PRESS RELEASE
Monday, March 21, 2016

Three KIYATEC abstracts accepted for poster presentations during 2016 AACR Conference in New Orleans

KIYATEC, a private company prioritizing accurate ex vivo prediction of patients' response to drug treatment, received acceptance of three posters for presentations during the upcoming AACR conference.

GREENVILLE, South Carolina, March 21, 2016– KIYATEC Inc. is pleased to announce that three abstracts have been accepted by the committee at the American Association for Cancer Research (www.aacr.org). KIYATEC scientific and business development staff will be in attendance at the conference. In addition to the poster presentations, also **KIYATEC will be exhibiting at Booth 1050**. Stop by to learn more about our 10+ years of 3D cell culture experience and the latest 3D co-culture model development projects focused on immuno-oncology applications.

Abstract #611: A Complex 3D Model of Glioblastoma for Patient-Specific Drug Response Profiling
Sunday Apr 17, 2016 1:00 PM - 5:00 PM
Location: Section 28 / Poster 12
Session Category: Tumor Biology
Session Title: Drug Testing in Cell Lines and 3D Models
Authors: Tessa DesRochers1, Lillia Holmes1, Lauren O'Donnell1, Qi Guo1, David Schammel2, Jeff Edenfield3, Charles C. Kanos3, Hal E. Crosswell1. [1]KIYATEC, Inc., Greenville, SC; [2]Pathology Consultants, Greenville, SC; [3]Greenville Health System, Greenville, SC

Abstract: Approximately 70,000 new cases of brain tumors will be diagnosed this year with glioblastoma (GBM) accounting for about 17% of those cases. Unfortunately the median survival for patients with GBM is only 14.6 months due to the complexity of the tumor, the pattern of diffuse spread within the brain, and the lack of effective therapeutic options. Complex in vitro tumor models developed in 3D better mimic in vivo biology, and, when utilizing patient-derived cells, may offer robust platforms for new drug development and patient-specific drug response profiling. We have previously shown this to be true with both breast and ovarian cancer and we have adopted this approach to modelling GBM ex vivo. We hypothesize that primary derived GBM tissues and stem cells cultured in complex 3D microenvironments can recapitulate the intra-tumor heterogeneity and drug resistance similar to that found in the clinical. To this end, we have developed 3D models of GBM that incorporate tumor cells, endothelial cells, and macrophages within a complex extracellular matrix and cultured them for up to 2 weeks under perfusion flow. We have created these models using both cell lines and primary patient cells and these 3D tissues have been analyzed for changes in viability (PrestoBlue and PicoGreen), marker expression (flow cytometry, Luminex, and protein arrays), macrophage differentiation and invasion (flow cytometry, multiphoton microscopy, and IHC), and methylation patterns (ChIP on chip arrays and methylation specific PCR). Initial optimization work using cell lines has revealed that culturing with endothelial cells and/or macrophages has an effect upon proliferation dependent upon the differentiation state of the macrophages. Additionally, co-culture conditions affect protein secretion and phosphorylation along with patterns of gene methylation. We have also examined how changes in the tumor microenvironment affect these different metrics by examining the role of hypoxic growth conditions. Work using patient cells isolated from

primary glioblastoma has focused upon drug response dependent upon the tumor microenvironment and the presence of cancer stem cells utilizing a neurosphere generation assay. We have demonstrated temozolamide resistance in tumors with early relapse after initial chemo-radiation, and sensitivity to EGFR inhibitor afatanib in tumors with EGFR amplification, suggesting clinical and molecular correlation or our ex vivo 3D drug response profiling using patient derived GBM models. Taken together, our data support the further development and use of complex, 3D co-cultures to better mimic GBM in a patient-specific manner. These models are currently being considered for preclinical assessment of novel therapies, including chimeric antigen receptor T cells, and may be a useful adjunct in precision medicine applications to improve patient outcomes.

Abstract #2531: Marker free isolation and expansion of cancer stem cells from small cell lung cancer

Monday, Apr 18, 2016, 1:00 PM - 5:00 PM
Location: Section 34 / Poster 20
Session Category: Tumor Biology
Session Title: Stemness Properties of Neuronal and Pediatric Tumors and New Approaches
Authors: Tessa M. DesRochers, Melissa Millard, Alina Lotstein, Lillia Holmes, Hal E. Crosswell.
KIYATEC, Inc., Greenville, SC

Abstract: Small cell lung cancer (SCLC) accounts for approximately 15% of all lung cancer cases and, contrary to the advances in diagnostics and therapeutic options for non small cell lung cancer, SCLC long term survival has failed to materially improve over the last 3 decades. Early dissemination with hematogenous metastasis, advanced stages at diagnosis, dramatic response to chemotherapy with early and aggressive relapses are the clinical hallmarks. Limited surgical samples, early palliation and limited second line treatment options negating the benefit of repeat biopsies have limited tissue available for research to understand the biology of this relatively rare tumor. Recent data has suggested the presence of rare populations of cells within the primary SCLC tumors which have stem cell-like properties. We hypothesize that novel in vitro culture platforms and methods can be used to isolate and expand cancer stem cells (CSCs) within SCLC tumors and that the expanded CSCs will yield a renewable cell source for research applications and for predictive drug response profiling. Many groups have relied upon cell surface markers such as CD133 and CD24 to identify the CSC population. Our approach is unique in that we have employed a marker-free approach utilizing a combination of chemical and functional isolation. We then use a combination of growth factors, extracellular matrix proteins, and oxygen tension in a 3D perfusion culture system to further isolate, purify and expand isolated cells with the goal of having quantities of functional SCLC stem cells for molecular, proteomic and functional in vitro and in vivo assays. Initial cell sources will include immortalized cell lines and patient derived xenograft lines, with eventual adaptation to primary patient samples. Using numerous techniques, we have qualified these cells based upon marker expression (flow cytometry, RT-PCR), dye exclusion, drug response, and limited dilution assays. Future experiments will confirm stemness with limited dilution tumorigenicity assays in vivo. Initial data suggest poor correlation of canonical CSC marker expression with clonogenic growth in cell lines, supporting the need for unbiased and functional isolation methods. These expanded populations of CSCs may be used to identify novel CSC markers and/or targets, mechanisms of resistance and screen and develop novel therapeutics Our ultimate goals are to be able to use our novel culture platforms and methods to expand CSCs for real time precision medicine applications to identify novel therapies in real time that can eradicate the CSC population and improve outcomes of patients with SCLC.

Abstract # 5119: Complex, 3D tissues for modeling the immune response in cancer and predicting the activity of immunotherapies

Wednesday Apr 20, 2016 8:00 AM - 12:00 PM
Location: Section 31 / Poster 5
Session Category: Tumor Biology
Session Title: Immunomodulation and Immunotherapy

Authors: Tessa M. DesRochers1, Lillia Holmes1, Qi Guo1, Lauren O'Donnell1, Stephen Shuford1, Larry Puls2, Jeff Elder2, Jeff Edenfield2, Hal E. Crosswell1. [1]KIYATEC, Inc., Greenville, SC; [2]Greenville Health System, Greenville, SC

Abstract: The immune system plays an active role in both the prevention and the promotion of cancer dependent upon its interaction with the tumor cells. The roles of both macrophages and T-cells in cancer progression have been heavily studied over the past few years. Macrophages have been found to be either tumor promoting or tumor preventing depending upon their differentiation status and the tumor microenvironment while the homing and cell destroying capabilities of T-cells have been manipulated to effect better, more specific tumor cell cytotoxicity through the development of therapies such as chimeric antigen receptor T-cells (CAR-T cells). Unfortunately the majority of research in the area of immune-oncology has relied upon either 2D cell culture or animal models. While a large amount of information has been learned from these models, it has been well established that 2D cell culture does not mimic in vivo biology and the immune system of mouse models differs from that of humans in numerous ways including T-cell subsets, cytokine receptors, and costimulatory molecule expression. To overcome these limitations, we have developed a number of 3D in vitro tissue models including multi-cell type models of glioblastoma (GBM), breast, and ovarian cancer. Our GBM model combines tumor cells, endothelial cells, and macrophages; the ovarian cancer model includes patient-derived tumor cells and macrophages; our breast cancer model incorporates 5 cell types including tumor cells, fibroblasts, adipocytes, endothelial cells, and macrophages. We have also begun preliminary work to incorporate patient-specific T-cells into these models. Preliminary work in GBM and breast has utilized a combination of cell lines and primary cells while ovarian modeling has relied solely upon primary patient cells from either solid tumors or ascitic fluid. These models have been characterized for tumor cell viability, biomarker expression, and drug response. We have noted changes in both tumor cell viability and biomarker expression dependent upon macrophage differentiation and have also observed the reciprocal effect of tumor cells upon the macrophages. Additionally, employing multiphoton microscopy we have identified the incorporation of macrophages into the 3D microtumors. Utilizing primary patient tumor cells and immune components, we can test these models for patient-specific responses to not only traditional chemotherapeutics, but also immunotherapies such as CAR-Ts and antibody drug conjugates.

About KIYATEC Inc.

KIYATEC prioritizes accurate ex vivo prediction of patients' response to drug treatment, with a focus on data correlation to human clinical outcomes. The company creates and utilizes live phenotypic 3D cell-based models for drug response profiling and applies them to generate information relevant to preclinical testing, clinical trials and clinical diagnostics applications. By accurately predicting patient drug response without ever exposing actual patients to drugs, KIYATEC will create informed drug selection that minimizes clinical trials' failures and maximizes patient outcomes in the clinic. For more information, please visit www.kiyatec.com or follow KIYATEC on Twitter (@KIYATEC).