Clinical prognostic factors to guide treatment strategy for HPV-positive oropharyngeal cancer using treatment outcomes of induction chemotherapy: A real-world experience

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Abstract. The role of induction chemotherapy (IC) in locally advanced oropharyngeal cancer (OPC) remains debatable, and suitable candidates for de-escalation treatment in these patients have not been fully identified. Therefore, the present study aimed to identify high-risk candidates for human papillomavirus (HPV)-positive OPC by analyzing patients who underwent IC followed by chemoradiotherapy (CRT) to guide optimal treatment strategies. Patients diagnosed with stage III-IVA OPC and treated with a minimum of two cycles of IC followed by CRT, between 2004 and 2020, were retrospectively reviewed. All the patients were restaged according to the American Joint Committee on Cancer, 8th edition. The overall response rate and survival outcomes associated with clinical factors based on HPV status were analyzed using univariate and multivariate analyses. The present study analyzed 105 patients with a median age of 60 years (range, 40-76 years). Among 105 patients, 40 (38.1%) were HPV-negative and 65 (61.9%) HPV-positive. In all patients, survival outcomes were notably poorer in patients aged ≥60 years (P=0.006) and those who did not achieve complete response post-CRT (P<0.001), irrespective of the HPV status. The median relative dose intensity of IC was ≥80%, indicating adequate treatment, regardless of age. In contrast to patients with HPV-negative OPC, age ≥60 years (P=0.011) and T4 stage (P=0.019) emerged as substantial poor prognostic factors for survival outcomes in patients with HPV-positive OPC. Patients with HPV-positive OPC were categorized into three groups based on the number of clinical factors at diagnosis (such as age and T4 stage). The progression-free and overall survival showed significant stratification across each group as the number of high-risk factors increased despite IC and CRT. The findings indicated that patients with these high-risk factors require a cautious therapeutic strategy even when they are diagnosed with HPV-positive OPC, and the role of combined modality, including IC, will need to be investigated in a randomized trial to be routinely incorporated into clinical practice.

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Introduction

It is predicted that there will be 58,450 new patients with head and neck squamous cell carcinoma (HNSCC) diagnoses in the United States in 2024 and more than 12,000 deaths (1). A comparable incidence was noted in Korea, where it constituted ~2.1% and ranked as the 10th most prevalent cancer among male patients (2). The most common cause of HNSCC is smoking, with human papillomavirus (HPV) contributing to oropharyngeal cancer (OPC) and Epstein-Barr virus linked to nasopharyngeal cancer among other associated causative factors. Notably, HPV-positive OPC is more prevalent among younger individuals, particularly in relatively high-income countries (3). Recently, there has been a growing trend in the prevalence of HPV-positive OPC among older individuals. A previous study demonstrated an increase in the median age at diagnosis from 53 to 58 years between 1998 and 2013, aligning with the observed trend in Korea (4-6). Furthermore, the incidence of HPV-positive OPC is increasing in both younger and older patients, with a noticeable shift in the burden towards older patients. However, it is noteworthy that most cases occur in patients aged <60 years.

HPV-positive OPC exhibits distinctive histopathological characteristics that impact clinical outcomes. The expression of viral E6 and E7 oncoproteins by HPV deactivates the tumor suppressor protein p53 and the retinoblastoma protein (pRb), respectively (7). Typically, HPV-positive OPC exhibits high expression of p16 protein and a lower prevalence of genetic mutations, such as those in the *TP53* (p53) gene, compared with HPV-negative OPC. These features are associated with chemo- or radiosensitivity (8). Moreover, HPV-positive OPC displays

a unique immune microenvironment characterized by $CD8^+$ T-cell activation and elevated programmed death-ligand 1 (PD-L1) expression, setting it apart from HPV-negative OPC. These cells can trigger an immune response; consequently, immunotherapy is used to treat HPV-positive OPC. The favorable clinical and biological characteristics of the HPV-positive OPC led to its downstaging in the 8th edition of the American Joint Committee on Cancer (AJCC) staging system (9). Moreover, distinct treatment strategies have been proposed in the National Comprehensive Cancer Network (NCCN) guidelines for HPV-positive OPC compared with HPV-negative OPC (10).

Given this context, a number of clinical studies have actively investigated de-intensified treatment approaches to mitigate the toxicity in patients with HPV-positive OPC (11,12). De-intensified treatments include measures such as reducing the chemotherapy dose, omitting induction chemotherapy (IC), lowering the radiation dose, and opting for less invasive surgery.

Clinical studies on de-intensified treatment have been attempted; however, there is insufficient evidence for selecting suitable patients. As a result, this approach is not widely used in clinical practice and not endorsed by the NCCN guidelines (13). Previously, it has been reported that patients with OPC with high p53 expression had inferior survival outcomes than those with low p53 expression among patients with HPV-positive OPC (14). These results suggested that even in patients with HPV-positive OPC, treatment results may differ due to a combination of variable etiologies, and it is necessary to identify high-risk factors that can predict survival and develop treatment strategies. These disparities between the theoretically favorable prognosis and practical evidence of HPV-positive OPC may derive from tumor heterogeneity in addition to staging, biological features and patient-specific characteristics.

Hence, in the present retrospective study, the authors aimed to identify clinical high-risk factors for HPV-positive OPC and conducted a survival analysis in patients with OPC treated with IC followed by chemoradiotherapy (CRT). The outcomes of the present study can provide insights into chemoresistance.

Patients and methods

Study population. Patients who underwent treatment for OPC at Chonnam National University Hwasun Hospital between June 2004 and October 2020 were retrospectively reviewed. Inclusion criteria were as follows: i) Patients with pathologically confirmed, HNSCC (stage II-III or stage IV M0); ii) underwent at least two cycles of IC followed by CRT; and iii) older than 19 years at the time of diagnosis. Patients with an initial diagnosis of stage IV M1 disease or nasopharyngeal cancer were excluded. Briefly, 100 (95%) were male and the median age at diagnosis was 60 years (range, 40-67). To be eligible for the study, their medical records needed to include information on staging using computed tomography (CT) or magnetic resonance imaging (MRI) along with ^{18}F-fluorodeoxyglucose positron emission tomography (^{18}F-FDG-PET) scans as necessary. Baseline patient demographics collected encompassed sex, age, Eastern Cooperative Oncology Group (ECOG) performance status score, body mass index (BMI) (15), smoking history, pathologic differentiation, chemotherapy regimen and cumulative radiation dose. To diagnose HPV-positive OPC, p16 and HPV status were confirmed through medical records. In cases where HPV was not tested, additional PCR using My HPV Chip Kit™ (AG Bio Diagnostics Co. Ltd.) was performed with archived tissue from Biobank of Chonnam National University Hwasun Hospital, a member of the Korean Biobank Network. The HPV DNA chip contained 12 types of high-risk HPV (HPV 16, 18, 31, 33, 35, 39, 45, 51, 52, 54, 56, 58) and 8 types of low-risk HPV (HPV 6, 11, 34, 40, 42, 43, 44, 54) strains. The PCR products from all samples were subjected to electrophoresis on 2.0% agarose gels, and size of HPV DNA products was 100 base pairs. After 10 μl of the amplified HPV product was denatured at 95˚C for 5 min, it was mixed with a hybridization solution and then applied to the DNA chip. Hybridization HPV DNA was visualized using a DNA chip scanner (GenePix 4000B; Molecular Devices, LLC). HPV amplicons were hybridized with the corresponding type of specific oligonucleotide probe and visualized on HPV DNA chip slides as double positive spots. Among patients diagnosed before 2017, eligible individuals were re-staged according to the AJCC 8th edition staging system based on HPV status. The protocol of the present study was approved (approval no. CNUHH-2023-232) by the Institutional Review Board of Chonnam National University Hwasun Hospital (Hwasun, Republic of Korea).

Treatment and tumor assessment. The patients received IC every 3 weeks, with a dose of 70 mg/m^2 docetaxel and 75 mg/m^2 cisplatin administered as a 4-h intravenous infusion on day 1. Additionally, 1,000 mg/m^2 5-fluorouracil was administered as a 24-h continuous infusion for 4 days. After IC, definitive treatment, such as CRT or surgery, was conducted based on the consensus of the multidisciplinary head and neck cancer team. CRT commenced within 4 weeks of IC completion, involving concurrent administration of cisplatin at a dose of 100 mg/m^2 every 3 weeks (on days 1, 22 and 43) or at a dose of 40 mg/m^2 weekly for 7 days (days 1, 8, 15, 22, 29, 36 and 43). Radiotherapy (RT) was administered using three-dimensional conformal radiation therapy (3D-CRT) or intensity-modulated RT (IMRT). Response was evaluated by the Response Evaluation Criteria in Solid Tumors 1.1 and involved assessing complete response (CR), partial response, stable disease and progressive disease after IC and 8 weeks post CRT completion (16). Alongside imaging studies, all patients underwent a physical examination by an otolaryngologist to validate the response evaluation. Patients who achieved a CR upon physical examination and CT or MRI scan, underwent ^{18}F-FDG-PET scans for confirmation 1 month after CR confirmation. Most patients were evaluated for response by CT, but in cases of radiation-induced swelling on CT, it was difficult to assess the response, thus in these cases, MRI was performed. After completing treatment, patients underwent monthly follow-ups with physical examinations. CT or MRI scans were conducted every 4 months for the initial 2 years, followed by biannual scans until disease progression.

Statistical analysis. Overall survival (OS) was characterized as the time from diagnosis to death. Progression-free survival

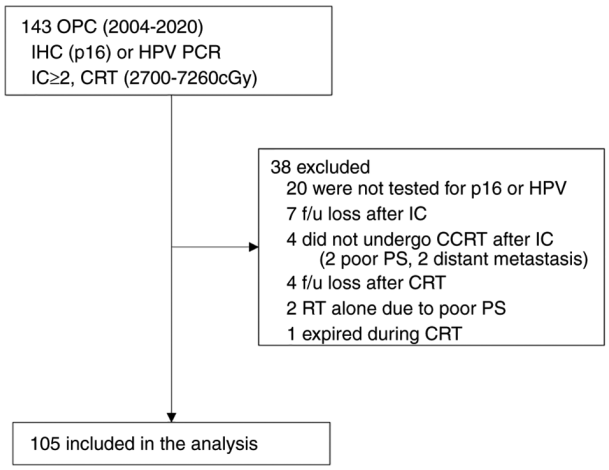

Figure 1. Flow diagram of patient selection. OPC, oropharyngeal cancer; IHC, immunohistochemistry; HPV, human papillomavirus; IC, induction chemotherapy; CRT, chemoradiotherapy; cGy, centigray; f/u, follow-up; RT, radiotherapy; PS, performance score.

(PFS) was defined as the time from diagnosis to death or the initially documented recurrence, further categorized as locoregional recurrence (tumor at the primary site or regional nodes) or distant metastasis. Patient characteristics were compared using the Chi-square test, Fisher's exact test and unpaired Student's t-test. Continuous variables are expressed as median, and categorical variables are presented in terms of frequency and percentage values. Survival analysis utilized Kaplan-Meier survival curves, and the log-rank test was used for comparison. Univariate and multivariate survival analyses were conducted using Cox proportional hazards. The level of significance adopted in all analysis was 5% with a 95% confidence interval and $P<0.05$ was considered to indicate a statistically significant difference. Statistical analyses were performed using R, version 4.2.2 (R Foundation for Statistical Computing; https://www.r-project.org/).

Results

Patient characteristics. The present study included 105 patients, comprising 40 HPV-negative and 65 HPV-positive individuals (Fig. 1). Baseline patient characteristics are presented in Table I. The median follow-up period for surviving patients was 60 months, and the median age at diagnosis was 60 years (range, 40-76 years). The study population primarily comprised males (95.2%), with 74.3% having a smoking history. In comparison to patients with HPV-negative OPC, those with HPV-positive OPC were younger (P=0.046), showed a lower percentage of N2 stage (P=0.024), had a less frequent smoking history (P=0.049), and a higher incidence of poorly differentiated tumors (P=0.001). Upon restaging according to the AJCC 8th edition staging system, 51 out of 65 patients (78%) with HPV-positive OPC were down-staged from their initial diagnosis, indicating a higher proportion of patients in the early stages of disease (P<0.01) compared with HPV-negative OPC. Specifically, 3 individuals were down-staged from stage III to stage II, 10 were down-staged from stage IV to stage III and 38 were downgraded from stage IV to stage II.

Treatment delivery and outcomes. Of the total participants, 102 (97.1%) underwent three cycles of IC. At the same time, 3 patients received additional cycles beyond the initial three to elicit further responses due to a minor response after the initial treatment. There was no significant difference in the relative dose intensity between patients with HPV-negative and HPV-positive OPC (HPV-negative vs. HPV-positive; docetaxel, 92.9 vs. 91.6%; cisplatin, 92.2 vs. 89.5; and 5-FU, 92.1 vs. 91.3%). Most patients (N=99, 94.3%) who completed the planned IC cycle advanced to CRT and 18 patients (17%) underwent surgery following CRT. A total of 6 patients discontinued the scheduled cycle of CRT; 2 experienced acute kidney injury and 4 declined planned CRT due to grade 3 oral mucositis, chemotherapy-induced nausea and vomiting, and odynophagia. Among them, 5 out of 6 patients who discontinued planned CRT had HPV-positive OPC. The median cumulative radiation dose was 66 Gray (Gy) (range, 27.0-72.6 Gy) and the median cumulative dose of cisplatin was 200 mg/m^2 (range, 60-300 mg/m^2). There were no significant differences observed between HPV-negative OPC and HPV-positive OPC, nor between older (≥60 years) and younger patients (<60 years) with a median age of 60 years. Additionally, all but three patients received radiation with a cumulative dose exceeding 50 Gy. RT modalities comprised 3D-CRT in 52 patients (50%) and IMRT in 53 (50%).

CR to IC was observed in 29 patients (27.6%), with 68 (64.8%) showing PR. Following CRT, CR was achieved in 87 patients (82.8%), and 12 (11.4%) exhibited PR. The overall response rates after IC followed by CRT, detailed in Table I, revealed no significant difference between HPV-negative and HPV-positive OPC (92.5 vs. 95.4%).

Survival analysis and patterns of recurrence. Throughout the follow-up period, recurrence was observed in 34 patients (32.4%), with 67.6% experiencing locoregional recurrence and 32.4% developing distant metastases (lung, liver, or bone). No significant differences were noted in the patterns of locoregional recurrence and distant metastasis between HPV-positive and HPV-negative OPC. The 3-year PFS for HPV-negative OPC was 57.4% [95% confidence interval (CI): 42.11-72.69] and 69.2% (95% CI: 58.03-80.37) in HPV-positive OPC. Additionally, 5-year OS was 60.9% in HPV-negative OPC (95% CI: 45.42-76.38) and 77.8% in HPV-positive OPC (95% CI: 67.41-88.19) (data not shown). While the 3-year PFS and 5-year OS exhibited a more favorable trend in HPV-positive OPC compared with HPV-negative OPC, neither the 3-year PFS (P=0.172) nor the 5-year OS (P=0.064) exhibited a statistically significant association with HPV status (Fig. 2A and B). To assess treatment outcomes based on HPV status and stage, patients were grouped into four categories: HPV-positive/stage II, HPV-negative/stage III, HPV-positive/stage II and HPV-negative/stage IV. A decline in survival outcomes with a higher stage for HPV-positive OPC and HPV-negative OPC in the 3-year PFS (P=0.005) and the 5-year OS (P=0.006) was observed (Fig. 2C and D). Notably, the survival outcomes of HPV-positive/stage III and HPV-negative/stage IV were comparable in the 3-year PFS and 5-year OS. Furthermore, the outcomes in HPV-positive/stage II and HPV-negative/stage III were similar, with no statistically significant differences among group. However, when the groups

Table I. Patient characteristics and response rate.

Characteristics	All (N=105)	HPV-negative (N=40)	HPV-positive (N=65)	P-value
Male	100 (95)	40 (100)	60 (92)	0.154
Age, years				
Median	60	62	57	
<60	55 (52)	16 (40)	39 (60)	[a]0.046
≥60	50 (48)	24 (60)	26 (40)	
WHO PS				
0	29 (28)	9 (22)	20 (31)	0.380
1	76 (72)	31 (78)	45 (69)	
BMI, kg/m^2				
Normal	45 (43)	17 (43)	28 (43)	0.136
Overweight	45 (43)	14 (35)	31 (48)	
Low	15 (14)	9 (23)	6 (9)	
Smoking status, pk-yrs				[a]0.049
No	27 (26)	6 (15)	21 (32)	
Yes	78 (74)	34 (85)	44 (68)	
<20	12 (15)	4 (12)	8 (18)	0.536
≥20	66 (85)	30 (88)	36 (82)	
Differentiated				[a]0.001
WD	29 (28)	19 (48)	10 (15)	
MD	40 (38)	15 (38)	25 (39)	
PD	22 (21)	5 (13)	17 (26)	
Unknown	14 (13)	1 (3)	13 (20)	
Stage				[a]<0.01
II	53 (51)	0 (0)	53 (82)	
III	18 (17)	6 (15)	12 (18)	
IV	34 (32)	34 (85)	0 (0)	
Tumor status				0.361
T1	8 (8)	4 (10)	4 (6)	
T2	40 (38)	18 (45)	22 (34)	
T3	37 (35)	10 (25)	27 (42)	
T4	20 (19)	8 (20)	12 (18)	
Node status				[a]0.024
N0	5 (5)	0 (0)	5 (8)	
N1	37 (35)	10 (25)	27 (41)	
N2	63 (60)	30 (75)	33 (51)	
Total radiation dose, cGy				0.819
<6,600	27 (26)	11 (27)	16 (35)	
≥6,600	78 (74)	29 (73)	49 (75)	
Response of IC				
ORR	92.4	92.5	90.3	>0.999
CR	29 (28)	11 (28)	18 (28)	>0.999
PR	68 (65)	26 (65)	42 (63)	
SD	8 (8)	3 (8)	5 (8)	
Response after CRT				
ORR	94.3	92.5	95.4	0.672
CR	87 (83)	32 (80)	55 (85)	0.088
PR	12 (11)	5 (13)	7 (11)	
SD	3 (3)	3 (8)	0 (0)	
PD	3 (3)	0 (0)	3 (5)	

[a]Indicates a statistically significant difference. Normal, 18.5-22.9 kg/m^2; Overweight, ≥23 kg/m^2; Low, <18.5 kg/m^2. HPV, human papillomavirus; WHO PS, World Health Organization performance status; pk-yrs, pack-years; WD, well differentiated; MD, moderately differentiated; PD, poorly differentiated; cGy, centigray; IC, induction chemotherapy; ORR, overall response rate; CR, complete response; PR, partial response; SD, stable disease; CRT, chemoradiotherapy; PD, progressive disease.

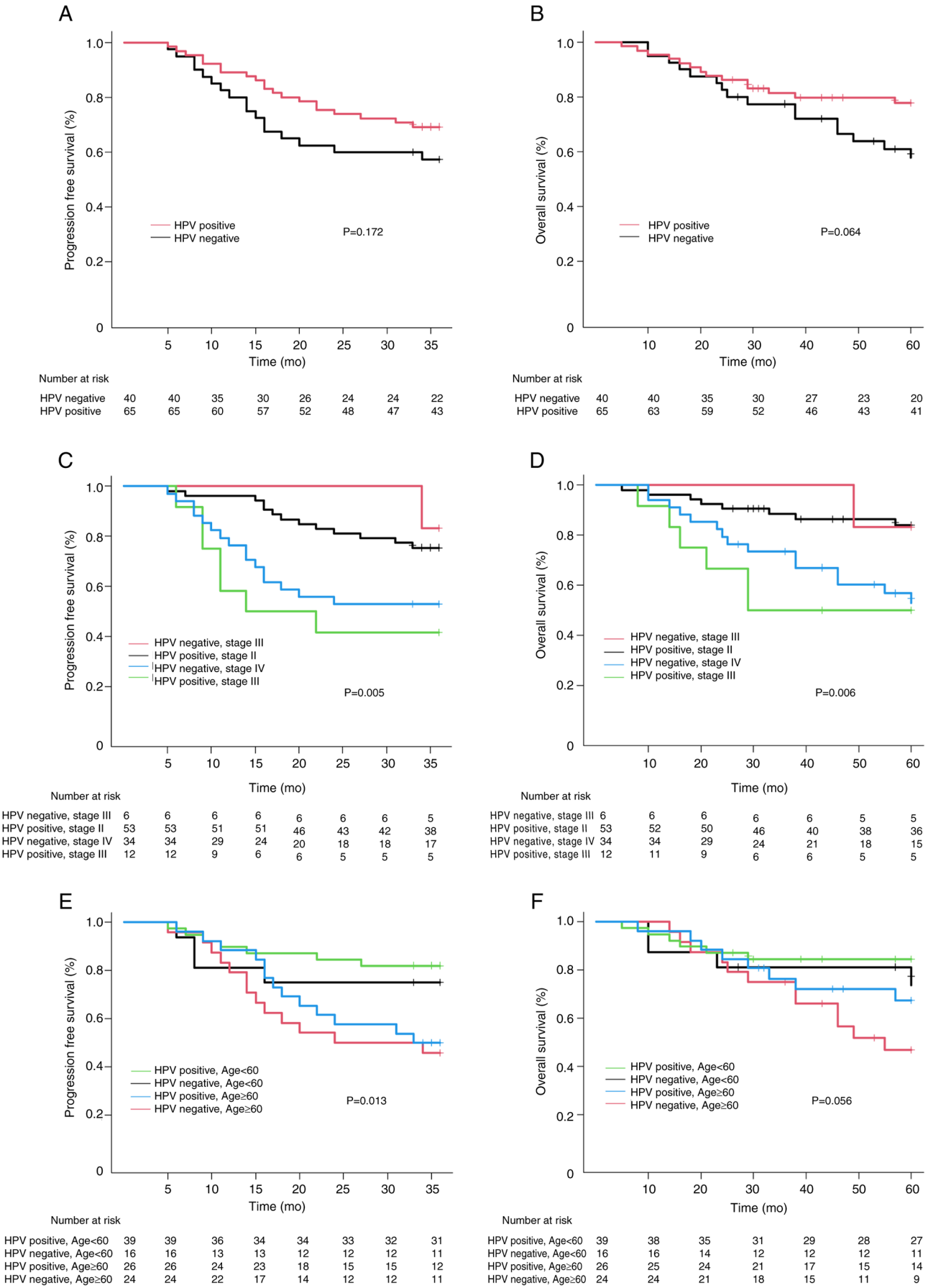

Figure 2. Kaplan-Meier estimates of (A) PFS and (B) OS in all patients with HPV-associated OPC. Kaplan-Meier estimates of (C) PFS and (D) OS in all patients with OPC with regard to HPV status and American Joint Committee on Cancer 8th staging of OPC. Kaplan-Meier estimates of (E) PFS and (F) OS in all patients with OPC with regard to HPV status and age. PFS, progression-free survival; OS, overall survival; HPV, human papillomavirus; OPC, oropharyngeal cancer; mo, months.

Table II. Uni- and multivariate analyses of survival outcomes in all patients, HPV-negative OPC and HPV-positive OPC for progression-free survival.

	All patients						HPV-negative OPC						HPV-positive OPC					
	Univariate			Multivariate			Univariate			Multivariate			Univariate			Multivariate		
Characteri stics	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI
Male	1.77	0.572	0.24-12.9										1.51	0.690	0.20-11.2			
Age, years																		
<60																		
≥60	[a]2.96	[a]0.003	[a]1.46-6.00	[a]2.70	[a]0.006	[a]1.33-5.50	2.35	0.136	0.76-7.21				[a]3.18	[a]0.014	[a]1.27-8.00	[a]3.38	[a]0.011	[a]1.32-8.63
WHO PS																		
0																		
1	1.31	0.441	0.66-2.61				0.64	0.407	0.23-1.83				0.79	0.627	0.32-1.99			
BMI, kg/m^2																		
Normal	1						1						1					
Overweight	0.53	0.089	0.26-1.10				0.40	0.402	0.12-1.31				0.66	0.386	0.26-1.68			
Low	0.96	0.924	0.38-2.40				0.81	0.806	0.25-2.62				0.93	0.921	0.20-4.23			
Smoking, pk-yrs																		
No	1						1						1					
<20	0.81	0.762	0.22-3.07				0.68	0.749	0.06-7.46				0.85	0.844	1.17-4.22			
≥20	1.43	0.375	0.65-3.16				1.40	0.656	0.62-6.17				1.22	0.686	0.46-3.26			
Tumor status																		
T0-T3																		
T4	2.02	0.058	0.98-4.18				0.84	0.858	0.26-3.11				[a]3.59	[a]0.007	[a]1.43-9.08	[a]3.16	[a]0.019	[a]1.21-8.27
Node status																		
N0-1[b], N1-N2b[c]																		
N2[b], N2c[c]	[a]2.10	[a]0.045	[a]1.02-4.34				2.63	0.057	0.97-7.14	[a]3.02	[a]0.035	[a]1.08-8.43	1.54	0.345	0.63-3.77			
Total radiation, cGy																		
<6,600	1.01	0.985	0.49-2.08				1.03	0.956	0.36-2.93				1.01	0.992	0.37-2.77			
≥6,600																		
After IC, non-CR																		
Overall response	1.90	0.127	0.83-4.32				2.10	0.245	0.60-7.30				1.77	0.306	0.59-5.31			
Primary	[a]1.95	[a]0.043	[a]1.02-3.73				2.09	0.137	0.79-5.49				2.03	0.114	0.84-4.89			
LNs	2.00	0.098	0.88-4.56				2.43	0.164	0.70-8.45				1.77	0.306	0.59-5.31			

Table II. Continued.

	All patients						HPV-negative OPC						HPV-positive OPC					
	Univariate			Multivariate			Univariate			Multivariate			Univariate			Multivariate		
Characteristics	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI
After CRT																		
CR																		
Non-CR	[a]4.71	[a]<0.01	[a]2.38-9.33	[a]4.29	[a]<0.01	[a]2.16-8.51	[a]4.12	[a]0.006	[a]1.50-11.3	[a]4.68	[a]0.004	[a]1.64-13.4	[a]5.15	[a]0.001	[a]2.03-13.0	[a]3.88	[a]0.005	[a]1.50-10.0

[a]Indicates a statistically significant difference; [b]All patients and HPV-positive OPC; [c]HPV-negative OPC. Normal, 18.5-22.9 kg/m^2; Overweight, 23≥kg/m^2; Low, <18.5 kg/m^2. HPV, human papillomavirus; OPC, oropharyngeal cancer; pk-yrs, pack years; HR, hazard ratio; CI, confidence interval; WHO PS, World Health Organization performance status; BMI, body mass index; cGy, centigray; CR, complete response; non-CR, non-complete response; IC, induction chemotherapy; LNs, lymph nodes; CRT, chemoradiotherapy.

A of HPV-negative/stage III and HPV-positive/stage II and B of HPV-negative/stage IV and HPV-positive/stage III were compared, both PFS and OS were significantly worse in group A, which had a relatively poor prognosis (P=0.001) (data not shown).

Risk factors associated with survival outcomes. To identify distinct clinical prognostic factors in HPV-positive OPC, factors related to survival outcomes based on HPV status were explored. It was found that individuals ≥60 years of age and non-CR after CRT were linked to a higher risk for PFS in all patients compared with patients <60 years of age and CR after CRT (Table II). Regarding OS, non-CR at the primary site following IC, and non-CR after CRT were identified as significant adverse factors, while being overweight emerged as a favorable factor (Table III). Although age had no statistical significance for OS in the multivariate analysis, this data revealed a close relationship between survival outcomes and age. Hence, the survival outcomes were examined based on the median age of 60 years (<60 vs. ≥60 years) to ascertain the association depending on HPV status. It was demonstrated that PFS (P=0.013) and OS (P=0.056) were poorer in patients aged ≥60 years than in those <60 years (Fig. 2E and F).

In addition to the aforementioned data, Tables II and III revealed significant clinical factors in HPV-negative and HPV-positive OPC. Non-CR after CRT emerged as an adverse risk factor for survival in HPV-negative OPC. In contrast to HPV-negative OPC, age ≥60 years, T4 stage and non-CR after CRT were identified as significant risk factors for PFS, while T4 stage and non-CR at the primary site after IC were associated with significantly worse survival outcomes for OS in patients with HPV-positive OPC. Utilizing these results, patients with HPV-positive OPC were categorized into three groups based on the number of clinical risk factors at diagnosis (age and T4 stage). As depicted in Fig. 3, the PFS and OS demonstrated significant stratification across each group as the number of clinical risk factors was increased.

Discussion

Theoretically, HPV-positive OPC is expected to exhibit more favorable survival outcomes than HPV-negative OPC due to its better responsiveness to chemotherapy (7,17). Contrary to previous studies (7,17), the data of the present study revealed no significant survival difference between the two groups. This could be because the tumorigenesis of HPV-positive OPC involves multiple confounding factors, such as smoking, alcohol consumption and environmental factors, alongside viral pathogens in head and neck cancer (18,19). Consequently, using a single factor such as HPV status to predict treatment response in all cases is challenging. Furthermore, interpersonal or intrapersonal heterogeneity can lead to varied treatment outcomes depending on host factors, even if it involves a single pathogen. To identify the clinical factors in patients with HPV-positive OPC that exhibited a poor prognosis comparable with those in patients with HPV-negative OPC, each group was analyzed. Therefore, it is crucial to consider known pathogens and additional clinical indicators when evaluating prognostic factors.

Table III. Uni- and multivariate analysis of survival outcomes in all patients, HPV-negative OPC and HPV-positive OPC for overall survival.

	All patients						HPV-negative OPC						HPV-positive OPC					
	Univariate			Multivariate			Univariate			Multivariate			Univariate			Multivariate		
Characteristics	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI
Male	1.44	0.720	0.19-10.6										0.99	0.992	0.13-7.57			
Age, years																		
<60																		
≥60	[a]2.34	[a]0.028	[a]1.09-5.01	2.06	0.068	0.95-4.49	2.27	0.159	0.73-7.07				2.03	0.190	0.70-5.86			
WHO PS																		
0																		
1	0.58	0.146	0.27-1.21				0.481	0.177	0.17-1.39				0.54	0.257	0.19-1.56			
BMI, kg/m^2																		
Normal	1			1			1						1					
Overweight	[a]0.29	[a]0.011	[a]0.12-0.76	[a]0.28	[a]0.009	[a]0.11-0.73	0.39	0.159	0.10-1.45				[a]0.26	[a]0.044	[a]0.07-0.96			
Low	1.26	0.613	0.52-3.03	1.51	0.370	0.62-3.69	1.17	0.776	0.38-3.61				1.07	0.936	0.23-4.94			
Smoking, pk-yrs																		
No	1						1						1					
<20	1.75	0.403	0.47-6.53				1.62	0.632	0.23-11.5				1.67	0.573	0.28-10.0			
≥20	1.71	0.283	0.64-4.52				1.19	0.818	0.27-5.34				1.77	0.390	0.48-6.56			
Tumor status																		
T0-T3																		
T4	1.93	0.112	0.86-4.33				0.65	0.570	0.15-2.87				[a]4.19	[a]0.008	[a]1.45-12.1	[a]3.15	[a]0.042	[a]1.04-9.53
Node status																		
N0-1[b], N1-N2b[c]																		
N2[b], N2c[c]	[a]2.93	[a]0.019	[a]1.19-7.16				2.18	0.150	0.75-6.30				1.79	0.297	0.59-5.34			
Total radiation, cGy																		
<6,600	0.98	0.969	0.44-2.21				1.23	0.725	0.39-3.80				0.89	0.847	0.28-2.85			
≥6,600																		
After IC, non-CR																		
Overall response	[a]3.03	[a]0.039	[a]1.06-8.69				1.85	0.338	0.53-6.49				6.01	0.084	0.79-46.0			
Primary	[a]3.43	[a]0.001	[a]1.66-7.10	[a]2.76	[a]0.015	[a]1.22-6.23	[a]3.41	[a]0.015	[a]1.27-9.13				[a]3.86	[a]0.016	[a]1.29-11.6	[a]5.64	[a]0.006	[a]1.66-19.2
LNs	2.44	0.069	0.93-6.38				1.45	0.517	0.47-4.51				6.01	0.084	0.79-46.0			

Table III. Continued.

	All patients						HPV-negative OPC						HPV-positive OPC					
	Univariate			Multivariate			Univariate			Multivariate			Univariate			Multivariate		
Characteristics	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI	HR	P-value	95% CI
After CRT																		
CR																		
Non-CR	[a]7.44	[a]<0.01	[a]3.58-15.5	[a]3.93	[a]0.001	[a]1.17-8.82	[a]7.83	[a]<0.01	[a]2.86-21.6	[a]6.56	[a]<0.01	[a]2.31-18.6	[a]6.43	[a]0.001	[a]2.20-18.8			

[a]Indicates a statistically significant difference; [b]All patients and HPV-positive OPC; [c]HPV-negative OPC. Normal, 18.5-22.9 kg/m^2; Overweight, ≥23 kg/m^2; Low, <18.5 kg/m^2. HPV, human papillomavirus; OPC, oropharyngeal cancer; pk-yrs, pack years; HR, hazard ratio; CI, confidence interval; WHO PS, World Health Organization performance status; BMI, body mass index; cGy, centigray; CR, complete response; non-CR, non-complete response; IC, induction chemotherapy; LNs, lymph nodes; CRT, chemoradiotherapy.

As aforementioned, the routine use of IC is not recommended in current standard guidelines because OPC typically responds well to chemotherapy and radiation (20). Nevertheless, IC has advantages of reducing the rate of distant metastasis, selecting chemosensitive patients to minimize subsequent therapies, and facilitating organ preservation. In previous randomized clinical trials with subgroup analyses of patients with and without OPC, no survival benefit was observed in the OPC group (21-23). Nevertheless, these trials had several limitations, such as the absence of HPV assessment, diverse tumor stages or IC regimens and lack of standard CRT. Despite these limitations, IC remains an attractive treatment option due to its potential to reduce bulky masses, thereby increasing the possibility of de-intensifying radiotherapy or enhancing operability, ultimately improving the likelihood of achieving CR (22).

Furthermore, it is established that the response to IC is a reliable predictor of survival outcomes in head and neck cancer, a trend consistent with that of OPC (24-27). Consequently, attempts to de-escalate CRT based on the IC response have been made in most clinical studies involving HPV-positive OPC, which tends to be more sensitive to radiation. This approach aims to mitigate adverse effects associated with radiation.

The ECOG E1308 study demonstrated that reduced-dose IMRT benefited patients who achieved clinical CR at the primary site after IC with cisplatin, paclitaxel and cetuximab. However, in a subgroup analysis, T4 stage, N2c [metastasis in bilateral or contralateral lymph node(s), non-larger than 6-cm in greatest dimension and extranodal extension (ENE) negative], or a smoking history exceeding 10 years demonstrated a statistically significant decrease in PFS (11). In the OPTIMA study, patients with HPV-positive OPC were classified into low-risk [T1-3, N0-2b (N0, no regional lymph node; N1 metastasis in a single ipsilateral lymph node, ≤3 cm in greatest dimension and ENE negative; N2a, metastasis in a single ipsilateral node >3 cm but not >6 cm in greatest dimension and ENE negative; N2b, metastases in multiple ipsilateral nodes, non-larger than 6 cm in greatest dimension and ENE negative), smoking history ≤10 years] and high-risk {T4 or N2c-N3 [N2c, metastasis in bilateral or contralateral lymph nodes, non-larger than 6 cm in greatest dimension and ENE negative; N3, metastasis in a lymph node larger than 6 cm in greatest dimension and ENE negative; or metastasis in any node(s) and clinically overt ENE positive], smoking history >10 years} categories. De-escalation treatment was subsequently administered with 50 Gy RT alone, 45 Gy CRT, or 75 Gy CRT based on the response to IC (12). De-escalated RT or CRT for HPV-positive OPC based on risk stratification and response to IC showed no significant difference in the 2-year PFS or OS, and toxicity could be minimized by reducing the RT dose. This study highlighted that risk-stratified patients with <50% response following IC exhibited comparable outcomes, with 28 and 27% for low- and high-risk patients, respectively.

While Ang *et al* (7) illustrated risk stratification based on the N stages of the AJCC 5th edition, the results of the present study indicated that the T stage remained a crucial prognostic factor for survival in HPV-positive OPC even after IC. This difference could be attributed to alterations in the AJCC staging system. In the earlier AJCC 5-7th staging systems, there was no distinction in the TNM stage system or treatment strategy for

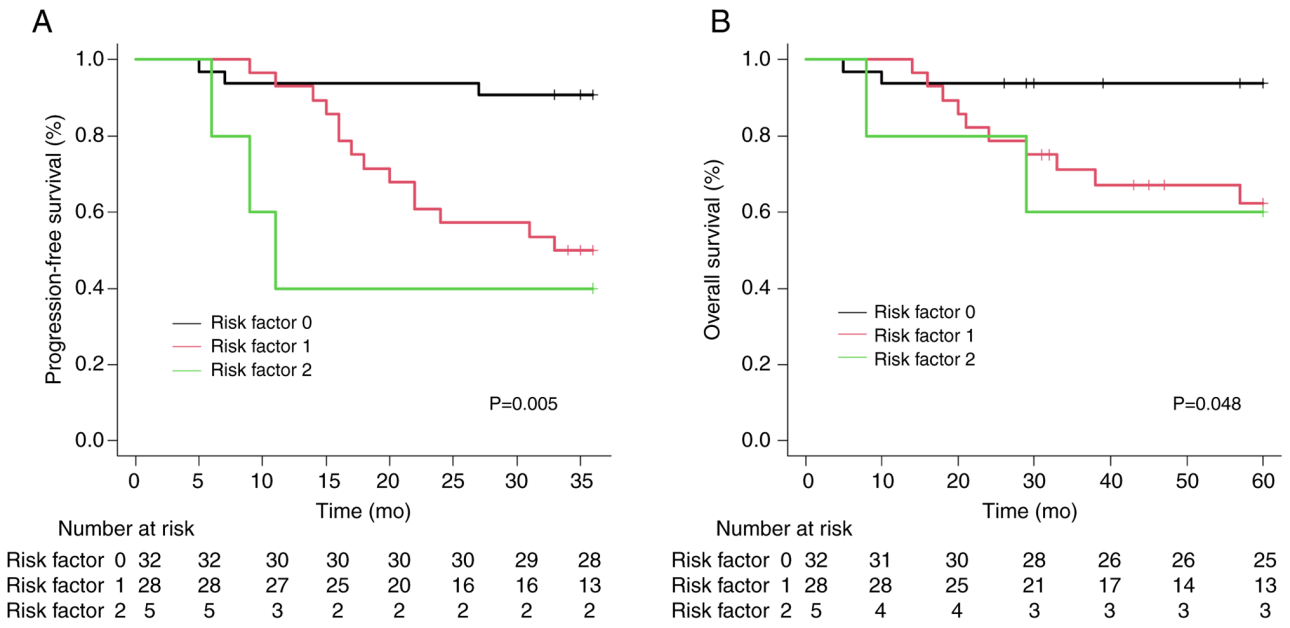

Figure 3. Kaplan-Meier estimates of (A) progression-free survival and (B) overall survival by stratifying across each group as the number of clinical risk factors at diagnosis (age and T4 stage) for human papillomavirus-positive oropharyngeal cancer. Risk factor 0, no number of risk factors; Risk factor 1, one of two risk factors; Risk factor 2, two of two risk factors; mo, months.

HPV-positive OPC. However, in the AJCC 8th staging system, the N stage of HPV-positive OPC has been modified and defined as multiple ipsilateral, equivalent to N1. By contrast, in the AJCC 7th staging system, this stage was identical to N2 in multiple ipsilateral regardless of HPV status. As a result, most patients in the present study were downstaged to stage II or III according to the AJCC 8th staging system. This prompts the question of whether the current staging system for HPV-positive OPC is sufficient to reflect the prognosis as depicted in Fig. 2C and D. Yoo *et al* (28) similarly highlighted an advanced T stage in the univariate analysis, indicating that patients may not be suitable for de-escalation treatment. T4 stage emerged as a risk factor for poor prognosis in other clinical trials that had previously attempted de-escalation treatment (11). Combining previous findings with the results of the present study, it has become evident that the T4 stage is a distinct high-risk factor in HPV-positive OPC. The N stage in HPV-positive OPC in the AJCC 8th staging system requires modification to enhance its applicability in clinical practice.

The present study revealed no differences in treatment outcomes, including overall response rate and survival between HPV-positive and HPV-negative OPCs. This is likely due to the improved treatment outcomes compared with those in previously published results for HPV-negative OPC. In a trial in the United States by Ang *et al* (7), the 3-year PFS and 3-year OS of HPV-negative OPC were 43.4 and 57.1%, respectively, in a retrospective analysis of the OPC patients who were treated with CRT. Compared with this report, a higher survival rate with a 3-year PFS of 57.4% and a 5-year OS of 60.9% was observed. Furthermore, after CRT, there was a high CR rate of 80% for the HPV-negative OPC in the present study. Therefore, the enhanced survival outcome in patients with HPV-negative OPC who received the IC followed by CRT may mitigate the survival gap between HPV-negative and HPV-positive OPCs. This result could explain why the two groups exhibited no significant difference in the survival rate.

To identify other clinical factors, several studies have explored smoking, a significant etiological factor for head and neck cancer, as a prognostic factor. However, the findings were inconsistent (29-31). A recent study has indicated that age-related molecular alterations and genomic, immunological and tumor differences may contribute to tumor responses in older adults (32). The present study similarly found no relationship between survival and smoking. This could be attributed to inaccuracies in medical records or individual variations in susceptibility to smoking. de la Iglesia *et al* (33) demonstrated that smoking status was not linked to tumor heterogeneity based on gene mutation burden. Instead, active smoking manifests an immunosuppressive effect by inhibiting the infiltration of cytotoxic T cells into the tumor. Hence, there is a need to identify a biomarker that can reflect the molecular and immunological changes induced by current smoking rather than relying solely on smoking history.

Extensive research has been conducted on genes influencing the prognosis and novel treatments for HPV-positive OPC (5). Khwaja *et al* demonstrated that elevated E6 expression was linked to a high risk of distant metastasis and poor survival outcomes (34). A recent study revealed that patients with HPV-positive OPC exhibiting high levels of FGF11, recognized for its oncogenic functions, had poor survival outcomes (35). *TP53*, identified in HPV-positive OPC in heavy smokers, has been linked to poor prognosis. However, it can function as a target for viral enzymes E6/E7, resulting in the degradation of TP53 (36,37). Hence, next-generation sequencing (NGS) enables the analysis of biomarkers related to tumor response to chemotherapy and facilitates the exploration of age-related biomarkers, aiding in the identification of patients at high risk for HPV-positive OPC and complementing traditional markers such as HPV or p16.

The present study has several limitations in terms of adopting a clinical treatment strategy based on the results. First,

the present study relied on retrospective data collected from a single institution. Therefore, further validation with data from multiple centers is necessary. However, the eligible patients in the present study received a consistent IC regimen and CRT method, minimizing treatment-related bias. Second, biomarkers such as p53 expression or NGS were not assessed. Finally, recently proposed as standard treatment, immune checkpoint inhibitors were not utilized during this period. Therefore, future studies are warranted to conduct these evaluations.

In conclusion, a clinical T4 stage and age ≥60 years at diagnosis were associated with a poor prognosis in patients with HPV-positive OPC. The innovative finding in the present study is that it is not appropriate to consider a de-escalated treatment strategy for high-risk patients, even for HPV-positive patients with OPC. In addition, further clinical trials using a combined modality, such as the introduction of IC to chemoradiotherapy are needed to improve survival.

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Availability of data and materials

The data generated in the present study may be requested from the corresponding author.

Authors' contributions

SHC conceived and designed the study. HJB, SHL, and SHC confirm the authenticity of all the raw data. HJB, HJK, SHL, HJS, JEH, WKB, IJC and SHC performed the data analysis and interpretation of the data. HJB and SHC drafted the manuscript. HJB, HJK, SHL, HJS, JEH, WKB, IJC and SHC critically revised the manuscript. All authors have read and approved the final manuscript.

Ethics approval and consent to participate

The study protocol was approved (approval no. CNUHH-2023-232) by the Institutional Review Board of Chonnam National University Hwasun Hospital (Hwasun, Republic of Korea).

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References

1. Siegel RL, Giaquinto AN and Jemal A: Cancer statics, 2024. CA Cancer J Clin 74: 12-49, 2024
2. Korea National Cancer CenterNational Cancer Registration Project Annual Report (2020 Cancer Registration Statistics). https://ncc.re.kr/cancerStatsView.ncc?bbsnum=638&searchKey=total&searchValue=&pageNum=1. Accessed May 19, 2023 (In Korean).
3. Faraji F, Rettig EM, Tsai HL, El Asmar M, Fung N, Eisele DW and Fakhry C: The prevalence of human papillomavirus in oropharyngeal cancer is increasing regardless of sex or race, and the influence of sex and race on survival is modified by human papillomavirus tumor status. Cancer 125: 761-769, 2019.
4. Kang EJ, Lee YG, Keam B, Choi JH, Kim JS, Park KU, Lee KE, Kim HJ, Lee KW, Kim MK, *et al*: Characteristics and treatment patterns in older patients with locally advanced head and neck cancer (KCSG HN13-01). Korean J Intern Med 37: 190-200, 2022.
5. Lechner M, Liu J, Masterson L and Fenton TR: HPV-associated oropharyngeal cancer: Epidemiology, molecular biology and clinical management. Nat Rev Clin Oncol 19: 306-327, 2022.
6. Rettig EM, Fakhry C, Khararjian A and Westra WH: Age profile of patients with oropharyngeal squamous cell carcinoma. JAMA Otolaryngol Head Neck Surg 144: 538-539, 2018.
7. Ang KK, Harris J, Wheeler R, Weber R, Rosenthal DI, Nguyen-Tân PF, Westra WH, Chung CH, Jordan RC, Lu C, *et al*: Human papillomavirus and survival of patients with oropharyngeal cancer. N Engl J Med 363: 24-35, 2010.
8. Cao X, Hou J, An Q, Assaraf YG and Wang X: Towards the overcoming of anticancer drug resistance mediated by p53 mutations. Drug Resist Updat 49: 100671, 2020.
9. Amin MB, Edge SB, Greene FL, Byrd DR, Brookland RK, Washington MK, Gershenwald JE, Compton CC, Hess KR, Sullivan DC, *et al* (eds): AJCC Cancer Staging Manual. 8th edition. Springer, New York, NY, 2017.
10. Pfister DG, Spencer S, Adelstein D, Adkins D, Anzai Y, Brizel DM, Bruce JY, Busse PM, Caudell JJ, Cmelak AJ, etal: Head and Neck Cancers, Version 2.2024, NCCN Clinical Practice Guidelines in Oncology. J Natl Compr Canc Netw, 2024.
11. Marur S, Li S, Cmelak AJ, Gillison ML, Zhao WJ, Ferris RL, Westra WH, Gilbert J, Bauman JE, Wagner LI, *et al* : E1308: Phase II trial of induction chemotherapy followed by reduced-dose radiation and weekly cetuximab in patients with HPV-associated resectable squamous cell carcinoma of the oropharynx-ECOG-ACRIN Cancer Research Group. J Clin Oncol 35: 490-497, 2017.
12. Seiwert TY, Foster CC, Blair EA, Karrison TG, Agrawal N, Melotek JM, Portugal L, Brisson RJ, Dekker A, Kochanny S, *et al*: OPTIMA: A phase II dose and volume de-escalation trial for human papillomavirus-positive oropharyngeal cancer. Ann Oncol 30: 1673, 2019.
13. Kang JJ, Yu Y, Chen L, Zakeri K, Gelblum DY, McBride SM, Riaz N, Tsai CJ, Kriplani A, Hung TKW, *et al*: Consensuses, controversies, and future directions in treatment deintensification for human papillomavirus-associated oropharyngeal cancer. CA Cancer J Clin 73: 164-197, 2023.
14. Kim MJ, Ki MS, Kim K, Shim HJ, Hwang JE, Bae WK, Chung IJ, Lee DH, Lee JK, Yoon TM, *et al*: Different protein expression associated with chemotherapy response in oropharyngeal cancer according to HPV status. BMC Cancer 14: 824, 2014.
15. World Health Organization and Regional Office for the Western Pacific: The Asia-Pacific perspective: redefining obesity and its treatment. Health Communications Australia, Sydney, pp 18, 2000. https://iris.who.int/handle/10665/206936. Accessed May 19, 2023.
16. Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, Dancey J, Arbuck S, Gwyther S, Mooney M, *et al*: New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1). Eur J Cancer 45: 228-247, 2009.
17. Chow LQM: Head and Neck Cancer. N Engl J Med 382: 60-72, 2020.
18. Chen SY, Massa S, Mazul AL, Kallogjeri D, Yaeger L, Jackson RS, Zevallos J and Pipkorn P: The association of smoking and outcomes in HPV-positive oropharyngeal cancer: A systemic review. Am J Otolaryngol 41: 102592, 2020.
19. Lai YH, Su CC, Wu SY, Hsueh WT, Wu YH, Chen HHW, Hsiao JR, Liu CH and Tsai YS: Impact of alcohol and smoking on outcomes of HPV-related oropharyngeal cancer. J Clin Med 11: 6510, 2022.
20. Ferrari D, Ghi MG, Franzese C, Codeca C, Gau M and Fayette J: The slippery role of induction chemotherapy in head and neck cancer: Myth and reality. Front Oncol 10: 7, 2020.
21. Cohen EE, Karrison TG, Kocherginsky M, Mueller J, Egan R, Huang CH, Brockstein BE, Agulnik MB, Mittal BB, Yunus F, *et al*: Phase III randomized trial of induction chemotherapy in patients with N2 or N3 locally advanced head and neck cancer. J Clin Oncol 32: 2735-2743, 2014.

22. Ghi MG, Paccagnella A, Ferrari D, Foa P, Alterio D, Codecà C, Nolè F, Verri E, Orecchia R, Morelli F, *et al*: Induction TPF followed by concomitant treatment versus concomitant treatment alone in locally advanced head and neck cancer. A phase II-III trial. Ann Oncol 28: 2206-2212, 2017.
23. Haddad R, O'Neill A, Rabinowits G, Tishler R, Khuri F, Adkins D, Clark J, Sarlis N, Lorch J, Beitler JJ, *et al*: Induction chemotherapy followed by concurrent chemoradiotherapy (sequential chemoradiotherapy) versus concurrent chemoradiotherapy alone in locally advanced head and neck cancer (PARADIGM): A randomised phase 3 trial. Lancet Oncol 14: 257-264, 2013.
24. Abdelmeguid AS, Teeramatwanich W, Roberts DB, Amit M, Ferraroto R, Glisson BS, Kupferman ME, Su SY, Phan J, Garden AS, *et al*: Neoadjuvant chemotherapy for locoregionally advanced squamous cell carcinoma of the paranasal sinuses. Cancer 127: 1788-1795, 2021.
25. Bossi P, Lo Vullo S, Guzzo M, Mariani L, Granata R, Orlandi E, Locati L, Scaramellini G, Fallai C and Licitra L: Preoperative chemotherapy in advanced resectable OCSCC: long-term results of a randomized phase III trial. Ann Oncol 25: 462-466, 2014.
26. Lee YG, Kang EJ, Keam B, Choi JH, Kim JS, Park KU, Lee KE, Kim HJ, Lee KW, Kim MK, *et al*: Induction chemotherapy as a prognostication index and guidance for treatment of locally advanced head and neck squamous cell carcinoma: The concept of chemo-selection (KCSG HN13-01). Cancer Res Treat 54: 109-117, 2022.
27. Zhang Q, Xu T, Shen C, Qian W, Ying H, He X, Wang Y, Ji Q, Hu C, Zhou X and Lu X: Response to induction chemotherapy predicts survival outcomes in oropharyngeal cancer. Cancer Med 12: 9175-9185, 2023.
28. Yoo SH, Ock CY, Keam B, Park SJ, Kim TM, Kim JH, Jeon YK, Chung EJ, Kwon SK, Hah JH, *et al*: Poor prognostic factors in human papillomavirus-positive head and neck cancer: Who might not be candidates for de-escalation treatment? Korean J Intern Med 34: 1313-1323, 2019.
29. Broughman JR, Xiong DD, Moeller BJ, Contrera KJ, Prendes BL, Lamarre ED, Ku JA, Burkey BB, Woody NM, Joshi NP, *et al*: Rethinking the 10-pack-year rule for favorable human papillomavirus-associated oropharynx carcinoma: A multi-institution analysis. Cancer 126: 2784-2790, 2020.
30. Fakhry C, Zhang Q, Nguyen-Tan PF, Rosenthal DI, Weber RS, Lambert L, Trotti AM III, Barrett WL, Thorstad WL, Jones CU, *et al*: Development and validation of nomograms predictive of overall and progression-free survival in patients with oropharyngeal cancer. J Clin Oncol 35: 4057-4065, 2017.
31. Huang SH, Xu W, Waldron J, Siu L, Shen X, Tong L, Ringash J, Bayley A, Kim J, Hope A, *et al*: Refining American Joint Committee on Cancer/Union for International Cancer Control TNM stage and prognostic groups for human papillomavirus-related oropharyngeal carcinomas. J Clin Oncol 33: 836-845, 2015.
32. Chatsirisupachai K, Lagger C and de Magalhaes JP: Age-associated differences in the cancer molecular landscape. Trends Cancer 8: 962-971, 2022.
33. de la Iglesia JV, Slebos RJC, Martin-Gomez L, Wang X, Teer JK, Tan AC, Gerke TA, Aden-Buie G, van Veen T, Masannat J, *et al*: Effects of tobacco smoking on the tumor immune microenvironment in head and neck squamous cell carcinoma. Clin Cancer Res 26: 1474-1485, 2020.
34. Khwaja SS, Baker C, Haynes W, Spencer CR, Gay H, Thorstad W, Adkins DR, Nussenbaum B, Chernock RD, Lewis JS Jr and Wang X: High E6 gene expression predicts for distant metastasis and poor survival in patients with HPV-positive oropharyngeal squamous cell carcinoma. Int J Radiat Oncol Biol Phys 95: 1132-1141, 2016.
35. Flon CH, Haeggblom L, Holzhauser S, Kostopoulou ON, Zupancic M, Dalianis T, Munck-Wikland E, Marklund L and Näsman A: High levels of FGF11 correlate with poor survival in patients with human papillomavirus (HPV)-positive oropharyngeal squamous cell carcinoma. Cancers (Basel) 15: 1954, 2023.
36. de Bakker T, Journe F, Descamps G, Saussez S, Dragan T, Ghanem G, Krayem M and Van Gestel D: Restoring p53 function in head and neck squamous cell carcinoma to improve treatments. Front Oncol 11: 799993, 2021.
37. Dogan S, Xu B, Middha S, Vanderbilt CM, Bowman AS, Migliacci J, Morris LGT, Seshan VE and Ganly I: Identification of prognostic molecular biomarkers in 157 HPV-positive and HPV-negative squamous cell carcinomas of the oropharynx. Int J Cancer 145: 3152-3162, 2019.