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Patterns of failure and prognosis in nasopharyngeal carcinoma according to Epstein-Barr virus DNA status

Lin-Feng Guo[1†], Guan-Zhong Lu[1†], Zhen-Zhen Lu[2], Yi-Feng Yu[1*] and San-Gang Wu[1*]

Abstract

Purpose To investigate the patterns of failure and prognosis in recurrent or metastatic nasopharyngeal carcinoma (rmNPC) according to Epstein-Barr virus-DNA (EBV-DNA) status.

Methods We included NPC patients who were diagnosed with locoregional recurrence (LRR) and/(or) distant metastasis (DM) between January 2017 and June 2024. Receiver operating characteristic analysis, Chi-square test, Wilcoxon rank sum test, Kaplan-Meier method, and Multivariate Cox regression analyses were used for statistical analysis.

Results This study involved 108 patients, including 105 (97.2%) who had EBV-DNA detectable at the initial diagnosis of NPC. Regarding progression patterns, 34 patients (31.5%) experienced only LRR, while 60 patients (55.6%) had only DM. LRR followed by DM was observed in 5 (4.6%) patients, DM followed by LRR occurred in 2 (1.8%) patients, and both LRR and DM were presented simultaneously in 7 (6.5%) patients. EBV-DNA positivity rates significantly differed between LRR and DM patients, at 76.9% and 97.1% respectively ($P=0.003$). A significant difference was also observed in EBV-DNA levels, with a median level of 413 copies/mL for LRR and 6,550 copies/mL for DM ($P<0.001$). While the EBV-DNA positivity rate did not differ significantly between oligometastatic disease and polymetastatic disease ($P=0.493$), the levels were significantly elevated in the polymetastatic disease group than the oligometastatic disease group ($P<0.001$). Multivariate analysis showed that liver metastasis ($P=0.012$) and EBV-DNA levels ≥ 3,525 copies/mL at progression ($P=0.009$) independently correlated with poorer overall survival.

Conclusions Our study provides substantial evidence linking higher EBV-DNA levels with disease failure patterns and identifies liver metastasis and EBV-DNA levels at disease progression as independent prognostic factors for poorer overall survival in rmNPC patients.

Keywords Nasopharyngeal carcinoma, EBV-DNA, Failure patterns, Outcome

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Introduction

Nasopharyngeal carcinoma (NPC) is an epithelioid carcinoma originating from the nasopharyngeal mucosa and is characterized by a unique geographical distribution. It is predominantly found in Southeast Asia, especially in Southern China [1, 2]. The widespread application of intensity-modulated radiotherapy (IMRT) and systemic therapy in NPC has significantly improved the efficacy of NPC treatment. However, approximately 20–25% of patients still experience disease failure following chemoradiotherapy [3, 4]. Distant metastasis (DM) is the primary mode of failure in NPC, accounting for about 70%, followed by locoregional recurrence (LRR), which accounts for approximately 30% [3, 4]. Common metastatic sites include the bone, lung, liver, and distant lymph nodes, with disease progression occurring via regional lymph nodes (lymphatic route) and primary tumors (hematogenous route). These metastases significantly impact the survival time and life quality of patients (5–6).

The development of NPC is closely associated with Epstein-Barr virus (EBV) infection. EBV-DNA has demonstrated high sensitivity and specificity as a tumor marker for detecting NPC [7]. Several studies have indicated that the levels of EBV-DNA before and after treatment hold significant prognostic value and can serve as an important marker for follow-up monitoring and treatment adjustment [8–10]. Despite several studies from endemic and non-endemic areas analyzing the patterns of DM and/or LRR in NPC, there remains a scarcity of research focusing on the relationship between EBV-DNA status and disease failure patterns [11–14]. Therefore, this study aimed to investigate the patterns of disease failure and survival outcomes in recurrent or metastatic nasopharyngeal carcinoma (rmNPC) according to EBV-DNA status.

Materials and methods

Patients

We retrospectively included patients diagnosed with rmNPC from January 2017 and June 2024 at the First Affiliated Hospital of Xiamen University. Patients who met the following criteria were included: (1) a confirmed pathological diagnosis of NPC; (2) complete pre-treatment and progression-time plasma EBV-DNA level data; (3) comprehensive patient clinicopathological characteristics and follow-up records; (4) a diagnosis with LRR and/(or) DM during the follow-up period. Patients with *de novo* metastatic disease, a history of secondary malignancies, or a lack of EBV-DNA records were excluded. This study was approved by the Ethics Committee of the First Affiliated Hospital of Xiamen University, and informed consent was waived as the study was retrospective in nature.

Variables

The following variables were included in the analysis: age at rmNPC diagnosis, gender, Eastern Cooperative Oncology Group (ECOG) performance status, histology subtypes, smoking history, drinking history, tumor (T) stage at initial NPC diagnosis, nodal (N) stage at initial NPC diagnosis, plasma EBV-DNA levels, sites of LRR, sites of DM, and the status of DM. All patients were staged according to the American Joint Committee on Cancer staging system 8th edition. The status of DM was categorized as oligometastatic disease (OM) (1–2 metastatic organs or 1–5 metastatic lesions) and polymetastatic disease (PM) (beyond OM) (15–16). The primary endpoint of our study was overall survival (OS), defined as the time interval from rmNPC diagnosis to death from any cause.

EBV-DNA quantification

Peripheral whole blood samples (10 mL) were collected. Circulating EBV-DNA was extracted from the plasma and quantified using droplet digital PCR. Plasma EBV-DNA levels were deemed detectable (positive) if above 0 copies/mL and undetectable (negative) if at 0 copies/mL [8]. The cut-off point for plasma EBV-DNA levels in patients with newly diagnosed NPC was classified as low-risk (<4000 copies/ml) and high-risk (≥4000 copies/ml) based on previous studies [17–19]. However, the optimal EBV-DNA cut-off point for patients with rmNPC remains unclear. To determine the optimal cut-off point for EBV-DNA levels in patients with disease progression, a receiver operating characteristic (ROC) curve was employed.

Statistical analysis

Categorical variables were compared using the chi-square test or Fisher's exact test. The ROC curve was employed to determine the optimal cut-off point for EBV-DNA level associated with OS, and the maximal area under the curve (AUC) value was chosen as the cut-off for model building. Differences in EBV-DNA levels at progression were assessed using the Wilcoxon Rank-Sum test. Survival estimations were performed using the Kaplan-Meier method with differences evaluated by the log-rank test. Univariate and multivariate Cox regression analyses were conducted to identify independent prognostic factors associated with OS. Variables exhibiting a P value of less than 0.10 in the univariate Cox regression model were included in the multivariate Cox proportional hazards analysis to identify the prognostic factors significantly associated with OS. All statistical analyses utilized the SPSS statistical software package (version 26.0; IBM Corporation, Armonk, NY, USA). Statistical significance was determined by a P-value of less than 0.05.

Results

Patient characteristics

This study involved 108 patients (Table 1), with a median age of 50 years (range, 23–79 years). At the time of initial diagnosis, 1 (0.9%) patient had stage I, 3 patients (2.8%) were at stage II, 53 (49.1%) patients were at stage III, and 51 (47.2%) patients were at stage IVa. In addition, 81 (75.0%) were classified as having stage T3-4 tumors and 78 (62.2%) were identified with stage N2-3 disease. Among these patients, 105 (97.2%) had EBV-DNA detectable at initial diagnosis of NPC, with a median level of 2,650 copies/mL (range, 58–310,000 copies/mL).

Table 1 Patient characteristics

Variables	*N* (%)
Gender	
Men	86 (79.6)
Women	22 (20.4)
ECOG status	
0	36 (33.3)
1	69 (63.9)
2	3 (2.8)
Age (years)	
≤50	51 (47.2)
>50	57 (52.8)
Histology subtypes	
WHO II	16 (14.8)
WHO III	92 (85.2)
Smoking history	
Yes	53 (49.1)
No	55 (50.9)
Drinking history	
Yes	37 (34.3)
No	71 (65.7)
Clinical stage at initial diagnosis	
I	1 (0.9)
II	3 (2.8)
III	53 (49.1)
IVa	51 (47.2)
T stage at initial diagnosis	
T1-2	27 (25.0)
T3-4	81 (75.0)
N stage at initial diagnosis	
N0-1	30 (27.8)
N2-3	78 (62.2)
Pre-treatment EBV-DNA levels (copies/mL)	
0	3 (2.8)
1-3999	63 (58.3)
≥4000	42 (38.9)
EBV-DNA levels at disease progression (copies/mL)	
0	11 (10.3)
1-3524	55 (50.9)
≥3525	42 (38.8)
The first line of treatment in LRR-only patients ($n=39$)	
Chemotherapy	20 (51.3)
Chemotherapy and immunotherapy	19 (48.7)
The first line of treatment in DM patients ($n=69$) [#]	
Chemotherapy	42 (60.9)
Chemotherapy and immunotherapy	27 (39.1)

WHO, World Health Organization; T, tumor; N, nodal; LRR, locoregional recurrence; DM, distant metastasis; ECOG, Eastern Cooperative Oncology Group

[#] includes 7 patients who experienced both locoregional recurrence and distant metastasis simultaneously

Patterns of disease progression and EBV-DNA levels

Regarding progression patterns (Table 2 and Supplementary Table 1), 34 patients (31.5%) experienced only LRR, while 60 patients (55.6%) had only DM. LRR followed by DM was observed in 5 (4.6%) patients, DM followed by LRR occurred in 2 (1.8%) patients, and both LRR and DM were presented simultaneously in 7 (6.5%) patients (Fig. 1A). The median time for first progression was 22.8 months for LRR (range, 7.7–86.2 months) and 15.8 months for DM (range, 3.6–86.2 months).

Among the 39 patients (36.1%) who initially progressed with LRR, 21 (53.8%) had recurrence confined to the nasopharynx, 15 (38.5%) in cervical lymph nodes including 3 patients had retropharyngeal lymph node recurrence, and 3 (7.7%) in both sites (Fig. 1A). EBV-DNA positivity rates for nasopharynx-only recurrence were 76.2% ($n=16$) with a median level of 291 copies/mL, for cervical lymph nodes recurrence 73.3% ($n=11$) with a median level of 1,250 copies/mL, and for both sites 100% ($n=3$) with a median EBV-DNA level of 580 copies/mL. No statistical differences were found in EBV-DNA levels ($P=0.444$) or positivity rates ($P=0.773$) among these groups (Table 2).

DM was noted in 69 patients (63.9%), involving 114 metastatic sites. The most frequent metastatic sites were bone ($n=36$, 52.2%), followed by liver ($n=30$, 43.5%), lung ($n=25$, 36.2%), and distant lymph nodes ($n=23$, 33.3%) (Fig. 1B). All patients with bone metastases were EBV-DNA positive (100%), with a median EBV-DNA level of 6,150 copies/mL. In the case of liver metastasis, all patients were EBV-DNA positive, with a median level of 12,650 copies/mL. Among lung metastasis patients, 23 out of 25 (92.0%) were EBV-DNA positive, with a median level of 6,550 copies/mL. For distant lymph node metastasis, all patients were EBV-DNA positive, with a median level of 15,575 copies/mL. There were no significant differences in EBV-DNA positivity rates ($P=0.086$) or levels ($P=0.715$) across these metastatic sites (Table 2).

EBV-DNA positivity rates significantly differed between LRR and DM patients, at 76.9% and 97.1% respectively ($P=0.003$). A significant difference was also observed in EBV-DNA levels, with a median level of 413 copies/mL for LRR and 6,550 copies/mL for DM ($P<0.001$) (Table 2). Similar results were observed in patients with LRR only ($n=39$), DM only ($n=62$), and

Table 2 Failure patterns with EBV-DNA levels and EBV-DNA positive rate at the time of progression

Failure patterns	*N* (%)	Number of patients with EBV-DNA positive (%)	Number of patients with EBV-DNA negative (%)	Median EBV-DNA levels (range, copies/mL)	*P**	*P*$^{+}$
Failure patterns (*n* = 108)						
LRR	39 (36.1)	30 (76.9)	9 (23.1)	413 (33–33,500)	0.003	< 0.001
DM [#]	69 (63.9)	67 (97.1)	2 (2.9)	6,550 (20 – 1,962,500)		
LRR sites (*n* = 39)						
Only nasopharynx	21 (53.8)	16 (76.2)	5 (23.8)	291 (33 – 2,820)	0.773	0.444
Only cervical lymph nodes	15 (38.5)	11 (73.3)	4 (26.7)	1,250 (78 – 33,500)		
Both	3 (7.7)	3 (100)	0 (0)	580 (41 – 1,670)		
Sites of DM (entire cohort) (*n* = 114)						
Bone	36 (52.2)	36 (100)	0 (0)	6,150 (50 – 1,962,500)	0.086	0.715
Liver	30 (43.5)	30 (100)	0 (0)	12,650 (388-1,962,500)		
Lung	25 (36.2)	23 (92.0)	2 (8.0)	6,550 (20 – 1,962,500)		
DLNs	23 (33.3)	23 (100)	0 (0)	15,575 (203-1,962,500)		
Sites of DM (single organ metastasis) (*n* = 41)						
Only bone	18 (26.1)	18 (100)	0 (0)	2,888 (50–237,750)	0.187	0.335
Only liver	10 (14.5)	10 (100)	0 (0)	4,875 (388 – 81,000)		
Only lung	10 (14.5)	8 (80.0)	2 (20.0)	3,413 (20–34,000)		
Only DLNs	3 (4.3)	3 (100)	0 (0)	1,250 (585-1,560)		
Metastatic status (*n* = 69)						
OM	35 (50.7)	33 (94.3)	2 (5.7)	1,545 (20–237,750)	0.493	< 0.001
PM	34 (49.3)	34 (100)	0 (0)	18,188 (50 – 1,962,500)		

* indicates the *P*-value between those with EBV-DNA positive and EBV-DNA negative using the chi-square test or Fisher's exact test

$^{+}$ indicates the *P*-value between the EBV-DNA levels among the groups using the Wilcoxon Rank-Sum test

[#] includes 7 patients who experienced both locoregional recurrence and distant metastasis simultaneously

LRR, locoregional recurrence; DM, distant metastasis; DLNs, distant lymph nodes; OM, oligometastatic disease; PM, polymetastatic disease

those with both LRR and DM concurrently (n=7), as shown in Supplementary Table 2.

Within the DM group (n=69), 35 patients (50.7%) were classified as having OM, and 34 patients (49.3%) as PM. At progression, 33 patients (94.3%) in the OM group were EBV-DNA positive, with a median level of 1,545 copies/mL. All PM patients (100%) were positive for EBV-DNA, with a median EBV-DNA level of 18,188 copies/mL. While the EBV-DNA positivity rate did not differ significantly between OM and PM (P=0.493), the levels were significantly elevated in the PM group compared to the OM group (P<0.001) (Table 2).

For patients with single organ metastasis (n=41), 18 (43.9%), 10 (24.4%), 10 (24.4%), and 3 (7.3%) had bone, liver, lung, and distant lymph node metastasis, respectively. However, there were no significant differences in EBV-DNA positivity rates (P=0.187) or levels (P=0.335) across these metastatic sites (Table 2).

Failure patterns in pre-treatment EBV-DNA positive patients

In our analysis of 105 patients with detectable EBV-DNA at initial diagnosis (Fig. 2A), 37 patients exhibited only LRR at the first progression. Among these, 30 patients (81.1%) retained positive EBV-DNA status, while 7 patients (18.9%) did not. Notably, 4 patients went on to develop DM at the second progression, all of whom tested positive for positive EBV-DNA. This group included two patients who were initially negative and two who were initially positive for EBV-DNA during the first progression.

Furthermore, 61 patients experienced only DM at first progression, with 60 (98.4%) showing positive EBV-DNA and 1 (1.6%) remaining negative. Two patients developed LRR at the second progression, both with persistent positive EBV-DNA from the first progression. In addition, 7 patients had concurrent LRR and DM at first progression, all of whom were positive for EBV-DNA.

Failure patterns in pre-treatment EBV-DNA negative patients

Three patients had undetectable EBV-DNA at the initial diagnosis of NPC (Fig. 2B). Of these, two developed LRR with negative EBV-DNA at the first progression, while one progressed to DM at the second progression. Only one patient manifested DM at first progression while maintaining a negative EBV-DNA status throughout the follow-up period.

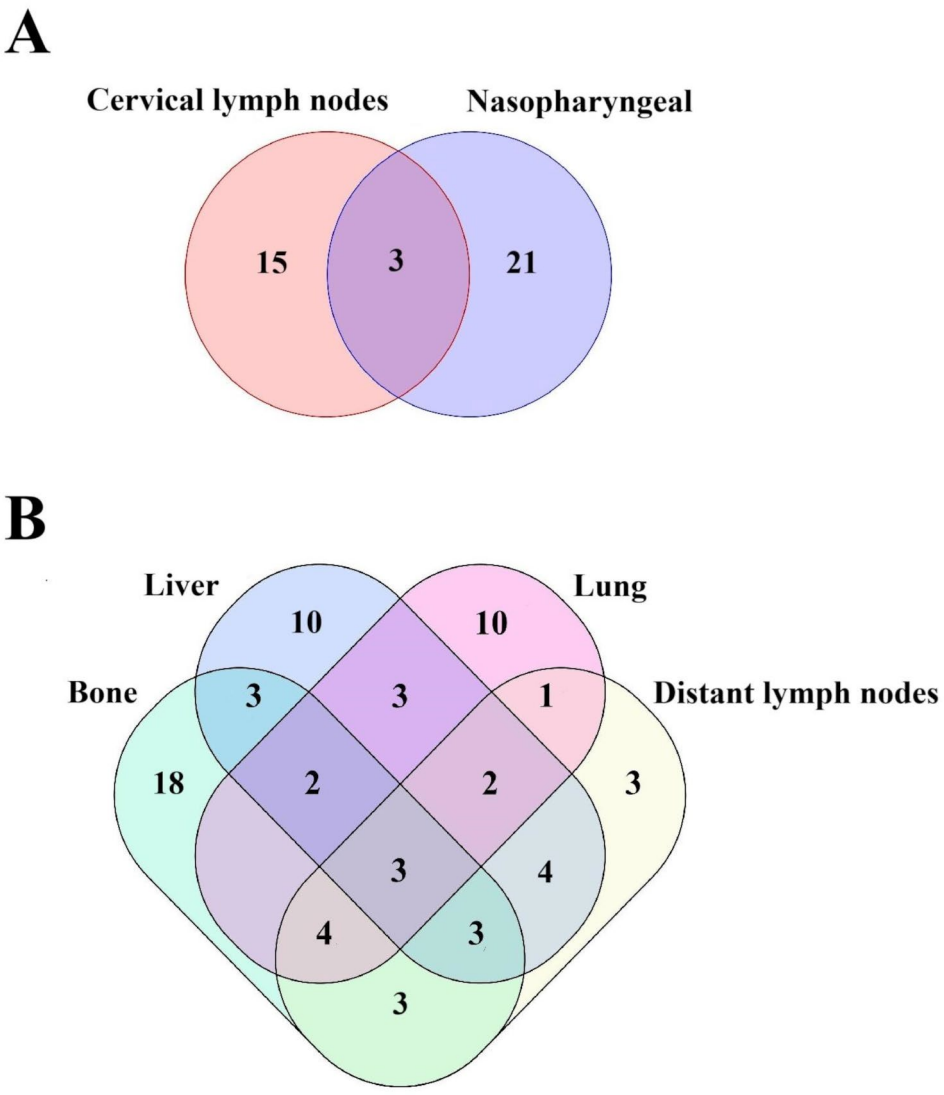

Fig. 1 The distribution of disease failure in patients (**A**: locoregional recurrence only [*n* = 39]; **B**: distant metastasis [*n* = 69, including 7 patients with concurrent locoregional recurrence and distant metastasis])

Survival and prognostic analysis

The median follow-up period for this cohort post-disease failure was 38.8 months (range, 0.5–90.8 months). The median OS was calculated at 38.1 months (range, 0.5–90.8 months), with 1-year, 2-year, and 3-year OS rates of 74.9%, 58.1%, and 52.9%, respectively.

Examining survival relative to metastatic sites, patients without bone metastasis exhibited a trend toward better OS compared to those with bone metastasis (median OS: 51.2 vs. 23.2 months, $P=0.152$), though this difference was not statistically significant (Fig. 3A). A significant survival advantage was observed in patients without liver metastasis compared to those with liver involvement (median OS: 54.0 vs. 14.3 months, $P<0.001$) (Fig. 3B). No significant survival differences were noted for lung (median OS: 43.6 vs. 22.8 months, $P=0.599$) or distant lymph node metastasis (median OS: 43.6 vs. 24.6 months, $P=0.912$).

The EBV-DNA level cut-off at progression was established at 3,525 copies/mL for predicting OS (Fig. 4). Patients with EBV-DNA levels ≥ 3,525 copies/mL at progression had significantly shorter OS compared to those with lower levels (median OS: 20.5 vs. 54.0 months, $P<0.001$) (Fig. 3C). While patients with OM tend to have longer OS than those with PM (median OS: 43.6 vs. 20.5 months), no significant difference was found ($P=0.191$) (Fig. 3D).

In the univariate analysis, liver metastasis (hazard ratio [HR] 2.854, 95% confidence interval [CI] 1.548–5.262, $P=0.001$) and the EBV-DNA levels ≥ 3,525 copies/mL at progression (HR 2.759, 95% CI 1.570–4.849, $P<0.001$) were significantly associated with OS. Multivariate

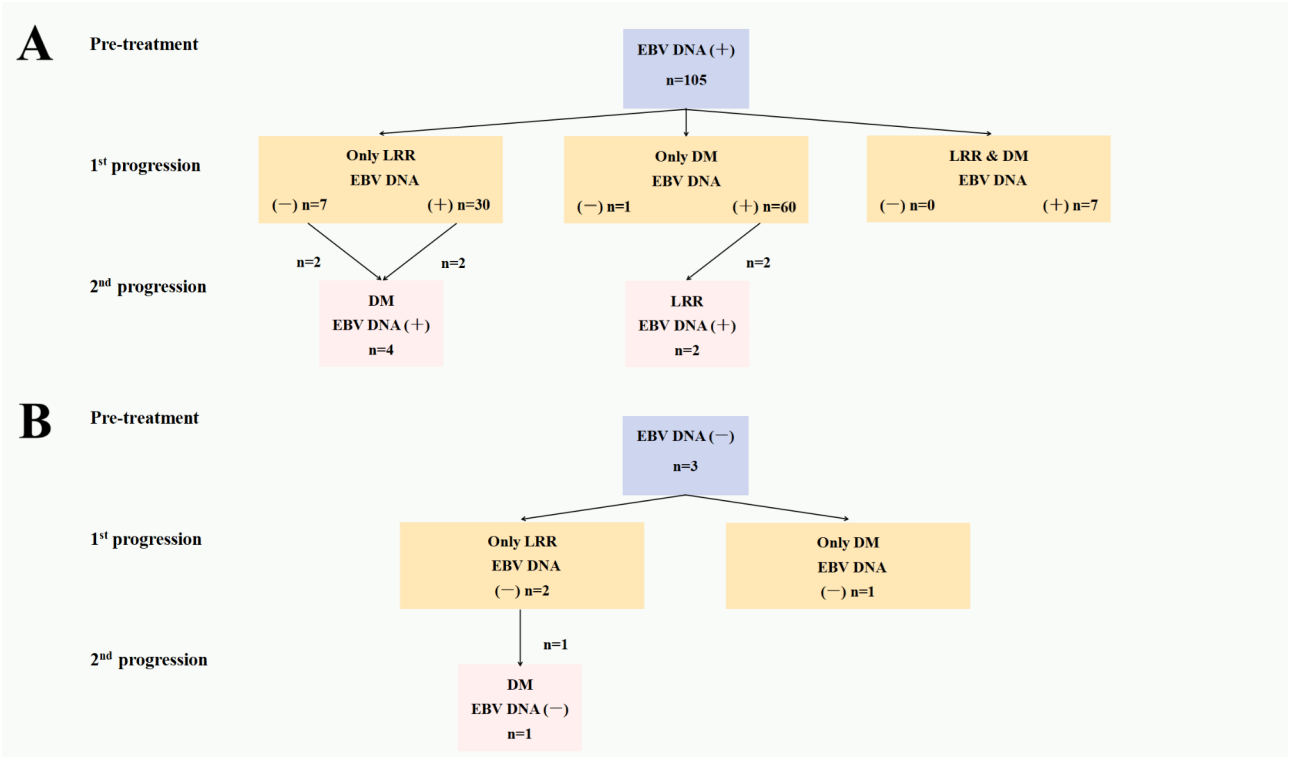

Fig. 2 Failure patterns according to EBV-DNA status in patients with pre-treatment positive EBV-DNA (**A**) and pre-treatment negative EBV-DNA (**B**)

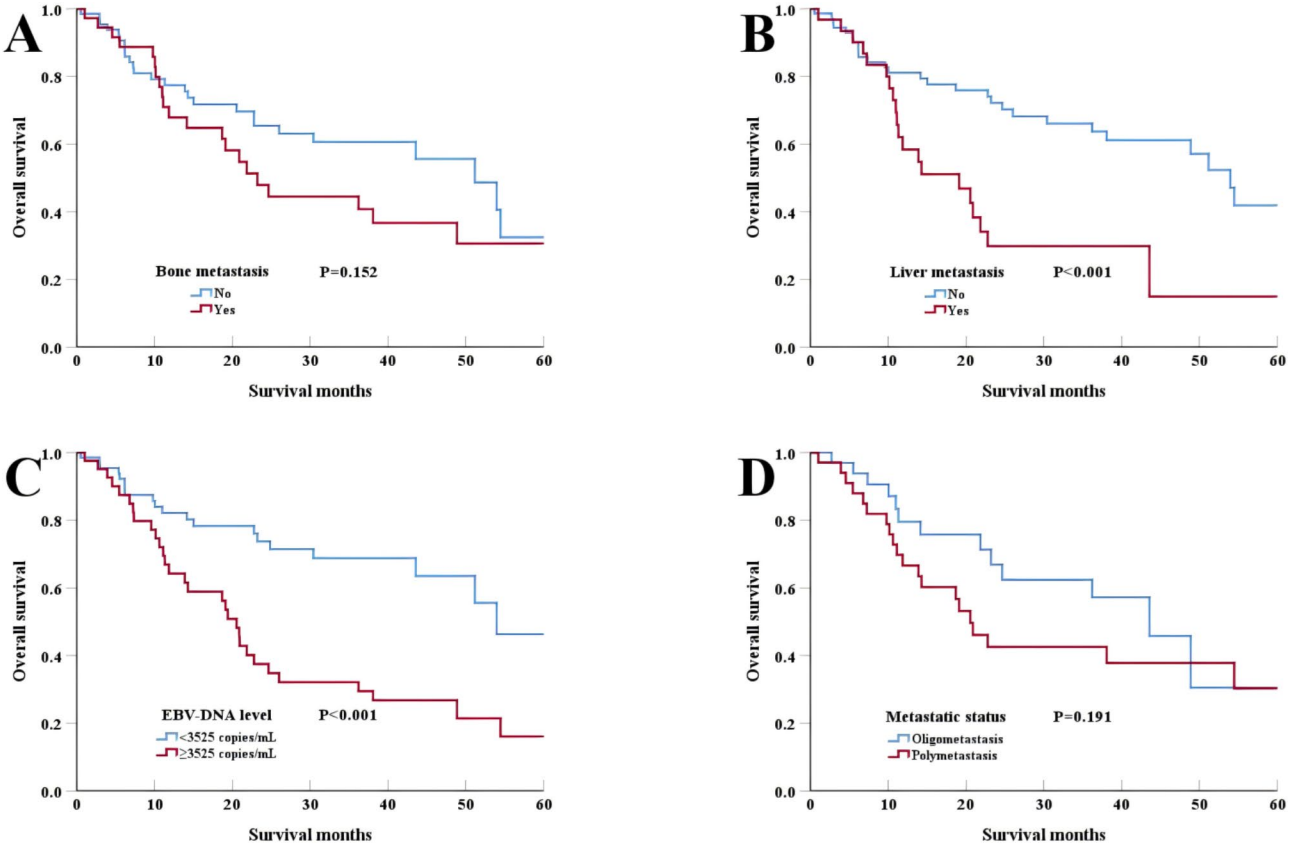

Fig. 3 Overall survival in patients (**A**, bone metastasis; **B**, liver metastasis; **C**, EBV-DNA levels; **D**, metastatic status)

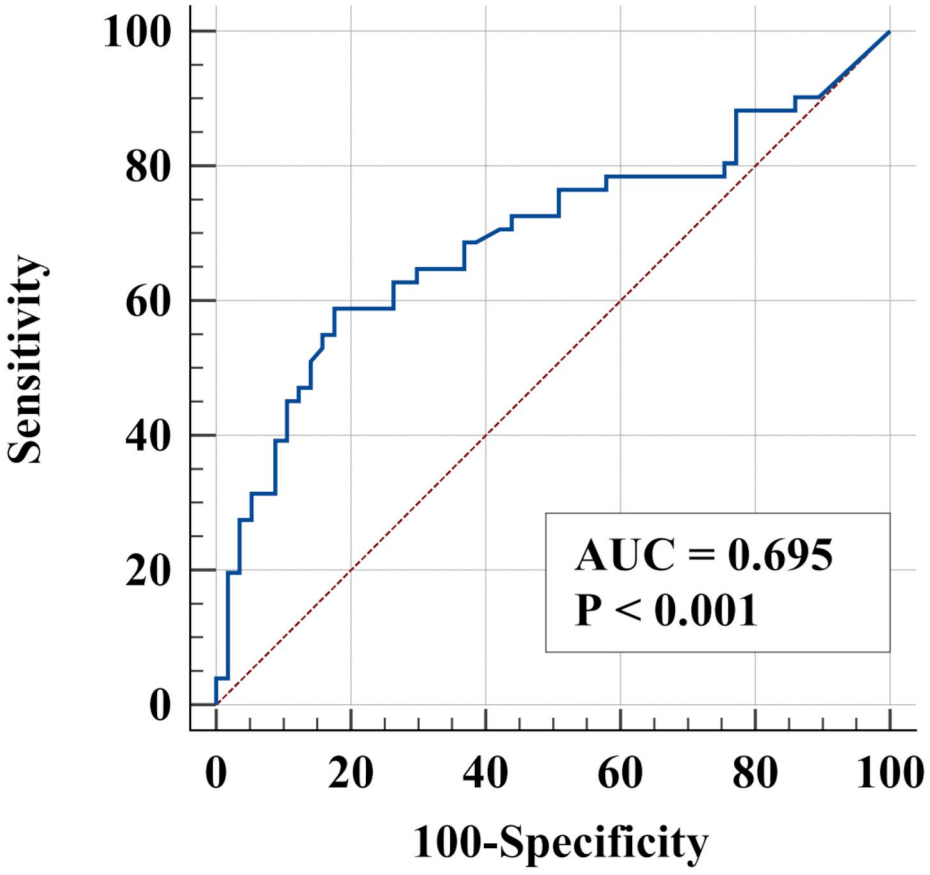

Fig. 4 Receiver operating characteristic curve analysis for assessing the optimal cut-off value of EBV DNA levels at disease progression on overall survival

analysis further confirmed that liver metastasis (HR 2.261, 95% CI 1.193–4.287, $P=0.012$) and EBV-DNA levels ≥ 3,525 copies/mL at progression (HR 2.239, 95% CI 1.226–4.089, $P=0.009$) independently correlated with poorer OS (Table 3).

Discussion

In this study, we explored the patterns of disease failure and survival outcomes in rmNPC with respect to EBV-DNA levels. Our findings indicated distinct patterns of disease failure based on EBV-DNA levels, which showed that patients with liver metastasis and higher EBV-DNA levels experienced significantly poorer OS.

EBV-DNA is derived from tumor cells and serves as an effective biomarker for detecting LRR and DM, highlighting its diagnostic and prognostic significance for NPC [12, 20]. Our study showed that 76.9% of LRR patients had positive EBV-DNA, compared to 97.1% of DM patients. Furthermore, there was a significant difference in EBV-DNA levels between the LRR and DM groups ($P<0.001$). This aligns with findings by Hong et al., who reported detectable EBV-DNA in 81.5% of patients with disease failure, with 65.4% positive in the LRR group and 96.4% in the DM group [21]. Hsu et al. also observed median EBV-DNA levels of 1,965 copies/mL in DM patients compared to 264 copies/mL in LRR patients [10], consistent with our results. Radiotherapy-induced local fibrosis and vascular occlusion can impede EBV-DNA release into circulation, explaining the elevated EBV-DNA levels in DM patients relative to those with LRR. The lower EBV-DNA positivity rate in LRR patients (76.9%) compared to those with DM or those undergoing initial treatment highlights EBV-DNA's potential as a biomarker for differentiating between distant and locoregional disease progression [22]. Elevated EBV-DNA levels may signify a higher systemic disease burden, indicating more aggressive tumor behavior and a propensity for metastasis beyond the primary site.

Our study found no significant association between elevated EBV-DNA levels and specific sites of organ metastasis or LRR. However, higher EBV-DNA levels may reflect the tumor burden and dynamics in rmNPC, suggesting its value as a marker for monitoring disease status and progression. These findings are consistent with literature emphasizing EBV-DNA's utility in NPC management [23–25]. Close surveillance should be considered

Table 3 Univariate and multivariate analysis of prognostic factors to overall survival

Variables	Univariate			Multivariate		
	HR	95% CI	P	HR	95% CI	P
Age (years)						
≤ 50	1			—		
> 50	1.391	0.802–2.415	0.240	—	—	—
Gender						
Male	1			—		
Female	0.770	0.361–1.638	0.497	—	—	—
Smoking history						
No	1			—		
Yes	1.514	0.864–2.651	0.147	—	—	—
Alcoholic history						
No	1			—		
Yes	0.905	0.507–1.615	0.737	—	—	—
Bone metastasis						
No	1			—		
Yes	1.989	0.856–2.671	0.155	—	—	—
Liver metastasis						
No	1			1		
Yes	2.854	1.548–5.262	0.001	2.261	1.193–4.287	0.012
Lung metastasis						
No	1			—		
Yes	1.193	0.615–2.307	0.599	—	—	—
DLNs metastasis						
No	1			—		
Yes	1.038	0.534–2.018	0.912	—	—	—
Failure patterns						
OM ± LRR	1			—		
PM ± LRR	1.447	0.750–2.792	0.271	—	—	—
Only LRR	0.919	0.447–1.892	0.819	—	—	—
DM [#]						
No	1			—		
Yes	1.283	0.687–2.396	0.434	—	—	—
LRR						
No	1			—		
Yes	0.830	0.463–1.488	0.531	—	—	—
EBV-DNA levels at progression (copies/mL)						
< 3525	1			1		
≥ 3525	2.759	1.570–4.849	< 0.001	2.239	1.226–4.089	0.009

OS, overall survival; HR, hazard ratio; CI, confidence interval; LRR, locoregional recurrence; DM, distant metastasis; DLNs, distant lymph nodes; OM, oligometastatic disease; PM, polymetastatic disease

[#] includes 7 patients who experienced both locoregional recurrence and distant metastasis simultaneously

for patients with elevated EBV-DNA, while less frequent monitoring may suffice for those with undetectable levels post-treatment, optimizing resources and reducing patient burden. Current NPC follow-up protocols include monitoring locoregional areas and distant organs like the lungs, liver, and bones [26, 27]. The ESMO guidelines recommend annual plasma EBV-DNA evaluation for recurrence diagnosis [26], while the Chinese Society of Clinical Oncology advises assessment every 3–6 months during the first three years post-treatment [27]. Our study shows most patients with detectable EBV-DNA at initial diagnosis retained this status after treatment failure. The median time to first LRR and DM was 22.8 and 15.8 months, respectively, suggesting that future follow-up models could benefit from an EBV-DNA-driven approach, especially in the first three years post-diagnosis. This strategy offers a non-invasive monitoring option, advantageous compared to traditional imaging that involves radiation or invasive procedures. However, a consistent standard for EBV-DNA detection in NPC is currently lacking [24, 28]. To effectively integrate EBV-DNA into clinical practice, standardized measurement

and interpretation protocols are necessary. Collaborative efforts across institutions might help develop consensus guidelines, ensuring consistent and accurate use of this biomarker in routine NPC management.

Interestingly, while patients with PM had significantly higher EBV-DNA levels than those with OM, OS did not differ significantly between these groups. Advances in systemic therapies, including targeted therapies, immunotherapies, and improved chemotherapeutic regimens [29–31], may have contributed to enhancing OS, regardless of metastasis extent.

In our study, three patients with initially negative EBV-DNA remained negative throughout treatment and disease progression, with OS durations of 4.3, 39.8, and 54.0 months, respectively. As negative EBV-DNA cases are relatively rare in the Chinese population, there is limited understanding of their clinical characteristics and the prognostic differences between EBV-DNA positive and negative patients. A large multi-center study suggested that EBV-DNA positive patients tended to achieve better survival compared to negative patients, potentially indicating a need for enhanced systemic therapies for the latter group [32]. Additionally, the systemic infection response index may provide more accurate risk stratification and prognosis predictions for patients with negative EBV-DNA. It highlights the need for increased attention to these patients, focusing on inflammatory and immune indicators, essential pre-treatment examinations, and close monitoring to develop precise treatment strategies tailored to individual conditions [33].

Liver metastasis is also a common site for DM in NPC [34, 35]. Multiple studies have identified liver metastasis as a major prognostic factor in rmNPC, with a significantly worse prognosis than other metastatic sites [34–36]. In our study, patients without liver metastasis had significantly better OS than those with liver metastasis (median OS: 54.0 vs. 14.3 months, $P<0.001$). The liver's extensive vascularization may facilitate the dissemination and proliferation of metastatic tumor cells. Although our study was conducted in the era of immunotherapy, the liver's immunosuppressive microenvironment might hinder the effectiveness of such treatments [33, 36]. Consequently, liver metastasis not only signifies a more aggressive disease phenotype but also presents unique biological challenges adversely affecting patient prognosis. Therefore, exploring more active and effective local and systemic treatment strategies for patients with liver metastasis is critical. Recent research focuses on enhancing immunotherapy effectiveness through approaches like stereotactic body radiation therapy [33, 37].

Our study also found that patients with rmNPC displaying elevated EBV-DNA levels (≥3,525 copies/mL) had significantly poorer OS. This aligns with previous research identifying EBV-DNA levels as a robust prognostic marker in NPC post-treatment and in rmNPC settings [23–25, 38, 39]. These findings suggest that EBV-DNA levels not only indicate tumor load but also the disease's aggressiveness. Monitoring EBV-DNA levels can aid in risk stratification and inform treatment decisions, potentially identifying patients who may benefit from more aggressive therapeutic strategies or closer post-treatment surveillance.

Several limitations of this study should be noted. Firstly, the retrospective design and potential selection biases may affect the generalizability of our findings. Secondly, the relatively small sample size in certain metastatic subgroups might limit the statistical power of our conclusions. Thirdly, the study did not account for the treatment modalities in patients experiencing disease failure, which may impact the results. Finally, variations in EBV-DNA testing standards across different studies may also affect the consistency of the outcomes.

Conclusion

In conclusion, our study provides substantial evidence linking higher EBV-DNA levels with disease failure patterns and identifies liver metastasis and EBV-DNA levels at disease progression as independent prognostic factors for poorer OS in rmNPC patients. These findings underscore the importance of EBV-DNA as a prognostic biomarker and advocate for personalized therapeutic strategies that consider metastatic profiles and viral dynamics. Future prospective studies with larger, more diverse cohorts are essential to validate these findings and further elucidate the role of EBV-DNA in NPC pathogenesis and progression.

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s13027-024-00631-1.

Supplementary Material 1

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Declared none.

Author contributions
Lin-Feng Guo, Guan-Zhong Lu, Zhen-Zhen Lu, and San-Gang Wu drafted the manuscript. Lin-Feng Guo and San-Gang Wu acquired the datasets. San-Gang Wu conceived the study. Lin-Feng Guo conducted the statistical analyses. Guan-Zhong Lu and San-Gang Wu participated in the study design. All authors read and approved the final manuscript.

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Data availability
No datasets were generated or analysed during the current study.

Declarations

Ethics approval and consent to participate

This study was approved by the Ethics Committee of the First Affiliated Hospital of Xiamen University, and we obtained informed consent from the patients.

Consent for publication

Not applicable.

Human and animal rights

This research was conducted on humans in accordance with the Helsinki Declaration of 1975, as revised in 2013 [http://ethics.iit.edu/ecodes/node/3931].

Competing interests

The authors declare no competing interests.

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References

1. Chen YP, Chan ATC, Le QT, Blanchard P, Sun Y, Ma J. Nasopharyngeal carcinoma. Lancet. 2019;394(10192):64–80. https://doi.org/10.1016/S0140-6736(19)30956-0. Epub 2019 Jun 6. PMID: 31178151.
2. Bray F, Laversanne M, Sung H, Ferlay J, Siegel RL, Soerjomataram I, Jemal A. Global cancer statistics 2022: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin. 2024;74(3):229–63. https://doi.org/10.3322/caac.21834.
3. Yin WJ, Mao W, Yang F, Wang MY, Liu JQ, Chen DP, Zhou LP, Chen SD, Jiang XZ, Ou YM, Wu Z, Qi B. Effect analysis of 847 nasopharyngeal carcinoma cases treated with intensity modulated radiation: experience and suggestions. Oral Oncol. 2024;158:107001. https://doi.org/10.1016/j.oraloncology.2024.107001.
4. Lee AWM, Ng WT, Chan JYW, Corry J, Mäkitie A, Mendenhall WM, Rinaldo A, Rodrigo JP, Saba NF, Strojan P, Suárez C, Vermorken JB, Yom SS, Ferlito A. Management of locally recurrent nasopharyngeal carcinoma. Cancer Treat Rev. 2019;79:101890. https://doi.org/10.1016/j.ctrv.2019.101890.
5. Lin M, Zhang XL, You R, Liu YP, Cai HM, Liu LZ, Liu XF, Zou X, Xie YL, Zou RH, Zhang YN, Sun R, Feng WY, Wang HY, Tao GH, Li HJ, Huang WJ, Zhang C, Huang PY, Wang J, Zhao Q, Yang Q, Zhang HW, Liu T, Li HF, Jiang XB, Tang J, Gu YK, Yu T, Wang ZQ, Feng L, Kang TB, Zuo ZX, Chen MY. Evolutionary route of nasopharyngeal carcinoma metastasis and its clinical significance. Nat Commun. 2023;14(1):610. https://doi.org/10.1038/s41467-023-35995-2.
6. Li AC, Xiao WW, Wang L, Shen GZ, Xu AA, Cao YQ, Huang SM, Lin CG, Han F, Deng XW, Zhao C. Risk factors and prediction-score model for distant metastasis in nasopharyngeal carcinoma treated with intensity-modulated radiotherapy. Tumour Biol. 2015;36(11):8349–57. https://doi.org/10.1007/s13277-015-3574-0.
7. Tsao SW, Tsang CM, Lo KW. Epstein-Barr virus infection and nasopharyngeal carcinoma. Philos Trans R Soc Lond B Biol Sci. 2017;372(1732):20160270. https://doi.org/10.1098/rstb.2016.0270.
8. Zheng H, Zhou P, Wang J, Yu YF, Zhou R, Lin Q, Wu SG. Prognostic effect of residual plasma Epstein-Barr viral DNA after induction chemotherapy for locoregionally advanced nasopharyngeal carcinoma. Cancer Med. 2023;12(14):14979–87. https://doi.org/10.1002/cam4.6132.
9. Lan K, Mao J, Sun X, Li S, Xie S, Sun R, Liu S, Mai H. Combined pre-treatment and middle-treatment Epstein-Barr virus DNA load contributes to prognostication and treatment modification in nasopharyngeal carcinoma patients. Ther Adv Med Oncol. 2024;16:17588359231221343. https://doi.org/10.1177/17588359231221343.
10. Hsu CL, Chan SC, Chang KP, Lin TL, Lin CY, Hsieh CH, Huang SF, Tsang NM, Lee LY, Ng SH, Wang HM. Clinical scenario of EBV DNA follow-up in patients of treated localized nasopharyngeal carcinoma. Oral Oncol. 2013;49(6):620–5. https://doi.org/10.1016/j.oraloncology.2013.02.006.
11. Xiao Z, Li K, Su F, Yang X, Zou H, Qu S. Nomogram model of survival prediction for nasopharyngeal carcinoma with lung metastasis: developed from the SEER database and validated externally. Front Oncol. 2024;14:1351578. https://doi.org/10.3389/fonc.2024.1351578.
12. Qu W, Li S, Zhang M, Qiao Q. Pattern and prognosis of distant metastases in nasopharyngeal carcinoma: a large-population retrospective analysis. Cancer Med. 2020;9(17):6147–58. https://doi.org/10.1002/cam4.3301.
13. Zhao L, Lin Q, Gu J, Zhang H, Chen H, Lin Q. A Nomogram for the prediction of prognosis in patients with distant metastases of nasopharyngeal carcinoma. Front Oncol. 2019;9:240. https://doi.org/10.3389/fonc.2019.00240.
14. Xiao XT, Zou SQ, Chen YP, Guo R, Tang LL, Sun Y, Ma J, Li WF. Patterns and prognosis of Local Recurrence of Nasopharyngeal Carcinoma after intensity-modulated Radiotherapy. J Cancer. 2024;15(2):456–65. https://doi.org/10.7150/jca.88148.
15. Liu H, Yang P, Jia Y. Optimal management of oligometastatic nasopharyngeal carcinoma. Eur Arch Otorhinolaryngol. 2022;279(2):567–76. https://doi.org/10.1007/s00405-021-06918-y.
16. Huang S, Jiang F, Cao C, Jin Q, Jin T, Hua Y, Chen X. Long-term efficacy analysis of radiotherapy and local management of metastases in patients with newly diagnosed oligometastatic nasopharyngeal carcinoma: a prospective, single-arm, single-center clinical study. Radiother Oncol. 2024;196:110265. https://doi.org/10.1016/j.radonc.2024.110265.
17. Guo SS, Yang JH, Sun XS, Liu LZ, Yang ZC, Liu LT, Liu SL, Li XY, Lv XF, Luo DH, Li JB, Liu Q, Wang P, Guo L, Mo HY, Sun R, Yang Q, Liang YJ, Jia GD, Zhao C, Chen QY, Tang LQ, Mai HQ. Reduced-dose radiotherapy for Epstein-Barr virus DNA selected staged III nasopharyngeal carcinoma: a single-arm, phase 2 trial. Eur J Cancer. 2023;194:113336. https://doi.org/10.1016/j.ejca.2023.113336.
18. Liu LT, Liang YJ, Guo SS, Xie Y, Jia GD, Wen DX, Tang LQ, Chen QY, Mai HQ. Identifying distinct risks of treatment failure in nasopharyngeal carcinoma: study based on the dynamic changes in peripheral blood lymphocytes, monocytes, N classification, and plasma Epstein-Barr virus DNA. Head Neck. 2022;44(1):34–45. https://doi.org/10.1002/hed.26897.
19. Liang YJ, Wen DX, Luo MJ, Tang LQ, Guo SS, Wang P, Chen QY, Liu LT, Mai HQ. Induction or adjuvant chemotherapy plus concurrent chemoradiotherapy versus concurrent chemoradiotherapy alone in paediatric nasopharyngeal carcinoma in the IMRT era: a recursive partitioning risk stratification analysis based on EBV DNA. Eur J Cancer. 2021;159:133–43. https://doi.org/10.1016/j.ejca.2021.09.045.
20. Tian Y, Huang WZ, Zeng L, Bai L, Han F, Lan Y. The failure patterns of Nasopharygeal Carcinoma after Intensity-Modulated Radiotherapy and implications for Surveillance. Cancer Manag Res. 2022;14:2813–23. https://doi.org/10.2147/CMAR.S347864.
21. Hong RL, Lin CY, Ting LL, Ko JY, Hsu MM. Comparison of clinical and molecular surveillance in patients with advanced nasopharyngeal carcinoma after primary therapy: the potential role of quantitative analysis of circulating Epstein-Barr virus DNA. Cancer. 2004;100(7):1429–37. https://doi.org/10.1002/cncr.20129.
22. Wang WY, Twu CW, Lin WY, Jiang RS, Liang KL, Chen KW, Wu CT, Shih YT, Lin JC. Plasma Epstein-Barr virus DNA screening followed by ^{18}F-fluoro-2-deoxy-D-glucose positron emission tomography in detecting posttreatment failures of nasopharyngeal carcinoma. Cancer. 2011;117(19):4452–9. https://doi.org/10.1002/cncr.26069.
23. Yoshizaki T, Kondo S, Dochi H, Kobayashi E, Mizokami H, Komura S, Endo K. Recent advances in assessing the clinical implications of Epstein-Barr Virus infection and their application to the diagnosis and Treatment of Nasopharyngeal Carcinoma. Microorganisms. 2023;12(1):14. https://doi.org/10.3390/microorganisms12010014.
24. Lee VH, Adham M, Ben Kridis W, Bossi P, Chen MY, Chitapanarux I, Gregoire V, Hao SP, Ho C, Ho GF, Kannarunimit D, Kwong DL, Lam KO, Lam WKJ, Le QT, Lee AW, Lee NY, Leung TW, Licitra L, Lim DW, Lin JC, Loh KS, Lou PJ, Machiels JP, Mai HQ, Mesía R, Ng WT, Ngan RK, Tay JK, Tsang RK, Tong CC, Wang HM, Wee JT. International recommendations for plasma Epstein-Barr virus DNA measurement in nasopharyngeal carcinoma in resource-constrained settings: lessons from the COVID-19 pandemic. Lancet Oncol. 2022;23(12):e544–51. https://doi.org/10.1016/S1470-2045(22)00505-8.
25. Lv J, Xu LX, Li ZX, Lin L, Wu CF, Quan TQ, Zhen ZC, Li WF, Tang LL, Mao YP, Chen L, Guo R, Zhang LL, Ai XL, Wu SY, Hao MY, Wei D, Li JB, Ma J, Chen YP, Zhou GQ, Sun Y. Longitudinal on-treatment circulating tumor DNA as a biomarker for real-time dynamic risk monitoring in cancer patients: the EP-SEASON study. Cancer Cell. 2024;42(8):1401–e14144. https://doi.org/10.1016/j.ccell.2024.07.001.
26. Bossi P, Chan AT, Licitra L, Trama A, Orlandi E, Hui EP, Halámková J, Mattheis S, Baujat B, Hardillo J, Smeele L, van Herpen C, Castro A, Machiels JP, ESMO Guidelines Committee. Electronic address: clinicalguidelines@esmo.org; EURACAN. Nasopharyngeal carcinoma: ESMO-EURACAN clinical

practice guidelines for diagnosis, treatment and follow-up†. Ann Oncol. 2021;32(4):452–65. https://doi.org/10.1016/j.annonc.2020.12.007.
27. Tang LL, Chen YP, Chen CB, Chen MY, Chen NY, Chen XZ, Du XJ, Fang WF, Feng M, Gao J, Han F, He X, Hu CS, Hu DS, Hu GY, Jiang H, Jiang W, Jin F, Lang JY, Li JG, Lin SJ, Liu X, Liu QF, Ma L, Mai HQ, Qin JY, Shen LF, Sun Y, Wang PG, Wang RS, Wang RZ, Wang XS, Wang Y, Wu H, Xia YF, Xiao SW, Yang KY, Yi JL, Zhu XD, Ma J. The Chinese Society of Clinical Oncology (CSCO) clinical guidelines for the diagnosis and treatment of nasopharyngeal carcinoma. Cancer Commun (Lond). 2021;41(11):1195–227. https://doi.org/10.1002/cac2.12218.
28. Lee AWM, Lee VHF, Ng WT, Strojan P, Saba NF, Rinaldo A, Willems SM, Rodrigo JP, Forastiere AA, Ferlito A. A systematic review and recommendations on the use of plasma EBV DNA for nasopharyngeal carcinoma. Eur J Cancer. 2021;153:109–22. https://doi.org/10.1016/j.ejca.2021.05.022.
29. Liu GY, Ye YF, Jiang YF, Chen GJ, Xia WX, Huang YS, Gao TS, Liu YM, Hou YT, Li JF, Liu JH, Lu N, Chen CL, Ke LR, Liang H, Bei WX, Li WZ, Dong SH, Liu Q, Xie C, Yao HR, Xiang YQ. Nab-paclitaxel, cisplatin, and capecitabine versus cisplatin and gemcitabine as first line chemotherapy in patients with recurrent or metastatic nasopharyngeal carcinoma: randomised phase 3 clinical trial. BMJ. 2024;385:e077890. https://doi.org/10.1136/bmj-2023-077890.
30. Yan K, Lim DW, Ma BBBY. Progress in the clinical development of investigational systemic agents for recurrent and metastatic nasopharyngeal carcinoma. Expert Opin Investig Drugs. 2024;1–10. https://doi.org/10.1080/13543784.2024.2401910.
31. Liu X, Shen H, Zhang L, Huang W, Zhang S, Zhang B. Immunotherapy for recurrent or metastatic nasopharyngeal carcinoma. NPJ Precis Oncol. 2024;8(1):101. https://doi.org/10.1038/s41698-024-00601-1.
32. Xiong Y, Yuan M, Liu Z, Huang J, Bi J, Pi G, Li Y, Li Y, He H, Verma V, Tian S, Han G. Long-term outcomes of nasopharyngeal carcinoma by Epstein-Barr Virus Status in the Chinese Population: a Multicenter Investigation. J Clin Med. 2023;12(8):3005. https://doi.org/10.3390/jcm12083005.
33. Yu J, Green MD, Li S, Sun Y, Journey SN, Choi JE, Rizvi SM, Qin A, Waninger JJ, Lang X, Chopra Z, El Naqa I, Zhou J, Bian Y, Jiang L, Tezel A, Skvarce J, Achar RK, Sitto M, Rosen BS, Su F, Narayanan SP, Cao X, Wei S, Szeliga W, Vatan L, Mayo C, Morgan MA, Schonewolf CA, Cuneo K, Kryczek I, Ma VT, Lao CD, Lawrence TS, Ramnath N, Wen F, Chinnaiyan AM, Cieslik M, Alva A, Zou W. Liver metastasis restrains immunotherapy efficacy via macrophage-mediated T cell elimination. Nat Med. 2021;27(1):152–64. https://doi.org/10.1038/s41591-020-1131-x.
34. Xu Y, Huang T, Mao M, Zhai J, Chen J. Metastatic patterns and prognosis of de novo metastatic nasopharyngeal carcinoma in the United States. Laryngoscope. 2021;131(4):E1130–8. https://doi.org/10.1002/lary.28983.
35. Yang H, Liu C, Luo S, Ma R, Zhou Y, Yin Y, Zhao L, Shi M. Prognostic analysis of 152 patients with distant metastasis after intensity-modulated radiotherapy for nasopharyngeal carcinoma. Ann Palliat Med. 2021;10(6):6824–32. https://doi.org/10.21037/apm-21-1279.
36. Wang FH, Wei XL, Feng J, Li Q, Xu N, Hu XC, Liao W, Jiang Y, Lin XY, Zhang QY, Yuan XL, Huang HX, Chen Y, Dai GH, Shi JH, Shen L, Yang SJ, Shu YQ, Liu YP, Wang W, Wu H, Feng H, Yao S, Xu RH. Efficacy, Safety, and Correlative Biomarkers of Toripalimab in previously treated recurrent or metastatic nasopharyngeal carcinoma: a phase II clinical trial (POLARIS-02). J Clin Oncol. 2021;39(7):704–12. https://doi.org/10.1200/JCO.20.02712.
37. Jiang C, Zhang ZH, Li JX. Consideration on immunotherapy of liver metastases of malignant tumors. World J Gastrointest Surg. 2024;16(8):2374–81. https://doi.org/10.4240/wjgs.v16.i8.2374.
38. Xu JY, Wei XL, Ren C, Zhang Y, Hu YF, Li JY, Chen JL, Wang YQ, Han F, Wang FH. Association of plasma Epstein-Barr Virus DNA with outcomes for patients with recurrent or metastatic nasopharyngeal carcinoma receiving anti-programmed cell death 1 immunotherapy. JAMA Netw Open. 2022;5(3):e220587. https://doi.org/10.1001/jamanetworkopen.2022.0587.
39. Jiang Y, Fang T, Lu N, Bei W, Dong S, Xia W, Liang H, Xiang Y. Anti-PD1 rechallenge in combination with anti-angiogenesis or anti-EGFR treatment beyond progression in recurrent/metastatic nasopharyngeal carcinoma patients. Crit Rev Oncol Hematol. 2023;190:104113. https://doi.org/10.1016/j.critrevonc.2023.104113.

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