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ORIGINAL RESEARCH

IncRNA small nucleolar RNA host gene 20 predicts poor prognosis in glioma and promotes cell proliferation by silencing P21

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Xiang-Sheng Li¹ Fa-Zheng Shen¹ Li-Yong Huang¹ Lei Hui¹ Rui-Hua Liu¹ Yan-Juan Ma² Bao-Zhe Jin¹

¹Department of Neurosurgery, The First Affiliated Hospital of Xinxiang Medical University, Xinxiang 453000, Henan Province, People's Republic of China; ²Department of Emergency, The First Affiliated Hospital of Xinxiang Medical University, Xinxiang 453000, Henan Province, People's Republic of China

Correspondence: Bao-Zhe Jin Department of Neurosurgery, The First Affiliated Hospital of Xinxiang Medical University, No 88 Weihui Health Road, Xinxiang 453100, Henan Province, People's Republic of China Email jinbaozhe2016@163.com



Background: In multiple cancers, long non-coding RNA small nucleolar RNA host gene 20 (IncRNA SNHG20) is generally dysregulated. In the present study, both the biological role and clinicopathological value of *lncRNA SNHG20* in glioma are explored.

Methods: Real-time PCR was employed to determine *lncRNA SNHG20* expression in glioma patients. The prognostic role of expression of *lncRNA SNHG20* was evaluated in a retrospective cohort study. In addition, the association between *lncRNA SNHG20* expression and the clinicopathological features of glioma patients, such as tumor recurrence, survival status, follow-up time, WHO grade, resection extent, tumor location, Karnofsky performance scale score, cystic change, tumor size, gender and age, was discussed. By constructing and transfecting siRNAs that targeted *lncRNA SNHG20* into the glioma U87 cells, the effects of *lncRNA SNHG20* on the proliferation and cell cycle of U87 cells were assessed through cell counting kit-8, colony formation and cell cycle assays, respectively. In addition, Western blot and real-time PCR measured the expression levels of P21 and CCNA1 in U87 cells after being transfected with SNHG20 siRNA.

Results: Our results suggested the high expression of *lncRNA SNHG20* in human glioma tissues compared with normal brain tissues, which was related to recurrence-free survival and poor overall survival in glioma patients. According to the existing retrospective cohort study, high *lncRNA SNHG20* expression was associated with tumor size, extent of resection, WHO grade, follow-up time, survival status and recurrence. Besides, knocking down the expression of IncRNA SNHG20 could inhibit the proliferation and colony formation abilities of glioma U87 cells through cell cycle arrest. Consequently, the expression of CCNA1 was inhibited, and the expression of P21 was up-regulated in U87 cells.

Conclusion: A high IncRNA SNHG20 expression level predicts the poor prognosis for glioma patients. Moreover, IncRNA SNHG20 can promote glioma proliferation through silencing P21 and thus *lncRNA SNHG20* is an independent potential prognostic biomarker for glioma patients. Keywords: IncRNA SNHG20, glioma, clinicopathological, prognosis, proliferation

Introduction

As the most common central nervous system malignant tumor, glioma has the characteristics of poor prognosis, aggressiveness, rapid progression and frequent recurrence.¹ In particular, the median survival time of patients with glioblastoma was shorter than 1 year.² Obviously, dismal was remained in the prognosis for glioma patients although there are a variety of treatment options, such as chemotherapy, radiotherapy and surgery.³ So far, few useful biomarkers have been used to monitor the burden and response to treatment of gliomas. Therefore, prognostic evaluation and

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early diagnosis of glioma are essential for early treatment and improvement of survival rate.

Small nucleolar RNA host gene 20 (*SNHG20*) was originally identified in hepatocellular carcinoma through microarray data, which was a recently identified lncRNA and located at chromosome 17q25.2 position in the human genome.⁴ Recent studies demonstrated that *lncRNA SNHG20* was dysregulated in multiple cancer and it exerts an important role in tumor growth, metastasis, invasion and poor survival.⁵ As a result, *lncRNA SNHG20* is of great importance to tumor progression. However, the role of *lncRNA SNHG20* in the prognosis and tumorigenesis of gliomas has not been fully clarified.

Admittedly, the function and prognostic value of *lncRNA SNHG20* in glioma were first examined in the present study. The study results indicated the up-regulation of *lncRNA SNHG20* expression in glioma tissues. Subsequently, the function and clinical significance of *lncRNA SNHG20* expression in human glioma were explored.

Patients and methods Patients and tissue samples

One hundred eight glioma patients undergoing an initial surgery at the First Affiliated Hospital of Xinxiang Medical University were enrolled into this study from 2011 to 2017. Additionally, epileptic resections obtained 12 normal brain tissues (NBTs). All glioma patients were diagnosed in the pathology department. All samples were stored and frozen in liquid nitrogen. Besides, all patients were naive to therapy before resection. The informed consent was provided by each patient, and the research samples obtained the approval of the Medical Ethics Committee of the First Affiliated Hospital of Xinxiang Medical University.

Quantitative real-time polymerase chain reaction (qRT-PCR) assay

The Trizol reagent was used to extract the total RNA from cells and glioma tissues. qRT-PCR detected the expression of *lncRNA SNHG20* mRNA through the one-step RT-PCR kit (Takara Bio, Inc., Otsu, Japan) according to the manufacturer's protocol. The primers were obtained from Genechem Co. Ltd (Invitrogen, Shanghai, People's Republic of China), and glyceraldehyde 3-phosphate dehydrogenase (GAPDH) was the internal control. The primers included *lncRNA SNHG20*: forward, 5'-ATGGCTATAAATAGATACACGC-3', and reverse, 5'-GGTACAAACAGGGAGGGA-3'; CCNA1: forward, 5'-ATTCATTAAGTGAAATTGTGC-3'; and reverse, 5'-CTTCCATTCAGAAACTTATTG-3'; P21: forward, 5'-CAGAGGAGGGAGGCGCCATGT-3'; and reverse, 5'-GGAAGGTAGAGCTTGGGCAG-3'; GAPDH: forward, 5'-GGAGTCAACGGATTTGGTCGTAT-3' and reverse, 5'-AGCCTTCTCCATGGTGGTGAAGAC-3'. The expression levels of *lncRNA SNHG20* mRNA, P21 and CCNA1 were quantified by the $2^{-\Delta\Delta CT}$ method and further normalized by the expression level of GAPDH mRNA.

Cell lines and transfection

Human glioma cell line U87 cells purchased from Shanghai Cell Bank (Shanghai, People's Republic of China) were cultured in DMEM (Thermo Fisher Scientific, Carlsbad, CA, USA), which were supplemented with 10% FBS (Invitrogen, Carlsbad, CA, USA) with 5% CO_2 at 37°C.

The negative control siRNA (NC siRNA) and siRNA of *lncRNA SNHG20* were purchased from Shanghai Genechem Biotechnology Co., Ltd. The 6×10^5 U87 cells achieved 70% confluence were put in a 6-well plate and infected with NC siRNA or *SNHG20* siRNA. The real-time PCR validated the efficiency of *SNHG20* siRNA transfection.

Western blotting

The tissues and cells were obtained under the instructions of the manufacturer, and a BCA Protein Assay Kit (Pierce, Rockford, IL, USA) was used to determine the protein concentration. The same amount of total protein was isolated by 12.5% SDS-PAGE and transferred to polyvinylidene difluoride (PVDF) membrane. The first antibody was purchased from ABCAM (Cambridge, UK) Trading Co., Ltd., including rabbit anti-P21, anti-CCNA1 and anti-GAPDH antibodies, while horseradish peroxidase combined goat anti rabbit IgG (purchased from Boster Biological Technology Co. Ltd, Wuhan, People's Republic of China) as secondary antibody. At the same time, GAPDH was used as internal control to normalize the expression level of *lncRNA SNHG20*.

Cell counting kit-8 (CCK-8) assay

U87 cells at exponential phase were collected, infected with *SNHG20* siRNA or NC siRNA, inoculated at a density of 3.5×10^3 cells/well, and cultured in the 96-well plates. After 0, 24, 48 and 72 hours, each well was added with $10 \,\mu$ L of CCK-8 solution. After 2 hours, a microplate reader (Thermo Fisher Scientific, Inc., Waltham, MA, USA) was adopted to measure the absorbance at 450 nm. Besides, a total of five repeats/groups were performed, three times independently.

Colony formation assay

After transfection with *SNHG20* siRNA or NC siRNA, glioma U87 cells were counted, plated onto the 6-well cell

culture plates at a density of 300 cells/well and incubated at 37° C for colony formation. Subsequently, the visible colonies were fixed with 4% paraformaldehyde for 30 minutes and stained with 0.1% crystal violet for another 30 minutes. Colony-forming efficiency was calculated as the number of colonies (with the diameter of >0.5 mm).

Flow cytometry

Glioma U87 cells were infected with *SNHG20* siRNA or NC siRNA and incubated at 37°C before being harvested. Then, the cells were washed twice with PBS and subsequently fixed with 75% cold ethanol. The cells were treated with a cell cycle staining kit and incubated in darkness for 30 minutes as directed by the manufacturer. Eventually, the cells were analyzed through flow cytometry.

Statistical analysis

In the current study, the data were conducted with statistical analysis using SPSS 22.0 software (SPSS Inc., Chicago, IL, USA). The measurement data expressed as mean \pm SD were analyzed through Student's *t*-test between two groups. The chi-squared test was carried out to analyze the enumeration data. The Kaplan–Meier analysis was performed to evaluate the survival, and the survival rate was evaluated through the long-rank test. In addition, the Cox regression analysis was also employed to perform univariate and multivariate analyses of survival data. The *P*-value of <0.05 was considered as statistically significant.

Ethics approval and consent to participate

According to the Declaration of Helsinki of 1964 and all subsequent revisions, this study was reviewed and approved by the Ethics committee of the First Affiliated Hospital of Xinxiang Medical University and all patients provided written informed consent according to the local ethics committee regulations.

Results IncRNA SNHG20 was upregulated in human glioma tissues

To determine the differential expression of *lncRNA SNHG20* between NBTs and glioma tissues, the *lncRNA SNHG20* expression levels were determined using real-time PCR assay in 108 glioma samples. The clinicopathological features of glioma patients were shown in Table 1. There was no significant difference in the age and gender distribution between the glioma and NBT groups. The results indicated that *lncRNA*

 Table I
 Clinicopathological characteristics of patient samples

 and expression of IncRNA SNHG20 in glioma

Characteristics	No of cases (%)				
Age (years)					
<50	51 (47.2)				
≥50	57 (52.8)				
Gender					
Female	52 (48.1)				
Male	56 (51.9)				
Tumor size (cm)					
<5	58 (53.7)				
≥5	50 (46.3)				
Cystic change	,				
Absence	61 (56.5)				
Presence	47 (43.5)				
KPS score					
≥80	61 (56.5)				
<80	47 (43.5)				
Tumor location					
Infratentorial	45 (41.7)				
Supratentorial	63 (58.3)				
Extent of resection					
GTR	61 (56.5)				
PR	47 (43.5)				
WHO grade					
1/11	43 (39.8)				
III/IV	65 (60.2)				
Follow-up time (months)	30.907±1.843				
Survival status					
Live	59 (54.6)				
Death	49 (45.4)				
Recurrence					
Absence	52 (48.1)				
Presence	56 (51.9)				
Expression of IncRNA SNHG20					
Low expression	54 (50.0)				
High expression	54 (50.0)				

Abbreviations: GTR, gross total resection; KPS, Karnofsky performance scale; PR, partial resection.

SNHG20 mRNA levels were significantly higher than those in NBT (Figure 1). The results suggested that *lncRNA SNHG20* may be involved in the development of glioma.

Association between *IncRNA SNHG20* expression and clinicopathological characteristics of glioma

For statistical analysis, the glioma patients were classified into the high *lncRNA SNHG20* expression group (n=54) and

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Figure I The *lncRNA SNHG20* expression levels in glioma tissues are outstandingly higher than those in normal brain tissues. Note: *P < 0.05 means vs normal brain tissues.

low IncRNA SNHG20 expression group (n=54) based on the median value of IncRNA SNHG20 expression level. Then, the association between IncRNA SNHG20 expression and clinicopathological characteristics in glioma patients was discussed. Table 2 demonstrates the association between the *lncRNA SNHG20* and the tumor size (P=0.002), extent of resection (P=0.012), WHO grade (P=0.0031), follow-up time (P=0.000), survival status (P=0.000) and recurrence (P=0.000). However, the expression of *lncRNA SNHG20* was not associated with tumor location, Karnofsky performance scale score, cystic change, gender and age $(P \ge 0.05)$. According to the Spearman analysis of the association between IncRNA SNHG20 and clinicopathological characteristics, the expression of IncRNA SNHG20 was associated with tumor size (P < 0.002), extent of resection (P = 0.011), WHO grade (P=0.031), follow-up time (P=0.000), survival status (P=0.000) and recurrence (P=0.000), as presented in Table 3.

High IncRNA SNHG20 expression was associated with poor prognosis for glioma

To investigate the effect of *lncRNA SNHG20* expression on the prognosis of gliomas, Kaplan–Meier analysis and log-rank test were performed in the current study. The results showed that the OS (Figure 2A, P < 0.0001) in glioma patients with high expression were significantly lower than those in glioma patients with low expression. Kaplan–Meier survival analysis of *lncRNA SNHG20* expression was performed in gliomas with different WHO grades and tumor sizes. According to the results, high expression of *lncRNA SNHG20* indicated poor OS in both different grades (Figure 2B and C, I < 0.01)

 Table 2 Association between IncRNA SNHG20 expression and clinicopathological characteristics of glioma patients

Characteristics	IncRNA SNHG20		P-value		
	expression				
	Low, no	High, no	1		
	of cases	of cases			
Age (years)					
<50	26	25	0.847		
≥50	28	29			
Gender					
Female	24	28	0.441		
Male	30	26	1		
Tumor size (cm)					
<5	37	21	0.002		
≥5	17	33			
Cystic change					
Absence	33	28	0.332		
Presence	21	26			
KPS score					
≥80	31	30	0.846		
<80	23	24			
Tumor location					
Infratentorial	21	24	0.558		
Supratentorial	33	30	1		
Extent of resection					
GTR	37	24	0.012		
PR	17	30			
WHO grade					
1/11	27	16	0.031		
III/IV	27	38			
Follow-up time (months)	38.056±2.82	23.759±1.96	0.000		
Survival status					
Live	42	17	0.000		
Death	12	37			
Recurrence					
Absence	36	16	0.000		
Presence	18	38			

Abbreviations: GTR, gross total resection; KPS, Karnofsky performance scale; PR, partial resection.

and different tumor sizes (Figure 2D and E, P < 0.01) glioma patients. Similar to previous results, high expression of *lncRNA SNHG20* indicated poor RFS in total (Figure 2F, P < 0.0001), different grades (Figure 2G and H, P < 0.01) and different tumor sizes (Figure 2I and J, P < 0.01) glioma patients.

All clinical data were conducted with multivariate and univariate Cox regression analyses. The results demonstrated that *lncRNA SNHG20* expression was an independent prognostic factor for OS (Table 4) and RFS (Table 5) in patients with glioma (P < 0.05).

Table	3	Spearman	analysis	of	correlation	between	IncRNA
SNHG2	0 a	nd clinicop	athologica	al			

Variables	IncRNA SNHG20 expression level			
	Spearman correlation	P-value		
Age (years)	0.019	0.849		
Gender	-0.074	0.446		
Tumor size	0.297	0.002		
Cystic change	0.093	0.336		
KPS score	0.019	0.848		
Tumor location	-0.056	0.562		
Extent of resection	0.243	0.011		
WHO grade	0.208	0.031		
Follow-up time	-0.343	0.000		
Survival status	0.465	0.000		
Recurrence	0.037	0.000		

Abbreviation: KPS, Karnofsky performance scale.

Inhibition of *IncRNA SNHG20* expression by *SNHG20* siRNA in human glioma U87 cells

SNHG20 siRNA and NC siRNA were transfected into human glioma U87 cells, so that the function of *lncRNA SNHG20* in glioma can be explored. qRT-PCR was used to analyze the knockdown effects. After transfection, the *lncRNA SNHG20* expression levels in U87 cells of *SNHG20* siRNA group were lower than NC siRNA group (Figure 3).

Downregulation of *IncRNA SNHG20* expression markedly inhibited glioma U87 cell proliferation and colony formation

CCK-8 assays were performed after glioma U87 cells transfected with *SNHG20* siRNA or NC siRNA, and the effect of *SNHG20* on glioma cell growth was examined. In Figure 4, the growth of U87 cells treated with *SNHG20* siRNA was markedly inhibited compared to NC siRNA group. The glioma U87 cell growth was significantly reduced by *SNHG20* siRNA cells on day 4 and day 5 in vitro. Similarly, after down-regulation of *lncRNA SNHG20*, colony formation was also dramatically reduced (P<0.01, Figure 5).

Inhibition of *IncRNA SNHG20* induces G0/ G1 phase arrest in glioma U87 cells

Due to the direct association between proliferation and cell cycle distribution, we investigated the cell cycle distribution of U87 cells after down-regulation of *lncRNA SNHG20*. The cell cycle of U87 cells was detected by flow cytometry. As shown in Figure 6, the *SNHG20* siRNA group displayed the following: (G0/G1 phase, 82.14% \pm 5.32%; S phase, 11.94% \pm 0.82%; and G2/M phase, 5.92% \pm 0.64%), and the NC siRNA group displayed the following distribution: (G0/G1 phase, 58.27% \pm 4.21%; S phase, 28.62% \pm 2.36%; and G2/M phase, 13.11% \pm 1.98%). With the absence of



Figure 2 (Continued)



Figure 2 Kaplan–Meier curves showing the survival of glioma patients with different IncRNA SNHG20 expression levels.

Notes: (A–E) OS curves stratified by *lncRNA SNHG20* expression in total (A), low-grade (B), high-grade (C), small tumor size (D) and lager tumor size (E) glioma patients. (F–J) RFS curves stratified by *lncRNA SNHG20* expression in total (F), low-grade (G), high-grade (H), small tumor size (I) and lager tumor size (J) glioma patients. Abbreviations: OS, overall survival; RFS, recurrence-free survival.

lncRNA SNHG20, the number of cells entering G0/G1 phase increased by 23.8%, and the number of cells entering S phase decreased by 16.7 cells (P<0.01). Knocking down *lncRNA SNHG20* expression significantly increased the proportion of cells in G0/G1 phase and decreased the percentage of cells in S phase and G2/M phase. The above results suggest that *lncRNA SNHG20* inhibits the proliferation of U87 cells through blocking the cell cycle progression in G0/G1 phase.

The change of cell cycle-associated proteins after transfected with SNHG20 siRNA in human glioma U87 cells

As *lncRNA SNHG20* affected cells proliferation through modulation of the cell cycle arrest, cell cycle-regulate gene expressions were examined at the transcriptional and translational levels. Furthermore, U87 cells were transfected with *SNHG20* siRNA or NC siRNA, and the P21 and CCNA1 expression levels were determined by real-time

Table 4 Univariate and multivariate analyse	s for overall survival in glioma patients
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Variable	Univariate analysis			Multivariate analysis		
	P-value	HR	95% CI	P-value	HR	95% CI
Age (years) (<50 vs ≥50)	0.373	1.296	0.732–2.293	0.405	1.297	0.703–2.393
Gender (Female vs male)	0.512	1.208	0.686–2.129	0.742	0.892	0.450–1.766
Tumor size (<5 vs ≥5 cm)	0.000	2.899	1.627–5.165	0.111	1.676	0.889–3.163
Cystic change	0.001	2.686	1.500-4.810	0.002	2.833	1.489–5.390
KPS score	0.001	2.812	1.565–5.051	0.095	1.797	0.903-4.574
Tumor location	0.145	1.543	0.862–2.764	0.073	1.737	0.949–3.179
Extent of resection	0.014	2.025	1.151–3.564	0.965	1.015	0.529–1.948
WHO grade	0.001	3.077	1.629–5.810	0.008	2.669	1.286-5.542
Expression of IncRNA SNHG20	0.000	5.774	2.945-11.319	0.000	4.722	2.189–10.186

Abbreviation: KPS, Karnofsky performance scale.

Variable	Univariate analysis			Multivariate analysis		
	P-value	HR	95% CI	P-value	HR	95% CI
Age (years)	0.340	1.298	0.760-2.215	0.460	1.239	0.703–2.184
Gender	0.237	1.384	0.808–2.371	0.421	1.281	0.701–2.340
Tumor size	0.000	2.872	1.663-4.959	0.042	1.87	1.023–3.418
Cystic change	0.011	2.028	1.176–3.499	0.045	1.791	1.014–3.165
KPS score	0.009	2.037	1.195–3.472	0.367	1.328	0.718–2.456
Tumor location	0.144	1.505	0.870–2.603	0.121	1.552	0.891–2.704
Extent of resection	0.011	1.985	1.170–3.367	0.600	1.169	0.653–2.092
WHO grade	0.003	2.366	1.344-4.163	0.053	1.858	0.993–3.480
Expression of IncRNA SNHG20	0.000	3.828	2.139–6.850	0.003	2.674	1.393–5.133

Table 5 Univariate and multivariate analyses for recurrence-free survival in glioma patients

Abbreviation: KPS, Karnofsky performance scale.

PCR and Western blotting. The results showed that P21 mRNA and protein levels were significantly increased in glioma U87 cells transfected with *SNHG20* siRNA compared to those transfected with NC siRNA. Furthermore, the results demonstrated that the CCNA1 protein levels were dramatically down-regulated after transfection with *SNHG20* siRNA compared with that after transfection with NC siRNA (Figure 7).

Discussion

IncRNA lacking a meaningful open reading frame with various important functions in disease such as posttranscriptional, transcriptional and epigenetic regulation is defined as transcribed RNA molecules which is >200 nucleotides in length that is lack.^{6,7} In addition, lncRNA dysregulation is concerned with different types of cancer.^{8–11} For instance, some lncRNAs exert a crucial part in metastasis, invasion

as well as cancer cell proliferation, showing that it might be a useful marker of cancer prognosis.^{12–14} Cancer still threats human health.¹⁵ The exact mechanism

of metastasis remains uncertain in cancer patients despite the fact that metastasis is an important indicator of a poor prognosis.^{16,17} At present, new molecular markers should be identified to predict tumor metastasis due to its important role in treating and predicting cancer.¹⁸ lncRNA is one of the molecular markers which can influence the development and occurrence of tumors. It has the potential of collecting biomarkers which are beneficial for monitoring and diagnosing tumors.¹⁹

lncRNA SNHG20 is proved to be an important oncogene in various human cancers according to previous studies.²⁰ According to the recent studies, it was confirmed that *lncRNA SNHG20* expression is aberrantly high expression in ovarian cancer tissues and *SNHG20* silencing could inhibit ovarian cancer progression through Wnt/β-catenin



Figure 3 SNHG20 siRNA is constructed, which can inhibit the *IncRNA* SNHG20 expression levels in glioma cells.

Notes: The knockdown efficiency of *IncRNA SNHG20* is assessed by qRT-PCR. *P < 0.05 means vs the NC siRNA group.

Abbreviation: qRT-PCR, quantitative real-time polymerase chain reaction.



Figure 4 IncRNA SNHG20 modulates the proliferation abilities of glioma U87 cells. Notes: CCK-8 assays show that IncRNA SNHG20 suppression inhibits the proliferation of glioma U87 cells. *P<0.05 means vs the NC siRNA group. Abbreviation: CCK-8, cell counting kit-8.



Figure 5 IncRNA SNHG20 modulates the colony formation abilities of glioma U87 cells. Notes: Colony formation is dramatically reduced following SNHG20 silencing. *P<0.05 vs the NC siRNA group.

signaling pathway.²¹ Moreover, according to Wang et al, *SNHG20* was significantly higher in osteosarcoma samples than that in the cancer-adjacent tissues, and high *lncRNA SNHG20* promoted the osteosarcoma tumorigenesis through mitochondrial apoptosis pathway.²² Li et al²³ reported that knockdown of *SNHG20* inhibited colorectal cancer cell proliferation, cell cycle progression, migration and invasion, revealing that *lncRNA SNHG20* might be a crucial prognostic factor for cancer patients. The function of *lncRNA* *SNHG20* in prognosis and progression of glioma patients still remains unknown.

The present study aimed to investigate the clinical significance and role of *lncRNA SNHG20* in glioma patients for the first time. First, our results showed that the expression of *lncRNA SNHG20* in human gliomas was higher than that in normal brain tissues. Stastistical analysis demonstrated that the expression of *lncRNA SNHG20* was closely related to tumor size, WHO grade, adjuvant therapy and recurrence.



Figure 6 Flow cytometry of the cell cycle distribution of glioma U87 cells transfected with SNHG20 siRNA or NC siRNA. Note: *P<0.05 vs the NC siRNA group.



Figure 7 IncRNA SNHG20 modulates the P21 (A–C) and CCNA1 (A, D, E) expression levels in glioma U87cells. Note: *P < 0.05 means vs the NC siRNA group.

Third, Kaplan-Meier analysis proved that OS and RFS were shorter in glioma patients undergoing high expression of IncRNA SNHG20. In addition, further multivariate and univariate survival analyses confirmed that IncRNA SNHG20 could be used as an independent prognostic biomarker in glioma patients. SNHG20 siRNA was constructed and transfected to glioma U87 cells to detect the function of IncRNA SNHG20 in glioma cells, and the expression levels of IncRNA SNHG20 gene in U87 cells in vitro were down-regulated. The results indicated that after knockdown of IncRNA SNHG20 expression, the proliferation and colony formation abilities of glioma U87 cells were inhibited through increasing the percentage of cells at G0/G1 phase. To understand the potential molecular mechanisms, the potential target proteins involved in cell cycle progression were assessed. Here, CCNA1 expression levels decreased due to the loss of IncRNA SNHG20 in glioma U87 cells. There existed inverse association between IncRNA SNHG20 expression and P21 expression. CCNA1 alters cell cycle progression has been proved to induce carcinogenesis.²⁴ P21 encoded by the CDKN1A gene on 6p21.2 in humans is a cyclin-dependent kinase inhibitor. P21 inhibits the complexes of CDK2 and CDK1 to mediate the p53-dependent cell cycle G1 phase arrest.^{23,25} In accordance with the results, IncRNA SNHG20 contributes to the proliferation of U87 cells via regulating CCNA1 and P21 expression. Nevertheless, the explicit mechanism of IncRNA SNHG20 should be evaluated by

further validation and functional evaluation in other glioma cell lines.

Conclusion

To conclude, *lncRNA SNHG20* is of great important to the progression and prognosis of glioma. Based on the high expression of *lncRNA SNHG20* in human glioma tissues, the high *lncRNA SNHG20* expression predicts dismal prognosis for glioma patients. Besides, *lncRNA SNHG20* expression can be downregulated by *SNHG20* siRNA, and the proliferation abilities of glioma U87 cells would be inhibited. Taken together, *lncRNA SNHG20* can be taken as an independent prognostic biomarker for glioma.

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Disclosure

The authors report no conflicts of interest in this work.

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