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Knockdown of lncRNA PVT1 Inhibits Glioma Progression by Regulating miR-424 Expression

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Plasmacytoma variability translocation 1 (PVT1), an oncogene, has been reported to be highly expressed in many tumors, including human glioma, gastric cancer, and non-small cell lung cancer. Functionally, it could also regulate the development of tumor cells. However, its specific roles and pathogenesis in human gliomas are still not clear. This study investigated the function and mechanism of PVT1 knockdown in the proliferation and malignant transformation of human gliomas. We first examined the expression levels of PVT1 and miR-424 in human glioma tissues and cell lines. We also used gene manipulation techniques to explore the effects of PVT1 knockdown on cell viability, migration, invasion, and miR-424. We found that PVT1 knockdown effectively inhibited cell viability, migration, and invasion of human glioma cells and increased miR-424 expression. Based on the negative correlation between PVT1 and miR-424, we then confirmed the direct interaction between PVT1 and miR-424 using RNA immunoprecipitation (RIP) and luciferase reporter assays. Further, we established a xenograft nude mouse model to determine the role and mechanism of PVT1 on tumor growth in vivo. In addition, PVT1 knockdown was shown to promote miR-424 in vivo. In summary, the present study demonstrated that PVT1 knockdown could negatively regulate miR-424 to inhibit human glioma cell activity, migration, and invasiveness. PVT1 knockdown could negatively regulate miR-424 to inhibit cellular activity, migration, and invasiveness in human gliomas, which explained the oncogenic mechanism of PVT1 in human gliomas. It also suggested that PVT1 might be a novel therapeutic target for human gliomas.

Key words: PVT1; miR-424; Human glioma

INTRODUCTION

The occurrence of cancer is the result of the participation of multiple genes, which are often manifested as activation of oncogenes and/or inactivation of tumor suppressor genes[1–3]. The differential expression of these key genes, regulatory sequences, or effectors induces the cells to go beyond growth control and then undergo cancer[4–6]. As noncoding RNAs longer than 200 nt, lncRNAs have been identified in various mechanisms such as genomic imprinting, chromatin modification, transcription processing, and posttranscriptional processing[7–9]. It is known that more and more lncRNAs have been found to be associated with various cancers, including plasmacytoma variability translocation 1 (PVT1), HOTAIR, and PCGEM1, especially in malignant tumors[10,11]. Gliomas are the most common and invasive primary tumors in the nervous system, accounting for the majority of primary malignant brain tumors[12,13]. Glioma is difficult to cure, especially glioblastoma[14,15]. Therefore, a large number of studies have investigated the molecular abnormalities of gliomas to learn more about the pathogenesis of gliomas. As one of the many molecules that have been shown to be involved in gliomas, the abnormal expression of lncRNAs in the process of carcinogenesis has attracted more and more attention[16,17]. Although many lncRNAs have been shown to play a crucial regulatory role in the extensive cellular processes of human diseases, the function of most lncRNAs remains unknown. Exploring the functional role

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of abnormal expression levels of lncRNAs in regulating the biological processes of gliomas will uncover new strategies for the mechanisms of tumor progression.

MicroRNA (miRNA), a small noncoding RNA of about 22 nucleotides (nt), targets the 3 -untranslated region (3 -UTR) of the corresponding RNA to regulate its expression and acts as a transcriptional regulator[18]. Various data indicate that miRNAs are important regulators of cell signaling pathways and often play the role of oncogenes or antioncogenes in cancer metastasis and progression[19,20]. Recently, studies have suggested that lncRNAs could target miRNA regulation in competing endogenous RNA (ceRNA) regulatory networks, and the interaction of lncRNAs and miRNAs might affect the expression and biological activity of miRNAs in cancer progression[21,22]. Given the importance of miRNAs in cancer, many previous studies have shown that miR-424 can regulate the differentiation, proliferation, and migration of human glioma cells, cervical cancer cells, and other tumor cells[23–26]. These previous reports on the role of miR-424 in tumors led us to assess the effect of PVT1 on miR-424. The aberrant expression of miR-424 implied a potentially critical role in the development and progression of malignancy. However, whether PVT1 could directly interact with miR-424 to influence the pathogenesis of gliomas remains to be explained.

In this research, we performed bioinformatics analysis using the online software starBase v2.0 to obtain potential miRNAs for PVT1 and found that PVT1 contains complementary binding sequences that bind to the miR-424 seed regions. Here we intended to investigate the link between PVT1 and miR-424 and further explore the impact of their interactions on the progression of human glioma cells.

MATERIALS AND METHODS

Tissue Samples and Cell Lines

All clinical specimens used in this study were from the First Affiliated Hospital of Henan University, including 26 human glioma tissues and adjacent noncancerous normal tissues. The study was approved by the ethics committee of the First Affiliated Hospital of Henan University, and written informed consent was obtained from each patient.

The human glial cell HEB and human glioma tumor cell lines (H4, LN-229, U251, and U87) were purchased from the American Type Culture Collection (ATCC, Rockville, MD, USA). HEB was cultured in RPMI-1640 (Thermo Fisher Scientific, Waltham, MA, USA) containing 10% fetal bovine serum (FBS), 2,000 U/ml penicillin G (1,000,000 U = 1 g), and 500 µg/ml streptomycin (Invitrogen, Carlsbad, CA, USA), and H4, LN-229, U251, and U87 cell lines were cultured in DMEM (Thermo Fisher Scientific) containing 10% FBS, 2,000 U/ml penicillin G, and 500 µg/ml streptomycin (Invitrogen). All experimental cells were cultured in a Therma incubator (Thermo Fisher Scientific) at 37°C and 5% CO_2.

Cell Transfection

Three siRNAs for PVT1 (si-PVT1#1, siPVT1#2, and si-PVT1#3), si-control, pcDNA-PVT1, pcDNA empty vector (pcDNA), miR-424, miR-control, PVT1 short hairpin RNA (sh-PVT1), and an empty lentivirus vector (sh-control) were from chemical synthesis of Sangon Biotech (Sangon Biotech, Shanghai, P.R. China). Using a six-well plate as an example, cells were seeded in six-well plates (Corning Incorporated, Corning, NY, USA) at 1×10^5/well and transfected with Lipofectamine 2000 reagent (Invitrogen). One hour before transfection, the medium was replaced with 1 ml of fresh serum-free medium and stored at 37°C in a 5% CO_2 incubator. The mixture of Lipofectamine 2000 and si-PVT1 was allowed to stand at room temperature for 20 min (volume/mass ratio of Lipofectamine 2000 and si-PVT1 was 1:1), and then added dropwise to the culture wells for 6 h. After transfection, cells were incubated in medium containing 10% fetal bovine serum. Forty-eight hours after transfection, the cells of interest were collected for further analysis. All experiments were repeated.

Quantitative Real-Time Polymerase Chain Reaction (qPCR)

qPCR was carried out in an Applied Biosystems 7500 System with Power SYBR Green PCR Master Mix (Applied Biosystems, Carlsbad, California, USA). -Actin and human U6 small nuclear RNA (snRNA) served as internal controls for PVT1 and miR-424, respectively. Our qPCR results were evaluated by the $2^{-\ CT}$ method. The specific primer sequences were as follows: PVT1, 5 -GCCCTCCAGCCTGATCTTTT-3 (forward) and 5 -TTCCACCAGCGTTATTCCCC-3 (reverse); -actin, 5 -CACTGGCATCGTGATGGA-3 (forward) and 5 -GGCCATCTCTTGCTCGAA-3 (reverse); miR-424, 5 -ATGGTTCAAAACGTGAGGCG T-3 (forward) and 5 -ACCTTCTACCTTCCCCACGA-3 (reverse); U6, 5 -CGCTTCGGCAGCACATATAC-3 (forward) and 5 -AAATATGGAACGCTTCACGA-3 (reverse).

Cell Viability Assay

Cell viability was measured using the MTT assay. Briefly, approximately 1×10^4 cells per well in a 96-well plate were transfected with si-PVT1, si-control, miR-424, miR-control, miR-424 + pcDNA, or miR-424 + pcDNA–PVT1. Forty-eight hours after transfection, 20 µl of MTT (5 mg/ml; Sigma-Aldrich, St. Louis, MO, USA) was

added to each well, and the cells were incubated at 37°C for 4 h. Subsequently, the supernatant was discarded, and 100 µl of dimethyl sulfoxide (DMSO; Sigma-Aldrich) was added to dissolve formazan crystals. Absorbance was measured at 490 nm with a SpectraMax Paradigm Multi-Mode Reader (Molecular Devices, Austria). All experiments were repeated.

Cell Migration Assay

In vitro cell migration assays were carried out to assess cell migration. The transfected 1×10^6 cells were resuspended in serum-free medium and placed in an insert containing 8-µm pores (Corning Incorporated). These inserts were placed in wells containing 10% FBS serum-containing medium. After 24 h of incubation, the cells on the upper surface of the membrane were removed. The cells that had migrated to the lower surface of the membrane were fixed with 70% methanol, which were then stained with 0.5% liquid crystal violet (Sigma-Aldrich) for 15 min and washed with 1× PBS to remove excess stains. Migratory cells on the bottom side of the membrane were counted in 10 different regions using an Olympus fluorescence microscope to obtain the average number of migrated cells per cell line.

Cell Invasion Assay

In vitro invasion assays were performed in 24-well cell culture plates using an 8.0-µm pore size membrane from Matrigel Invasions Chamber (BD Biosciences, San Jose, CA, USA). Briefly, transfected cells (1×10^6/ml) were seeded in the upper chamber of each well. In the lower chamber, 10% FBS was added as an inducer. The plate was then incubated for 24 h. After incubation, non-invasive cells on the upper surface were removed, and cells invaded in the lower chamber were fixed with 70% ethanol. The cells were then stained with 0.5% liquid crystal violet (Sigma-Aldrich) for 15 min and washed with 1× PBS to remove the excess stain. The invasive cells on the bottom side of the membrane were counted in 10 different regions using an Olympus fluorescence microscope to obtain the average number of migrating cells per cell line.

Luciferase Reporter Assay

Wild-type or mutated PVT1 sequences containing predicted miR-424 binding sites were amplified by PCR and subcloned into pGL3-basic vector (Promega, Madison, WI, USA) to generate reporter vector pGL3–PVT1-WT (PVT1-WT) and pGL3–PVT1MUT (PVT1-MUT). For the luciferase reporter assay, cells were seeded in 24-well plates, and 0.2 µg of the constructed luciferase plasmid was cotransfected with 50 ng of luciferase. pRL-TK (Promega), and miR-424 or miR control vectors were compared using Lipofectamine 2000 (Invitrogen). After 48 h of transfection, these cells were collected and assayed for luciferase activity using a dual-luciferase reporter gene detection system according to the manufacturer's instructions (Promega). Relative luciferase activity = firefly luciferase activity/*Renilla* luciferase activity.

RNA Immunoprecipitation

RNA immunoprecipitation (RIP) experiments were performed using the RNA-binding protein immunoprecipitation kit (Millipore, Billerica, MA, USA) according to the manufacturer's protocol. Cells were lysed using a complete RIP lysis buffer, and 100 µl of whole cell extracts were left ready. This reserve extract was then incubated together with human anti-argonaute 2 (Ago2) antibody-coated magnetic beads moistened with RIPA buffer for 8 h at 4°C. Normal mouse IgG was used as a negative control. Samples were washed with wash buffer and incubated with protease Kat at 55°C for 30 min to isolate RNA–protein complexes from the beads. The immunoprecipitated RNA was then extracted and used for qPCR analysis.

In Vivo Experiments

Female athymic BALB/C nude mice (5 weeks old) were purchased from the Guangdong Medical Experimental Animal Center (Guangzhou, P.R. China) and maintained under pathogen-free conditions. U251 cells stably transfected with sh-control or sh-PVT1 (1×10^6) were injected subcutaneously into the ventral side of nude mice ($n = 6$ mice/group). The tumor volume was measured every 7 days, and the volume was calculated using the formula: length × width × width/2. After 28 days, the mice were sacrificed under anesthesia, and xenograft tumor tissues were removed for testing. The animal experimental protocol was approved by the Animal Protection and Research Committee of Kaifeng Central Hospital.

Statistical Analysis

The SPSS version 19.0 software (SPSS, Chicago, IL, USA) was used to analyze the related data with *t*-tests or analysis of variance. A value of $p < 0.05$ was considered statistically significant.

RESULTS

Knockdown of PVT1 Notably Upregulated the Expression of miR-424 in Glioma Cells

To investigate the relationship between PVT1 and miR-424 in human gliomas, we first studied their expression in primary human glioma tissues by qPCR. Compared with the adjacent normal tissues, the expression of PVT1 in human glioma tissues was visibly increased (Fig. 1A), and miR-424 was exceptionally decreased in human

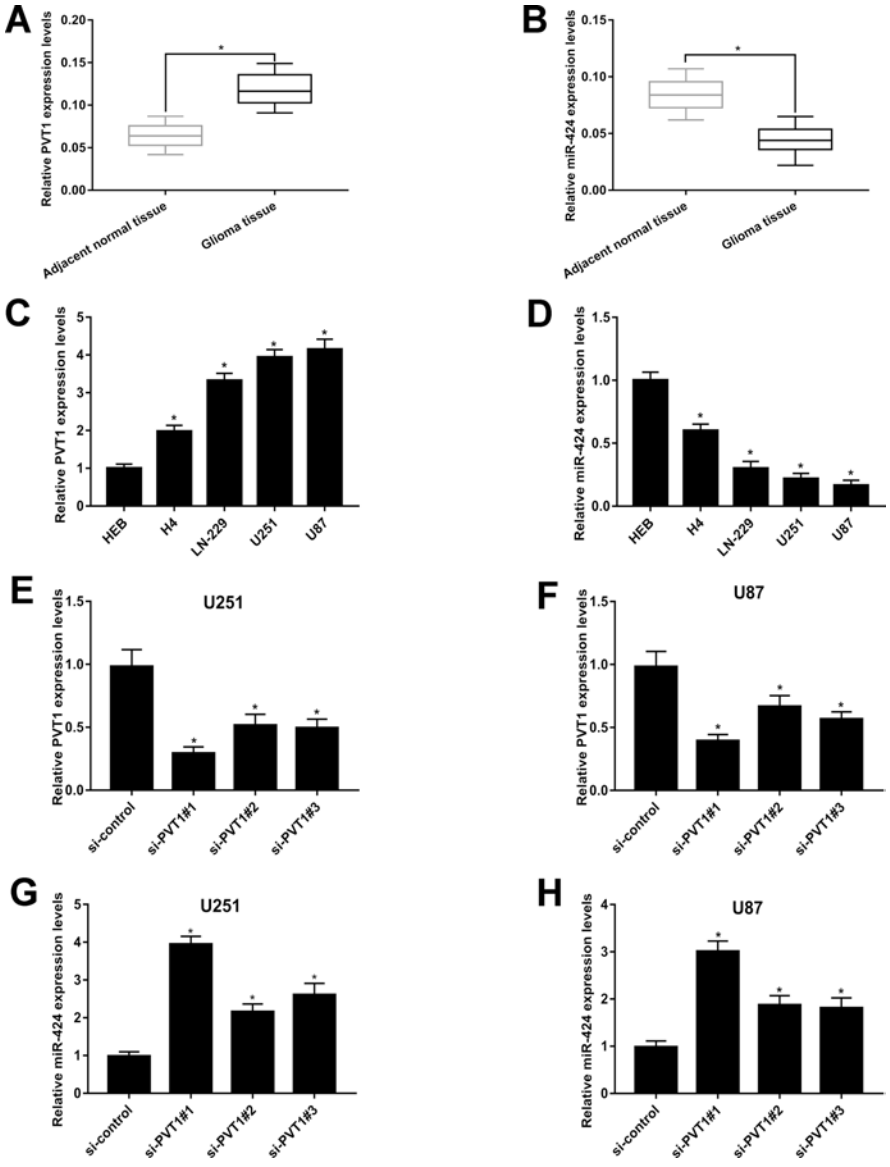

Figure 1. The knockdown of plasmacytoma variability translocation 1 (PVT1) significantly upregulated the expression of miR-424 in glioma cells. The expression of PVT1 (A) and miR-424 (B) in primary human glioma tissues was analyzed by quantitative real-time polymerase chain reaction (qPCR). qPCR analysis of PVT1 (C) and miR-424 (D) expression in four glioma tumor cell lines (H4, LN-229, U251, and U87) and normal human brain glial cell line as a control. PVT1 expression was detected by qPCR in U251 (E) and U87 (F) cells transfected with si-control, si-PVT1#1, si-PVT1#2, and si-PVT1#3. miR-424 expression was measured by qPCR in U251 (G) and U87 (H) cells transfected with si-control, si-PVT1#1, si-PVT1#2, and si-PVT1#3. $*p < 0.05$. Data shown are mean ± SD from three independent experiments.

glioma tissues (Fig. 1B). We then explored their expression in four human glioma cell lines (H4, LN-229, U251, and U87) and human glial cell line HEB by qPCR. In human glioma cells, particularly in U251 and U87 cells, the expression of PVT1 was evidently upregulated compared to HEB (Fig. 1C), and the expression of miR-424 was abnormally decreased (Fig. 1D). Therefore, we chose U251 and U87 cell lines for follow-up experiments. Further, we transfected U251 and U87 cells with si-control, si-PVT1#1, si-PVT1#2, and si-PVT1#3, respectively, and confirmed knockdown efficiency by qPCR. As shown in Figure 1E and F, si-PVT1#1, si-PVT1#2, or si-PVT1#3 could effectively reduce PVT1 expression in U251 and U87 cells, while si-PVT1#1 displayed higher knockdown efficiency. Moreover, the qPCR results exhibited that si-PVT1#1, si-PVT1#2 or si-PVT1#3, especially si-PVT1#1, also effectively increased the miR-424 expression in U251 (Fig. 1G) and U87 (Fig. 1H) compared to

the si-control Therefore, si-PVT1#1 was selected for the subsequent experiments.

Knockdown of PVT1 Could Partially Depress the Cell Viability, Migration, and Invasiveness in Glioma Cells

To further explore the functional role of PVT1 in human glioma cells, we used gene knockdown techniques to reduce PVT1 expression. We initially analyzed the effect of PVT1 knockdown on cell viability by MTT assay. These results suggested that knockdown of PVT1 considerably inhibited the viability of U251 and U87 cells (Fig. 2A). Next, we also found that U251 and U87 cells transfected with si-PVT1 expressed obvious inhibition of cell migration compared with the si-control group (Fig. 2B). Furthermore, our data showed that PVT1 knockdown attenuated invasiveness in U251 and U87 cells compared to the si-control group (Fig. 2C). The above results indicated that knockdown of PVT1 partially prohibited the progression of human glioma cells.

There Is a Direct Crosstalk or Interaction Between PVT1 and miR-424

We first examined the correlation between PVT1 and miR-424 and found a negative correlation between PVT1

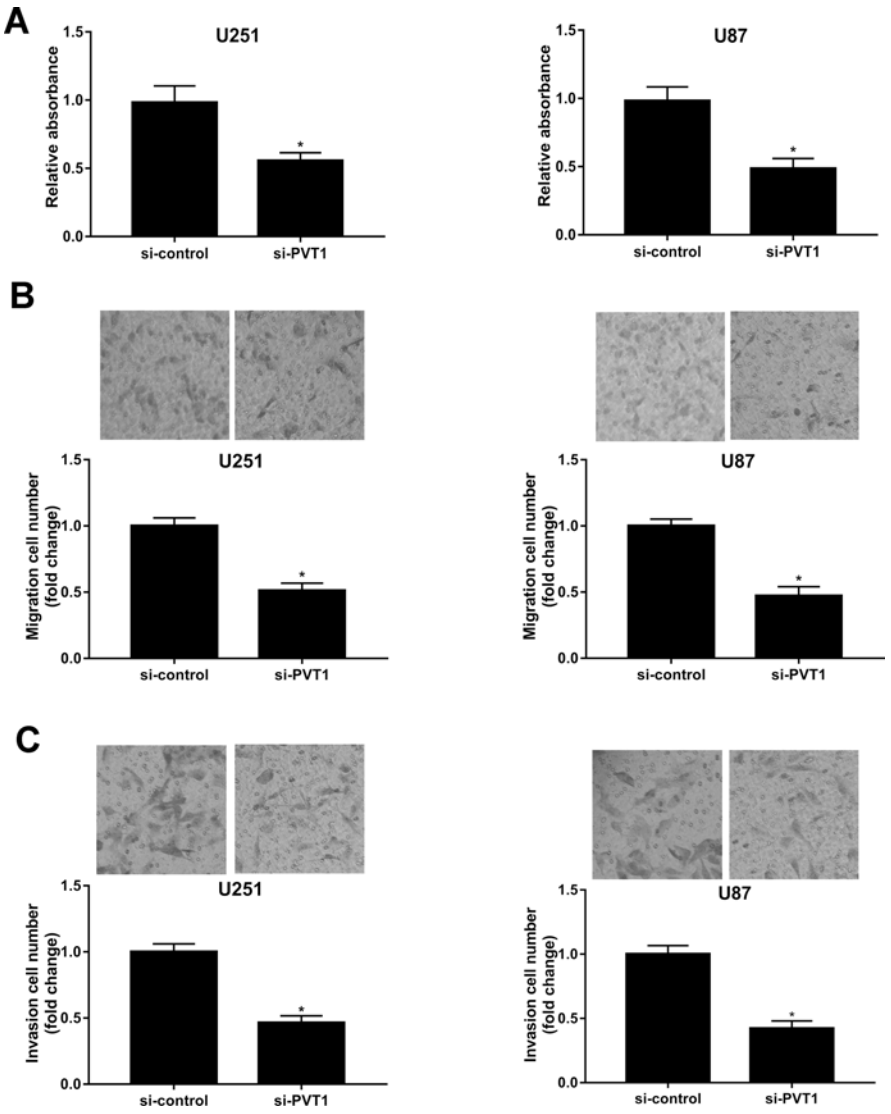

Figure 2. Knockdown of PVT1 could partially inhibit the cell viability, migration, and invasiveness of glioma cells. U251 and U87 cells were transfected with si-PVT1 or si-control and incubated for 48 h. (A) Cell viability of transfected U251 and U87 cells was measured by MTT. (B) The migration of transfected U251 and U87 cells was examined by cell migration assay. (C) Cell invasion assays were used to detect cell invasiveness of transfected U251 and U87 cells. *$p < 0.05$. Data shown are mean ± SD from three independent experiments.

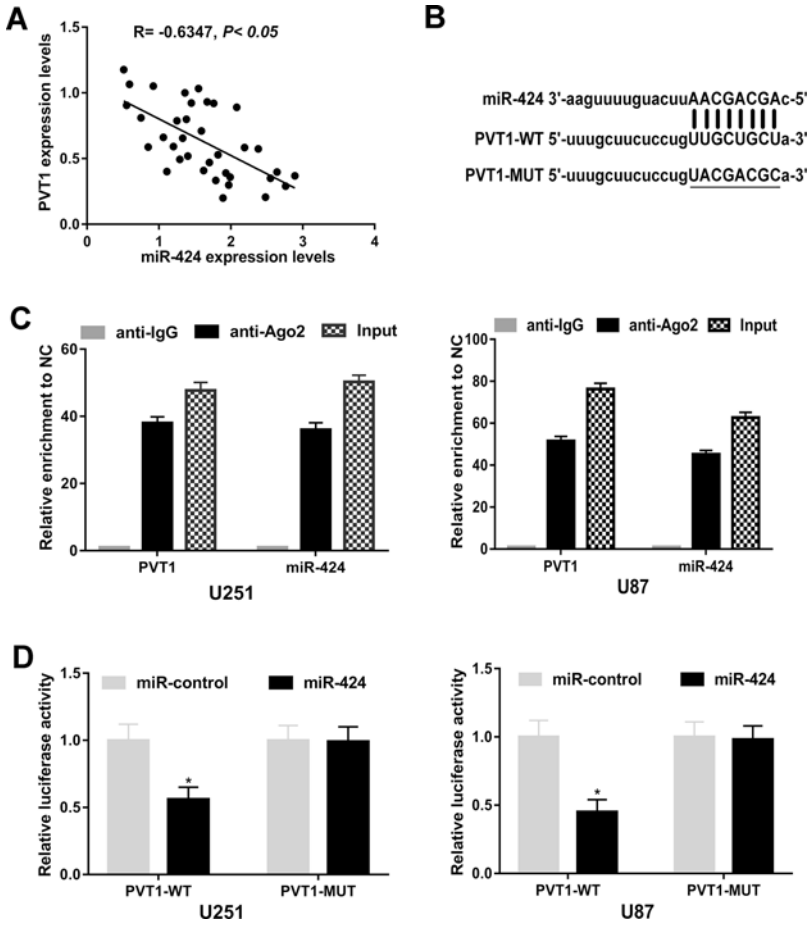

Figure 3. There is a direct, negative correlation between mRNA levels of PVT1 and miR-424. (A) The expression of PVT1 and miR-424 was negatively correlated in glioma cells. (B) The prediction of wild-type or mutated miR-424 binding sites in PVT1 was based on the Starbase v2.0 software. (C) RNA immunoprecipitation (RIP) assays were performed using IgG, Ago2 antibodies, or 10% input from U251 and U87 cell extracts, and the expression of PVT1 and miR-424 RNAs in the immunoprecipitates was detected by qPCR. NC refers to a negative transfection. (D) Luciferase activity was detected in U251 and U87 cells after cotransfection of luciferase reporter plasmid (PVT1-WT or PVT1 MUT) and miR-424 or miR-control. $*p < 0.05$. Data shown are mean ± SD from three independent experiments.

and miR-424 expression in glioma cells (Fig. 3A). Based on the negative correlation between PVT1 and miR-424 expression, we speculated potential miRNAs of PVT1 and found that PVT1 contains complementary sequences to the miR-424 seed region (Fig. 3B). To support this hypothesis, we performed an RIP assay in U251 and U87 cells using the antibody Ago2 to verify whether PVT1 and miR-424 had a common acting basis. The RIP assays showed that both PVTl and miR-424 were significantly enriched in Ago2 conjugates compared to the control group (Fig. 3C). These results indicated that PVT1 and miR-424 were in a common functional complex. In addition, luciferase reporter assays confirmed that the ectopic expression of miR-424 partially attenuated the luciferase activity of PVT1-WT but did not affect the luciferase activity of PVT1-MUT (Fig. 3D). Collectively, these data revealed that there was a direct crosstalk or interaction between PVT1 and miR-424.

The PVT1-miR-424 Axis Was Involved in the Progression of Human Glioma Cells

Given that PVT1 could influence expression of miR-424, we further explored the effect of PVT1–miR-424 axis on the activity, migration, and invasiveness of human glioma cells, and U251 and U87 cells were transfected with miR-control, miR-424, pcDNA, pcDNA–PVT1, miR-424 + pcDNA or miR-424 + pcDNA–PVT1. As shown in Figure 4A, qPCR confirmed that pcDNA–PVT1 strikingly increased PVT1 expression in U251 and U87 cells compared to the pcDNA group. Furthermore, MTT assays disclosed that overexpression of miR-424 visibly reduced cell viability of U251 and U87 cells

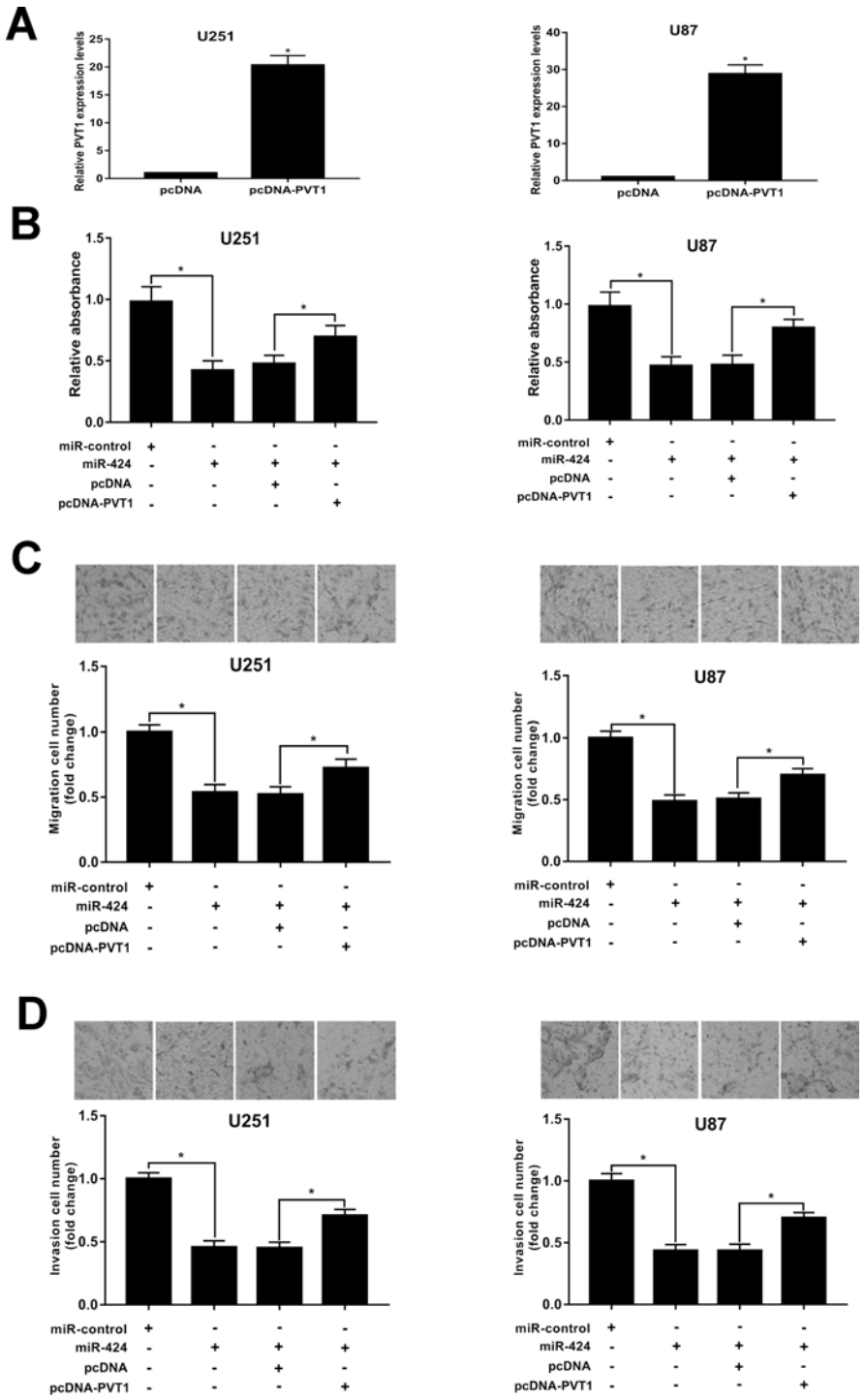

Figure 4. Overexpression of PVT1 partially reverses the viability, migration, and invasion of miR-424 on glioma cells. (A) The relative expression levels of PVT1 in U251 and U87 cells transfected with pcDNA or pcDNA–PVT1 were determined by qPCR. (B) The relative absorbance of cell viability was measured after cotransfection of miR-control, miR-424, miR-424 + pcDNA, and miR-424 + pcDNA–PVT1 in U251 and U87 cells. (C) The relative migration of cells was detected in U251 and U87 cells cotransfected with miR-control, miR-424, miR-424 + pcDNA, and miR-424 + pcDNA–PVT1. (D) The relative invasiveness of cells was examined in U251 and U87 cells cotransfected with miR-control, miR-424, miR-424 + pcDNA, and miR-424 + pcDNA–PVT1. *$p < 0.05$. Data shown are mean ± SD from three independent experiments.

compared with miR-control group, whereas PVT1 overexpression partly reversed the effect of miR-424 (Fig. 4B). We then further found that miR-424-transfected U251 and U87 cells dramatically reduced cell migration compared with the miR-control, whereas introduction of exogenous PVT1 partially compensated miR-424-induced cell migration inhibition (Fig. 4C). In addition, we used cell invasion assays to examine the effect of the PVT1–miR-424 axis on cell invasiveness (Fig. 4D). The results revealed that overexpression of miR-424 inhibited the invasiveness of U251 and U87 cells. Conversely, PVT1 overexpression promoted their invasiveness to some extent. Taken together, these results demonstrated that the PVT1–miR-424 axis was involved in the progression of human glioma cells, and PVT1 overexpression could antagonize the regulation of miR-424 biological function.

The PVT1–miR-424 Axis Regulated Tumor Growth In Vivo

The apparent influence of PVT1 on the progression of glioma cells via the PVT1–miR-424 axis in vitro prompted us to further study the effects of PVT1 on tumor development and xenograft formation in nude mice. U251 cell lines stably transfected with sh-control or sh-PVT1 were injected into nude mice. Twenty-eight days after the injection, the mice were sacrificed. Tumor volumes were measured every 7 days, and tumors were exercised and weighed 28 days after injection. The results indicated that knockdown of PVT1 obviously depressed tumor growth in vivo compared to the sh-control group, including the mean tumor volume (Fig. 5A) and weight (Fig. 5B). Notably, we used qPCR to detect miR-424 expression in xenograft tissues. PVT1 knockdown also significantly upregulated miR-424 expression in vivo and was consistent with previous results in vitro (Fig. 5C). In short, these data supported that PVT1 knockdown could affect the growth of human glioma cells through the PVT1–miR-424 axis in vivo.

DISCUSSION

In this study, we first investigated that PVT1 knockdown significantly upregulated the expression of miR-424 in human glioma cells. We also found that PVT1 knockdown could affect cell viability, migration, and invasion of human glioma cells. Next, our research gave fresh proof that PVT1 could directly interact with miR-424 and inhibit its expression. Moreover, we ascertained

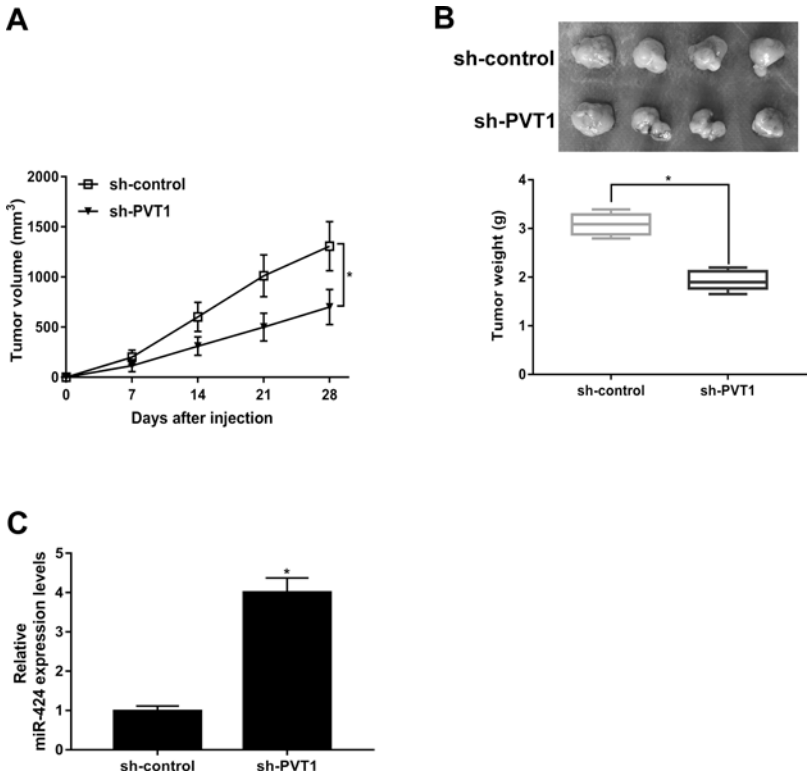

Figure 5. Knockdown of PVT1 suppressed tumor growth in vivo by upregulating miR-424. (A) Downregulation of PVT1 distinctly reduced tumor volume compared with sh-control. (B) At 28 days after injection, downregulation of PVT1 markedly reduced tumor weight compared to the sh-control group. (C) The relative expression levels of miR-424 were analyzed by qPCR in solid tumors. *$p < 0.05$. Data shown are mean ± SD from three independent experiments.

that PVT1 could regulate the cell viability, migration, and invasion of human glioma cells through the PVT1–miR-424 axis. In addition, similar results were observed in nude mice. These studies demonstrated that PVT1 and miR-424 had antagonistic effects in the ceRNA regulatory network of human glioma cells.

Increasing evidence has verified that lncRNAs and miRNAs played crucial roles in the development and progression of human cancers[27–29]. In particular, upregulation of PVT1 has been found in many types of tumors and acts as an oncogenic lncRNA. For instance, Ding et al. revealed that lncRNA PVT1 was associated with tumor progression and predicted recurrence in hepatocellular carcinoma patients[30], and Verduci et al. demonstrated that PVT1 could mediate the carcinogenesis of head and neck squamous cells[31]. In non-small cell lung cancer, knockdown of lncRNA PVT1 could inhibit tumorigenesis by regulating miR-497[32]. In fact, lncRNAs could affect the proliferation process of tumor cells, which often involve abnormal changes in their sequence, spatial structure, or expression level[33,34]. To the best of our knowledge, we are the first to discover that lncRNA PVT1 could regulate the progression of tumor cells in human glioma cell lines by inhibiting miR-424. Similarly, there was a significant negative correlation between lncRNA PVT1 and miR-424 in nude mouse tissue samples. In addition, PVT1 was involved in the progress of PVT1-miR-424 axis-regulated glioma cells, which is consistent with previous reports[35,36].

PVT1 has been confirmed as an oncogene in human glioma cells, but the underlying mechanism of PVT1 in human glioma remains to be elucidated. In recent years, lncRNA–miRNA–mRNA has been widely recognized as a novel mode of regulation, in which lncRNAs function as miRNA target genes or target conjugates to bind or compete with target miRNAs, thereby affecting miRNA expression[37]. For instance, PVT1 could competitively bind to miR-195 and inhibit its expression[38], and PVT1 acted as a ceRNA that negatively regulated miR-424 in cervical cancer cell[26]. Moreover, PVT1 competitively linked miR-186 and inhibit its function in gastric cancer[39]. To further investigate the relationship and role of PVT1 and miR-424 in human glioma tumors, we used genetic manipulation techniques to knock down PVT1 to explore the relationship between PVT1 and miR-424. Our study confirmed that miR-424 was downregulated in human glioma tumors and that increased expression of miR-424 could promote cell viability, migration, and invasion when PVT1 was knocked out. In addition, PVT1 knockdown was certified to promote miR-424 in vivo. These results indicated that PVT1 exerted its oncogenic role in human gliomas by regulating miR-424 expression.

In general, our results indicated that PVT1 expression level was upregulated and negatively correlated with miR-424 expression level in human glioma cells. Furthermore, PVT1 knockdown could negatively regulate miR-424 to inhibit human glioma cell activity, migration, and invasiveness. This finding explained that PVT1, as an lncRNA, had a molecular mechanism of carcinogenesis in human glioma. Therefore, PVT1 might be a new therapeutic target for human glioma. A better understanding of this mechanism could guide to find novel therapeutic strategies that inhibit malignant progression of human glioma cells.

PVT1 knockdown could negatively regulate miR-424 to inhibit human glioma cell activity, migration, and invasiveness. This finding explained that PVT1, as an lncRNA, had a molecular mechanism of carcinogenesis in human glioma. Therefore, PVT1 might be a new therapeutic target for human glioma. A better understanding of this mechanism could help to find novel therapeutic strategies that inhibit malignant progression of human glioma cells.

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