Received: 29 July 2022 | Accepted: 27 October 2022

DOI: 10.1002/jmv.28285

SHORT COMMUNICATION

JOURNAL OF MEDICAL VIROLOGY WILEY

Comprehensive analysis of HHV-6 and HHV-7-related gene signature in prognosis and response to temozolomide of glioma

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Funding information

National Natural Science Foundation of China; Medico-Engineering Cooperation Funds from University of Electronic Science and Technology of China; Fundamental Research Funds for the Central Universities

Abstract

Human herpesvirus (HHV)-6 and HHV-7 have been detected in central nervous system and glioma tissue, while their exact role in glioma remains uncertain. Omics profiles and clinical information were downloaded from public databases, including The Cancer Genome Atlas cohort for training set and the Chinese Glioma Genome Atlas cohorts for validation sets. Differentially expressed genes between HHV-6 and HHV-7 infected or noninfected glioma patients were screened for establishing the HHV-6 and HHV-7 infection (HI) model through Lasso regression analysis. Bioinformatics methods were used to analyze the correlation between HI scores and prognosis, metastasis in glioma patients. Predictable efficacy of HI in temozolomide-resistance and HI-related genetic signatures were also explored. The HI model was constructed as: Risk score = (0.014709*DIRAS3) + (0.029787*TEX26) + (0.223492*FBXO39) + (0.074951*MYBL1) + (0.060202*HILS1). The five gene signature showed good performance in predicting survival time for glioma patients, while higher HI score is correlated with malignant features. Moreover, DNA mismatch repair genes were augmented in glioma patients with higher HI score as well as nonresponse to temozolomide treatment, which was in parallel with the transcriptomic result of temozolomide-resistant glioma cell. Targeting the five gene signature is beneficial for prognosis of glioma patients, especially in glioma patients underwent temozolomide treatment.

KEYWORDS

DNA mismatch repair, glioma, human herpesvirus 6/7, prognosis, temozolomide

Luoyi Chen, Xinchen Zhao and Yuyang Liu are co-first authors.
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Abbreviations: BBB, blood-brain barrier; CGGA, Chinese Glioma Genome Atlas; DEGs, differentially expressed genes; EBV, Epstein-Barr virus; GSCA, gene set cancer analysis; GSEA, gene set enrichment analysis; HBV, hepatitis B virus; HCMV, human cytomegalovirus; HCV, hepatitis C virus; HHV, human herpesvirus; HPV, human papilloma virus; IL-6/8, interleukin 6/8; LASSO, least absolute shrinkage and selection operator; MMR, mismatch repair; ROC, receiver-operating characteristic; TCGA, The Cancer Genome Atlas; TMZ, Temozolomide; TNF, tumor necrosis factor.

1 | INTRODUCTION

Glioma represents the most common and aggressive primary brain tumor, labeled with poor outcome and high recurrence rate. Resistance to standard treatment (e.g., Temozolomide, radiotherapy) or innovative therapy (e.g., immune therapy, targeted therapy) restricts clinical benefit in numerous patients, the median survival time of them is only 12–15 months.[1] With the development of sequencing technology, growing numbers of viral related features have been illustrated in aggressive or drug-resistant processes of tumor. To date, various viruses have been unearthed to be participated in types of human cancers. In particular, viruses bearing integrative ability such as human cytomegalovirus (HCMV), human papilloma virus (HPV), Epstein-Barr virus, and hepatitis B virus have been positively detected in glioma tissue.[2]

Human herpesvirus (HHV) are contagious double-stranded DNA viruses, while nine species of HHV can infect humans. HHV-6 and HHV-7 belong to *Roseolovirus*, which are named by being the sixth or seventh isolated herpesvirus, sharing partial homology with HCMV. HHV6 is comprised of HHV-6A and HHV-6B, which establishes acute, incessant and permanent infection especially in immunocompromised host.[3] Both DNA and protein of HHV6 have been detected in glioma tissues, while more production of interleukin 6/8 (IL-6/8), tumor necrosis factor α, and transforming growth factor β (TGF-β) were detected in the cyst fluid specimens from HHV-6-positive glioma patients.[4] Besides, a substantial proportion (36.7%) of intracephalic HHV-7 DNA has been confirmed in healthy adults.[5] Other viral components of HHV-7 including messenger RNA (mRNA) and protein were also positively detected in brain tissue, especially restricted to oligodendrocytes.[6] However, the role of HHV infection (HI) in malignancy of glioma remains unclear. In this article, we explored correlation between HHV-6 and HHV-7 and glioma as well as Temozolomide (TMZ)-resistance, which is supposed to be prognostic valuable for glioma patients.

2 | METHODS

2.1 | Public data acquisition

The omics data and clinical information of glioma patients were downloaded from the Cancer Genome Atlas (TCGA; https://portal.gdc.cancer.gov/) and the Chinese Glioma Genome Atlas (CGGA; http://www.cgga.org.cn/) databases.[7] Quantification data of HHV-6 and HHV-7 infection in glioma patients were collected from previous literature, which scanned and quantified viral contents by using controlled raw sequencing data of cancer patients from TCGA database.[8,9] As mentioned in these references, a score of normalized reads per million was defined to hit over the total reads of each virus in the samples. Raw data of viral contents was downloaded from the online source: https://gdc.cancer.gov/about-data/publications/panimmune.

2.2 | Omics data analysis

Differentially expressed genes (DEGs) were analyzed by using transcriptional sequencing data and using the "DESeq. 2" and "ggplot2" packages in R platform, with the definition of foldchange >1.5 and false discovery rate (FDR) < 0.05. Gene Ontology analysis was presented using these DEGs to illustrate the differentially expressed pathways or symptoms through gene set enrichment analysis (GSEA) software.[10]

2.3 | Establishment and verification of HI gene signature

DEGs between HHV-6 and HHV-7 infected or noninfected glioma patients were included for least absolute shrinkage and selection operator (LASSO) analysis by utilizing "glmnet" package.[11] Variable selection and shrinkage were carried out. Through tenfold cross-verification, the penalty parameter (λ) of HHV-6 and HHV-7 infection (HI) model was determined, when corresponded to the lowest partial likelihood deviance. The HI score was calculated based on the formula constructed of expression level of candidate genes and their regression coefficient: HI score = esum (regression coefficient × gene's expression). Glioma patients were grouped into HI-High or HI-Low in line with the median value of HI scores. Univariate or Multivariate Cox regression models were established sequentially for assessing the predictive independency of candidate genes. Time-dependent receiver-operating characteristic (ROC) curve analyses were performed for evaluating the predictive power of the gene signature with "pROC" package.

2.4 | TMZ-resistance related gene network and drug sensitivity analysis

The medication and clinical follow-up information of patients were integrated to perform Kaplan–Meier analysis using the "survival" package. The Pearson's correlation coefficient between HI score and clustered genes were calculated by using the "corrplot" package. The correlation map was constructed by using the "gephi" software. Gene panels of DNA mismatch repair (MMR) were summarized in previous literature.[12] Drug sensitivity correlated to major MMR genes were calculated using Pearson correlation method via gene set cancer analysis platform.[13]

2.5 | Transcriptomic sequencing

The TMZ-resistant U87MG cell line (U87MG-TMZ-R) was constructed as mentioned in our previous research.[14] Total RNA of U87MG and U87MG-TMZ-R cells were extracted with Trizol reagent, for isolating poly(A)-containing mRNA and noncoding RNA with

beads bearing oligo(dT). Sequenced reads were trimmed for adaptor sequence and masked for low-complexity or low-quality sequences, then mapped to the hg19 whole genome using HISAT2. The sequencing experiment was performed by HaploX Genomic Center.

3 | RESULTS

3.1 | Identification of HHV-6 and HHV-7 associated risk factors in glioma

In an attempt to investigate the possible correlation between virus infection and malignancy of glioma, we obtained the clinical information and normalized viral loads in 513 low grade of glioma patients from previous literature. Among them, positive HHV-6 and HHV-7 infection was detected in 24 patients. As presented in Table 1, there is no alteration in gender, age, pathological grade between HHV-6 and HHV-7 infected or noninfected LGG patients. To be noticed, HHV-6 and HHV-7 infected patients display higher IDH mutation but lower 1p/19q co-deletion (codel) compared to noninfected patients, suggesting a distinguished genetic pattern. To unearth their transcriptomic difference, we preprocessed their transcriptomic data and screened DEGs. With foldchange >1.5 and FDR value <0.05, a total of 82 upregulated and 578 downregulated genes were identified in infected group (Figure 1A and Table S1). Subsequently, LASSO analysis was performed by using these candidate DEGs to generate coefficients of them (Figure 1B and Table S2). According to the minimum value of λ ($\lambda_{min} = 0.074$), 10 genes were selected as the most valuable prognostic genes (Figure 1C), and hazard ratio for each gene was calculated by Univariate Cox model (Figure 1D). For further briefing their role, statistical significance of these genes was calculated by using Multivariate Cox (proportional hazards) model (Figure 1E,F). With Pr(>|z|) <0.05, five genes were finally defined as risk factors to construct the risk formula: Risk score = (0.014709*DIRAS3) + (0.029787*TEX26) + (0.223492*FBXO39) + (0.074951*MYBL1) + (0.060202*HILS1). Through ROC analysis, we revealed the risk predictive formula possessed a good prognostic performance with area under the ROC curves (AUCs) at 1, 2, 3 years of 0.89, 0.90, 0.90, respectively (Figure 1G). Result of Kaplan–Meier survival analysis also suggested higher risk scores were associated with poorer overall survival (OS) rates in LGG patients (Figure 1H).

3.2 | Validation of the signature in CGGA cohorts

Based on this formula, other two GBM cohorts from CGGA database were obtained for validation (Table S3). Risk scores (named as HHV index, HI) of each patient were calculated with the formula as described above. In each cohort, GBM patients were stratified into high- (HI-High) or low-risk (HI-Low) subgroups. As presented in Figure 2A and 2D, more death events were enriched in HI-High groups. Moreover, patients in HI-High group are prominently associated with shorter OS time, according to Kaplan–Meier survival analysis (Figure 2B and 2E). AUCs of HI in both two cohorts also indicated diagnostic accuracy (Figure 2C and 2F).

Beyond this, correlation between HI and other features were evaluated in GBM patients. There was a slightly increase of HI in older patients (>60 years) than younger patients (≤60 years), but no difference between female or male patients (Figure 2G–I). HI can also reflect malignancy of GBM, since HI increased along with higher pathological grades. Patients with IDH mutant showed relative lower HI, while patients with 1p/19q codel showed relative higher HI. However, when we compared HI score between glioma patients infected with HHV and noninfected patients, no significant difference was observed (Figure S1).

3.3 | HI effects in predicting outcome of TMZ-based treatment

Considering that TMZ-based chemotherapy is the standard treatment for GBM patients, we explored prognostic value HI model in glioma patients underwent TMZ treatment excluding other drugs. According to clinical profiles, patients from TCGA were grouped into responsive group including CR/PR/SD to TMZ treatment, and nonresponsive group namely progressive. Significant elevated HI was observed in responsive group than nonresponsive group, hinting that HI may function to predict resistance to TMZ (Figure 3A). Besides, higher HI in these TMZ-treated patients was connected to an obvious poor survival rate, indicating that HI score act as a negative predictor of outcome to TMZ treatment (Figure 3B). For validation, the survival analysis was also performed in patients from CGGA database. As presented in Figure 3F, hazard ratio between HI-High and HI-Low patients were 1.567 and 5.168, respectively. Even though the *p* value of HR in CGGA-1 cohort indicated difference was insignificant, which might be due in insufficient number of patients after screen.

Next, patients received TMZ treatment were grouped as HI-High and HI-Low based on the median HI score. DEGs were screened out by comparing their transcriptomic data (Figure 3C). Through GSEA, we found that transcriptomic discrepancy presented in two groups, including MMR, homologous recombination, cell cycle, oxidative phosphorylation, taurine and hypotaurine metabolism, and ribosome (Figure 3D). As illustrated in the mutation landscape, missense or nonsense mutation on oncogenic genes were more abundant in in responsive group but rarer in nonresponsive group, such as IDH1, TP53, and ATRX (Figure 3E). Furtherly, we calculated correlation between HI and key factors involved in classical TMZ-resistance related pathways. Consistently, genes participated in DNA repair pathway showed a tight association with HI score (Figure 3F).

Therefore, we matched expression of these genes and HI scores, and observed that gene panel of MMR increased along with the rising of HI scores, such as MLH1, PMS2, etc (Figure 4A). Almost MMR genes were augmented in patients with higher HI scores, as compared from 3 cohorts (Figure 4B). These observations indicated

TABLE 1 Demographic features of the study population

Categories	HHV-6A noninfected	HHV-6A infected	HHV-6B noninfected	HHV-6B infected	HHV-7 noninfected	HHV-7 infected	HHV-6 & HHV-7 noninfected	HHV-6 & HHV-7 infected
Gender								
Female	225/509 (44.2%)	3/4 (75%)	228/510 (44.7%)	0/3 (0%)	220/494 (44.5%)	8/19 (42.1%)	217/489 (44.4%)	11/24 (45.8%)
Male	284/509 (55.8%)	1/4 (25%)	282/510 (55.3%)	3/3 (100%)	274/494 (55.5%)	11/19 (57.9%)	272/489 (55.6%)	13/24 (54.2%)
Age								
≤60	449/509 (88.2%)	3/4 (75%)	449/510 (88%)	3/3 (100%)	435/494 (88.1%)	17/19 (89.5%)	58/489 (11.9%)	3/24 (12.5%)
>60	60/509 (11.8%)	1/4 (25%)	61/510 (12%)	0/3 (0%)	59/494 (11.9%)	2/19 (10.5%)	431/489 (88.1%)	21/24 (87.5%)
Grade								
G2	212/451 (47%)	4/4 (100%)	214/452 (46.9%)	2/3 (66.7%)	209/437 (47.8%)	7/18 (38.9%)	204/432 (47.2%)	12/23 (52.2%)
G3	239/451 (53%)	4/4 (100%)	238/452 (53.1%)	1/3 (33.3%)	228/437 (52.2%)	11/18 (61.1%)	228/432 (52.8%)	11/23 (47.8%)
IDH								
Wildtype	93/506 (18.4%)	1/4 (25%)	94/507 (18.5%)	0/3 (0%)	93/491 (18.9%)	1/19 (5.3%)	92/486 (18.9%)	2/24 (8.3%)
Mutant	413/506 (81.6%)	3/4 (75%)	413/507 (81.5%)	3/3 (100%)	398/491 (81.1%)	18/19 (94.7%)	394/486 (81.1%)	22/24 (91.7%)
1p/19q								
Non-codel	341/489 (69.7%)	3/4 (75%)	342/489 (69.9%)	2/3 (66.7%)	325/494 (65.8%)	19/19 (100%)	322/489 (65.8%)	22/24 (91.7%)
Codel	168/489 (30.3%)	1/4 (25%)	168/489 (30.1%)	1/3 (33.3%)	169/494 (34.2%)	0/19 (0%)	167/489 (34.2%)	2/24 (8.3%)
Median OS (days)	2433	2660	2660	1106	2660	2875	2433	2660
Median PFS (days)	1262	1070	1306	1106	1306	987	1262	987

Note: Age, gender, pathogenic grades, IDH mutation, 1p/19q status, overall survival time and progression-free interval time of each population were presented.

Abbreviations: HHV, human herpesvirus; OS, overall survival.

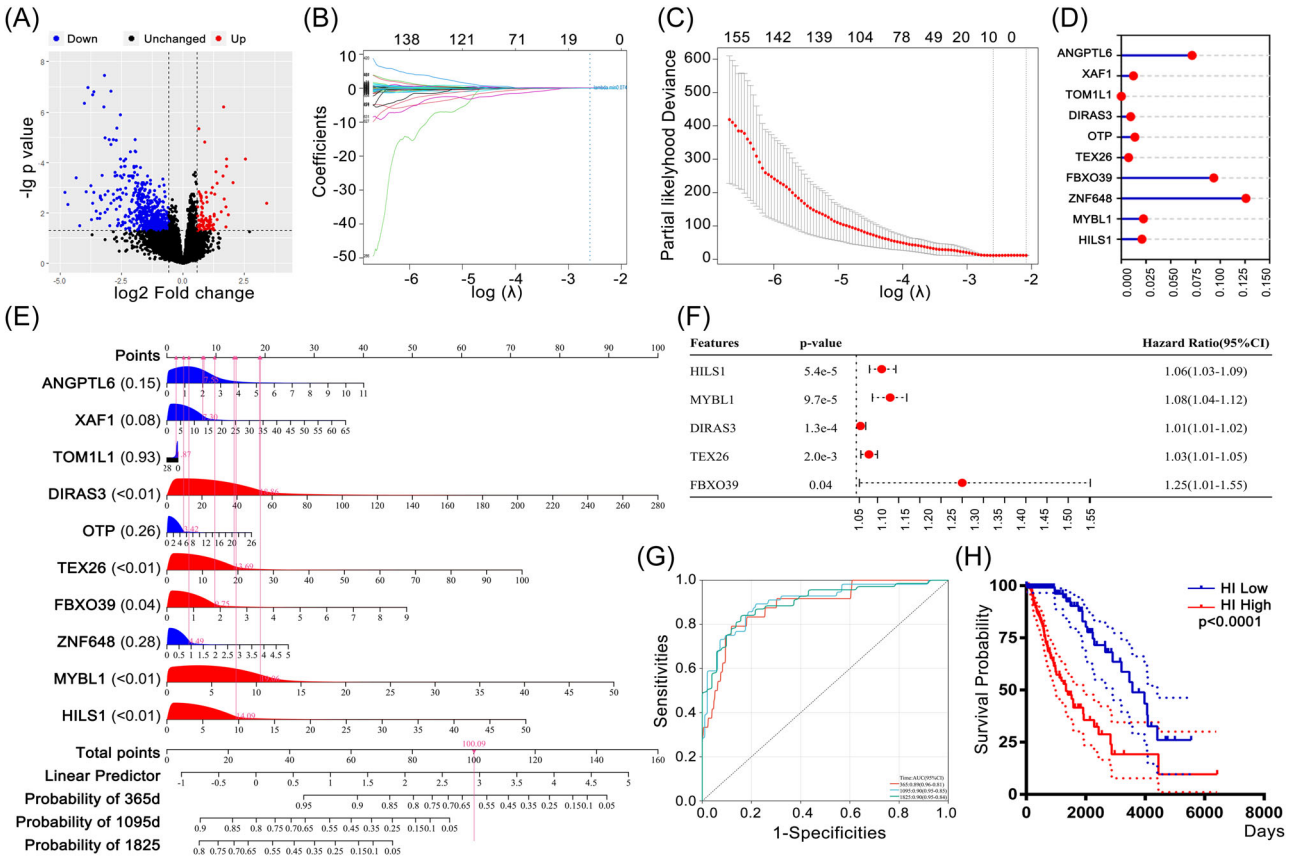

FIGURE 1 The landscape of different virus infection in GBM patients. (A) Comparison of transcriptomic difference between HHV-6 and HHV-7 infected or uninfected patients with glioma. As presented in the volcano plot, the red dots and bule dots indicated upregulated and downregulated genes in infected patients compared to noninfected patients. (B) Establishment of the Lasso model by using HHV-6 and HHV-7 associated DEGs. The changing trajectory of each independent variable was presented (abscissa: corrected lambda; ordinate: coefficient of the independent variable). (C) Partial likelihood deviance for the LASSO coefficient profiles. The log value of the independent variable lambda was presented (abscissa: confidence interval of each lambda; ordinate: errors in cross validation). (D) Univariate Cox regression analysis revealed the association between each candidate genes with OS. (E) Nomogram was drawn to exhibit Multivariate Cox regression analysis revealed the association of 10 candidate genes with OS. Five genes with $p < 0.05$ were screened out for further exploration. (F) Forest plot was drawn to exhibit Multivariate Cox regression analysis of five candidate genes with OS. (G) The 1-, 3-, and 5-year ROC curve in glioma patients from TCGA cohort based on HI model. (H) The KM curve of the HI model-based stratification in TCGA cohort. DEG, differentially expressed genes; HHV, human herpesvirus; HI, HHV infection; KM, Kaplan–Meier; LASSO, least absolute shrinkage and selection operator; OS, overall survival; ROC, receiver operating characteristic; TCGA. The Cancer Genome Atlas

that a stronger repair system in patients with higher HI score, contributing to correct the genetic mistakes introduced by TMZ and leading to TMZ-resistance. On this basis, we explored promising therapeutic agents against patients with high HI score and expression of MMR genes. Based on their gene signature, 28 GSDC-derived and 30 CTRP-derived compounds were identified, which were supposed to be intolerant in TMZ-resistant GBM patients (Figure 4C).

Additionally, we compared transcriptomic data between U87MG cells and U87MG cells resistant to TMZ (U87MG-TMZ-R), which were established in our previous research (Figure 4D and Table S4). Pathways related to cancer and virus infection (HSV-1 or HPV) varied between wildtype cells and TMZ-R cells (Figure 4E). As predicted, HI scores were significantly higher in TMZ-R cells (Figure 4F). Taken together, HI gene signature is valuable to determine outcome in GBM patients, especially in GBM patients underwent TMZ treatment.

4 | DISCUSSION

Crosstalk between cancer and pathogenic microorganisms is a long-lasting issue, while it's estimated that viruses may attributed to 15% of human cancers worldwide.[15] Once infected, certain types of viruses contribute to oncogenesis in various ways: encoding or secreting oncogenic components; presenting persistent inflammation; promoting genomic instability or other genetic alterations in host cells.[16] In this study, we focused on exploring connection between HHV-6 and HHV-7 and glioma.

Several types of cells in the brain can be infected with HHV and trigger neurological dysfunction. Positive detection of HHV-6 has been reported in patients with GBM, which also demonstrated glial-tropic features of HHV-6 in CNS tumors. HHV6 early antigens (gp41) and late antigens (gp116/64/54) have been detected in both primary

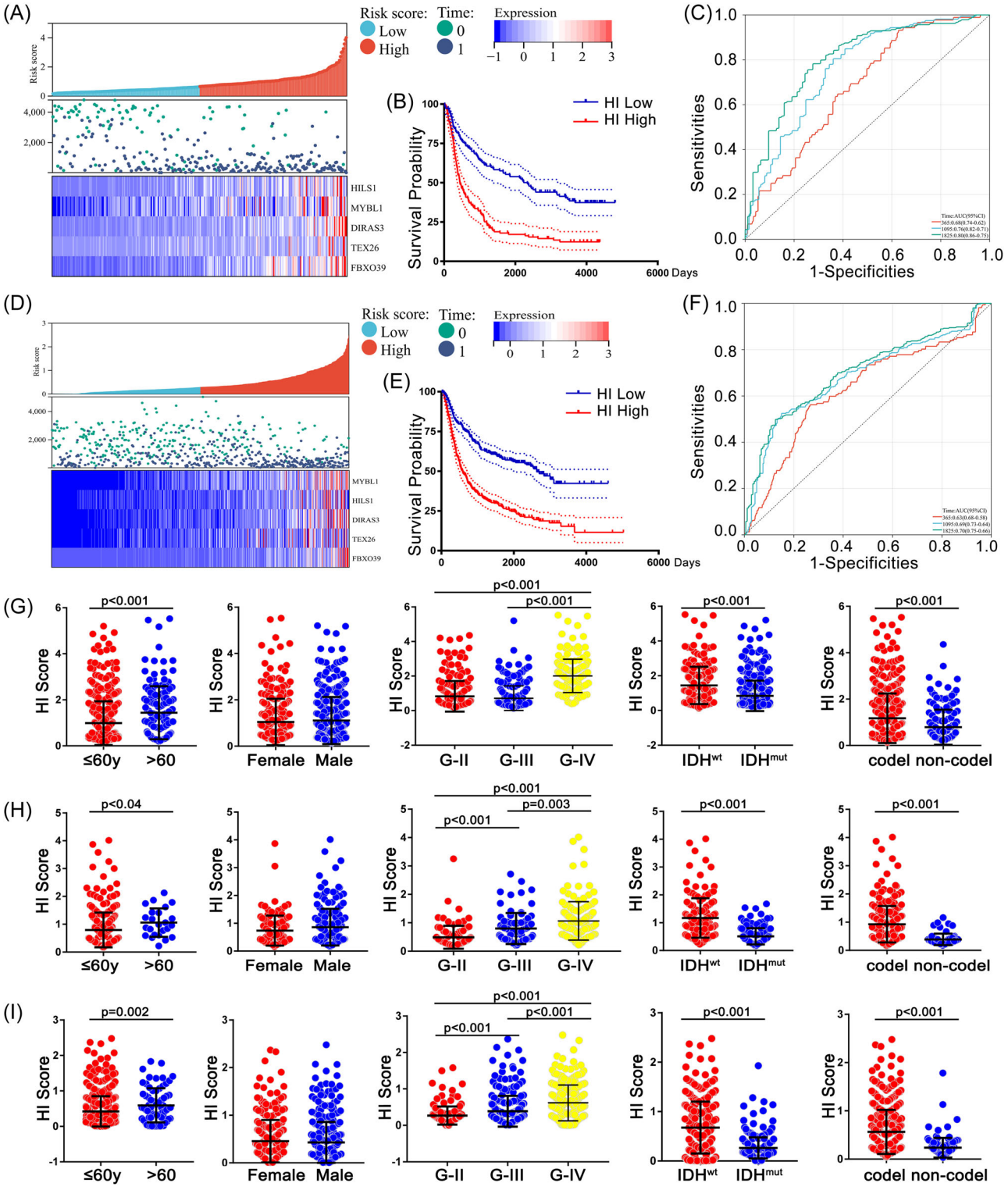

FIGURE 2 HI score is associated with poor prognosis of glioma patients. (A, D) HI score distribution, sample survival, and five gene expression in HI-High and HI-Low groups in CGGA-1 and CGGA-2 cohorts. (B, E) The KM curve of the HI model-based stratification in CGGA-1 and CGGA-2 cohorts. (C, F) ROC curve calculated using HI model in CGGA-1 and CGGA-2 cohorts. (G–I) Comparison of HI scores in glioma patients grouped by different features (age, gender, pathogenic grades, IDH mutation, 1p/19q status), who are collected from TCGA-LGG (G), CGGA-1 (H), CGGA-2 (I) cohorts respectively. CGGA, Chinese Glioma Genome Atlas; HHV, human herpesvirus; HI, HHV infection; KM, Kaplan–Meier; LASSO, least absolute shrinkage and selection operator; OS, overall survival; ROC, receiver operating characteristic; TCGA. The Cancer Genome Atlas

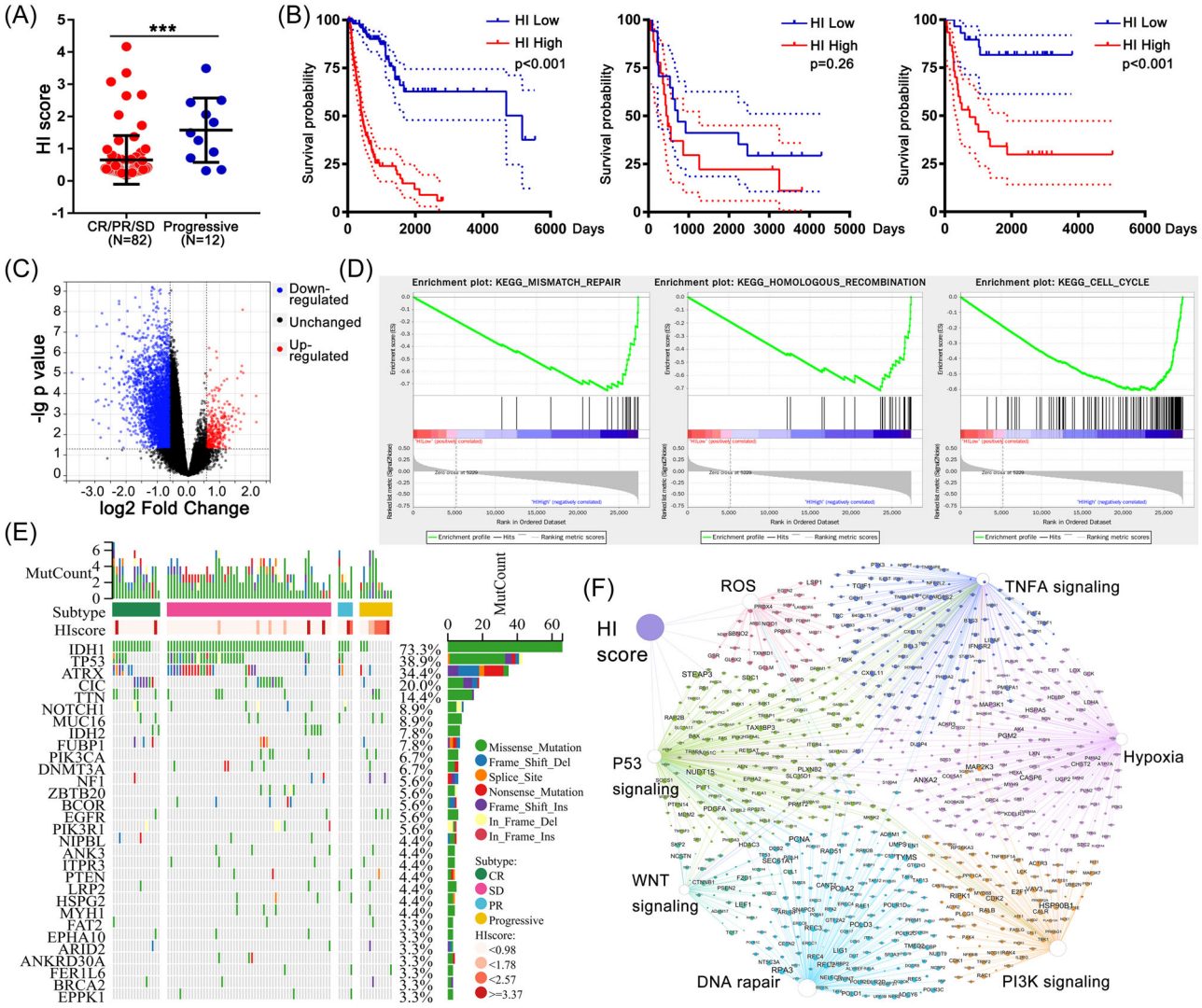

FIGURE 3 Validation of prognostic value of HI in TMZ-treated glioma patients. (A) Comparison of HI scores in glioma patients responsive (CR/PR/SD) or nonresponsive (progressive) to TMZ treatment. CR: complete response; PR: partial response; SD: stable disease. (B) The KM curve of the HI score-based stratification glioma patients from TCGA and CGGA databases. (C) Comparison of transcriptomic difference between HI-High and HI-Low glioma patients underwent TMZ-based therapy. As presented in the volcano plot, the red dots and bule dots indicated upregulated and downregulated genes in patients with higher HI score compared to patients with lower HI score. (D) Top enriched pathways related to DEGs between HI-high and HI-low glioma patients underwent TMZ-based therapy were analyzed by GSEA. (E) SNV waterfall plot showing the mutation distribution of top mutated genes and a classification of variant SNV types in glioma patients received TMZ-based therapy. (F) Network shows interaction between HI score and gene clusters involved in TMZ-resistance. Font sizes of gene names indicated correlation between their expression levels with HI score. CGGA, Chinese Glioma Genome Atlas; DEG, differentially expressed genes; GSEA, gene set enrichment analysis; HHV, human herpesvirus; HI, HHV infection; KM, Kaplan–Meier; LASSO, least absolute shrinkage and selection operator; OS, overall survival; ROC, receiver operating characteristic; TCGA. The Cancer Genome Atlas; TMZ, Temozolomide

and recurrent CNS tumors, which is more frequently in glial tumor.[17,18] Wild intracranial spread of HHV6 is somehow accomplished by upregulated CD46 in glioma, which serves as a major receptor of HHV6.[19,20] Integration of HHV-6 or HHV-7 DNA in CNS have already been confirmed, while these HHV- 6 infection associated CNS complications are common in oncology patients.[21] There is an inherited condition in which complete HHV-6 genome being integrated into telomere of chromosome, named as inherited chromosomally integrated HHV-6 (iciHHV-6), occurred in about 1% humans but more in lymphoma patients. HHV-6A and HHV-6B share almost genes with each other. These genes are termed U1 to U100, and open reading frames within the direct repeats are designated as repeats DR1–DR7.[22] DR7 of HHV6 enhanced migration, invasion and angiogenesis of glioma cells, participated in glioma development and progression, which is consistent with the observation that higher grades of glioma patients have higher HI scores.[23] Furthermore, production of a batch of cytokines such as IL-6, IL-8, and TGF-β are upregulated after DR7 stimulation or HHV6 latent infection, which are involved in pathogenesis of glioma.[4,24] Compared to HHV-6, correlation between HHV-7 and glioma remains limited, while

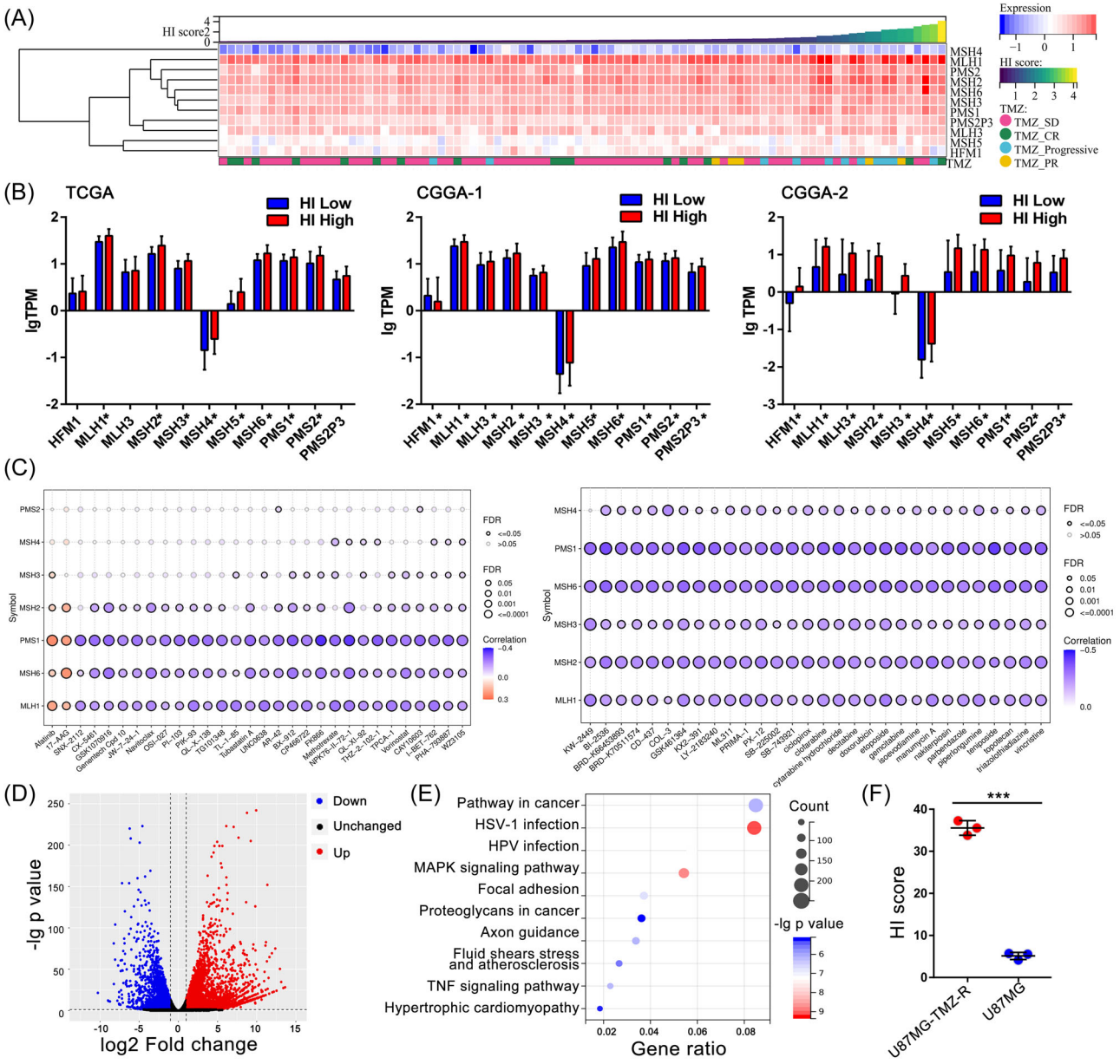

FIGURE 4 Correlation between HI score and MMR gene signatures as well as TMZ resistance. (A) The heatmap exhibited expression of MMR genes and HI scores in glioma patients underwent TMZ-based therapy. (B). Comparison of MMR genes expression in HI-High or HI-Low glioma patients from TCGA, CGGA databases, respectively. (C) Drug sensitivity analysis of MMR genes related IC_{50} drug data from Genomics of Drug Sensitivity in Cancer (GDSC) and Cancer Therapeutics Response Portal (CTRP). Red bubbles represented positive correlations while blue bubbles represented negative correlations. The top 30 ranked drugs were included. (D) Comparison of transcriptomic difference between U87MG and U87MG-TMZ-R cells. As presented in the volcano plot, the red dots and bule dots indicated upregulated and downregulated genes in TMZ-resistant U87MG cells compared to U87MG cells. (E) Gene ontology analysis between U87MG and U87MG-TMZ-R cells. Top enriched biology pathways related to DEGs between TMZ-resistant U87MG cells and U87MG cells were presented. (F) Comparison of HI scores between U87MG-TMZ-R cells and U87MG cells. CGGA, Chinese Glioma Genome Atlas; DEG, differentially expressed genes; HHV, human herpesvirus; HI, HHV infection; KM, Kaplan–Meier; LASSO, least absolute shrinkage and selection operator; MMR, mismatch repair; OS, overall survival; ROC, receiver operating characteristic; TCGA. The Cancer Genome Atlas; TMZ, Temozolomide

potential association between HHV-7 infection with CNS disorders such as limbic encephalitis has been reported.[25]

Additionally, we examined the correlation of HI and glioma patients' outcome towards TMZ treatment. HI scores were higher in patients who are resistant to TMZ-based treatment, as well as in TMZ-resistant U87MG cells than control group. For multiple herpesviruses, viral DNA is presented in nucleus of infected cells, but maintain with suppressed replication or transcription during latently infection.[26] DNA MMR proteins are recruited to HSV-1 replication compartments, which is required for efficient HSV-1

replication.[27] In particular, MLH1 and MSH2 act successionally to guarantee recruiting or synthesizing viral genome. Our results also showed that a set of MMR genes (e.g., MLH1/3, MSH2/3, PMS1/2) were significantly augmented in glioma patients with higher HI scores, which was in consistent with previous findings that knockdown of MLH1 reversed TMZ sensitization in different GBM cell lines.[28] As for the observation of undifferentiated HI scores between glioma patients infected with HHV and noninfected patients, small number of infected patients might be responsible for that: number of patients infected with HHV-6A = 4, HHV-6B = 3, HHV-7 = 19. Besides, weight coefficients of those genes were majorly dependent on their contribution to prognosis in glioma patients rather than degree of HI.

Taken together, we constructed a prognosticative approach of gene signature related to HHV-6 and HHV-7 infection, which provided new insights into crucial MMR machinery in viral infection and response to TMZ in glioma.

AUTHOR CONTRIBUTIONS

Ying Shi and Chuan Xu: conception and design. **Luoyi Chen, Yuyang Liu, Mengwan Wu**: collection and assembly of data. **Xinchen Zhao and Shurong Li**: data analysis and interpretation. **Luoyi Chen, Xinchen Zhao, Yuyang Liu**: manuscript writing. All authors read and approved the final manuscript.

ACKNOWLEDGMENTS

We want to express our thanks to the researchers who performed sequencing of glioma patients and shared the precious data, especially Professor Marcus L. Ware and Professor Erik K. Flemington. The results here are based upon data generated by the TCGA Research Network (https://portal.gdc.cancer.gov/) and CGGA database (http://www.cgga.org.cn/). This study was supported by the National Natural Science Foundation of China (no. 81873048) to CX; Medico-Engineering Cooperation Funds from University of Electronic Science and Technology of China (no. ZYGX2021YGCX004) to CX; the National Natural Science Foundation of China (no. 82203539) to YS; Medico-Engineering Cooperation Funds from University of Electronic Science and Technology of China (No. ZYGX2021YGCX018) to YS; and the Fundamental Research Funds for the Central Universities (no. ZYGX2020KYQD002) to YS.

CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT

Raw transcriptomic data of glioma patients were downloaded from the TCGA and CGGA databases. The datasets supporting the conclusions of this article are available in the Supporting Information Material.

ETHICS STATEMENT

Ethics approval and consent to participate the data of this study are from TCGA and CGGA database, and do not involve animal experiments and human specimens, no ethics-related issues.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Chen L, Zhao X, Liu Y, et al. Comprehensive analysis of HHV-6 and HHV-7-related gene signature in prognosis and response to temozolomide of glioma. *J Med Virol*. 2022;95:e28285. doi:10.1002/jmv.28285