introduction and batch correction analyses by using external 293T cells as spike in controls during our single-cell and single-nucleus data generation steps to determine the most suitable method for batch-effect removal. Our analysis of human pLGGs at the single-cell and single-nucleus resolution provides critical insight into the heterogeneous biological activities that constitute these tumors.

**LGG-59. IDENTIFYING HIDDEN DRIVERS OF LOW-GRADE GLIOMA TUMOR GROWTH**

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Genomic drivers of pediatric low-grade gliomas (pLGGs) converge on alterations that activate the MAPK pathway. However, expression of individual driver oncogenes fails to induce tumor formation with high penetrance and, paradoxically, expression of these oncogenes suppresses growth in vivo. This is in agreement with the post-mortem tumor growth rate in patients, suggests that there are “hidden drivers” beyond a single driver oncogene that are necessary to support tumor growth. The goal of this project is to leverage high-throughput functional genomics strategies to identify these hidden drivers of pLGG tumor growth. Additionally, we hypothesize that secreted factors from the tumor microenvironment may drive pLGG tumor growth in part by modulating differentiation. In total, genes which cooperate with pLGG driver oncogenes to promote tumor growth may represent a new class of therapeutic targets and may explain the complex patterns of tumor growth that are observed in patients.

**LGG-60. DEVELOPMENT AND IMPLEMENTATION OF A COMPLEMENTARY DIAGNOSTIC RAPID STRATEGY TO DETECT TARGETABLE PATHWAYS IN PEDIATRIC GLIOMA PATIENTS**

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Pediatric Low grade gliomas are mainly driven by MAPK alterations including mutations in BRAF (BRAF fusions and BRAFV600) and FGFR. This has led to the study of BRAF, MEK and more recently FGFR inhibitors resulting in variable responses. We hypothesized that differing levels of MAPK, MAPK coupled with alternate pathway activation may be driving this variability. To address this, we designed a custom NanoString assay that integrates proteomic and transcriptomic profiling of 4 key cancer-relevant pathways (MAPK, PI3K-AKT-mTOR, JAK-STAT, and NFkB) with robust results on formalin-fixed paraffin embedded tissue, including archival samples up to approximately 15 years old. We validated this assay using 15 gold standard cell lines with defined changes in each pathway, including both isogenic activating mutations and perturbation with inhibitors. These findings were confirmed using data from the Cancer Cell Line Encyclopedia. The panel was further validated using a cohort of 40 low grade glioma samples with available RNAseq data where the RNA expression signatures had high concordance between assays. We have currently run the assay on over 200 surgical tumor samples, including 206 gliomas, 15 ependymomas, 11 medulloblastomas, 14 high grade gliomas and 10 control normal brain specimens. Findings indicate significant variability in pathway activations between tumors, although pLGG overall have higher MAPK activation scores than control tissue and other tumor types, a subset of these tumors have increased activity in PIK3CA, JAK and NFkB pathways, underscoring the importance of integrating transcriptomic and proteomic information in precision oncology treatments. Finally, single cell RNA sequencing data from pilocytic astrocytomas demonstrates significant heterogeneity in pathway activation states within the tumor cells, as well as high pathway activations in some tumor associated microglia. This raises further research questions regarding the role of tumor heterogeneity in treatment failures and the impact of targeted therapies on the tumor immune microenvironment.

**LGG-62. WEIGHT CHANGE IN PEDIATRIC PATIENTS TREATED WITH MEK INHIBITORS: A RETROSPECTIVE CHILDREN’S HOSPITAL LOS ANGELES EXPERIENCE**

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**BACKGROUND:** MEK inhibition is an emerging treatment strategy in pediatric tumors characterized by activation of the Ras-Raf-MEK-ERK pathway, including low-grade glioma (LGG) and neurofibromatosis 1 (NF1)-associated tumors. Preliminary clinical experience suggests that MEK inhibitors (MEKi) may be associated with weight gain in children, which has not been a reported toxicity in adults. **METHODS:** 35 patients > 1 and < 23 years old treated at CHLA with MEK inhibitors (MEKi) between October 2013 and May 2019 were identified. Data was collected at t = 0 (baseline), t = 3 months, t = 6 months, t = 12 months, and t = 24 months, as well as pre- and post-treatment time points. Weight change was categorized as no change (in Z-score > 0.25, +0.25), weight gain (change in Z-score > 0.25), and weight loss (change in Z-score < -0.25). Results: Weight gain > +0.25 was seen in 11 (34.4%) and 8 (25%) patients, respectively, after 6 months on therapy. Weight gain reversed in 4 out of 5 patients with post-treatment data. There was no clear association between weight outcome and hypothesized covariates (including hypothalamic location and NF1 status). Notably, significant weight gain was seen across baseline weight spectrum, including patients who had underweight and severely overweight BMI percentiles at baseline. **CONCLUSION:** Our findings preliminarily suggest that MEK inhibition may be associated with clinically significant weight change, especially weight gain, in a subset of children and young adults. Reversal upon drug cessation suggests a causal relationship. Further prospective and mechanistic investigation is needed.

**LGG-63 MEK INHIBITOR-ASSOCIATED RETINOPATHY (MEKAR) IN A PEDIATRIC PATIENT WITH AN OPTIC PATHWAY GLIOMA**

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Pediatric low-grade glioma (LGG) and plexiform neurofibroma (PN) universally have up-regulation of the Ras-mitogen-activated protein kinase (MAPK) pathway. Recent phase I and II clinical trials evaluating MEK inhibitors for the treatment pediatric LGG and PN report efficacy and tolerable side effects, including no reported ophthalmologic toxicity. Contrary to the pediatric experience, adult trials using MEK inhibitors describe severe
eral ophthalmalic side effects, including MEK inhibitor-associated retinopathy (MEKAR), also termed central serous-like retinopathy. MEKAR is defined as accumulation of subretinal fluid. It occurs in up to 90% of adults with Mlocked patients, usually occurring without symptoms and typically resolving without MEK inhibitor dose adjustment. We report a case of MEKAR in 15 year old boy with an optic pathway pilocytic astrocytoma with duplication of BRAF (7q34). Baseline ophthalmic exam showed 20/20 vision in his right eye with loss of the temporal hemifield and no light perception vision in the left eye. Nine months into treatment with Selumetinib his ophthalmologic exam and optical coherence tomography (OCT) showed asymptomatic bilateral subretinal fluid. Selumetinib was held for 2 weeks resulting in resolution of the subretinal fluid. Selumetinib was resumed at the prior dose and MEKAR recurred 2 weeks later but then permanently resolved 4 weeks later despite remaining on Selumetinib. Review of the literature discovered a single publication of 2 pediatric patients with optic pathway glioma who developed MEKAR around 6-7 months after initiating Selumetinib, which resolved after stopping Selumetinib. One patient was symptomatic and despite resolution of symptoms, Selumetinib was not resumed. The other patient was asymptomatic and resumed Selumetinib, but redeveloped MEKAR 8 months after restarting Selumetinib. Based on adult experience and the limited pediatric experience outlined above, we recommend pediatric patients with asymptomatic and mild-moderate symptomatic MEKAR undergo close monitoring without Selumetinib dose interruption or modification unless symptoms progress.

Abstracts

**MEDB-02. THE IDENTIFICATION AND FUNCTIONAL CHARACTERIZATION OF CIRCULAR RNA CIRC_63706 IN SONIC HEDGEHOG MEDULLOBLASTOMAS**
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Circular RNAs (circRNAs) are increasingly recognized as stable and specific biomarkers and therapeutic targets in many cancers, but little is known about their function, subtype specificity, and biomarker potential in medulloblastomas. Medulloblastoma is a central nervous system tumor that predominantly affects children and always requires aggressive therapy. Understanding and identifying novel disease-related molecular mechanisms and pathways are essential for developing optimal and novel therapies. To identify medulloblastoma subgroup-specific circRNAs, we subjected RNA-seq data from 173 clinical medulloblastoma samples representing the four subgroups to a statistical and machine learning (random forest classification) pipeline. Circular RNA circ_63706 expression was specific to the sonic hedgehog (SHH) group, which was confirmed through in situ hybridization analysis of clinical tissue samples. Functional characterization of circ_63706 by siRNAs and shRNAs demonstrated that cell proliferation, invasion, and apoptosis are perturbed in circ_63706 cells and inhibited in vivo tumor growth. These novel medulloblastoma-specific circRNAs are emerging as important oncogenes that not only provide valuable mechanistic insights into how medulloblastomas develop but also how they can be used as biomarkers and therapeutic targets. These results pave the way for the specific identification and personalized treatment of different medulloblastoma subgroups.

**MEDB-03. MEDULLOBLASTOMA CEREBROSPLINAL FLUID REVEALS HYPOXIC INDICATORS (METABOLITES AND LIPIDS) AND CANCER-SPECIFIC RNAs**
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Medulloblastoma (MB) is the most common malignant brain tumor in children. There remains an unmet need for diagnostics to sensitively detect the disease, particularly recurrences. Cerebrospinal fluid (CSF) provides a window into the central nervous system, and liquid biopsy of CSF could provide a less-invasive means for disease diagnosis. There has yet to be an integrated analysis of the transcriptomic, metabolomic, and lipidomic changes occurring in the CSF of children with MB. CSF samples from patients with (n=40) or without (n=11; no cancer) MB were subjected to metabolomics and lipidomics to identify differentially expressed markers of cancer. Our findings indicate that CSF from MB patients has distinct metabolomic and lipidomic profiles compared to those without cancer, with several potential biomarkers identified. These results suggest that cerebrospinal fluid could be a valuable diagnostic tool for medulloblastoma, offering potential for personalized medicine and improved patient outcomes.

**MEDB-04. MANAGEMENT OF THE RISK OF MEDULLOBLASTOMA**
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Medulloblastoma (MB) is a pediatric brain tumor that affects children aged 0-19 years, and it is the most common malignant brain tumor in children. Early detection and treatment are crucial for improving outcomes. The identification of high-risk features in patients with MB can help guide treatment decisions and improve patient outcomes. In this study, we assessed the risk of MB in a cohort of patients aged 0-19 years with acute lymphoblastic leukemia (ALL) or acute myeloid leukemia (AML). We found that patients with MB had a significantly higher risk of developing MB compared to the general population. Our findings highlight the importance of implementing screening protocols for MB in high-risk patients with ALL or AML, and the need for continued research to develop more effective treatment strategies for MB.

**MEDB-05. HEDGEHOG MEDULLOBLASTOMAS**
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Hedgehog (SHH) pathway activation is a frequent event in medulloblastoma, and it plays a critical role in tumor formation and progression. In this study, we investigated the impact of SHH pathway activation on medulloblastoma progression and survival. We found that SHH pathway activation is associated with poorer survival outcomes in medulloblastoma patients. Our findings suggest that targeting the SHH pathway may be a promising strategy for improving survival rates in medulloblastoma patients.