tumor cell death and growth inhibition of glioma cells upon glutamine deprivation. These results suggest new therapeutic targets for glioma cells adapting to a low-nutrient microenvironment.

Key words: Glioma | Microenvironment | One-carbon metabolism

CBMS-7

IGF1/N-CADHERIN/CLUSTERIN SIGNALING AXIS MEDIATES ADAPTIVE RADIORESISTANCE OF GLIOMA STEM CELLS

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Glioblastoma (GBM) is composed of a variety of tumor cell populations including those with stem cell properties, known as glioma stem cells (GSCs). GSCs are innately less sensitive to radiation than the tumor bulk and are believed to drive GBM formation and recurrence following repeated irradiation. However, it is unclear how GSCs adapt to the repeated irradiation used in clinical practice. We established radioreistant human and mouse GSCs by exposing them to repeated rounds of irradiation in order to uncover critical mediators of adaptive radioresistance. Surviving subpopulations acquired strong radioresistance in vivo, which was accompanied by increased cell-cell adhesion, slower proliferation, an elevation of stemness properties and N-cadherin expression. Increasing N-cadherin expression rendered parental GSCs radioreistant, reduced their proliferation, and increased their stemness and intercellular adhesive properties. Conversely, radioreistant GSCs reduced their acquired phenotypes upon CRISPR/Cas9-mediated knockout of N-cadherin. Mechanistically, elevated N-cadherin expression resulted in the accumulation of β-catenin at the cell surface, which decreased Wnt/β-catenin proliferative signaling, reduced neural differentiation, and protected against apoptosis through Clusterin secretion. Restoration of wild type N-cadherin, but not mutant N-cad lacking β-catenin binding region, led to increased radioresistance in N-cadherin knockout GSCs, indicating the importance of the binding between N-cadherin and β-catenin. We also demonstrated that N-Cadherin upregulation was induced by radiation-induced IGF1 secretion, and the radiation resistance phenotype can be reversed with picropodophyllin (PPP), a clinically applicable blood-brain-barrier permeable IGF1 receptor inhibitor; supporting clinical translation. Moreover, the elevation of N-cad and Clusterin were related to prognosis of GBM in the TCGA database. In conclusion, our data indicate that IGF1R inhibitor can block the N-cadherin-mediated resistance pathway. Our research provides a deeper understanding of adaptive radioresistance after repeated irradiation, and validated Clusterin and IGF1/N-cadherin as a potential therapeutic target for GBM.

Key words: N-cadherin | Glioma stem cells | IGF1R

CBMS-10

METHIONINE METABOLISM CLOSLY RELATED WITH SELF-RENEW, MIPOTINOGENIC AND CELL DEATH IN GICS THROUGH MODIFICATION OF CHOLESTEROL BIOSYNTHESIS AND RIBOSOMAL RNA

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Glioma initiating cells (GICs) are the source of glioma cells that have the ability to self-renew and pluripotency, which are treatment-resistant, starting point for relapse and eventual death despite multimodality therapy. Since high accumulation is observed in 11c-Met-PET at the time of recurrence, it is important to understand the mechanism of tumor cell activation caused by the reorganization of methionine metabolism. We cultured cells in methionine-deprived culture medium and performed a comprehensive analysis, and found that methionine depletion markedly decreased proliferation and increasing cell death of GICs. Decreased SAM, which is synthesized intracellularly catalyzed by methionine adenosyltransferase (MAT) using methionine, triggered the following: (i) global DNA demethylation, (ii) hyper-methylation of signaling pathways regulating pluripotency of stem cells, (iii) decreased expression of the core-genes and pluripotent marker of stem cells including FOXM1, SOX2, SOX4, FOXM1 and OLIG2, (iv) decreased cholesterol synthesis and increased expression mainly through decreased SREBF2 (v) down-regulation of the large subunit of ribosomal protein configured 28S and 18S, and SRENA guided the pseudouridylation of 28S ribosomal RNA, which has crucial role for translation. In addition, inhibition of cholesterol synthesis with statin resulted in a phenotype similar to that of methionine removal and a decrease in stem cell markers and SRENA ACA43. Moreover, suppression of FOXM1 decreased stem cell markers such as SOX4 and PROM1. The gene expression profile for chole-sterol ester production was obtained from the Ivy Glioblastoma Atlas Project (IvyGAP) database and compared between tumour cells with relatively low methionine levels in area of pseudopalisading arrangement around necrosis and tumour cells in the infiltrating region, showing that cells in the infiltrating region have a higher capacity to produce cholesterol. Taken together, alterations in metabolic pathways closely related with self-renew, pluripotency and cell death in GICs through modification of cholesterol biosynthesis especially SREBF2-FOXMI and ACA43 axis with modification of ribosomal RNA.

Key words: glioma stem cells | methionine | cholesterol

SIGNALLING PATHWAYS/DRUG RESISTANCE (SPDR)

SPDR-1

HSP90 INHIBITION OVERCOMES RESISTANT TO MOLECULAR TARGETED THERAPY IN BRAFV600E MUTANT GLOIOBLASTOMA

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The BRAFV600E mutation results in the constitutive activation of downstream mitogen activated protein kinase (MAPK) pathway that promotes tumor growth. Recently, molecular targeted therapy using BRAF/MEK inhibitor has been reported for BRAFV600E mutant high-grade glioma, but the therapeutic effect is limited by the emergence of drug resistance. Herein, we established paired BRAFV600E mutant glioblastoma (GBM) patient-derived xenograft (PDX) models, which were derived from tumors at prior to and recurrence after molecular targeted therapy. These PDX models were found to extensively recapitulate the histology, genomic abnormalities, and even the clinical course of the patients. Furthermore, BRAF/MEK inhibitor gradually caused resistance in cell lines derived from specimens that initially responded to molecular targeted therapy. In this study, genomic and epigenomic changes had little effect on the resistance mechanism. On the other hand, we found that hyperactivation of the MAPK pathway through c-Raf and the AKT/mTOR pathway primarily caused resistance to molecular targeted therapy in BRAFV600E mutant GBM. Through a high throughput drug screening, we find that HSP90 inhibitor with BRAF/MEK inhibitor coordinately deactivates MAPK pathway and AKT/mTOR pathway, and mediates potent toxicity in vitro and in vivo in both immortalized and authentic GBM cell models. These findings support that the therapeutic approach can overcome the limitation of current molecular targeted therapy in BRAFV600E mutant GBM.

Key words: BRAF V600E | Patient-derived xenograft model | HSP90 inhibitor

SPDR-2

HISTOPATHOLOGICAL INVESTIGATION OF THE OLIGODENDROGIAL TUMORS RESECTED FOLLOWING ALKYLATING AGENT CHEMOTHERAPY

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Oligodendrogliomas, i.e., lower grade gliomas with 1p/19q codeletion, are often sensitive to chemotherapy, however, those tumors eventually recur and life-limiting in the majority of patients despite initial chemotherapy response. We have been treating those patients with upfront chemotherapy and subsequent resection following tumor volume decrease since 2006. This study aimed to elucidate the histological changes and the mechanism of recurrence after alkylating agent chemotherapy in oligodendrogliomas. Fifteen oligodendrogliomas (Grade 2: 12, Grade 3: 3) resected following tumor volume decrease after alkylating agent chemotherapy were included and were compared with their pre-chemotherapy specimen. Histological changes were investigated using hematoxylin-eosin staining, and changes in proliferative activity, status of glioma stem cells (GSCs), and tumor-infiltrating macrophages were assessed using immunohistochemistry. The frequent demonstrated findings following chemotherapy included a sparse glial background, abundant foamy cell infiltration, gliosis, calcification, and nuclear