JBUON 2021; 26(4): 1549-1555
ISSN: 1107-0625, online ISSN: 2241-6293 • www.jbuon.com
Email: editorial_office@jbuon.com

ORIGINAL ARTICLE

Effects of triptolide on radiosensitivity of human glioma cells and its mechanism

Mengting Shu[1], Chenguang Li[2], Chen Zhong[2], Tongyi Huang[1], Xinzhuang Wang[3], Yang Tan[1], Wenyang Zhao[3], Xiaoyan Xie[1]

[1]Department of Medical Ultrasonics, Division of Interventional Ultrasound, Institute of Diagnostic and Interventional Ultrasound, the First Affiliated Hospital of Sun Yat-sen University, Guangzhou, China. [2]Department of Neurosurgery, the Second Affiliated Hospital of Zhejiang University, Hangzhou, China. [3]Department of Neurosurgery, the First Affiliated Hospital of Harbin Medical University, Nangang District, Harbin, China.

Summary

Purpose: *To study the effect of triptolide (TP) on radiosensitivity of human glioma U251 cells and its mechanism, so as to provide new ideas and methods for the radiotherapy of glioma.*

Methods: *U251 cells were treated with 10, 50, 100 nmol/L TP at different concentrations and irradiated with 0, 2, 4, 6, 8 Gy X-ray. The radiosensitivity of cells in each group were detected by MTT. U251 cells were then divided into control group, 10 nmol/L TP group, 4 Gy radiation group, 10 nmol/L TP + 4 Gy radiation group. The formation ability of U251 cells in each group was detected by colony formation assay. Flow cytometry was used to detect cell cycle and apoptosis in each group. Western blot was used to detect the changes of PI3K/Akt signal pathway in each group.*

Results: *When 10, 50, 100 nmol/L TP were combined with 2, 4, 6, 8 Gy X-ray, the proliferation inhibition rate of U251 cells in each group increased significantly ($p<0.05$); compared with 10 nmol/L TP alone group and 4 Gy radiation alone group, the colony formation ability rate of U251 cells in 10 nmol/L TP + 4Gy radiation combined group decreased significantly ($p<0.05$), the cell cycle was blocked in G1 phase, and the apoptosis rate was significantly reduced ($p<0.05$). The level of p-pi3k and p-Akt decreased significantly ($p<0.05$).*

Conclusions: *Triptolide could significantly increase the radiosensitivity of human glioma U251 cells and play a role by inhibiting the PI3K/Akt signal pathway.*

Key words: *triptolide, glioma, radiosensitivity, apoptosis, cell cycle, PI3K/Akt signaling pathway*

Introduction

Human glioma is the most common intracranial primary tumor, and its incidence accounts for more than 70% of intracranial malignancies [1]. At present, great progress has been made in the clinical understanding and treatment of malignant gliomas, but because of its rapid malignant progression, high invasiveness and infiltration, and since most gliomas grow in important functional areas of the brain, surgery is often unable to completely remove the tumor, and the prognosis of many patients is poor with a low cure rate [2,3]. Most patients with glioma require adjuvant radiation therapy postoperatively to enhance the local control rate and increase patient survival [4]. Radiotherapy is currently an important non-surgical treatment for glioma, while many glioma patients are less sensitive to radiation, due to the biological characteristics of glioma cells and the influence of the local environment of the tumor, while the inability of radiotherapy to effectively kill tumors leads to poor efficacy [5]. Therefore, searching for safe and effective methods for radiotherapy sen-

Corresponding author: Xiaoyan Xie, MD. Department of Medical Ultrasonics, Division of Interventional Ultrasound, Institute of Diagnostic and Interventional Ultrasound, the First Affiliated Hospital of Sun Yat-sen University, 58 Zhong Shan Rd 2, Guangzhou, Guangdong 510080, China.
Tel: +86-020-87765183, Email: xiexyan@mail.sysu.edu.cn
Received: 20/06/2021; Accepted: 08/07/2021

sitization becomes an urgent need for malignant glioma treatment in the clinic.

Triptolide (TP), a natural product extracted from the traditional Chinese herb Tripterygium Wilfordii Hook F, is also the main active ingredient in preparations such as Tripterygium Wilfordii Hook F [6]. Pharmacological studies have found that triptolide with broad-spectrum and potent antitumor activity can significantly inhibit the pathogenesis and progression of lung, breast, and prostate cancer and other malignant tumors [7,8]. The latest study found that triptolide combined with radiation significantly inhibited nasopharyngeal carcinoma (NPC) CNE-2 cell proliferation and NPC tumorigenesis in nude mice in both *in vivo* and *in vitro* assays, and speculated that triptolide may play a sensitizing role in NPC radiotherapy [9,10].

However, there has been no study on whether triptolide plays a role in radiotherapy of human gliomas. Therefore, in this paper, we focused on human glioma U251 cells to observe the effect of triptolide on their radiosensitivity and provide a new idea and method for radiation therapy of glioma.

Methods

Main materials

Human glioma U251 cells were purchased from the cell resource bank of Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences (Shanghai, China); Triptolide (HPLC > 98%) was obtained from Shanghai Yuanye biological company (Lot No. yy90324) (Shanghai, China); Dulbecco's Modified Eagle Medium (DMEM) cell culture medium, 0.25% pancreatin EDTA (ethylenediaminetetraacetic acid), fetal bovine serum (FBS) were purchased from GIBCO (Rockville, MD, USA); MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide), phosphate buffered saline (PBS) reagents were purchased from Beyotime biological company (Beijing, China); Cell cycle detection kit, annexin-V/PI apoptosis kit were purchased from Beijing Dingguo bio company (Beijing, China); Polyclonal rabbit anti human phosphorylated phosphatidylinositol-3-hydroxykinase (p-PI3K), phosphorylated serine / threonine kinase (p-Akt), and internal reference protein (GAPDH) antibodies were purchased from ABCAM (Cambridge, MA, USA); Horseradish peroxidase (HRP)-labeled Goat anti rabbit IgG secondary antibody and ECL enhanced chemiluminescence reagent were purchased from Beijing Zhonghuajinqiao company (Beijing, China).

Cell culture and radiation methods

Frozen human glioma U251 cells were removed and quickly lysed in a 37°C water bath, collected by centrifugation at 800 R/min for 5 min, and resuspended in DMEM cell culture medium containing 10% FBS before being placed in a 37°C, 5% CO_2 incubator. The growth status of the cells was observed under an inverted microscope every 12 h, and after the cells reached a confluence of 80%, they were digested with pancreatin EDTA containing 0.25% for centrifugal passage. U251 cells in logarithmic growth phase and in good condition were taken for subsequent experiments, and glioma cells receiving radiotherapy were stimulated with different doses of X-ray irradiated cells.

MTT assay

U251 cells in logarithmic growth phase were collected and diluted into a cell suspension of 10^4 cells/ml with DMEM cell culture medium and seeded into 96-well plates at 200 µl per well and placed into a cell culture incubator for 4-h adherent growth. After removing the supernatant, the cell culture medium containing 0, 10, 50, and 100 nmol/L TP was changed and incubated for 12 h to adhere and were then exposed to X-ray irradiation (intensity, 6 MeV) at the following doses: 0 Gy, 2 Gy, 4 Gy, 6 Gy, and 8 Gy at room temperature. After 24 h of irradiation, the supernatant was aspirated from each well and 150 µl of MTT solution (0.5 mg/ml) and incubation continued for 4 h. Then, 50 µl of dimethyl sulfoxide (DMSO), was added with shaking for 15 min, and then the optical density (OD) value of each group of cells at A490 nm was measured on a microplate reader. Five replicate wells were set for each group of cells, and the inhibition rate of cell proliferation was calculated for each group = [(1-value of a of treatment well/value of a of no treatment well) × 100%].

Cell monoclonal experiments

Based on the MTT assay results described above, glioma U251 cells were divided into four groups: control group (without any treatment), 10 nmol/L TP treatment (cultured with cell culture medium containing 10 nmol/L TP alone), 4 Gy radiation (cultured with 4 Gy X-ray radiation alone), 10 nmol/L TP + 4 Gy radiation combined group (cultured after treatment with 10 nmol/L TP containing cell culture medium followed by 4 Gy of X-ray radiation). The collected logarithmic growth phase U251 cells were diluted with culture medium into 200 per well and seeded in collagen coated cell culture plates, and the supernatant was removed after 4 h of static culture. The incubation continued for 96 h following the above grouping treatment, after which the supernatant medium was discarded. One ml of paraformaldehyde was added to each well for 20 min fixation, then stained with 5% crystal violet for 15 min, washed with PBS once and photographed and observed, and the cell monoclonal formation rate (the ability of a single tumor cell to form a clonal population in culture) of each group was counted = (the number of cell monoclonal formation / the number of seeded cells in the plate) × 100%.

Flow cytometry assay

U251 cells in logarithmic growth phase were trypsinized and collected by centrifugation at 800 rpm for 5 min after the above grouping. A portion of U251 cells were washed once with 1 ml of PBS buffer added to each well and then fixed with 2 ml of prechilled 70% ethanol at 4°C for 30 min, the supernatant was removed by

centrifugation again, and after one PBS wash 50 μg/ml propidium iodide (PI) solution was added and incubated for 30 min in the dark at room temperature, followed by addition of 200 μL of PBS and then mixed. U251 cell cycle changes in each group were detected by BD Calibur flow cytometer (BD Biosciences, Franklin Lakes, NJ, USA). The cells treated with the above grouping were harvested, washed with PBS once and resuspended by adding 10 μl of FITC labeled annexin-V and PI, mixed and incubated in the dark at room temperature for 30 min, then washed with 1 ml of PBS once, and similarly subjected to flow cytometry to determine the apoptosis rate changes in each group.

Western blot assay

The treated U251 cells from each group described above were digested with pancreatin, and the cells were collected by centrifugation in PBS. To each set of cells, 500 μL of strong ice-mixed RAPI cell lysate was utilized to lyse for 5 min to extract total protein from cells. The total protein concentration in the cells of each group was measured using a bicinchoninic acid (BCA) kit. Thirty μg total protein of per group were loaded for SDS-PAGE gel electrophoresis for 2 h, then blocked with 5% nonfat dry milk for 10 min, washed with TBST solution and transferred to polyvinylidene fluoride (PVDF) membranes (Millipore, Billerica, MA, USA), which were incubated overnight at 4°C with polyclonal rabbit anti human p-pi3k, p-Akt, GAPDH primary antibodies (1:1000). The next day, after three more washes with TBST solution, monoclonal peroxidase labeled goat anti rabbit IgG secondary antibody (1:5000) were incubated for 2 h at room temperature, followed by three more washes with TBST solution. Finally, the PVDF membranes were treated with ECL chemiluminescence reagent for 5 min, exposed, photographed, and analyzed by quantity one software for relative expression of each protein of interest, using GAPDH protein as an internal reference.

Statistics

All data were statistically analyzed using SPSS 22.0 (IBM, Armonk, NY, USA), and data that conformed to a normal distribution were expressed as mean values ± standard deviation. Comparisons among multiple groups were performed by one-way analysis of variance (ANOVA), and pairwise comparisons between groups were tested for significance using the SNK-q method, which was considered significant when $p<0.05$.

Results

Triptolide can significantly increase the proliferation inhibition rate of glioma U251 cells under X-ray

MTT assay showed that different concentrations of triptolide (TP) could significantly increase the proliferation inhibition rate of glioma U251 cells under X-ray irradiation (Table 1). Statistical results showed that when the dose of 0 nmol/L TP reached 4 Gy, the proliferation inhibition rate of U251 cells increased significantly ($p<0.05$). When the concentration of TP was higher than 10 nmol/L, the proliferation inhibition rate of U251 cells increased significantly ($p<0.05$). When TP of 10 nmol/L, 50 nmol/L and 100 nmol/L were combined with X-ray of 2 Gy, 4 Gy, 6 Gy and 8 Gy, the proliferation inhibition rates of U251 cells in each group were significantly increased ($p<0.05$), indicating that TP of different concentrations could significantly increase the radiosensitivity of U251 cells.

Triptolide can significantly inhibit the monoclonal ability of glioma U251 cells under X-ray irradiation

The results showed that 10 nmol/L TP combined with 4 Gy X-ray could significantly inhibit the monoclonal formation ability of glioma U251 cells (Figure 1). The results showed that the mon-

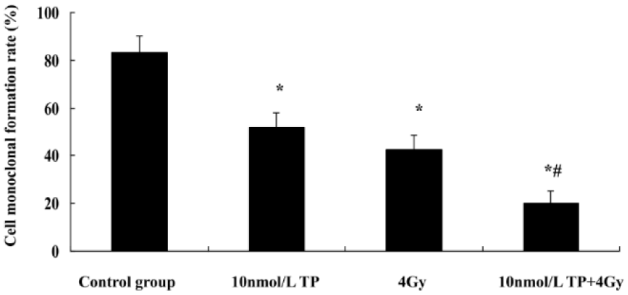

Figure 1. Effect of a combination of triptolide and 4 Gy X-ray on the ability of glioma U251 cells to form monoclonal colonies (x±s, n=5). *indicates $p<0.05$ *vs.* control group; #indicates $p<0.05$ *vs.* the 10 nmol/L TP group, 4 Gy group.

Table 1. Effect of combination of different concentrations of TP and different doses of X-ray on the proliferation inhibition rate of glioma U251 cells by MTT assay (x±s) %, n=5

Group	*0 Gy*	*2 Gy*	*4 Gy*	*6 Gy*	*8 Gy*
0 nmol/L TP	6.14±2.03	11.90±3.96	24.37±3.84*	38.31±4.03*	54.14±5.29*
10 nmol/L TP	18.36±2.16#	20.34±3.84*#	35.56±4.66*#	51.96±5.20*#	66.92±5.01*#
50 nmol/L TP	26.48±2.33#	33.49±4.82*#	48.24±5.07*#	63.33±4.29*#	75.63±5.14*#
100 nmol/L TP	38.48±3.27#	45.69±5.24*#	59.55±5.27*#	71.75±5.60*#	89.48±5.38*#

* indicates $p<0.05$ *vs.* 0 Gy group; #indicates $p<0.05$ *vs.* the 0 nmol/L TP group

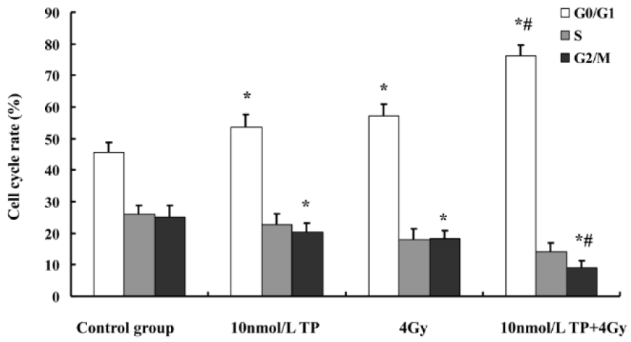

Figure 2. Effect of triptolide and 4 Gy X-ray in combination on cell cycle of glioma U251 cells results (x±s, n=5). *indicates p<0.05 *vs.* control group; #indicates p<0.05 *vs.* the 10 nmol/L TP group, 4 Gy group.

oclonal formation rate of U251 cells in 10 nmol/L TP group and 4 Gy X-ray group was significantly lower than that in the control group (p<0.05). Compared with 10 nmol/L TP alone group and 4 Gy X-ray alone group, the monoclone formation rate of U251 cells in 10 nmol/L TP + 4 Gy X-ray combined group was also significantly decreased (p<0.05).

Triptolide can significantly inhibit the cell cycle of U251 glioma cells under X-ray irradiation

Flow cytometry showed that 10 nmol/L TP combined with 4 Gy X-ray could significantly block the cell cycle in G1 phase of glioma U251

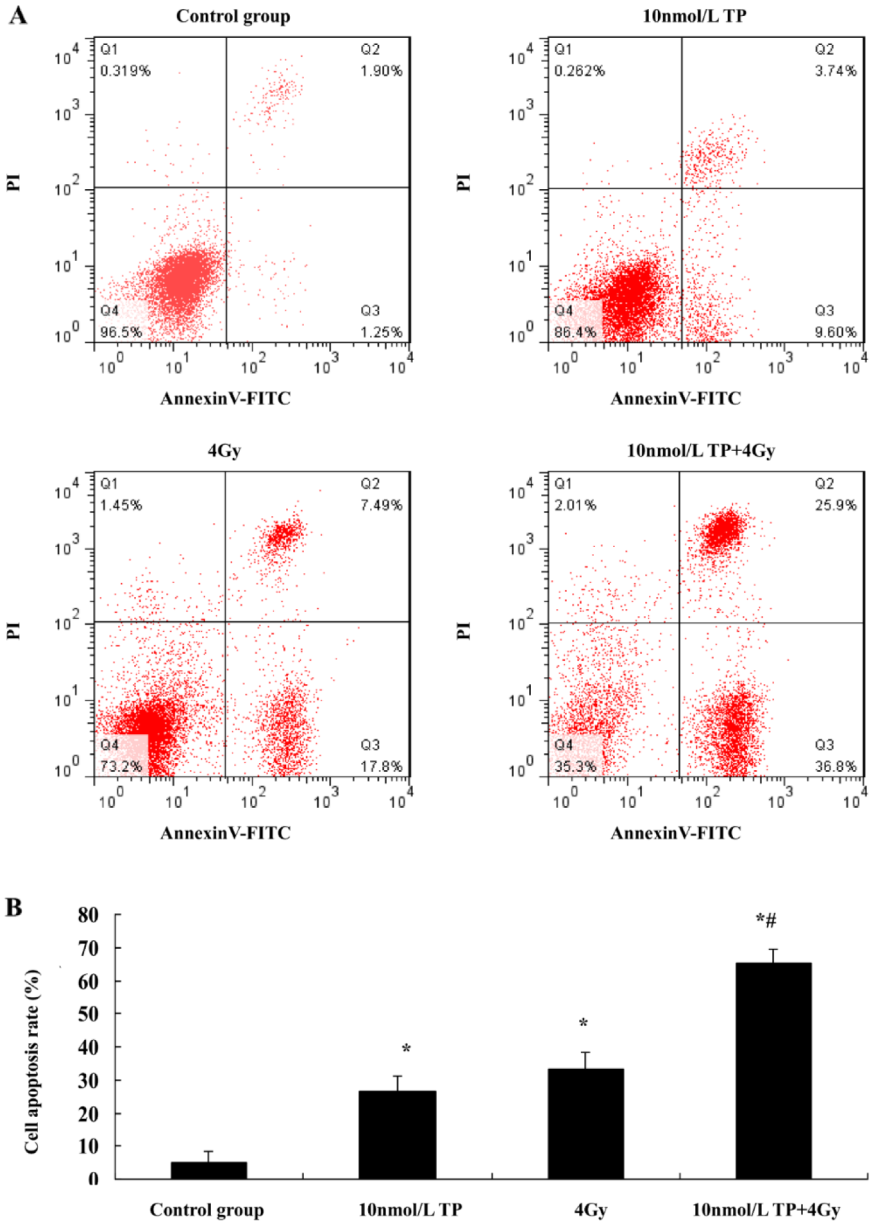

Figure 3. Apoptosis of glioma U251 cells treated with a combination of triptolide and 4 Gy X-ray (x±s, n=5). **A:** Apoptosis was detected by flow cytometry; **B:** Apoptosis rate statistics. *indicates p<0.05 *vs.* control group; #indicates p<0.05 *vs.* the 10 nmol/L TP group, 4 Gy group.

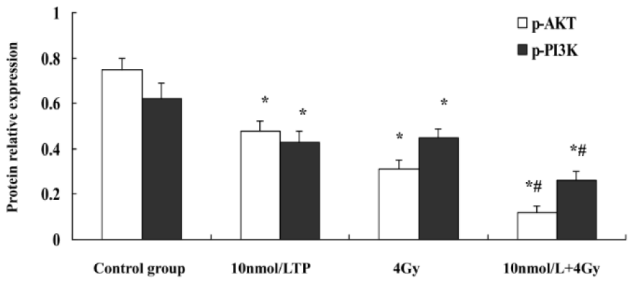

Figure 4. Effect of triptolide combined with 4Gy X-ray on the expression of p-AKT and p-PI3K in glioma U251 (x±s, n=5). *indicates p<0.05 *vs.* control group; #indicates p<0.05 *vs.* the 10 nmol/L TP group, 4 Gy group.

cells (Figure 2). The results showed that compared with the control group, the G1 phase of U251 cell cycle in 10 nmol/L TP group and 4Gy X-ray group were significantly increased, while the G2 phase was significantly decreased (p<0.05). Compared with 10 nmol/L TP group and 4 Gy X-ray group, the G1 phase of U251 Cell Cycle in 0 nmol/L TP + 4 Gy X-ray group increased more significantly, and the G2 phase also decreased significantly (p<0.05).

Triptolide can significantly promote the apoptosis of glioma U251 cells induced by X-ray

Flow cytometry showed that 10 nmol/L TP combined with 4 Gy X-ray could significantly promote the apoptosis of glioma U251 cells (Figure 3). Statistics showed that the apoptosis rate of U251 cells in 10 nmol/L TP group and 4 Gy X-ray group was significantly higher than that in the control group (p<0.05). Compared with 10 nmol/L TP group and 4 Gy X-ray group, the apoptosis rate of U251 cells in 10 nmol/L TP + 4 Gy X-ray group increased significantly (p<0.05).

Triptolide can significantly inhibit PI3K/Akt signaling pathway in glioma U251 cells under X-ray irradiation

Western blot analysis showed that 10 nmol/L TP combined with 4 Gy X-ray could significantly inhibit the phosphorylation levels of Akt and PI3K in glioma U251 cells, indicating that TP combined with X-ray could inhibit Akt/PI3K signaling pathway in U251 cells (Figure 4). The results showed that the levels of p-Akt and p-pi3k in U251 cells treated with 10 nmol/L TP alone and 4Gy X-ray alone were significantly lower than those in the control group (p < 0.05). Compared with the 10 nmol/L TP alone group and the 4 Gy X-ray alone group, the levels of p-Akt and p-pi3k in U251 cells in 10 nmol/L TP + 4 Gy X-ray combined group were also significantly decreased (p<0.05).

Discussion

Radiotherapy is a targeted killing method of tumor cells using irradiation sources rays such as X-rays, γ-rays etc and it is currently one of the important means of treating malignant tumors [11]. The biological characteristics of glioma cells are complex, their growth is rapid, the tumor invading the human brain is often poorly demarcated from normal tissue, surgery is often unable to completely remove the tumor, and patients have a poor prognosis with postoperative adjuvant chemoradiotherapy and are prone to recurrence [12]. Also, the reduced sensitivity of gliomas to radiation is one of the important reasons affecting their prognosis, and clinically, even increasing the local radiation dose does not further reduce the recurrence of disease [13]. Therefore, how to improve the radiosensitivity of gliomas is a a hotspot problem needing answers for clinical glioma treatment.

Current research has found that medicinal plants are rich in natural active products, while products with antitumor activity mostly have radiosensitizing utility, which has the advantages of multi-pathway, multi-target, multi-utility, little toxic side effects, and plays an important complex role in tumor adjuvant radiotherapy [14]. On the one hand, active ingredients in traditional Chinese medicine (TCM), are able to enhance the sensitivity of radiation therapy, aid in tumor treatment and prevent recurrence, while, on the other hand, they have a regulatory effect on the body as a whole, and are able to alleviate the toxic effects of radiotherapy [15]. In recent years, Tripterygium wilfordii as an antitumor therapeutic drug, often combined with Salvia miltiorrhiza, Astragalus membranaceus, and Glycyrrhiza, is not only widely used in China, but also in a considerable number of Asian countries such as Korea and Japan [16]. As an active diterpenoid extracted from Tripterygium Wilfordii Hook f (rupintrivir hook F.), its inhibitory effects on tumors such as gastric cancer, pancreatic cancer, breast cancer, adrenal cancer, and neuroblastoma have been reported, but little is known about its effects on radiosensitivity [17]. Cao et al [18] found that low concentrations (12.5 ng/ml) of TP significantly enhanced the toxic effects of chemotherapeutic agents such as cisplatin on HepG2 cells, and could reverse cisplatin resistance in HepG2 cells. At the same time, new studies have shown that TP combined with irradiation can significantly inhibit human nasopharyngeal carcinoma cell proliferation and tumor growth, and combined with irradiation can be used as a new choice for nasopharyngeal carcinoma treatment [19], but at present, there has been no study on whether TP has a synergis-

tic effect on the radiotherapy sensitivity of human glioma cells. In this study, we used human glioma U251 cells and used different concentrations of TP to act on U251 cells under different doses of X-ray irradiation, and detected that all could significantly enhance the inhibition of cell proliferation and had concentration effect in U251 cells under X-ray irradiation, proving that TP could enhance the sensitivity of glioma U251 cells to X-ray radiation. Also, it has consistency with the above studies.

Clinical radiobiology shows that if the cells can maintain unlimited proliferation ability after radiation it is an important standard to judge the curative effect of radiation, and the application of cell monoclonal test can prove whether the cells treated by radiation still have reproductive integrity [20]. In this study, we selected low concentration of TP and X-ray to act on U251 cells separately and in combination, and detected the ability of cell monoclonal formation. We found that TP can significantly inhibit the ability of U251 cells monoclonal under X-ray irradiation, which confirmed that TP can improve the radiation efficacy of glioma U251 cells. In the study of Wan et al [21], different concentrations of TP were applied to acute leukemia cells, and it was found that TP could significantly promote the apoptosis of HL-60 cells, showed a significant concentration effect and had a blocking effect on the G1 phase of HL-60 cell cycle. In this study, flow cytometry also found that TP can block the cell cycle in G1 phase of U251 cells under X-ray irradiation, and can significantly promote the apoptosis rate of U251 cells under X-ray irradiation, which also indicates that TP has a synergistic effect on U251 cells under X-ray irradiation.

PI3K/Akt signaling pathway plays a very important role in the occurrence and development of various tumors, and it is also closely related to tumor radiosensitivity [22]. It is generally believed that abnormal activation of PI3K/Akt signaling pathway exists in most malignant tumors, such as glioma, and downstream pathway genes are activated through Akt phosphorylation, which plays a role in inhibiting tumor cell apoptosis and promoting cell cycle progression. Therefore, selective inhibition of PI3K/Akt signaling pathway by pharmacological or genetic means can significantly increase the radiosensitivity of tumor cells [23,24]. It has been found that LY294002, a PI3K inhibitor, can decrease the survival rate of SGC-7901 cells under irradiation and induce cell cycle arrest at G1/G2 phase, while LY294002 mainly plays a role in inducing apoptosis by reducing p-Akt [25]. In this study, the levels of p-pi3k and p-Akt in U251 cells induced by 10 nmol/L TP and 4 Gy X-ray decreased significantly. When TP was combined with X-ray, the levels of p-pi3k and p-Akt in U251 cells decreased more significantly, indicating that TP enhanced the radiosensitivity of glioma U251 cells by inhibiting PI3K/Akt signaling pathway. However, the specific mode of action and the changes of downstream signaling pathway need further investigations.

Conclusions

In conclusion, this study confirmed that TP can significantly improve the radiosensitivity of human glioma cells and play a role by inhibiting PI3K/Akt signaling pathway. TP can be used as a potential adjuvant of radiotherapy, which provides a new treatment strategy for glioma.

Conflict of interests

The authors declare no conflict of interests.

References

1. He X, Fan S. hsa-miR-212 modulates the radiosensitivity of glioma cells by targeting BRCA1. Oncol Rep 2018;39:977-84.
2. Hou W, Song L, Zhao Y, Liu Q, Zhang S. Inhibition of Beclin-1-Mediated Autophagy by MicroRNA-17-5p Enhanced the Radiosensitivity of Glioma Cells. Oncol Res 2017;25:43-53.
3. Li T, Ma J, Han X et al. MicroRNA-320 Enhances Radiosensitivity of Glioma Through Down-Regulation of Sirtuin Type 1 by Directly Targeting Forkhead Box Protein M1. Transl Oncol 2018;11:205-12.
4. Visvanathan A, Patil V, Arora A et al. Essential role of METTL3-mediated m(6)A modification in glioma stem-like cells maintenance and radioresistance. Oncogene 2018;37:522-33.
5. Mei Z, Yang Y, Li Y et al. Galanin suppresses proliferation of human U251 and T98G glioma cells via its subtype 1 receptor. Biol Chem 2017;398:1127-39.
6. Li XY, Wang SS, Han Z et al. Triptolide Restores Autophagy to Alleviate Diabetic Renal Fibrosis through the miR-141-3p/PTEN/Akt/mTOR Pathway. Mol Ther Nucleic Acids 2017;9:48-56.
7. Chen C, Yang S, Zhang M et al. Triptolide mitigates radiation-induced pneumonitis via inhibition of alveolar macrophages and related inflammatory molecules. Oncotarget 2017;8:45133-42.

8. Li R, Lu K, Wang Y et al. Triptolide attenuates pressure overload-induced myocardial remodeling in mice via the inhibition of NLRP3 inflammasome expression. Biochem Biophys Res Commun 2017;485:69-75.
9. Noel P, Von Hoff DD, Saluja AK, Velagapudi M, Borazanci E, Han H. Triptolide and Its Derivatives as Cancer Therapies. Trends Pharmacol Sci 2019;40:327-41.
10. Noel P, Hussein S, Ng S et al. Triptolide targets super-enhancer networks in pancreatic cancer cells and cancer-associated fibroblasts. Oncogenesis 2020;9:100.
11. Rekers NH, Olivo PV, Yaromina A et al. The immunocytokine L19-IL2: An interplay between radiotherapy and long-lasting systemic anti-tumour immune responses. Oncoimmunology 2018;7:e1414119.
12. Lu J, Li D, Zeng Y et al. IDH1 mutation promotes proliferation and migration of glioma cells via EMT induction. JBUON 2019;24:2458-64.
13. Malouff TD, Peterson JL, Mahajan A, Trifiletti DM. Carbon ion radiotherapy in the treatment of gliomas: a review. J Neurooncol 2019;145:191-9.
14. Jiang X, Cao G, Gao G, Wang W, Zhao J, Gao C. Triptolide decreases tumor-associated macrophages infiltration and M2 polarization to remodel colon cancer immune microenvironment via inhibiting tumor-derived CXCL12. J Cell Physiol 2021;236:193-204.
15. Datan E, Minn I, Xu P et al. A Glucose-Triptolide Conjugate Selectively Targets Cancer Cells under Hypoxia. iScience 2020;23:101536.
16. Philips BJ, Kumar A, Burki S, Ryan JP, Noda K, D'Cunha J. Triptolide-induced apoptosis in non-small cell lung cancer via a novel miR204-5p/Caveolin-1/Akt-mediated pathway. Oncotarget 2020;11:2793-806.
17. Chen H, Chen Q, Jiang CM et al. Triptolide suppresses paraquat induced idiopathic pulmonary fibrosis by inhibiting TGFB1-dependent epithelial mesenchymal transition. Toxicol Lett 2018;284:1-9.
18. Cao LJ, Li HD, Yan M et al. The Protective Effects of Isoliquiritigenin and Glycyrrhetinic Acid against Triptolide-Induced Oxidative Stress in HepG2 Cells Involve Nrf2 Activation. Evid Based Complement Alternat Med 2016;2016:8912184.
19. Lou D, Lou Z, Lin Y et al. ATB(0,+)-targeted delivery of triptolide prodrugs for safer and more effective pancreatic cancer therapy. Bioorg Med Chem Lett 2021;33:127728.
20. Pedersen KH, Kunugi KA, Hammer CG, Culberson WS, DeWerd LA. Radiation Biology Irradiator Dose Verification Survey. Radiat Res 2016;185:163-8.
21. Wan CK, Wang C, Cheung HY, Yang M, Fong WF. Triptolide induces Bcl-2 cleavage and mitochondrial dependent apoptosis in p53-deficient HL-60 cells. Cancer Lett 2006;241:31-41.
22. Hu XF, He XT, Zhou KX et al. The analgesic effects of triptolide in the bone cancer pain rats via inhibiting the upregulation of HDACs in spinal glial cells. J Neuroinflammation 2017;14:213.
23. Li W, Yang Y, Hu Z, Ling S, Fang M. Neuroprotective effects of DAHP and Triptolide in focal cerebral ischemia via apoptosis inhibition and PI3K/Akt/mTOR pathway activation. Front Neuroanat 2015;9:48.
24. Kim SH, Kang JG, Kim CS et al. Synergistic cytotoxicity of BIIB021 with triptolide through suppression of PI3K/Akt/mTOR and NF-kappaB signal pathways in thyroid carcinoma cells. Biomed Pharmacother 2016;83:22-32.
25. Zhang Y, Li Z, Fan X et al. PRL-3 promotes gastric cancer peritoneal metastasis via the PI3K/AKT signaling pathway in vitro and in vivo. Oncol Lett 2018;15:9069-74.