sinonasal carcinomas frequently harbor TP53 alterations along with EGFR and KRAS pathway mutations. This supports the hypothesis that malignant transformation in SP may follow a conventional head and neck carcinogenesis pathway rather than an HPV-driven mechanism.

The Nature Scientific Reports, Kwon et al., 2024 (22) study demonstrated that p53 immunoexpression strongly correlates with TP53 mutations in inverted papillomas, making it a reliable marker for malignant transformation. Importantly, these alterations were detectable even in the papilloma component, supporting a stepwise progression to carcinoma. In contrast, p16 expression was inconsistent and did not reliably indicate HPV-driven disease, limiting its value as a surrogate marker.

The Medicina (MDPI), Enache et al., 2024 (23) study showed that benign inverted papillomas typically exhibit low, basal p53 expression, while increased p53 staining and higher Ki-67 index are associated with dysplasia and progression. Although p16 showed diffuse positivity, it lacked specificity for HPV. The study concluded that p53, combined with morphology and proliferation markers, is more useful than p16 alone in predicting malignant potential.

Emerging evidence suggests two potential molecular pathways in Schneiderian papilloma-associated carcinogenesis: an HPV-driven pathway and a TP53-driven pathway (12,15,16). In HPV-driven tumors, p16 overexpression and low p53 levels are expected due to viral-mediated degradation of p53 protein. Conversely, in HPV-negative tumors, TP53 mutation results in p53 accumulation and variable p16 expression. The present study supports the predominance of the TP53-driven pathway in this population, as evidenced by low HPV positivity and strong p53 overexpression in malignant cases. The frequent p16 positivity in benign papillomas without HPV detection suggests dysregulation of the Rb pathway independent of viral oncogenesis. Possible mechanisms include epigenetic alterations, chronic inflammation, or environmental carcinogens.

Clinical Implications

The divergent patterns—p16 expression common in benign lesions regardless of HPV status, p53 overexpression emerging with dysplasia and carcinoma—suggest distinct roles for these biomarkers. p16 positivity should not be interpreted as evidence of HPV causation in sinonasal Schneiderian papillomas. Conversely, p53 overexpression correlates strongly with histopathological progression, supporting its utility in risk stratification.

Clinically, our findings support careful histopathological evaluation of all Schneiderian papillomas for dysplastic changes, with consideration of p53 immunohistochemistry as an adjunct in ambiguous cases. Patients with p53-overexpressing dysplastic papillomas may warrant more aggressive surgical resection and closer surveillance, given their apparent increased risk for malignant progression [10].

Comparison with Global Data

The low HPV prevalence observed in this study contrasts with certain Western cohorts reporting moderate HPV detection rates (12). Geographic variation, differences in viral subtype distribution, and methodological factors may explain these discrepancies. In Indian and South Asian populations, HPV involvement in sinonasal papillomas appears to be relatively low compared to oropharyngeal carcinomas (limited regional data available). This highlights the importance of population-specific molecular profiling rather than generalized assumptions based on Western data.

Study Limitations

Limitations include small sample size (30 cases), single-institution retrospective design, absence of long-term follow-up, and lack of confirmatory TP53 sequencing. HPV genotyping was limited to consensus L1 detection without comprehensive typing of all potentially relevant genotypes.

Study Strengths and Future Directions

This study provides valuable regional data correlating histopathology, immunohistochemistry, and HPV status. However, the absence of molecular TP53 mutation analysis and small sample size are limitations. Future multicentric studies incorporating PCR-based HPV typing, RNA-based detection of transcriptionally active virus, and genomic sequencing would provide deeper mechanistic insight.

CONCLUSIONS

p16 expression is common in Schneiderian papillomas but does not correlate with HPV status, limiting its utility as a surrogate marker for viral etiology. In contrast, p53 overexpression shows a strong, graded association with histopathological progression, suggesting potential as a biomarker for identifying lesions at increased malignant transformation risk. The very low HPV detection rate supports the concept that HPV does not play a major etiological role in most inverted papillomas.

REFERENCES

1. Wenig BM. Schneiderian papillomas. In: Wenig BM. Atlas of Head and Neck Pathology. Philadelphia: Elsevier; 2016. p. 84–94.
2. Cheung FMF, Lau TWS, Cheung LKN, Li ASM, Chow SK, Lo AWI. Schneiderian papillomas and carcinomas. Ear Nose Throat J. 2010;89:1–6.
3. Nudell J, Chiosea S, Thompson LDR. Carcinoma ex-Schneiderian papilloma. Head Neck Pathol. 2014;8:269–286.
4. Altavilla G, Staffieri A, Busatto G, et al. Expression of p53 and HPV DNA in inverted papilloma. Acta Otolaryngol. 2009;129:1242–1249.
5. Yamashita Y, Hasegawa M, Deng Z, et al. HPV infection and expression of p53 and p16 in sinonasal disease. Infect Agents Cancer. 2015;10:23.
6. Sano T, Oyama T, Kashiwabara K, et al. Overexpression of p16 associated with HPV infection. Pathol Int. 1998;48:580–585.
7. Levine AJ. p53, the cellular gatekeeper. Cell. 1997;88:323–331.
8. Lin GC, Scheel A, Akkina S, et al. p16 and p53 staining patterns in inverted papilloma. Int Forum Allergy Rhinol. 2013;3:885–889.
9. Lawson W, Patel ZM. The evolution of management for inverted papilloma. Otolaryngol Clin North Am. 2009;42:481–499.
10. Syrjänen K. HPV in sinonasal papillomas: systematic review. Acta Otolaryngol. 2013;133:1–9.
11. Bishop JA, Ogawa T, Chang X, et al. HPV-related carcinoma of sinonasal tract. Am J Surg Pathol. 2013;37:185–192.
12. Lawson W, Ho BT, Shaari CM, et al. Inverted papilloma: meta-analysis of HPV detection. Laryngoscope. 2008;118:119–124.
13. Lewis JS Jr, Beadle B, Bishop JA, et al. Human papillomavirus testing in head and neck carcinomas. Arch Pathol Lab Med. 2018;142:559–597.
14. Poeta ML, Manola J, Goldwasser MA, et al. TP53 mutations and survival in head and neck SCC. N Engl J Med. 2007;357:2552–2561.
15. Udager AM, McHugh JB, Goudsmit CM, et al. Genomic profiling of sinonasal squamous cell carcinoma. Clin Cancer Res. 2015;21:2601–2609.

16. WHO Classification of Head and Neck Tumours. 5th ed. Lyon: IARC; 2022.
17. Vorasubin, N., Vira, D., Suh, J. D., Bhuta, S., Wang, M. B., & Blackwell, K. E. (2013). Schneiderian papillomas: Comparative review of exophytic, oncocytic, and inverted types. American Journal of Rhinology & Allergy, *27*(4), 287-292.
18. Radiopaedia. (2024). Schneiderian papilloma. Radiopaedia.org. Retrieved from https://radiopaedia.org/articles/schneiderian-papilloma
19. Thompson, L. D. R. (2021). Middle ear and temporal bone papilloma: A clinicopathologic study and comprehensive literature review of 57 cases. Head and Neck Pathology, *15*(4), 1212-1220.
20. Nudell, J., Chiosea, S., & Thompson, L. D. (2014). Carcinoma ex-Schneiderian papilloma (malignant transformation): A clinicopathologic and immunophenotypic study of 20 cases. Head and Neck Pathology, *8*(3), 269-286.
21. Radiologica. (2023). Inverting papilloma. Radiologica Knowledge Base.
22. Kwon, S., Kim, JW., Kim, E.S. et al. Assessment of TP53 and CDKN2A status as predictive markers of malignant transformation of sinonasal inverted papilloma. Sci Rep 14, 14286 (2024).
23. Enache I, Anghelina F, Ciolofan M-S, Mogoant CA, Stepan AE, Vilcea I-C, Obleaga CV, Osman A. Endoscopic and Immunohistochemical Aspects in an Inverted Papilloma with Unilateral Total Obstruction of the Nasal Fossa. Journal of Mind and Medical Sciences. 2024; 11(2):541-546.