



ORIGINAL RESEARCH

The prognostic significance of human papilloma virus in sinonasal squamous cell carcinoma

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Abstract

Background: Human papilloma virus (HPV) has been implicated in the pathology of oropharyngeal head and neck cancers, but its role in sinonasal squamous cell carcinoma (SNSCC) has not been well established.

Methods: Thirty-two patients with SNSCC diagnosed between 2011 and 2018 were identified and stratified by HPV status and viral serotype, as determined by PCR. Endpoints including recurrence, metastases and survival were analyzed using the Kaplan-Meier method.

Results: Seventeen (53%) patients were HPV-positive and 15 (47%) were HPV-negative. The median follow-up time of living patients was 30.7 months (range 4-123 months). Survival did not differ by HPV status, but HPV+ tumors were more likely to locally recur and metastasize. When stratifying by treatment type, the lowest rate of recurrence occurred in patients receiving surgery and chemoradiation.

Conclusion: A significant proportion of sinonasal tumors appear to be associated with HPV. Testing for HPV might be justified in all cases of sinonasal cancers. Further investigation is warranted to better understand the role of HPV in SNSCC.

KEYWORDS

human papilloma virus (HPV), prognosis, sinonasal carcinoma

1 | INTRODUCTION

Sinonasal carcinomas are rare malignancies, comprising about 3% of head and neck cancers with an incidence of less than 1 per 100 000 in the United States.¹ Although the incidence has been declining in recent

years, clinical outcomes remain poor.² While there is significant histologic heterogeneity among these tumors, the most common histology is sinonasal squamous cell carcinoma (SNSCC), which accounts for 60% to 75% of sinonasal carcinomas.² The pathogenesis of SNSCC is not yet clearly understood, but several environmental exposures, such as wood dust and certain chemicals, have been identified as risk factors. The association, however, is stronger for sinonasal adenocarcinoma.³ Furthermore, unlike squamous cell carcinomas arising in other sites of the head

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TABLE 1 Patient and disease characteristics by HPV status

		P-value	HPV+ (n = 17)	HPV- (n = 15)
Site	Nasal cavity	.03	15	8
	Sinonasal		2	7
Gender	Male	.53	12	9
	Female		5	6
Age	Average age (range)	.08	56.8 (45-83)	64.7 (46-87)
	SD		9.4	12.8
Histology	Basaloid	.88	3	2
	Conventional		4	2
	Keratinizing		6	3
	Non-keratinizing		7	5
	Papillary		4	5
	Poorly differentiated		2	3
Smoking	Current smoker	.93	5	5
	Former smoker (and passive exposure)		5	4
	Never smoker		7	5
	Average pack years		14.3	16.3
Clinical stage	I	.83	4	4
	II		3	1
	III		5	5
	IV		5	5
Pathological Stage	I	.3	5	1
	II		1	3
	III		4	2
	IV		4	2
Follow up (months)	Median	.19	27.2	36.4
	SD		18.1	33.2
	Range		4.2-69.4	3.6-120.8
Recurrence	Local recurrence	.6	4	2 (neither received RT)
	Distant recurrence		2	2

Abbreviation: HPV, human papilloma virus.

TABLE 2 HPV subtype by primary site

HPV subtype	Nasal cavity	Sinonasal	All sites total
16 only	5	1	6
18 only	2	0	2
33 only	2	0	2
35 only	1	0	1
45 only	1	0	1
56 only	1	0	1
69 only	1	0	1
11, 16	1	0	1
16, 33	1	0	1
16, 18, 45	0	1	1
Cases with multiple HPV subtypes	2	1	3

Abbreviation: HPV, human papilloma virus.

and neck, where it is a strong risk factor, smoking has been associated with only a slightly increased risk of SNSCC.³

Human papilloma virus (HPV) has been implicated in a significant fraction of head and neck cancers, and has a highly specific site predilection for the oropharynx and watershed areas immediately surrounding the oropharynx. Several studies in recent years have demonstrated that HPV-positive tumors comprise a significant percentage of SNSCCs.⁴ While HPV-positivity has been shown to be a positive prognostic indicator in oropharyngeal cancer,⁵ the role, biologic behavior, and prognosis of HPV in SNSCC has not been established. Efforts to do so have been impeded by heterogeneity in HPV testing methods, particularly since some assays have been unable to identify the clinical relevance of HPV infections due to the inability to link HPV detection with evidence of its biologic activity.⁶

Clinical outcomes related to HPV-positivity in SNSCC remain unclear. Previous studies have had conflicting results regarding a survival benefit associated with HPV-positivity.⁷⁻¹⁰ Larger series and established high quality pathologic and molecular studies are needed to better understand the biology, natural history, and impact of therapy of this disease to inform treatment decisions and prognoses. In an

effort to accomplish this, we studied a retrospective set of patients with SNSCC treated at our institution and compared the demographics, HPV subtype, TNM staging, treatment modalities, risk factors, and cancer control outcomes between HPV-positive and HPV-negative sinonasal carcinoma tumors.

2 | METHODS

2.1 | Patient selection

All patients with sinonasal cancer between January 1, 2011 and September 17, 2018 were identified from our departmental database using ICD-9 and ICD-10 diagnosis codes. This study was approved by our Institutional Review Board (IRB).

Thirty-seven patients with squamous cell carcinoma histology and follow up information were identified and stratified by HPV status. Five patients in total were excluded from this analysis: 3 patients because of absent tissue, and 2 who had p16 positive tests only (insufficient tissue was available for PCR). We determined HPV status

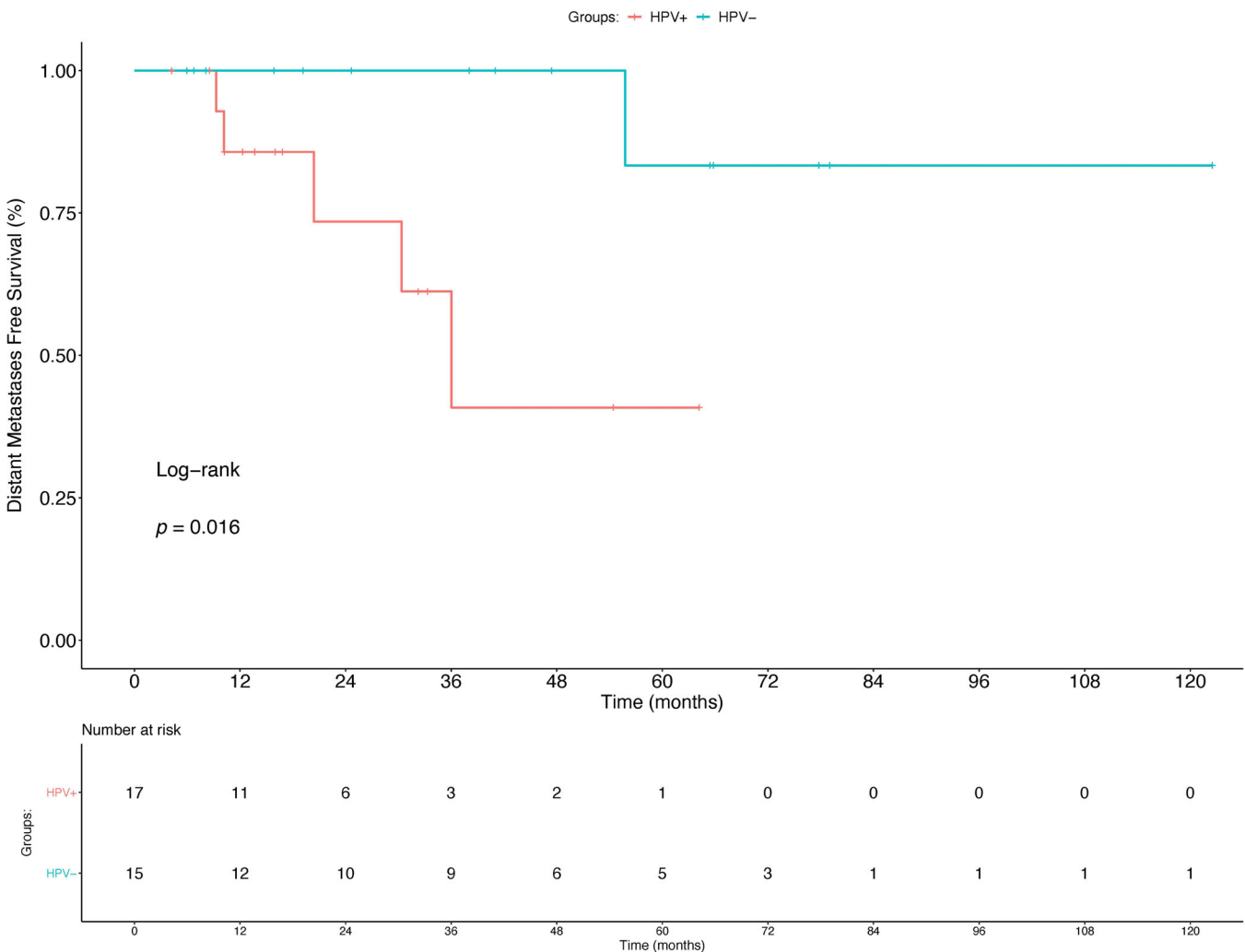


FIGURE 1 Time to metastasis by HPV status. HPV, human papilloma virus

and subtype by PCR for the remaining 32 patients. For HPV PCR testing, The Maxwell 16 FFPE Tissue LEV DNA Kit (Promega, Madison, Wisconsin) was used for DNA extraction from tissue sections according to the manufacturer's instructions. Type specific primers and probes for HPV 16 or HPV 18 (targeting the E6 region) were used to detect HPV 16 or HPV 18 by real time PCR. In the event HPV16/18 specific PCR was negative, Sanger Sequencing was performed to detect other HPV genotypes after amplification with GP5+ and GP6+ primer pairs. HPV subtypes other than HPV 16 or 18 were determined by aligning sequences in GenBank. Of the 32 included patients, 17 (53%) were HPV-positive and 15 (47%) were HPV-negative.

2.2 | Analysis

Demographics, TNM stage, histological subtype, treatment characteristics, treatment toxicities, risk factors, recurrence, metastasis, and

survival outcomes were compared between HPV-positive and HPV-negative tumors. Treatment toxicities were quantified using Radiation Oncology Therapy Group (RTOG) and Common Terminology Criteria for Adverse Events (CTCAE) scales.

2.3 | Statistical methods

Endpoints including recurrence, metastases, and death were calculated using actuarial methods and Kaplan Meier survival curves were compared using the log-rank test. Cancer control endpoints were based on the most recent follow-up visit. Baseline characteristics were compared using the chi-square test and the Mann Whitney *U* test for categorical and continuous variables, respectively. The null hypothesis was rejected for $P < .05$. Statistical analyses were conducted using Statistical Package for Social Sciences (SPSS) software (IBM SPSS Statistics for Windows, Version 22.0. Armonk, NY: IBM Corp) and R Studio Version 1.1.383.

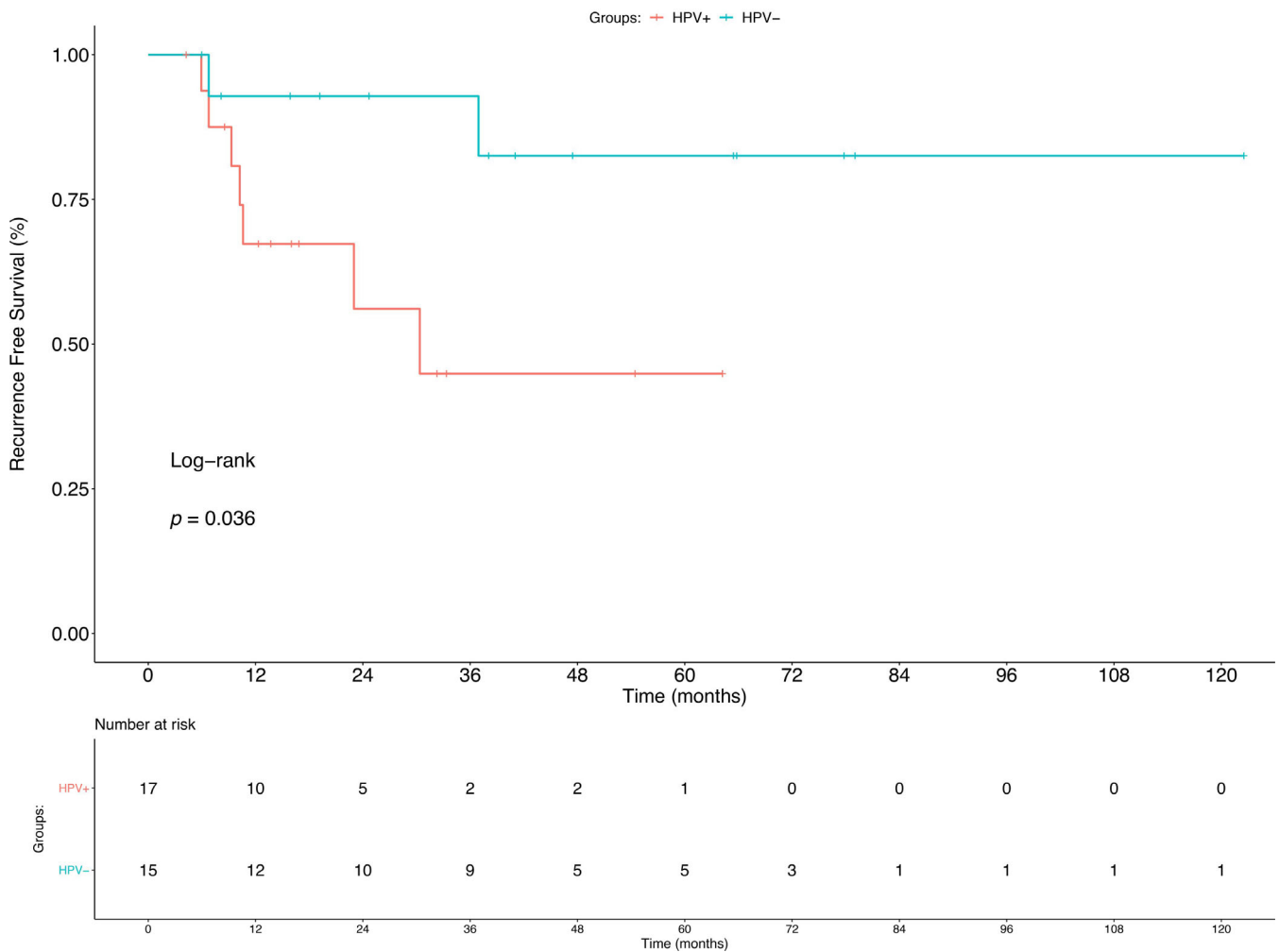


FIGURE 2 Time to local recurrence by HPV status. HPV, human papilloma virus

3 | RESULTS

The median age for the 32 patients in the present study was 58.0 years old (range 45-87 years). The median follow-up time for living patients was 30.7 months (range 4-123 months).

There were no significant differences between the HPV-positive and HPV-negative groups for gender, race, median age, smoking status, clinical stage, pathologic stage, or follow-up time. A greater proportion of nasal cavity tumors were HPV-positive as compared with sinonasal tumors ($P = .03$). Patient and disease characteristics are listed in Table 1.

All 17 HPV-positive patients had a known subtype (Table 2). The high risk subtypes represented included 16, 18, 33, 35, 45, 56, and 69. Of the 17 patients with HPV+ tumors in this study, 9 (53%) were subtype 16 and 3 (18%) were subtype 18. Three of 17 (18%) of tumors were positive for multiple subtypes, including HPV 16 and 18 among other subtypes. The combinations with multiple subtypes

included one tumor each with subtypes 16, 18, and 45; 16 and 11; and 16 and 33. The histologic types are summarized in Table 1.

HPV-positive tumors were more likely to metastasize ($P = .016$), as shown in Figure 1, and locally recur ($P = .036$), as shown in Figure 2. There were no significant differences between the HPV-positive and HPV-negative groups in overall survival ($P = .17$), as shown in Figure 3. While a statistically significant difference in distant metastasis-free survival (Figure 4) was noted among patients with high-risk HPV (HPV 16 and 18), patients with low-risk HPV (HPV subtypes other than 16 and 18), and patients who were HPV negative ($P = .041$), no differences were seen in local recurrence-free survival (Figure 5, $P = .11$) or overall survival (Figure 6, $P = .21$). Of the 9 recurrences, 6 (of 23) were nasal cavity tumors and 3 (of 9) were sinonasal tumors. Recurrences are summarized in Table 1.

When stratifying by treatment type, the lowest rate of local regional recurrence occurred in patients receiving surgery and adjuvant chemoradiation (2/17 patients, 12%), followed by surgery and

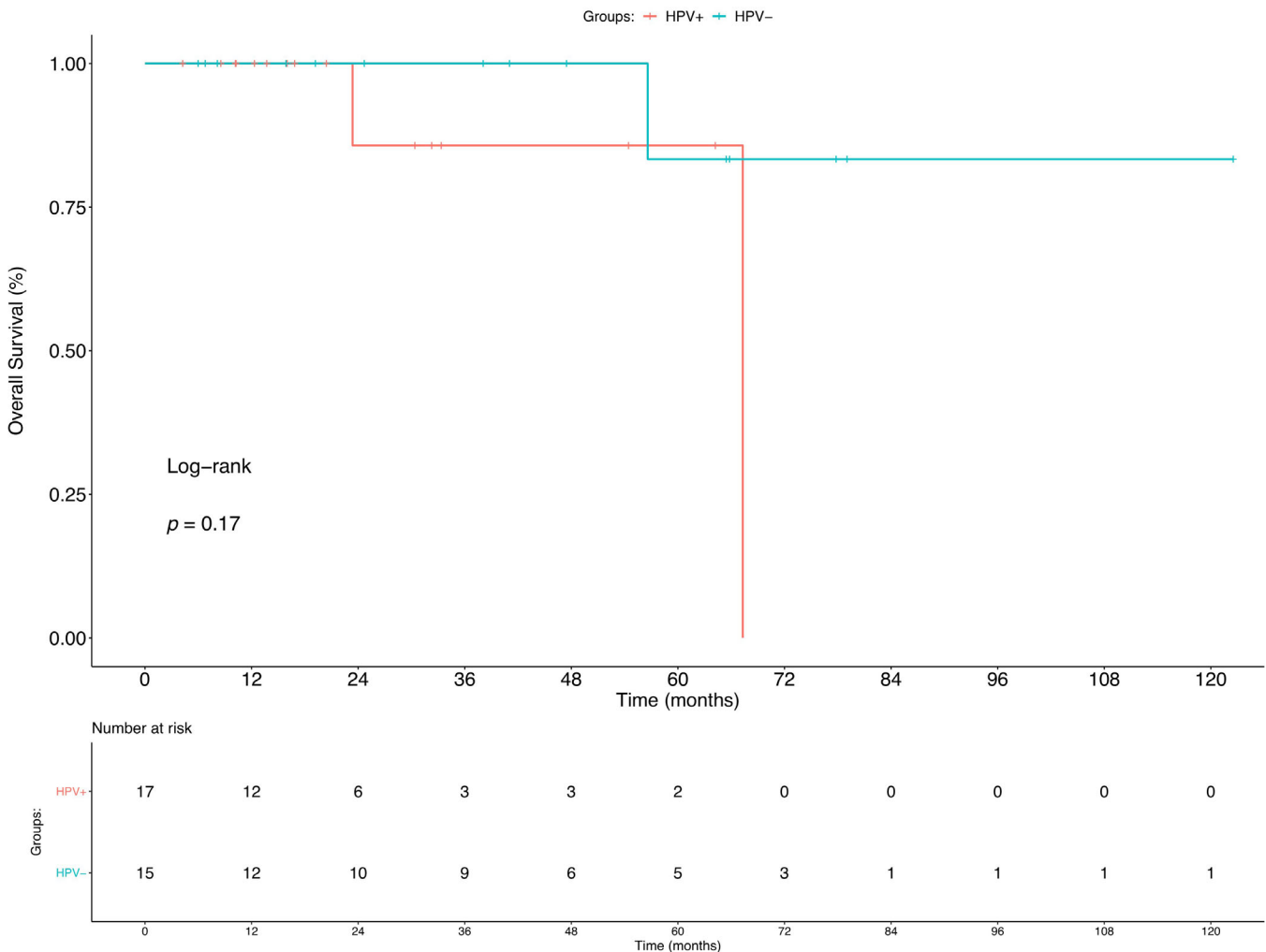


FIGURE 3 Overall survival by HPV status. HPV, human papilloma virus

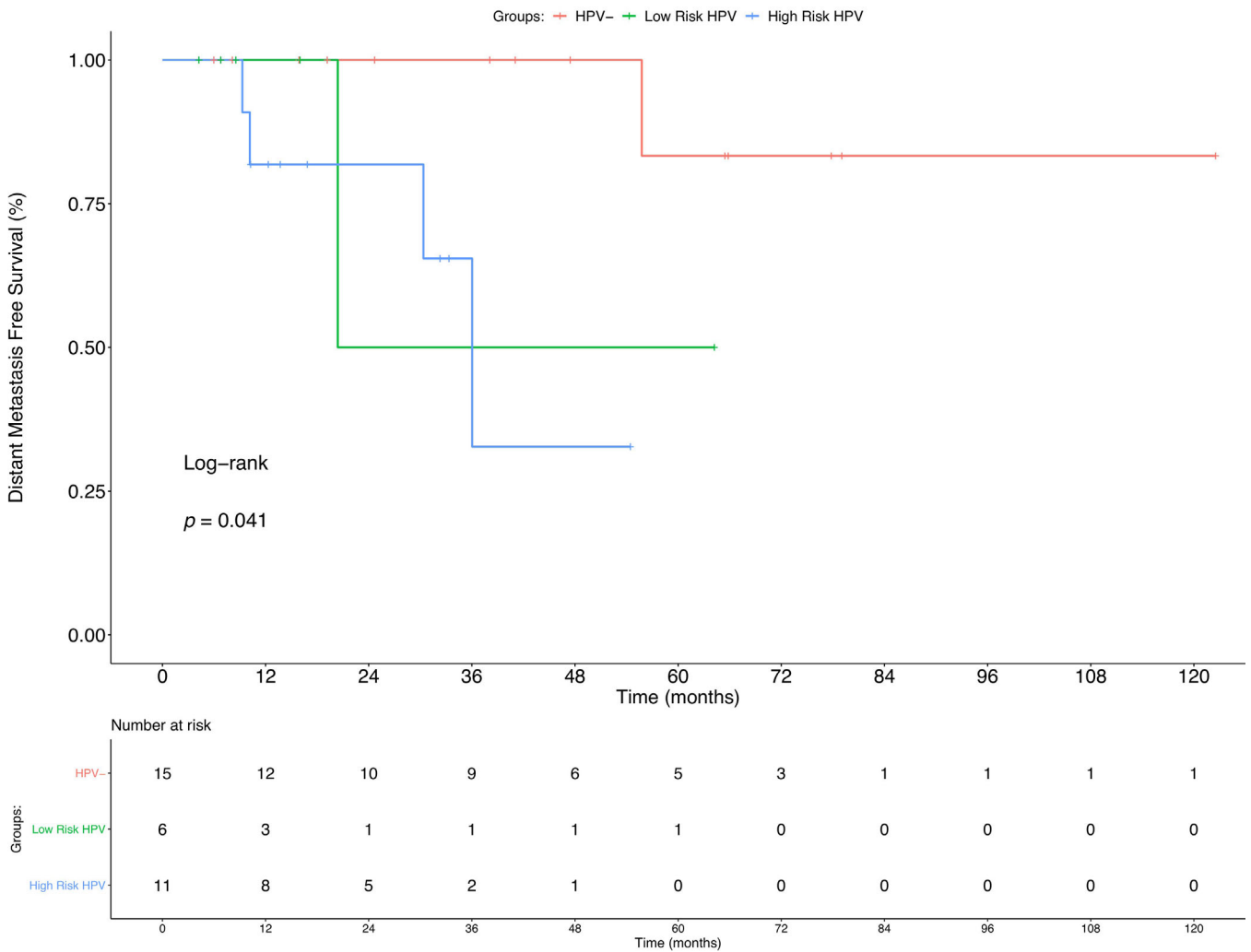


FIGURE 4 Time to metastasis by HPV high-risk subtype, HPV low-risk subtype, and HPV negative status. HPV, human papilloma virus

radiation (2/9 patients, 22%). The highest rate occurred in those who received surgery alone (3/5, 60% patients). Of the 26 patients that were treated with adjuvant radiation therapy (with or without chemotherapy), there was only 1 in-field failure, which occurred in a patient with sinonasal carcinoma who had received proton therapy.

4 | DISCUSSION

While HPV is a well-established positive prognostic indicator in oropharyngeal carcinoma, the prognostic value of HPV in sinonasal cancers has not been delineated, with previous studies showing conflicting results regarding a survival benefit associated with HPV-positive status in SNSCC. In this study, we characterized a population of patients with SNSCC at a single institution and analyzed disease control and survival outcomes based on HPV status. We found no significant differences in survival based on HPV status in this small cohort. HPV positive tumors were more likely to locally recur and

metastasize distantly, suggesting that HPV-positivity did not incur an advantage in this cohort. Treatment modality was also related to outcomes in this study, with possibly better local control observed in patients who received radiation therapy as compared to those who did not.

There are currently limited data available describing the prevalence and outcomes of SNSCC related to HPV status. Several of these small retrospective studies have associated HPV-positive status with better outcomes. For example, Alos et al and Larque et al demonstrated improved progression-free survival and overall survival in patients with HPV-positive tumors.^{7,8} Other studies have not shown a statistically significant difference in survival based on HPV status, although some demonstrated a trend toward better survival in patients with HPV-positive tumors.^{9,11} A larger analysis using the National Cancer Database (NCDB) by Kilic et al, which included 770 cases of SNSCC diagnosed from 2010 to 2014, found that HPV-positivity was associated with a better prognosis on multivariate analysis. They additionally found that nasal cavity tumors were more likely

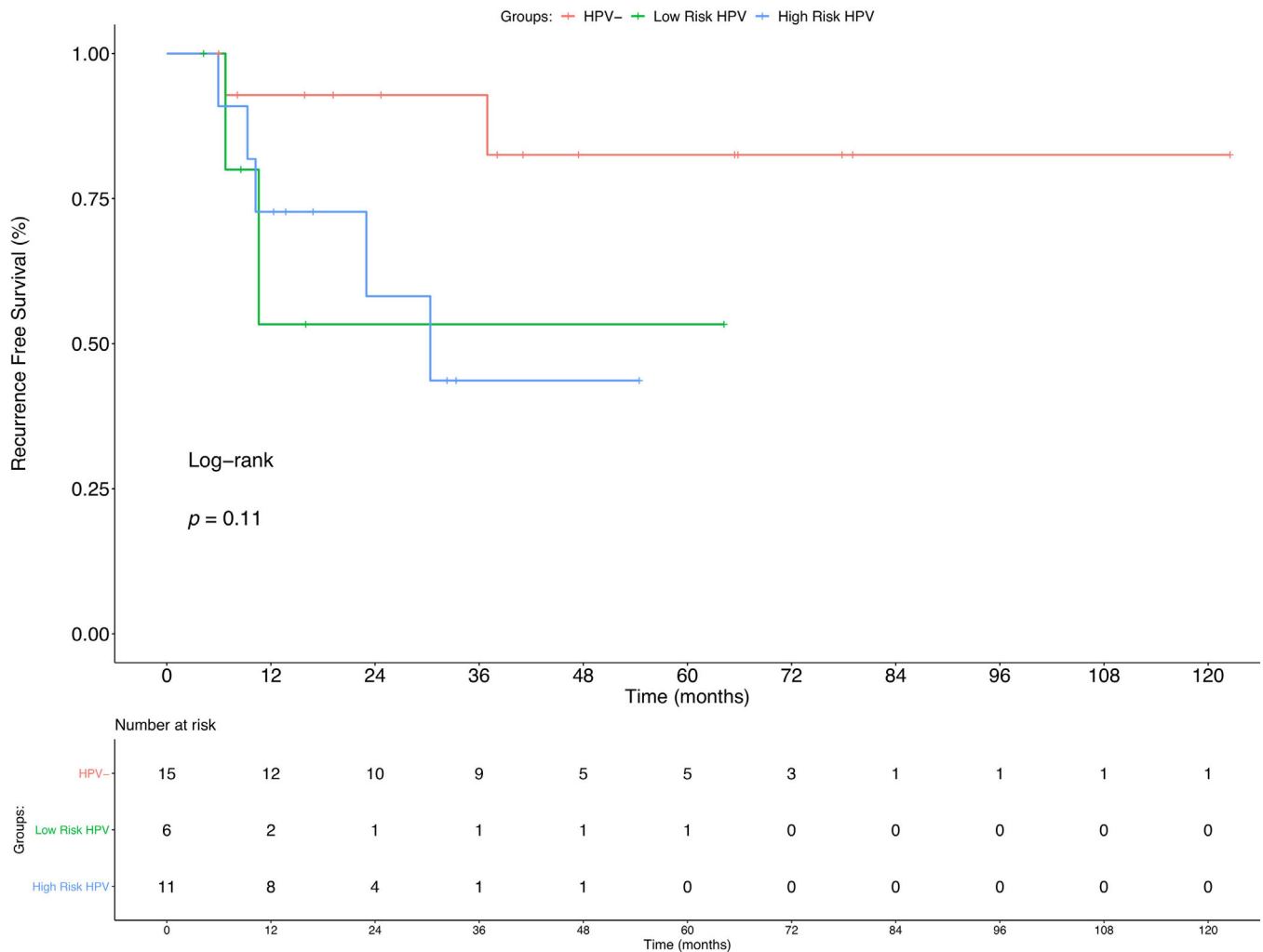


FIGURE 5 Time to local recurrence by HPV high-risk subtype, HPV low-risk subtype, and HPV negative status. HPV, human papilloma virus

to be HPV-positive than sinus tumors, with 49% of nasal cavity tumors found to be HPV-positive compared to 18% of sinus tumors.¹² A recent analysis of cases of SNSCC in the NCDB by Oliver et al found that HPV-positivity was associated with significant increases in overall survival in multivariable regression and propensity-score matched analyses. However, the authors noted that routine testing of HPV in patients with SNSCC is not commonly performed, limiting the conclusions that could be made concerning the link between HPV-positivity and overall survival.¹⁰

In this study, 65% of nasal cavity tumors were HPV-positive compared to 22% of sinus tumors. Among HPV-positive tumors, those of the nasal cavity had lower local recurrence rates than those with sinus tumors, although the number of patients was too small for appropriately powered statistical analysis. Differences in tumor site may explain the variability in outcomes seen in previous studies. While many studies did not report subgroup analyses by tumor site, Kiliç et al reported better overall survival in HPV-positive nasal cavity tumors but not in ethmoid or maxillary sinus tumors.¹²

Notably, over 50% of the patients in the current study had HPV-positive tumors, which was mainly determined by PCR rather than by p16 positive status. In the aforementioned studies, 20% to 33% of patients were found to have HPV-positive tumors.^{7,9,12} Other studies have also found similar rates. Udager et al found an HPV-positivity rate of 27.8% among SNSCCs, while Cabal et al found a rate of 20%.^{13,14} A 2013 systemic review and meta-analysis by Syrjänen and Syrjänen examining 492 SNSCCs across 35 studies found an HPV-positivity rate of 27%,¹⁵ while the previously mentioned study by Oliver et al found an HPV-positivity rate of 31.5%.¹⁰ A 2019 analysis by Sahane et al, however, detected HPV in 4/35 SNSCCs, for an HPV-positivity rate of 11.4%.¹⁶ While it is unclear why the rate of HPV-positivity is considerably higher in our study as compared to other published work, it may reflect the referral patterns to and the specific patient populations treated by our department.

This study was a small, single institution study, which may limit its generalizability. The study was underpowered to detect true differences between the HPV-positive and negative groups, including in

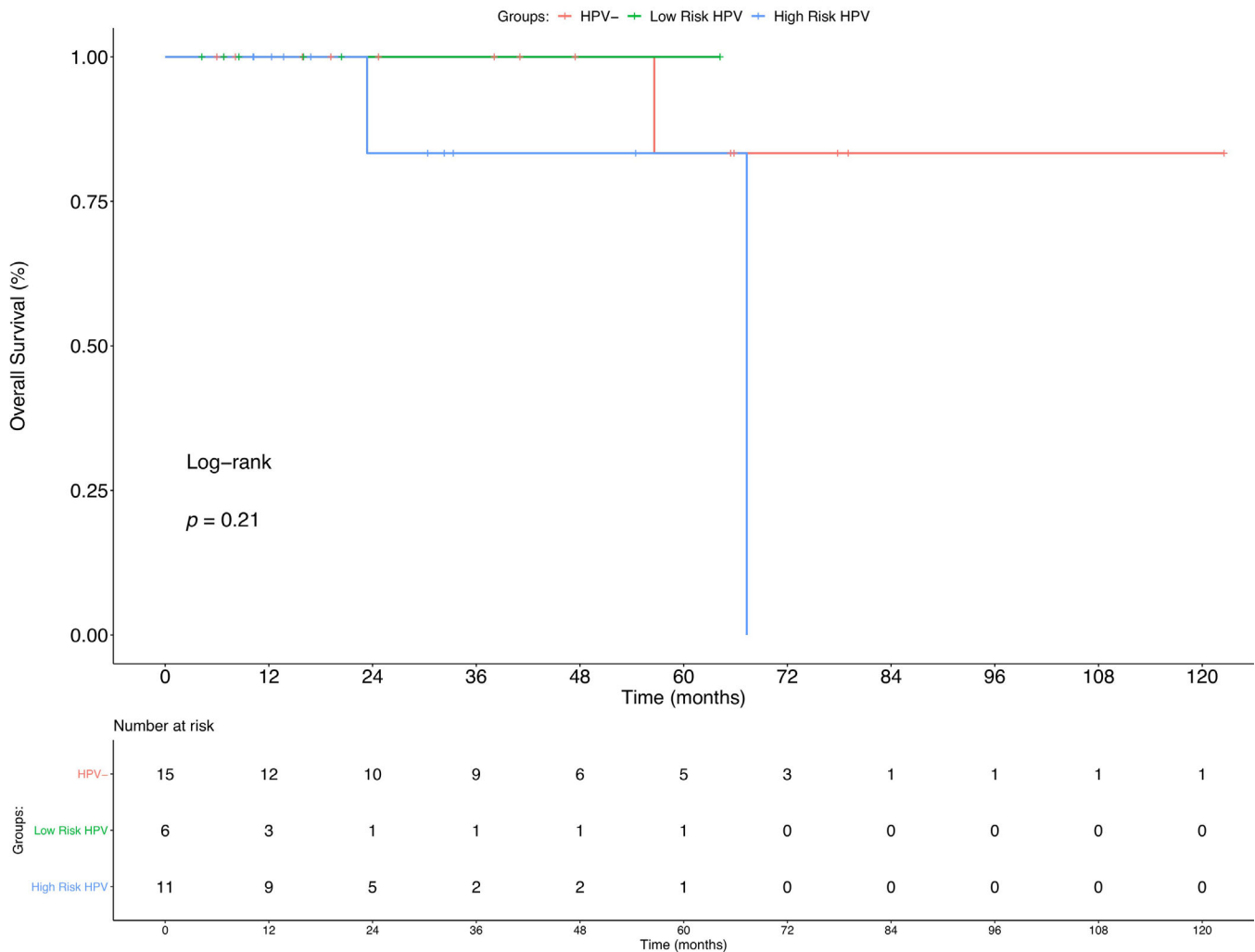


FIGURE 6 Overall survival by HPV high-risk subtype, HPV low-risk subtype, and HPV negative status. HPV, human papilloma virus

overall survival, even if they did exist. Its strengths lie in the fact that we were able to capture treatment-level details to describe differences in outcomes based on variations in treatment modalities, which has not been well described in previous studies. Additionally, this is one of the first studies to determine HPV status by PCR rather than using p16 positivity, which may not be an accurate proxy.

In conclusion, a significant proportion of sinonasal tumors appear to be associated with HPV, and testing for HPV might be justified in all cases of sinonasal cancers. Further research is needed to better elucidate the prognostic role of HPV status to guide management of SNSCC.

CONFLICT OF INTEREST

The authors declare no potential conflict of interest.

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REFERENCES

- Dutta R, Dubal PM, Svider PF, Liu JK, Baredes S, Eloy JA. Sinonasal malignancies: a population-based analysis of site-specific incidence and survival. *Laryngoscope*. 2015;125(11):2491-2497. <https://doi.org/10.1002/lary.25465>.
- Sanghvi S, Khan MN, Patel NR, Yeldandi S, Baredes S, Eloy JA. Epidemiology of sinonasal squamous cell carcinoma: a comprehensive analysis of 4994 patients. *Laryngoscope*. 2014;124(1):76-83. <https://doi.org/10.1002/lary.24264>.
- Llorente JL, López F, Suárez C, Hermsen MA. Sinonasal carcinoma: clinical, pathological, genetic and therapeutic advances. *Nat Rev Clin Oncol*. 2014;11(8):460-472. <https://doi.org/10.1038/nrclinonc.2014.97>.
- Lewis JS, Westra WH, Thompson LDR, et al. The sinonasal tract: another potential "hot spot" for carcinomas with transcriptionally-active human papillomavirus. *Head Neck Pathol*. 2014;8(3):241-249. <https://doi.org/10.1007/s12105-013-0514-4>.
- Gillison ML, Koch WM, Capone RB, et al. Evidence for a causal association between human papillomavirus and a subset of head and neck cancers. *J Natl Cancer Inst*. 2000;92(9):709-720. <https://doi.org/10.1093/jnci/92.9.709>.
- Syrjänen KJ. HPV infections in benign and malignant sinonasal lesions. *J Clin Pathol*. 2003;56(3):174-181. <https://doi.org/10.1136/jcp.56.3.174>.

7. Alos L, Moyano S, Nadal A, et al. Human papillomaviruses are identified in a subgroup of sinonasal squamous cell carcinomas with favorable outcome. *Cancer*. 2009;115(12):2701-2709. <https://doi.org/10.1002/cncr.24309>.
8. Larque AB, Hakim S, Ordi J, et al. High-risk human papillomavirus is transcriptionally active in a subset of sinonasal squamous cell carcinomas. *Mod Pathol*. 2014;27(3):343-351. <https://doi.org/10.1038/modpathol.2013.155>.
9. Bishop JA, Guo TW, Smith DF, et al. Human papillomavirus-related carcinomas of the sinonasal tract. *Am J Surg Pathol*. 2013;37(2):185-192. <https://doi.org/10.1097/PAS.0b013e3182698673>.
10. Oliver JR, Lieberman SM, Tam MM, et al. Human papillomavirus and survival of patients with sinonasal squamous cell carcinoma. *Cancer*. 2020;126(7):1413-1423. <https://doi.org/10.1002/cncr.32679>.
11. Laco J, Sieglóvá K, Vošmiková H, et al. The presence of high-risk human papillomavirus (HPV) E6/E7 mRNA transcripts in a subset of sinonasal carcinomas is evidence of involvement of HPV in its etiopathogenesis. *Virchows Arch*. 2015;467(4):405-415. <https://doi.org/10.1007/s00428-015-1812-x>.
12. Kılıç S, Kılıç SS, Kim ES, et al. Significance of human papillomavirus positivity in sinonasal squamous cell carcinoma. *Int Forum Allergy Rhinol*. 2017;7(10):980-989. <https://doi.org/10.1002/alr.21996>.
13. Udager AM, McHugh JB, Goudsmit CM, et al. Human papillomavirus (HPV) and somatic EGFR mutations are essential, mutually exclusive oncogenic mechanisms for inverted sinonasal papillomas and associated sinonasal squamous cell carcinomas. *Ann Oncol*. 2018;29(2):466-471. <https://doi.org/10.1093/annonc/mdx736>.
14. Cabal VN, Menendez M, Vivanco B, et al. EGFR mutation and HPV infection in sinonasal inverted papilloma and squamous cell carcinoma. *Rhinology*. 2020;58(4):368-376. <https://doi.org/10.4193/Rhin19.371>.
15. Syrjänen K, Syrjänen S. Detection of human papillomavirus in sinonasal carcinoma: systematic review and meta-analysis. *Hum Pathol*. 2013;44(6):983-991. <https://doi.org/10.1016/j.humpath.2012.08.017>.
16. Sahnane N, Ottini G, Turri-Zanoni M, et al. Comprehensive analysis of HPV infection, EGFR exon 20 mutations and LINE1 hypomethylation as risk factors for malignant transformation of sinonasal-inverted papilloma to squamous cell carcinoma. *Int J Cancer*. 2019;144(6):1313-1320. <https://doi.org/10.1002/ijc.31971>.

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