323

Generation of a functional precision medicine pipeline which combines comparative transcriptomics and tumor organoid modeling to identify bespoke treatment strategies for glioblastoma†

Megan R. Reed[1], A. Geoffrey Lyle[2], Annick De Loose[1], Katrina Learned[2], Cecile Rose T. Vibat[3], Christopher P. Wardell[1], Robert L. Eoff[1], Olena M. Vaske[2] and Analiz Rodriguez[1]
[1]University of Arkansas for Medical Sciences, [2]University of California Santa Cruz and [3]KIYATEC

OBJECTIVES/GOALS: A functional precision medicine platform to identify therapeutic targets for a glioblastoma patient with Li Fraumeni syndrome was performed. Comparative transcriptomics identified druggable targets and patient derived organoids and a 3D-PREDICT drug screening assay was used to validate the pipeline and identify further therapeutic targets. METHODS/STUDY POPULATION: A comparative transcriptomics pipeline was used to identify druggable genes that are uniquely overexpressed in our patient of interest relative to a cancer compendium of 12,747 tumor RNA sequencing datasets including 200 GBMs. Mini-ring patient derived organoid-based drug viability assays were performed to validate the comparative transcriptomics data. Additionally, a spheroid-based drug screening assay (3D-PREDICT) was performed and used to identify further therapeutic targets. RESULTS/ANTICIPATED RESULTS: Using comparative transcriptomics STAT1 and STAT2 were found to be significantly overexpressed in our patient, indicating ruxolitinib, a Janus kinase 1 and 2 inhibitor, as a potential therapy. Druggable pathways predicted using comparative transcriptomics corresponded with ruxolitinib sensitivity in a panel of patient derived organoids screened with this compound. Cells from the LFS patient were among the most sensitive to ruxolitinib compared to patient-derived cells with lower STAT1 and STAT2 expression levels. Additionally, 3D-PREDICT screening identified the mTOR inhibitor everolimus as a potential candidate. These two targeted therapies were selected for our patient and resulted in radiographic disease stability. DISCUSSION/SIGNIFICANCE: This research illustrates the use of comparative transcriptomics to identify druggable pathways irrespective of actionable DNA mutations present. Our results are promising and serve to highlight the importance of functional precision medicine in tailoring treatment regimes to specific patients.

324

The role of CCN3 in lung endothelial identity and function

Kalpana Betageri[1], Nunzia Caporarello[2] and Daniel Tschumperlin[2]
[1]Mayo Clinic Graduate School of Biomedical Sciences and [2]Mayo Clinic Rochester, MN, USA

OBJECTIVES/GOALS: Idiopathic Pulmonary Fibrosis (IPF) is a fatal disease of lung scarring. Aberrant vascular remodeling is a contributor to IPF progression. We have identified CCN3 as an endothelial gene that is upregulated in resolving but not in persistent lung fibrosis in mice. Here we tested the role of CCN3 in lung microvascular endothelial function. METHODS/STUDY POPULATION: RNAi loss of function experiments were used to evaluate the role of CCN3 in lung endothelial biology. Human lung microvascular endothelial cells (HLMECs) were transfected with human CCN3 siRNA and analyzed via qPCR to assess expression of endothelial transcripts, via wound healing assay for assessment of migratory function, and via 2D tube formation assay for assessment of angiogenic function. To ascertain the effect of HLMECs on Normal Human Lung Fibroblasts (NHLFs), conditioned media (CM) from endothelial cells with control and CCN3 siRNA was applied to TGFÎ² primed fibroblasts and qPCR was used to measure expression levels of pro-fibrotic transcripts. Recombinant human CCN3 protein was subsequently used to confirm the gain of function role of CCN3 in lung endothelial biology using a subset of these assays. RESULTS/ANTICIPATED RESULTS: CCN3 is a secreted matricellular protein thought to be involved in angiogenesis, cell adhesion, cell migration, and inflammatory responses in endothelial cells. In other organs, CCN3 suppresses expression of fellow matricellular protein, CCN2 (CTGF); importantly, CCN2 is a known pro-fibrotic mediator of aberrant tissue remodeling in the fibrotic lung. Upon CCN3 knockdown in HLMECs, we observed reduced transcripts for inflammatory and pro-fibrotic genes, along with impaired endothelial function in wound healing and angiogenesis assays. CM from CCN3 knockdown endothelial cells enhanced the pro-fibrotic effects of TGFÎ² in NHLFs. Addition of recombinant CCN3 to HLMECs generated, conditioned media that reduced fibroblast pro-fibrotic activation. DISCUSSION/SIGNIFICANCE: We have shown that matricellular protein-CCN3 plays a fundamental role in endothelial identity and function and could be a promising therapeutic target in IPF. A future goal is to restore levels of genes such as CCN3 in the aged vasculature in the setting of lung fibrosis to test their capacity to promote vascular repair and fibrosis resolution.

325

Blood pressure and the kidney cortex transcriptome response to high sodium diet challenge in female nonhuman primates

Angelica M. Riojas[1]; Kimberly D. Spradling-Reeves[2]; Robert E. Shade[3]; Sobha R. Puppala[2]; Clinton L. Christensen[4]; Shifra Birnbaum[4]; Jeremy P. Glenn[4]; Cun Li[5]; Hossam Shaltout[6]; Shannan Hall-Ursone[3] and *Laura A. Cox[2,3]
[1]Research Imaging Institute, UT Health San Antonio, San Antonio, Texas, USA, [2]Center for Precision Medicine, Department of Internal Medicine, Wake Forest School of Medicine, Winston-Salem, North Carolina, USA, [3]Southwest National Primate Research Center, Texas Biomedical Research Institute, San Antonio, Texas, USA, [4]Molecular Services Core, Texas Biomedical Research Institute, San Antonio, Texas, USA, [5]Department of Animal Science, University of Wyoming, Laramie, Wyoming, USA and [6]Hypertension and Vascular Research Center, Wake Forest School of Medicine, Winston-Salem, North Carolina, USA

OBJECTIVES/GOALS: The goal of this study was to understand the impact of a high sodium diet on gene networks in the kidney that correlate with blood pressure in female primates, and translating findings to women. METHODS/STUDY POPULATION: Sodium-naÃ¯ve female baboons (n=7) were fed a low-sodium (LS) diet for 6 weeks followed by a high sodium (HS) diet for 6 weeks. Sodium intake, serum 17 beta-estradiol, and ultrasound-guided kidney biopsies for RNA-Seq were collected at the end of each diet. Blood pressure was continuously measured for 64-hour periods throughout the study by implantable telemetry devices. Weighted gene coexpression network analysis was performed on RNA-Seq data to identify transcripts correlated with blood pressure on each diet. Network analysis was performed on transcripts highly

https://doi.org/10.1017/cts.2022.180 Published online by Cambridge University Press