

multiplex PCR-based NGS panel was negative for alteration of the SMO, PTCH1 and CTBNN1 genes. Further molecular characterization via methylation profiling demonstrated the sonic hedgehog (SHH) molecular subtype. Prior to initiation of chemotherapy, renal ultrasound was performed and identified congenitally absent right kidney; audiology evaluation was unremarkable. Discussion: In patients with Gorlin syndrome, cases of unilateral renal agenesis in association with germline SHH-pathway mutations have been reported [1]. SHH signaling is implicated in multiple steps in the development of the urinary system [2]. Outside Gorlin syndrome, however, to the best of our knowledge unilateral renal agenesis coinciding with SHH-driven MB has not been reported. Our patient notably lacks any clinical stigmata of Gorlin syndrome (skeletal abnormalities, skin pits, macrocephaly) and does not exhibit the characteristic germline genetic abnormalities that define GS (PTCH1 mutation or 9q22.3 microdeletion). There are important treatment implications for patients with the constellation of abnormalities we describe here, particularly regarding the requisite frequent monitoring of renal function during multi-agent chemotherapy courses. Our patient tolerated chemoradiation well, and is currently on maintenance chemotherapy with favorable course to date.

#### EMBR-20. ELONGATION CONTROL OF MRNA TRANSLATION DRIVES GROUP 3 MEDULLOBLASTOMA

Alberto Delaidelli<sup>1</sup>, Gian Luca Negri<sup>1</sup>, Que Xi Wang<sup>1</sup>, Albert Huang<sup>1</sup>, Simran Sidhu<sup>1</sup>, Joyce Zhang<sup>1</sup>, Yue Zhou Huang<sup>1</sup>, Betty Yao<sup>1</sup>, Sofya Langman<sup>1</sup>, Andrii Vislovukh<sup>1</sup>, Volker Hovestadt<sup>2</sup>, Michael Taylor<sup>3</sup>, Gabriel Leprévier<sup>4</sup>, and Poul Sorensen<sup>1</sup>; <sup>1</sup>BC Cancer Research Centre, Vancouver, BC, Canada, <sup>2</sup>Massachusetts General Hospital, Boston, MA, USA, <sup>3</sup>Arthur and Sonia Labatt Brain Tumor Research Centre, Toronto, ON, Canada, <sup>4</sup>Heinrich Heine University, Duesseldorf, Germany

Medulloblastoma (MB) is the most common pediatric intracranial tumor and leading cause of childhood related cancer deaths. Group 3 affiliation and genetic amplifications of the MYC oncogene are predictors of adverse outcome in MB, underscoring a dire need for novel and more effective therapeutic approaches. The let-7 family of small non-coding RNAs (miRNAs) is known to inhibit tumor progression and regulate metabolism by targeting and degrading several cellular mRNAs, including MYC. Indeed, let-7 miRNAs are frequently repressed in several cancer types, including in MYC-driven MB. We previously reported that the mRNA translation elongation regulator eukaryotic Elongation Factor-2 Kinase (eEF2K) is a pivotal mediator of cancer cell adaptation to nutrient deprivation. In the current work, we identified a potential binding site for let-7 miRNAs on the eEF2K 3' untranslated region (UTR). In addition, eEF2K mRNA and let-7 miRNA expressions negatively correlate in MB, suggesting a potential regulation of the former by the latter. Let-7 miRNAs transfection decreases eEF2K mRNA and protein levels (by ~40–50%). Down-regulation of luciferase activity by let-7 miRNAs is impaired upon mutation of the let-7 binding site on the eEF2K 3'UTR. Inhibition of eEF2K significantly reduces survival of MYC-amplified MB cell lines under nutrient deprivation, altering their mRNA translation rates. Knockout of eEF2K increases survival of MYC-amplified MB xenografts when mice are kept under caloric restricted diets. We conclude that let-7 miRNAs degrade the eEF2K mRNA by binding to its 3'UTR, indicating that let-7 repression in MYC-driven MB is partially responsible for increased eEF2K levels. Moreover, the let-7-eEF2K axis constitutes a critical mechanism for MYC-driven MB adaptation to acute metabolic stress, representing a promising therapeutic target. Future therapeutic studies will aim to combine eEF2K inhibition with caloric restriction mimetic drugs, as eEF2K activity appears critical under metabolic stress conditions.

#### EMBR-21. CLINICALLY TRACTABLE OUTCOME PREDICTION OF GROUP 3/4 MEDULLOBLASTOMA BASED ON TPD52 IMMUNOHISTOCHEMISTRY: A MULTICOHORT STUDY

Alberto Delaidelli<sup>1</sup>, Christopher Dunham<sup>2</sup>, Maria Rita Santi<sup>3</sup>, Gian Luca Negri<sup>1</sup>, Joanna Triscott<sup>4</sup>, Olga Zheludkova<sup>5</sup>, Andrey Golanov<sup>6</sup>, Marina Ryzhova<sup>6</sup>, Konstantin Okonechnikov<sup>7</sup>, Daniel Schrimpf<sup>7</sup>, Damian Stichel<sup>7</sup>, Andreas von Deimling<sup>7</sup>, Marcel Kool<sup>7</sup>, Stefan Pfister<sup>7</sup>, Vijay Ramaswamy<sup>8</sup>, Andrey Korshunov<sup>7</sup>, Michael Taylor<sup>8</sup>, and Poul Sorensen<sup>1</sup>; <sup>1</sup>BC Cancer Research Centre, Vancouver, BC, Canada, <sup>2</sup>BC Children's Hospital, Vancouver, BC, Canada, <sup>3</sup>Children's Hospital of Philadelphia, Philadelphia, PA, USA, <sup>4</sup>University of Bern, Bern, Switzerland, <sup>5</sup>St. Luke's Clinical Research Center for Children, Moscow, Russian Federation, <sup>6</sup>Neurosurgical NN Burdenko Institute, Moscow, Russian Federation, <sup>7</sup>German Cancer Research Center (DKFZ), Heidelberg, Germany, <sup>8</sup>The Arthur and Sonia Labatt Brain Tumour Research Centre, Toronto, ON, Canada

Background: International consensus and the 2021 WHO classification recognize eight molecular subtypes among Group 3/4 medulloblastoma (representing ~60% of tumors). However, very few clinical centers worldwide possess the technical capabilities to determine DNA-methylation pat-

terns or other molecular parameters of high-risk for Group 3/4 tumors. As a result, biomarker-driven risk stratification and therapy assignment constitutes a major challenge in medulloblastoma research. Here, we identify an immunohistochemistry (IHC) marker as a clinically tractable method for improved medulloblastoma risk-stratification. Patients and Methods: We bioinformatically analyzed published medulloblastoma transcriptomes and proteomes identifying a potential biomarker TPD52, whose IHC prognostic value was validated across three Group 3/4 medulloblastoma clinical cohorts (n = 387) treated with conventional therapies. Risk stratification and prediction capability were computed utilizing uni- and multivariate survival analysis. Newly developed risk classifiers including TPD52 IHC were compared to state-of-the-art risk stratification schemes in terms of prediction error, area under the time-dependent receiver operating characteristic (ROC) curves and C-statistic. Biomarker-driven prognostic stratification models identified were cross validated in different cohorts. Results: TPD52 IHC positivity represents a significant independent predictor of early relapse and death for Group 3/4 medulloblastoma (HRs between 3.67–26.7 [95% CIs between 1.00–706.23], p = 0.05, 0.017 and 0.0058). Cross-validated survival models incorporating TPD52 IHC with clinical features outperformed existing disease risk-stratification schemes, and reclassified ~50% of patients into more appropriate risk categories. Finally, TPD52 immunopositivity is a predictive indicator of poor response to chemotherapy (HR 12.66 [95% CI 3.53–45.40], p < 0.0001), suggesting important implication for therapeutic choices. Conclusion: The current study redefines the approach to risk-stratification in Group 3/4 medulloblastoma. Integration of TPD52 IHC in classification algorithms significantly improves outcome prediction and can be rapidly adopted for risk stratification on a global scale, independently of advanced but technically challenging molecular profiling techniques.

#### EMBR-22. RATIONAL DEVELOPMENT OF SYNERGISTIC THERAPIES ALONGSIDE BMI1 INHIBITION FOR GROUP 3 MEDULLOBLASTOMA

David Bakhshinyan<sup>1</sup>, Ashley A Adile<sup>1</sup>, Chitra Venugopal<sup>1</sup>, Kevin Brown<sup>2</sup>, Katherine Chan<sup>2</sup>, Maleeha A Qazi<sup>1</sup>, Chirayu Chokshi<sup>1</sup>, William D Gwynne<sup>1</sup>, David Tieu<sup>2</sup>, Jason Moffat<sup>2</sup>, and Sheila Singh<sup>1</sup>; <sup>1</sup>McMaster University, Hamilton, ON, Canada, <sup>2</sup>University of Toronto, Toronto, ON, Canada

Medulloblastoma (MB) is the most common pediatric brain tumor. Of its four distinct molecular subgroups, Group 3 MBs are associated with increased risk of recurrence, metastasis and overall poor patient outcome. In recent years, small molecule inhibitors targeting BMI1 have shown to be efficacious against several types of malignant tumors including pediatric MB. Although *in vivo* studies provide a promising proof-of-concept for the therapeutic targeting of BMI1 in Group 3 MB, mice that receive treatment eventually succumb to their disease. These results suggest that additional mechanisms may underlie the maintenance of MB and underscores the main obstacle in treating a constantly evolving tumor. After initial preclinical validation of BMI1 inhibitor PTC-596, DNA barcoding clonal tracking technology was leveraged to profile *in vivo* clonal dynamics of Group 3 MB in response to the established chemoradiotherapy regimen alone and in combination with PTC-596. Comparison of clonal composition of the tumors extracted from the brains and spines post-treatment revealed the persistence of a small number of clones with the ability to escape therapy and drive subsequent tumor expansion. In order to better understand molecular susceptibilities of MB cells post BMI1 inhibition, we undertook an *in vitro* genome-wide CRISPR/Cas9 screening to identify context-specific MB regulatory pathways to be synergistically targeted along with BMI1. By comparing the results of the *in vitro* genome wide CRISPR/Cas9 screen to the essential genes in human neural stem cells (hNSCs), we identified several context specific regulators of mTOR, AKT and PLK1 pathways. The combined treatment alongside PTC-596 has demonstrated synergistic efficacy against MB cells with minimal toxicity to hNSCs *in vitro* and is currently being evaluated in preclinical studies. This study provides the foundation for clinical validation of small-molecule inhibitors synergistic with PTC-596 to improve the durability of remissions and extend survival of patients with treatment-refractory Group 3 MB.

#### EMBR-23. KIF11 DEPENDENCY ON P53 MUTATIONAL STATUS IN MEDULLOBLASTOMA

Shiyang Huang<sup>1,2</sup>, Sekar Karthik<sup>1</sup>, Qi Lin<sup>3</sup>, YuChen Du<sup>3</sup>, Ching C Lau<sup>4</sup>, Adesina Adekunle<sup>5</sup>, Jack MF Su<sup>4,6</sup>, Angela Major<sup>7</sup>, M. Tarek Elghetany<sup>7,6</sup>, Kam-Man Hui<sup>1</sup>, Xiaonan Li<sup>3,8</sup>, and Wan-Yee Teo<sup>9,10,11,12,13</sup>; <sup>1</sup>Humphrey Oei Institute of Cancer Research, National Cancer Center Singapore, Singapore, Singapore, <sup>2</sup>Pediatric Brain Tumor Research Office, SingHealth-Duke-NUS Academic Medical Center, Singapore, Singapore, Singapore, <sup>3</sup>Ann & Robert H. Lurie Children's Hospital of Chicago, Chicago, IL, USA, <sup>4</sup>Department of Pediatrics, Division of Hematology-Oncology, Texas