

Curative Radiotherapy for Oropharyngeal Squamous Cell Carcinoma Stratified by p16 Status, According to the Eighth Edition American Joint Committee on Cancer Staging Manual: A Retrospective Study

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Abstract. *Background/Aim:* The prevalence of oropharyngeal squamous cell carcinoma (OPSCC) is increasing worldwide. This retrospective study aimed to investigate the clinical outcomes of patients with OPSCC undergoing definitive radiotherapy, stratified according to their p16 status. *Patients and Methods:* A retrospective analysis was conducted on consecutive patients with OPSCC treated with curative external beam radiotherapy between May 2015 and September 2023. Clinical staging was determined by the eighth edition AJCC staging manual for p16 positive and negative OPSCC. All patients were treated with radiotherapy using a simultaneous integrated boost (SIB) with helical tomotherapy. The fractionation scheme, with or without chemotherapy, for the primary site and nodal lesions consisted of 2 Gy per fraction for a total dose of 70 Gy in 35 fractions over seven weeks. *Results:* This study included 76 patients with a median age of 66 years. With a median follow-up time of 32.6 months, the 3-year progression-free survival rate was significantly higher in p16 positive patients compared to p16 negative patients (79.6% vs. 42.5%, $p < 0.001$). Concerning 54 patients with p16-positive tumors, the overall survival rates indicated excellent clinical

outcomes for stage I, II, and III with results of 100%, 100%, and 88.1%, respectively. *Conclusion:* This retrospective study revealed the clinical outcomes of patients with OPSCC treated with radical radiotherapy, emphasizing the significance of p16 status. While acknowledging the limitations of the retrospective nature of this study, future prospective studies with larger cohorts and extended follow-up periods are needed to enhance evidence quality.

The worldwide prevalence of oropharyngeal squamous cell carcinoma (OPSCC) is increasing, and its oncogenesis is associated with tobacco exposure, alcohol consumption, and human papillomavirus (HPV) infection (1-3). HPV is implicated in the pathogenesis of a subset of OPSCC. Over the past decade, OPSCC linked to HPV has emerged as a distinct entity, separating it from conventional head and neck cancers (4, 5). This distinction is evident in various aspects including epidemiology, genetics, tumor behavior, and treatment prognosis (6).

Immunohistochemistry for p16 expression stands as the most commonly employed surrogate marker of HPV-related oropharyngeal carcinoma (7, 8). The p16 status was utilized in the staging of oropharyngeal carcinoma in the eighth edition of the American Joint Committee on Cancer (AJCC) Staging system (9). Recently, Mehanna *et al.* reported that individuals diagnosed with oropharyngeal cancer showing inconsistency between p16 and HPV status (either p16 negative/HPV+ or p16 positive/HPV-) have an intermediate prognosis between p16 positive/HPV+ and p16 negative/HPV- oropharyngeal cancer based on individual patient data from multicenter and multinational data analyses (10).

Current evidence suggests that OPSCC cases with positive p16 status exhibit a favorable response to treatment and improved overall survival compared to their p16 negative counterparts. However, there remains a need for more comprehensive investigations to delineate the nuances of the

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Key Words: Radiotherapy, oropharyngeal cancer, squamous cell carcinoma, retrospective study.

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clinical outcomes associated with definitive radiotherapy in the context of p16 status. This retrospective study aimed to bridge this gap by examining the clinical outcomes of patients with OPSCC who underwent definitive radiotherapy, stratified according to their p16 status.

Patients and Methods

We retrospectively analyzed consecutive patients diagnosed with OPSCC who underwent curative external beam radiotherapy with or without chemotherapy between May 2015 and September 2023. The inclusion criteria were histologically confirmed primary OPSCC, treatment with curative-intent radiotherapy, the absence of distant metastasis at diagnosis, and no prior definitive treatment or radiotherapy for the head and neck region. Exclusion criteria involved cases treated with radiotherapy with palliative intent and insufficient medical records to confirm or determine the treatment modality.

Clinical staging adhered to the eighth edition of the AJCC Staging Manual and was chosen for its demonstrated clinical utility in this study. Adverse events were graded using the National Cancer Institute Common Toxicity Criteria for Adverse Events, version 5.0. We used simultaneous integrated boost (SIB) radiotherapy with helical tomotherapy (Accuray, Sunnyvale, CA, USA) and image-guided radiotherapy (IGRT). One patient underwent computed tomography (CT) for radiation therapy planning while wearing a thermoplastic mask. The CT image data were reconstructed with a 2 mm slice thickness for the tomotherapy planning systems.

Clinical target volume 1 (CTV1) was defined as a microscopic invasive lesion of the primary tumor and the involved lymph nodes. CTV2 was defined as the ipsilateral side of positive lymph node metastasis in prophylactic irradiation (PI). CTV3 was defined as the volume remaining in the PI area. Each planning target volume (PTV) was defined as an extension of the CTV by 3 mm, which was cropped to a distance of 3 mm from the skin surface. The prescription dose for PTV1 was 70 Gy in 35 fractions, for PTV2 59.5 Gy in 35 fractions, and for PTV3 was 54 Gy in 35 fractions.

Statistical analysis was performed using the R statistical package. Fisher's exact test was used to compare patient characteristics between groups. Overall survival (OS), progression-free survival (PFS), and cancer-specific survival (CSS) rates were calculated using the Kaplan-Meier method from the first day of the initial therapy. PFS was defined as the time from the initial therapy to the first evidence of radiological or clinical tumor progression or death from any cause. Statistical significance was set at $p < 0.05$.

Results

Seventy-six patients were included in this retrospective study. The patients' attributions were presented in Table I. The median age at treatment was 66 years (range=40-84 years). In p16 positive and negative groups, the majority of individuals were male, with 73% and 83%, respectively. The median follow-up time was 32.6 months, ranging from 1.6 to 89.8 months. The 3-year OS rates for p16-negative and positive patients with OPSCC were 59.2% [95% confidence interval (CI)=28.7-80.2%] and 95.7% (95%CI=83.8-98.9%), respectively, demonstrating a statistically significant difference ($p=0.002$) (Figure 1A).

Table I. Patient attributes among a cohort of 76 individuals undergoing definitive radiotherapy for oropharyngeal squamous cell carcinoma stratified according to p-16 status.

Variables	Number (Percentage)		
	p16-	p16+	p-Value
Age			
Median	70	66	0.958
Range	45-83	40-84	
Sex			
Male	16 (73%)	45 (83%)	0.346
Female	6 (27%)	9 (17%)	
ECOG Performance Status			
0	14 (64%)	47 (87%)	0.0194
1	8 (36%)	6 (11%)	
2	-	1 (2%)	
Tumor subsite			
Tonsillar complex	10 (45%)	38 (70%)	0.0103
Base of tongue	7 (32%)	15 (28%)	
Soft palate	2 (9%)	1 (2%)	
Pharyngeal wall	3 (14%)	-	
Clinical stage			
I	3 (14%)	23 (43%)	<0.001
II	3 (14%)	10 (19%)	
III	5 (23%)	21 (39%)	
IV A/B	11 (50%)	-	
Chemotherapy			
No	7 (32%)	10 (19%)	0.00846
Induction	-	11 (20%)	
Concurrent	15 (68%)	25 (46%)	
Induction and concurrent	-	8 (15%)	
Synchronous or metachronous cancers			
No	9 (41%)	42 (78%)	0.00304
Yes	13 (59%)	12 (22%)	

The 3-year PFS for p16 negative and positive patients with OPSCC were 42.5% (95%CI=21.5-62.2%) and 79.6% (95%CI=64.0-89.0%) respectively, demonstrating a statistically significant difference ($p < 0.001$) (Figure 1B). The results of the univariate and multivariate analyses of PFS are summarized in Table II. In the univariate analysis, p16 status was associated with PFS [hazard ratio (HR)=0.261; 95%CI=0.111-0.611; $p=0.002$]. In the multivariate analysis, p16 status still remained the factor associated with PFS (HR=0.231; 95%CI=0.068-0.786; $p=0.019$).

Fifty-four patients were identified as p16-positive in this study. Their ages ranged from 40 to 84 years, with a median age of 66. Males constituted the majority of patients ($n=45$). The clinical stages were stage I in 23 patients, stage II in 10 patients, and stage III in 21 patients, according to the 8th AJCC staging system. The OS rates of stages I, II, and III were 100%, 100%, and 88.1% (95%CI=60.2-96.9%), respectively (Figure 2A). During the follow-up period,

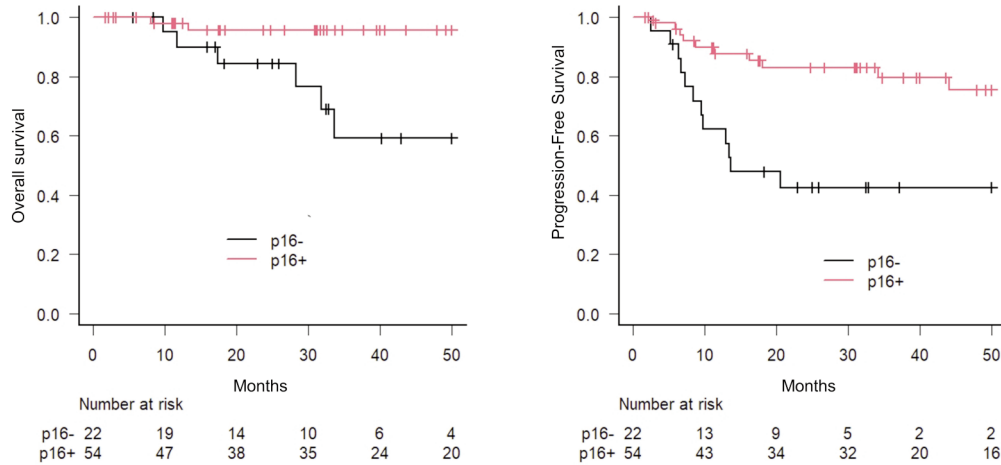


Figure 1. Kaplan–Meier curves for overall survival (A) and progression-free survival (B) after curative radiotherapy for the patients with oropharyngeal squamous cell carcinoma stratified by p-16 status.

Table II. Univariate and multivariate analyses for progression-free survival after definitive radiotherapy.

Covariables	Value	Univariate		Multivariate	
		Hazard ratio [95%CI]	p-Value	Hazard ratio [95%CI]	p-Value
Age	≤65 vs. >65 years old	0.681 [0.294-1.578]	0.370	0.282 [0.095-0.841]	0.023
Sex	Male vs. Female	0.710 [0.210-2.403]	0.582	0.972 [0.240-3.941]	0.968
Performance status	0 vs. 1-2	6.203 [2.623-14.670]	<0.001	4.356 [1.569-12.090]	0.005
Tumor subsite	Tonsillar complex vs. others	0.601 [0.235-1.538]	0.288	0.475 [0.170-1.331]	0.157
Clinical stage	I-II vs. III-IV A/B	2.092 [0.889-4.923]	0.091	1.426 [0.515-3.949]	0.495
Chemotherapy	No vs. Yes	0.384 [0.156-0.948]	0.038	0.466 [0.120-1.815]	0.271
Other cancers	No vs. Yes	1.378 [0.587-3.233]	0.462	0.540 [0.181-1.613]	0.270
p-16 status	p16- vs. p16+	0.261 [0.111-0.611]	0.002	0.231 [0.068-0.786]	0.019

cancer recurrence was observed in 10 of the 54 patients (18.5%). The PFS rate for three years was 89.9% (95%CI=65.3-97.4%) in stage I, 74.1% (95%CI=28.9-93.0%) in stage II, and 72.2% (95%CI=45.4-87.4%) in stage III (Figure 2B). The most frequent recurrence pattern was local recurrence, which was observed in four patients, followed by regional lymph node metastasis in three patients, and distant metastasis in three patients (Table III). Distant metastasis sites were identified in the lungs of two patients, and one patient exhibited multiple metastases involving both the lungs and liver.

Recurrence was identified in 11 of 22 patients with p16 negative patients. The most frequent recurrence pattern was distant recurrence in four patients, and all distant sites were the lungs. Regional lymph node and local recurrences were observed in three patients each, and the other patient exhibited complex recurrence involving both local and regional components.

Discussion

Our investigation aimed to assess the clinical outcomes of patients undergoing an SIB for OPSCC, stratified by p16 status. Our findings are consistent with those of previous studies. Gorphe *et al.* investigated 888 patients with T1-4, N0-3, and M0 oropharyngeal carcinoma (OPC) who underwent curative treatment at their center between 2011 and 2020 (11). The results showed that among these patients, 451 were p16-positive and 437 were p16-negative. The 5-year survival rate differed significantly between the two groups, with rates of 82.4% for p16 positive patients and 44% for p16 negative patients ($p<0.0001$, HR=0.24). Based on the results of the NRG Oncology RTOG 1016 trial, we investigated whether replacing cisplatin with cetuximab could reduce treatment toxicity in patients with HPV-positive OPSCC (12). The 5-year OS and PFS of the cisplatin arm were 84.6% and 78.4%, respectively.

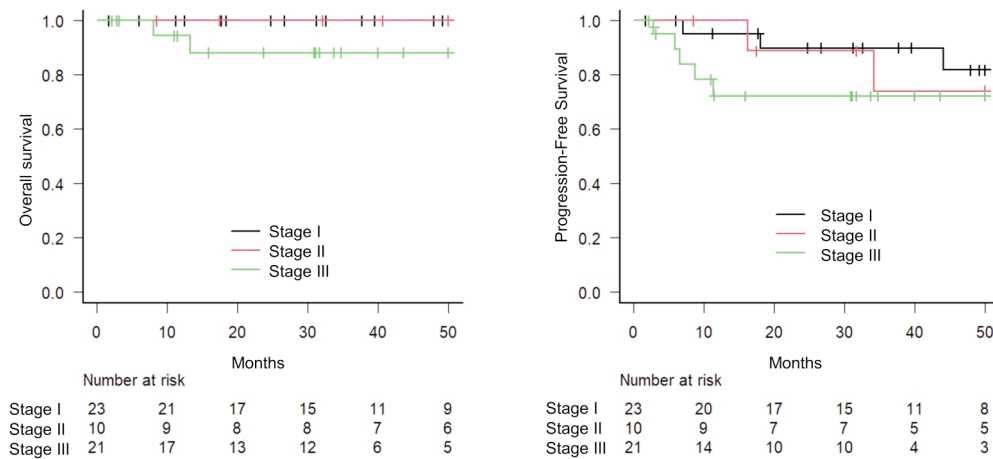


Figure 2. Kaplan–Meier curves for overall survival (A) and progression-free survival (B) after curative radiotherapy for patients with p-16 positive oropharyngeal squamous cell carcinoma stratified by clinical stage.

The concept of deintensification is a developing treatment strategy for individuals with HPV-associated OPSCC, aiming to uphold excellent oncological results while reducing the adverse effects associated with treatment. In a randomized phase II trial (ORATOR-2) involving 61 patients with HPV-associated OPSCC, dose-reduced definitive radiation (60 Gy of RT with weekly cisplatin) was compared between transoral robotic surgery (TORS) plus neck dissection and dose-reduced adjuvant RT (13). Despite an early closure due to two treatment-related deaths from TORS, dose-reduced definitive RT demonstrated improved two-year OS (100% *versus* 89%) and PFS (100% *versus* 84%) compared to TORS plus dose-reduced adjuvant RT, with similar rates of grade ≥ 2 toxicity (67% *versus* 71%) and overall quality of life at one year between the two treatment arms. Carbon-ion radiotherapy also emerges as a promising treatment avenue for oropharyngeal non-squamous cell carcinoma (14).

In a phase II trial, 43 patients diagnosed with HPV-associated OPSCC underwent deintensified chemoradiation with 60 Gy of intensity-modulated radiation therapy (IMRT) concurrently with weekly cisplatin (15). The trial demonstrated excellent clinical outcomes with three-year outcomes showing 100% rates of local control and a 95% OS rate.

Posttreatment surveillance is important for promptly identifying disease recurrence, enabling timely initiation of salvage therapy, and potentially improving patient outcomes. Chera *et al.* investigated the effectiveness of monitoring plasma circulating tumor HPV DNA (ctHPV-DNA) during the post-treatment surveillance of patients with HPV-associated OPC (16). Their findings indicated that two consecutive positive ctHPV-DNA tests have a high positive predictive value (94%) and negative predictive value (100%) for identifying disease recurrence, potentially facilitating earlier salvage therapy initiation.

Table III. First recurrence patterns following the administration of definitive radiotherapy.

Patterns of recurrence	Number (Percentage)	
	p16–	p16+
No recurrence	11 (50%)	44 (81%)
Local recurrence		
Local recurrence only	3 (14%)	4 (7%)
Local and regional recurrence	1 (5%)	-
Regional recurrence	3 (14%)	3 (6%)
Distant recurrence	4 (18%)	3 (6%)

The strength of our study is the homogeneity of the treatment modality, with SIB radiotherapy with helical tomotherapy using the same treatment protocol. However, our study has several limitations that warrant consideration. Reliance on retrospective data introduces limitations in terms of data completeness, accuracy, and the ability to control for all potential confounding variables. This may have affected the robustness of the statistical analyses and the interpretation of the results. Longer follow-up durations would provide a more comprehensive understanding of the survival patterns and late toxicities associated with treatment.

This retrospective study aimed to elucidate the clinical outcomes of patients with OPSCC treated with radical radiotherapy. The limitations of retrospective studies should be considered when interpreting their results and applying them in clinical practice. Future prospective studies with larger cohorts and longer follow-up periods are required to address these limitations and provide more robust evidence.

Conflicts of Interest

The Department of Comprehensive Radiation Oncology, to which Masanari Minamitani and Shingo Ohira belong, is an endowment department, supported by an unrestricted grant from Elekta K. K. and Chiyoda Technol Corporation. However, no funding was received for conducting this study.

Authors' Contributions

A.K., M.M., S.O., and H.Y. collectively devised the study and provided support in conducting data analyses. A.K. took the lead in composing the initial draft of the manuscript. M. M. played a significant role in analyzing and interpreting the data. Additionally, M.M., S.O., and H.Y. actively participated in preparing and critically reviewing the manuscript. The final version of the manuscript received approval from all Authors, who also committed to being accountable for all aspects of the work.

References

- Al-Jamaei AAH, Helder MN, Forouzanfar T, Brakenhoff RH, Leemans CR, de Visscher JGAM, van Dijk BAC: Age-group-specific trend analyses of oropharyngeal squamous cell carcinoma incidence from 1989 to 2018 and risk factors profile by age-group in 2015-2018: a population-based study in The Netherlands. *Eur J Cancer Prev* 31(2): 158-165, 2022. DOI: 10.1097/cej.0000000000000678
- Win Myint TT, McIvor N, Douglas R, Tin Tin S, Elwood M: Incidence, trends, and survival of oropharyngeal squamous cell cancer in Aotearoa New Zealand, 2006-2020. *Cancer Epidemiol* 85: 102393, 2023. DOI: 10.1016/j.canep.2023.102393
- Dalianis T: Human papillomavirus (HPV) and oropharyngeal squamous cell carcinoma. *Presse Med* 43(12): e429-e434, 2014. DOI: 10.1016/j.lpm.2014.08.010
- Smahel M, Nunvar J: Bioinformatics analysis of immune characteristics in tumors with alternative carcinogenesis pathways induced by human papillomaviruses. *Virol J* 20(1): 287, 2023. DOI: 10.1186/s12985-023-02241-6
- Tan Y, Wang Z, Xu M, Li B, Huang Z, Qin S, Nice EC, Tang J, Huang C: Oral squamous cell carcinomas: state of the field and emerging directions. *Int J Oral Sci* 15(1): 44, 2023. DOI: 10.1038/s41368-023-00249-w
- Ang KK, Harris J, Wheeler R, Weber R, Rosenthal DI, Nguyen-Tân PF, Westra WH, Chung CH, Jordan RC, Lu C, Kim H, Axelrod R, Silverman CC, Redmond KP, Gillison ML: Human papillomavirus and survival of patients with oropharyngeal cancer. *N Engl J Med* 363(1): 24-35, 2010. DOI: 10.1056/NEJMoa0912217
- El-Naggar AK, Westra WH: P16 expression as a surrogate marker for hpv-related oropharyngeal carcinoma: A guide for interpretative relevance and consistency. *Head Neck* 34(4): 459-461, 2012. DOI: 10.1002/hed.21974
- Doll C, Steffen C, Beck-Broichsitter B, Richter M, Neumann K, Pohrt A, Lohneis P, Lehmann A, Heiland M, Stromberger C, Cordes A, Johrens K, Raguse JD: The prognostic significance of p16 and its role as a surrogate marker for human papilloma virus in oral squamous cell carcinoma: an analysis of 281 cases. *Anticancer Res* 42(5): 2405-2413, 2022. DOI: 10.21873/anticancer.15719
- Machczyński P, Majchrzak E, Niewinski P, Marchlewska J, Golusiński W: A review of the 8th edition of the AJCC staging system for oropharyngeal cancer according to HPV status. *Eur Arch Otorhinolaryngol* 277(9): 2407-2412, 2020. DOI: 10.1007/s00405-020-05979-9
- Mehanna H, Taberna M, von Buchwald C, Tous S, Brooks J, Mena M, Morey F, Grønhoj C, Rasmussen JH, Garset-Zamani M, Bruni L, Batis N, Brakenhoff RH, Leemans CR, Baatenburg de Jong RJ, Klussmann JP, Wuerdemann N, Wagner S, Dalianis T, Marklund L, Mirghani H, Schache A, James JA, Huang SH, O'Sullivan B, Nankivell P, Broglie MA, Hoffmann M, Quabius ES, Alemany L, HNCIG-EPIC group: Prognostic implications of p16 and HPV discordance in oropharyngeal cancer (HNCIG-EPIC-OPC): a multicentre, multinational, individual patient data analysis. *Lancet Oncol* 24(3): 239-251, 2023. DOI: 10.1016/s1470-2045(23)00013-x
- Gorphe P, Classe M, Ammari S, Garcia G, Even C, Casiraghi O, Breuskin I, Tao Y, Temam S, Blanchard P, Moya-plana A: Patterns of disease events and causes of death in patients with HPV-positive versus HPV-negative oropharyngeal carcinoma. *Radiother Oncol* 168: 40-45, 2022. DOI: 10.1016/j.radonc.2022.01.021
- Gillison ML, Trotti AM, Harris J, Eisbruch A, Harari PM, Adelstein DJ, Jordan RCK, Zhao W, Sturgis EM, Burtneck B, Ridge JA, Ringash J, Galvin J, Yao M, Koyfman SA, Blakaj DM, Razaq MA, Colevas AD, Beitler JJ, Jones CU, Dunlap NE, Seaward SA, Spencer S, Galloway TJ, Phan J, Dignam JJ, Le QT: Radiotherapy plus cetuximab or cisplatin in human papillomavirus-positive oropharyngeal cancer (NRG Oncology RTOG 1016): a randomised, multicentre, non-inferiority trial. *Lancet* 393(10166): 40-50, 2019. DOI: 10.1016/S0140-6736(18)32779-X
- Palma DA, Prisman E, Berthelet E, Tran E, Hamilton S, Wu J, Eskander A, Higgins K, Karam I, Poon I, Husain Z, Enepekides D, Hier M, Sultanem K, Richardson K, Mlynarek A, Johnson-Obaseki S, Odell M, Bayley A, Dowthwaite S, Jackson JE, Dzienis M, O'Neil J, Chandarana S, Banerjee R, Hart R, Chung J, Tenenholtz T, Krishnan S, Le H, Yoo J, Mendez A, Winquist E, Kuruvilla S, Stewart P, Warner A, Mitchell S, Chen J, Parker C, Wehrli B, Kwan K, Theurer J, Sathya J, Hammond JA, Read N, Venkatesan V, MacNeil SD, Fung K, Nichols AC: Assessment of toxic effects and survival in treatment deescalation with radiotherapy vs transoral surgery for HPV-associated oropharyngeal squamous cell carcinoma: The ORATOR2 phase 2 randomized clinical trial. *JAMA Oncol* 8(6): 1-7, 2022. DOI: 10.1001/jamaoncol.2022.0615
- Kaneko T, Suefuji H, Koto M, Demizu Y, Saitoh JJ, Tsuji H, Okimoto T, Ohno T, Shioyama Y, Nemoto K, Nakano T, Kamada T: Multicenter study of carbon-ion radiotherapy for oropharyngeal non-squamous cell carcinoma. *In Vivo* 35(4): 2239-2245, 2021. DOI: 10.21873/invivo.12496
- Chera BS, Amdur RJ, Tepper JE, Tan X, Weiss J, Grilley-Olson JE, Hayes DN, Zanation A, Hackman TG, Patel S, Sheets N, Weissler MC, Mendenhall WM: Mature results of a prospective study of deintensified chemoradiotherapy for low-risk human papillomavirus-associated oropharyngeal squamous cell carcinoma. *Cancer* 124(11): 2347-2354, 2018. DOI: 10.1002/cncr.31338
- Chera BS, Kumar S, Shen C, Amdur R, Dagan R, Green R, Goldman E, Weiss J, Grilley-Olson J, Patel S, Zanation A, Hackman T, Blumberg J, Patel S, Thorp B, Weissler M, Yarbrough W, Sheets N, Mendenhall W, Tan XM, Gupta GP: Plasma circulating tumor HPV DNA for the surveillance of cancer recurrence in HPV-associated oropharyngeal cancer. *J Clin Oncol* 38(10): 1050-1058, 2020. DOI: 10.1200/JCO.19.02444

Received January 1, 2024

Revised January 22, 2024

Accepted January 23, 2024