

FSMP-14. METABOLIC SYMBIOSIS IN GLIOBLASTOMA: LACTATE DEHYDROGENASES TAKE THE LEAD

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Glioblastoma (GBM) is a common and devastating brain tumor, associated with a low median survival, despite standard therapeutic management. Among its major features, GBMs are highly angiogenic, infiltrative, and exhibit paradoxically an elevated glycolysis. Most of differentiated cells convert glucose into pyruvate that enters into the Krebs cycle to maximize energy production in the presence of oxygen. For cancer cells, glucose uptake and catabolism are increased regardless of oxygen level. However, tumor cell energy needs are important, mainly for rapid growth, which require a much faster production flow. Lactate dehydrogenase (LDH) are involved at this step. LDHA converts pyruvate into lactate, and generates NAD⁺ to maintain glycolysis. Thus, lactate is exported into the extracellular compartment inducing an acidification of the microenvironment. Moreover, LDHB, another LDH isoform, metabolizes lactate into pyruvate for generating energy in mitochondria. LDHB is generally expressed in oligodendrocytes or neurons, but also in GBM cells. Though LDHA has already been studied in many cancers including GBM, the simultaneous role of LDH enzymes have not yet been investigated in GBM development. Our results showed that hypoxia significantly increased LDHA expression and lactate production, but no changes were observed for LDHB. In presence of lactate, cell invasion was significantly increased. *In vitro* results showed that, under hypoxic condition, double sgLDHA/B cell growth and invasion was dramatically decreased in comparison to control cells, mainly caused by an increase in apoptosis. *In vivo* experiments showed that double impairment of LDHA and B significantly reduced tumor growth and cell invasion, and induces a massive increase in mouse survival. Considered for a long time as a metabolic waste, lactate is shown here to play a critical role in the tumor niche. This study highlighted GBM adaptability through the LDH isoforms and their involvement in GBM development.

FSMP-15. EVALUATING THE ROLE OF LONG-CHAIN FATTY ACID METABOLISM IN PROMOTING GLIOBLASTOMA GROWTH

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Glioblastomas (GBM) or Stage IV gliomas, are the most aggressive of primary brain tumors and are associated with high mortality and morbidity. Patients diagnosed with this lethal cancer have a dismal survival rate of 14 months and a 5-year survival rate of 5.6% despite a multimodal therapeutic approach, including surgery, radiation therapy, and chemotherapy. Aberrant lipid metabolism, particularly abnormally active *de novo* fatty acid synthesis, is recognized to have a key role in tumor progression and chemoresistance in cancers. Previous studies have reported a high expression of fatty acid synthase (FASN) in patient tumors, leading to multiple investigations of FASN inhibition as a treatment strategy. However, none of these have developed as efficacious therapies. Furthermore, when we profiled FASN expression using The Cancer Genome Atlas (TCGA) we determined that high FASN expression in GBM patients did not confer a worse prognosis (HR: 1.06; p-value: 0.51) and was not overexpressed in GBM tumors compared to normal brain. Therefore, we need to reexamine the role of exogenous fatty acid uptake over *de novo* fatty acid synthesis as a potential mechanism for tumor progression. Our study aims to measure and compare fatty acid oxidation (FAO) of endogenous and exogenous fatty acids between GBM patients and healthy controls. Using TCGA, we have identified the overexpression of multiple enzymes involved in mediating the transfer and activation of long-chain fatty acids (LCFA) in GBM tumors compared to normal brain tissue. We are currently conducting metabolic flux studies to (1) assess the biokinetics of LCFA degradation and (2) establish exogenous versus endogenous LCFA preferences between patient-derived primary GBM cells and healthy glial and immune cells during steady state and glucose-deprivation.

FSMP-17. GLOBAL METABOLOMIC PROFILING OF GLIOBLASTOMA MULTIFORME REVEALS METABOLIC VULNERABILITIES IN RESPONSE TO RADIATION THERAPY

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Glioblastoma multiforme (GBM), the most aggressive primary brain tumor, originates in astrocytes and oligodendrocytes and yields a median survival time of less than 2 years and a 5-year survival of 2.5%. There has been little in the way of treatments and novel approaches are needed to

combat the poor prognosis of GBM. Recent studies have established that GBM cells exhibit metabolic reprogramming to adapt to diverse metabolic gradients within heterogeneous tumor microenvironments. Using an unbiased metabolomics approach, we investigated metabolic changes both pre- and post-ionizing radiation across several patient-derived GBM cell lines. Surprisingly, acute high dosage of ionizing radiation resulted in significant changes in the synthesis of aminolevulinic acid (ALA), a non-proteinogenic amino acid. Fractionation of radiation therapy resulted in dose-dependent changes in the heme synthesis pathway within these cells. Using an orthotopic xenograft mouse model of GBM, we identify several enzymatic vulnerabilities *in vivo* and discuss a novel combinatorial therapeutic approach of radiation and targeted pharmacological intervention. Our findings reveal the fundamental biosynthetic changes that GBMs adopt when exposed to ionizing irradiation as well as the benefits of a combinatorial approach.

FSMP-18. COMPREHENSIVE METABOLIC PROFILING OF HIGH MYC MEDULLOBLASTOMA REVEALS KEY DIFFERENCES BETWEEN IN VITRO AND IN VIVO GLUCOSE AND GLUTAMINE USAGE

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Reprogramming of cellular metabolism is a hallmark of cancer. Altered metabolism can overcome unfavorable conditions, allowing cancer cells to proliferate and invade in different tumor microenvironments. Medulloblastoma is the most common malignant brain tumor of children. Genomic amplification of MYC is a hallmark of a subset of poor-prognosis medulloblastoma. However, the metabolism of high MYC amplified medulloblastoma subgroup remains underexplored. We performed comprehensive metabolic studies of human MYC-amplified medulloblastoma by comparing the metabolic profiles of tumor cells in different environments – *in vitro*, in flank xenografts and in orthotopic xenografts. Principal component analysis showed that the metabolic profiles of brain and flank high-MYC medulloblastoma tumors clustered closely together and separated away from normal brain and the high-MYC medulloblastoma cells in culture. Compared to normal brain, MYC-amplified medulloblastoma orthotopic brain tumor xenografts showed upregulation of nucleotide, amino acid and glutathione pathways. Glucose was the main carbon source for the nucleotide synthesis and the TCA cycle *in vivo*. The glutaminase ii pathway was the main pathway utilizing glutamine in MYC-amplified medulloblastoma. In brain and flank xenografts, glutathione was the most abundant upregulated metabolite. Glutamine derived glutathione was synthesized through glutamine transaminase K (GTK) enzyme *in vivo*. The glutamine analog 6-diazo-5-oxo-L-norleucine (DON) significantly inhibited glutathione, amino acid, and nucleotide synthesis. In conclusion, we found that MYC-amplified medulloblastoma relied on glutamine metabolism in synthesizing glutathione *in vivo*. Glutamine antagonists may have therapeutic applications in human patients.

FSMP-19. SEX DIFFERENCES IN REDOX REGULATION UNDERLIE GLUTAMINE DEPENDENCY IN MALE GLIOBLASTOMA

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Glioblastoma (GBM) is the most aggressive primary brain tumor in adults. GBM occurs more commonly in males but female patients survive significantly longer. Understanding the molecular mechanisms underlying this clinical sex disparity could support novel treatment strategies to improve outcomes for GBM patients. In this regard, we found that male and female GBM patient tissues differ in their metabolite profiles and that male GBM exhibit a higher abundance of amino acid metabolites. We confirmed these findings in a murine model of GBM. Furthermore, we found that male GBM cells are more sensitive to amino acid deprivation. This male-specific dependency on amino acids is almost entirely driven by amino acids involved in reactive oxygen species (ROS) regulation and glutathione synthesis. We found that male GBM cells are more sensitive to depletion of glutathione, which resulted in a significant increase in ROS and cell death in male GBM cells. Moreover, assays of glutathione oxidation demonstrated that male GBM cells exist in a chronically oxidized state. GLS1 mediates the conversion from glutamine to glutamate, a crucial component of glutathione. We found that male GBM cells are more sensitive to GLS1 inhibition with the clinical inhibitor CB-839. This correlated with significantly increased ROS and glutathione levels as well as significantly decreased TCA cycle metabolites in male GBM. Lastly, we found that the TCA cycle me-