TMOD-26. CYTOMEGALOVIRUS PROMOTES GLIOBLASTOMA GROWTH VIA PDGF-D DRIVEN PERICYTE RECRUITMENT AND ANGIOGENESIS

Harald Krenzlin, Prajna Behera, Carmela Passaro, Mykola Zdioruk, Korneel Grauwet, C David James, Hirotaka Ito, Charles Cobbs, Charles Cook, E Antonio Chiocca, Sean Lawler
Abstracts

Ultimately, our patient-derived iPSC-based approach may enable personalized precision medicine strategies against glioma.

TMOD-25. MODELING IDH1-MUTATED GLIOMAS: GENERATION, CHARACTERIZATION AND THERAPEUTIC SENSITIVITIES OF SEVEN PATIENT-DERIVED IDH1-MUTANT GLIOMA CELL LINES

INTRODUCTION: In spite of significant attention IDH mutations have attracted, in vitro and in vivo model systems with endogenous IDH mutations remain scarce. Development of these models is crucial not only for unraveling the molecular mechanisms that contribute to this disease entity, but especially for the development of new therapeutic interventions. METH- ODS: Fresh glioma tissue was obtained directly from the operating theater, dissociated and cultured under serum-free conditions. The presence of IDH mutations was verified at multiple passages using Sanger sequencing. D2-HG levels were measured through mass spectrometry. Cell proliferation was assessed using a cell counting, cell viability measured with an ATP-based viability assay. Drug screens were carried out with an FDA-approved Oncology Drugs Set from the NIH and multiple IDH mutant-specific inhibitors. RESULTS: Over 800 consecutive low grade and high grade glioma surgical samples were processed for cell culture. For seven tumors our model, MCMV can be seen in tumor cells and vascular pericytes, a finding critical for the culture in vitro. All cultures were derived from astrocytic tumors. From one patient both the primary grade II astrocytoma and recurrent grade IV glioblastoma formed successful IDH-mutant cultures. IDH mutant specific inhibitors showed modest effects on cell viability, indicating limited dependence on D2-HG for growth. Drug screening revealed a subset of compounds that decrease viability at clinically-feasible drug concentrations which may merit further investigation. Whole-genome sequencing in parallel with matched frozen-tissue tumor tissue, as well as RNA sequencing analyses are underway. CONCLUSION: We established a set of cell cultures derived from seven IDH mutant gliomas and characterized these on both genetic and transcriptional levels, and investigated drug sensitivity and D2-HG dependence. This unique set can be utilized to investigate novel therapeutic strategies.

TMOD-26. CYTOMEGALOVIRUS PROMOTES GLOBLASTOMA GROWTH VIA PDGF-D DRIVEN PERICYTE RECRUITMENT AND ANGIOGENESIS

Harald Krenzlin1, Prajna Behera2, Carmela Passaro2, Mykola Ziuziork2, Korneel Ganewer1, C. David Behera3, Hidetsugu Ito1, Charles Cobbs4, Charles Coulter2, Priyanka Chochia1, and Edward Struyf2, Gaja Salomons1, Sieger Leenstra1, Pam French2 and Martine Lamfers1

Human cytomegalovirus (HCMV) is highly prevalent, and like other herpes viruses, can persist for life in its host in a latent state. However, HCMV can be severely pathogenic in immunocompromised individuals. Initially granulocytic HCMV proteins and nuclear capsid antigens have been identified in 90% of patients with the incurable brain tumor glioblastoma (GBM) as well as some other cancers. Accumulating data supports the clinical relevance of HCMV in GBM, with some encouraging responses reported with HCMV-targeted immunotherapies. Although various HCMV proteins increase cell proliferation and invasion, a mechanistic link between HCMV and cancer in vivo has not been established, and the role of HCMV in GBM remains a subject of debate. In the current report we show that peripheral murine CMV (MCMV) infection induces a pro-angiogenic secretome, increasing tumor growth, pericyte accumulation, angiogenesis and tumor blood flow in a murine GBM model. Specifically, we identify platelet-derived growth factor-d (PDGF-D) as a CMV-induced factor essential for tumor growth. In our model, MCMV can be seen in tumor cells and vascular pericytes, a finding that we confirm in human GBM specimens. The anti-viral drug cidofovir improves survival in MCMV-infected mice, inhibiting MCMV activation, PDGF-D expression, pericyte recruitment and tumor angiogenesis. Together these data provide the first mechanistic explanation of how CMV poten- tiates GBM growth in vivo, identify PDGF-D as a potential therapeutic target, and support the application of anti-viral approaches for GBM therapy.

TMOD-27. HUMANIZED MICROBIOME MOUSE MODELS TO ENHANCE IMMUNOTHERAPY IN GLOBLASTOMA

Brendan McFarland, Kory Dees, Rebecca Little, William Van Der Pol, Ety Vbenenste, Casey Morrow and L. Burt Nabors; University of Alabama at Birmingham, Birmingham, AL, USA

Cancer immunotherapies, including the checkpoint inhibitors, demonstrate remarkable success in patients with melanoma, but have not shown a similar efficacy in patients with glioblastoma (GBM). Recently, the com- position of the gut microbiome has been shown to correlate with GBM patient response to immune checkpoint inhibitors in melanoma and other cancers suggesting a favorable composition of the gut microbiome is needed to produce an optimal response to checkpoint inhibitors and subsequent anti-tumor immune responses. We propose that the gut microbiome of GBM patients plays a role in resistance to immunotherapies. To investigate this, we have collected and analyzed the gut microbiome from GBM patients (short-term and long-term survivors) and healthy controls by microbial 16S and metagenomic sequencing. These results will determine microbiome differences between GBM patients and controls, as well as differences within each patient as the disease progressed. Furthermore, all GBM pre-clinical studies to date have been performed in mouse models using mouse gut microbiota. We have previously found that human fecal samples can be transplanted into gnotobiotic (germ-free) mice to successfully colonize the mouse GI tract with human microbes. Herein, we have established humanized microbiome mice utilizing human GBM donor fecal samples previously obtained and analyzed. This model is critical because it allows us to study the relationship between the human microbiome and GBM, and the responsiveness to ther- apies using the syngeneic GL261 intracranial model. We are currently using these humanized microbiome mouse models to analyze pre-clinical testing of anti-PD-1 in GBM, and endpoints to assess efficacy will include survival times, tumor growth, and examining the periphery and tumor environment for phenotype(s) of infiltrating immune cells. These studies will enhance our understanding of the mechanism of GBM patient's microbiome in the resist- ance to immunotherapies and lead to new therapeutic strategies to alter the microbiome composition to enhance immunotherapy for GBM.

TMOD-28. MYC OVEREXPRESSSION DRIVES MEDULLOBLASTOMA FROM HUMAN NEUROEPITHELIAL STEM CELLS

Sona Hutier1, Holger Weishaupt2, Gabriela Rosin2 and Fredrik Swartling3

1Uppsala University, Uppsala, Sweden, 2Uppsala University, Uppsala, Sweden

Medulloblastoma (MB) is the most prevalent malignant brain tumor in children. Based on molecular genetic profiling, this disease can be classified into four major subgroups which display distinct clinical features. Group 3 MB is associated with overexpression of an amplifiable MYC gene and rarely show any mutations in the tumor suppressor protein p53. Patients with MYC-driven MB have a particularly high risk of recurrence and are associated with extremely poor prognosis. Thus, modeling MYC-driven MB is critical for the development and testing of potential new treat- ment approaches for these high-risk MBs. Here we show the first human MB model developed from human hindbrain neuroepithelial stem (NES) cells and induced pluripotent stem cell-derived NES (iPS-NES) by lentiviral overexpression of wild-type MYC. Following orthotopic transplantation into im- mune-deficient mice, MB tumors are formed in the hindbrain showing progression. MYC-expressing tumors are comprised of poorly differentiated cells with high expression of E2A and driven MB is critical for the development and testing of potential new treat- ment approaches for these high-risk MBs. Here we show the first human MB model developed from human hindbrain neuroepithelial stem (NES) cells and induced pluripotent stem cell-derived NES (iPS-NES) by lentiviral overexpression of wild-type MYC. Following orthotopic transplantation into immu- nodeficient mice, MB tumors are formed in the hindbrain showing progression. MYC-expressing tumors are comprised of poorly differentiated cells with high expression of E2A and E47. This model will be useful for testing potential new treatment strategies against glioma.

TMOD-29. MOLECULAR CHARACTERIZATION OF GLIOMA PATIENT-DERIVED ORTHOTOPIC XENOGRAPHS: FROM BASIC RESEARCH TO PRECLINICAL STUDIES

Ann-Christin Haß1, Anna Golobiewska1, Anais Oudin1, Linsey Houben1, Daniel Steib2, Francisco Azua3, Tony Kaoma4, Arnaud Muller4, Frank Hertel1, Michel Mittlebronn1, Rolf Bjerkvig1 and Simone Niclou1

1NorLux Neuro-Oncology Laboratory, Department of Oncology, Luxembourg Institute of Health, Luxembourg, Luxembourg, 2Department of Genetics, Laboratoire National de Santé, Dudelange, Luxembourg, 3Genomics and Proteomics Research Unit, Department of Oncology, Luxembourg Institute of Health, Luxembourg, Luxembourg, 4Department of Neuro-Oncology, National Department of Luxembourg, Centre Hospitalier de Luxembourg, Luxembourg, Luxembourg, Luxembourg, 5Department of Anatomical and Molecular Pathology, Laboratoire National de Santé, Dudelange, Luxembourg

It is well recognized that long term cell cultures are poor models to study human cancer, largely because of loss of clonal heterogeneity, accumulation or loss of genomic alterations and adaptation to a highly artificial environment. Human derived orthotopic xenografts (PDOX) based on human embryonic three-dimensional tumor spheroids from human glioma samples are proposed to represent a reliable and clinically-relevant animal model. We have generated a living biobank of PDOX models from 34 glioma patients (grade III and IV), including longitudinal patient samples with matched recurrent tumors. Using an efficient orthotopic xenografting procedure we obtain an overall tumor

v274 NEURO-ONCOLOGY • NOVEMBER 2018