

the survival of TP53 mutant cells that can be targeted in TNBC. We have identified Kif11 as one such target and aim to further investigate its function in TP53 mutant TNBC. **METHODS/STUDY POPULATION:** We conducted a dual in silico/in vivo screen that identified Kif11 inhibition as preferentially inhibiting the growth of TP53 mutant TNBC. We obtained data on TP53 mutational status, KIF11 mRNA expression levels, and clinical characteristics from TCGA, METABRIC, and CCLE datasets. We treated breast cancer cell lines with the KIF11 inhibitor SB-743921. Cell counts were obtained through staining with DAPI or Hoechst and imaging on the ImageXPRESS PICO. We detected cell death by DRAQ7 staining and flow cytometry analysis following Annexin V-PI staining. To investigate mitotic spindle organization, we performed immunofluorescent staining with an anti-tubulin antibody and DAPI co-staining. Cell cycle analysis was performed through flow cytometry. **RESULTS/ANTICIPATED RESULTS:** KIF11 is highly expressed in TP53 mutant and TNBC clinical samples. High KIF11 expression is associated with poorer clinical outcomes. Kif11 inhibition suppresses growth of both TP53 mutant and wild-type breast cancer cells, but preferentially induces the death of TP53 mutant cells as detected by DRAQ7 and Annexin V/PI staining. Kif11 inhibition induces a G2-M block and growth inhibition in TP53 wild-type cells. On the other hand, following treatment with the Kif11 inhibitor SB-743921, TP53 mutant cells undergo mitotic spindle dysfunction leading to the formation of multinucleated cells and cell death. **DISCUSSION/SIGNIFICANCE:** These results demonstrate that Kif11 is a promising therapeutic target in aggressive, TP53 mutant TNBCs. Kif11 inhibitors, including SB-743921, have been tested in human trials, and are well tolerated, but it is unclear which patients would most benefit. Our studies show that Kif11 inhibitors may be most useful in patients with TP53 mutant TNBCs.

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The role of leucine-rich PPR motif-containing protein (LRPPRC) in myelin lipid metabolism*

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OBJECTIVES/GOALS: Leigh Syndrome, French Canadian-Type (LSFC) is a neurometabolic disorder caused by mutation of mitochondria-related gene, LRPPRC. White matter lesions and demyelination in central nervous system are common in LSFC. LRPPRC is enriched in myelinating glial cells, yet its role is not known. Our goal is to elucidate its mechanistic role in myelination. **METHODS/STUDY POPULATION:** We crossed C57BL/6N mice bearing a LRPPRC-loxP allele with mice bearing a Plp-CreERT2 allele. Mice with the Plp-CreERT2 allele expresses a tamoxifen-inducible Cre under the control of the Plp promoter, which drives expression in oligodendrocytes. Using these strains, we can target the deletion of LRPPRC, via tamoxifen injection, in both newly formed myelin and mature myelin. Plp-CreERT2; LRPPRC^L/L (LRPPRC-KO) or control littermate mice will be injected for LRPPRC deletion at developmental and maturation stages of myelin. Immunofluorescence and electron microscopy of isolated brain tissues will be used for myelin integrity analysis. Cognitive functions of the mice will be measured via behavioral tests. Lastly, we will submit tissues for lipidomic analyses to observe any lipid metabolite variation. **RESULTS/ANTICIPATED RESULTS:** Behavioral and motor defects would be expected in LRPPRC-KO mice performing in cognitive function tasks across myelin maturation stages. Electron microscopy-based structure analysis of optic nerve, corpus

callosum, and spinal cord should reveal thin or loss of myelin on the axons of LRPPRC-KO compared to control. Immunofluorescence staining of major myelin structural proteins, including myelin proteolipid protein (PLP), myelin basic protein (MBP), and myelin-associated glycoprotein (MAG) would be expected have lower levels in LRPPRC deficient tissues. Since myelin is a lipid-rich species, we would also expect lipid concentrations to be affected. LRPPRC-KO lipidomic analyses of myelin-related lipids should depict lower levels in comparison to control, which would imply dysfunctional lipid metabolism. **DISCUSSION/SIGNIFICANCE:** There are limited studies in ameliorating neural deficits caused by LS and LSFC. Successful completion of this project would help elucidate the functions of LRPPRC in myelination and lipid metabolism and potentially provide insights for developing novel therapeutic strategies for alleviating the demyelination and neural deficits in LSFC.

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The Role of the IL-6-IGF-II Axis in Systemic Sclerosis-Associated Lung Fibrosis

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OBJECTIVES/GOALS: Interleukin (IL)-6 is produced in excess in Systemic Sclerosis (SSc). Likewise, microarray analysis of Insulin-like Growth Factor (IGF)-II-treated NL fibroblasts revealed increased expression of the basic helix-loop-helix transcription factor, BHLHB2. Our goal is to delineate the role of BHLHB2 in the fibrotic response to IGF-II and IL-6. **METHODS/STUDY POPULATION:** Primary lung fibroblasts were cultured from human lung tissues at 37°C and 5% CO₂. Cell cultures were stimulated with IL-6. Gene expression was measured using quantitative PCR (qPCR). IGF-II mRNA expression levels after IL-6 stimulation were compared with those of the housekeeping gene PPIB (Peptidylprolyl Isomerase B). Western blot was performed on nuclear and chromatin-bound subcellular fractions from treated lung fibroblasts. BHLHB2 protein levels were assayed in response to IGF-II in comparison to PBS as vehicle control. **RESULTS/ANTICIPATED RESULTS:** Results: Our results show that IL-6 increases IGF-II levels in fibroblasts. In turn, IGF-II increases BHLHB2 nuclear localization. We further show that IL-6 increases BHLHB2 levels and its nuclear localization in lung fibroblasts. Our findings are novel since the role of the transcription factor BHLHB2 in the IL-6 induced/IGF-II-mediated fibrotic response in SSc lung disease remains unexplored. **DISCUSSION/SIGNIFICANCE:** Our findings may provide a rationale for combination therapy to block IL-6 and IGF-II function concomitantly and thus halt the progression of SSc pulmonary fibrosis (PF). Our findings may have wide implications for lung fibrosis associated with various diseases, since SSc-PF, is characterized by the activation of common fibrotic pathways.

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A Pilot Study of Tear Cytokine Profiling in a Patient with Ocular Graft versus Host Disease

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OBJECTIVES/GOALS: Ocular graft versus host disease (oGVHD) affects ~50% of individuals after an allogeneic hematopoietic stem cell transplant for treating blood cancers. oGVHD results in severe