have furthermore identified alterations in more than 100 IncRNAs in DIPG. METHODS: To identify IncRNAs required for proliferation of patient-derived DIPG cancer cells, we designed two custom genome-scale IncRNA libraries, a genome-wide CRISPR-Cas9 knockout library consisting of 45,766 single guide RNAs (sgRNAs). Additionally, we generated a genome-wide CRISPR interference pooled library consisting of 43,608 sgRNAs targeting IncRNA transcription start sites (TSS), RNaseI-resistant RNAs in vitro bioassays in cell lines revealing IncRNA dependencies. Candidate dependencies in our CRISPR-Cas9 knockout screen include LOC100507412, LOC105379524, and LINCC02193. CONCLUSION: Genome-wide IncRNA CRISPR knock-out and CRISPR interference screens are a novel approach for the unbiased identification of IncRNAs that are required for pediatric high-grade glioma proliferation. Further validation of specific IncRNAs is required, and these IncRNA dependencies represent potential novel therapeutic targets.

HGG-39. INTEGRATIVE ANALYSES OF BRAFV600E MUTATED GLIOMAS: FROM GENETIC TO METABOLIC CHARACTERISTICS
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BRAF p.v600e mutation is encountered in brain tumors, mostly low grade pediatric diffuse gliomas (LGGs) and glioblastomas (GBMs) such as gangliogliomas (GG) or pleomorphic xanthoastrocytomas (PXA). Less frequent, this mutation is present in high grade glial or glioneuronal tumors such as pleomorphic xanthoastrocytomas with anaplasia, anaplastic ganglioglioma, anaplastic diffuse astrocytoma or glioblastoma. Recently, few publications were highlighting differently the impact of BRAF mutation and CDKN2A deletion, as independent prognostic factors linked to a worst outcome in low grade forms. We studied retrospectively a monocentric cohort of 17 LGGs (14 GBMs) and 7 BRAF p.v600e HGG. The patients were aged below 20 years. We focused on extended tumors’ biology assessment (MethyEpic 850K, Next-Generation sequencing, RNA sequencing and metabolomics), as well as tumor immune phenotype and microenvironment. Among the LGGs, only one had a CDKN2A deletion and one a gain on chromosome 5. All except two LGGs had a complete surgical resection. Four of them were treated by chemotherapies but underwent relapses. All HGGs had a surgical resection followed by a first line chemotherapy (Lomustine, 5-fluoracil and radiotherapy. Five patients relapsed rapidly, benefiting from targeted therapy with vemurafenib and/or biotherapy associating dabrafenib plus trametinib. Among those HGGs, we had both subgroups: “de novo” tumors and patients with a history of LGG tumors. Both were responding well to targeted treatments. The biology uncovers in all HGGs a loss of CDKN2A gene and/ or protein. Additionally to this gene abnormality, specific transcriptomic expressions were associated to therapeutic response and immune microenvironment. Epigenetic modulation was linked to specific metabolic switches when BRAF p.v600e gliomas were getting higher grade features (e.g., glutaminolysis, serinolysis and phospholipidic metabolism). Those characteristics seem to be able to predict in LGG p.v600e potential evolution.

HGG-40. NF1 MOSAICISM IN A CMRRD-PATIENT WITH A GLOBLASTOMA
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Constitutional Mismatch Repair Deficiency (CMMRD) and Neurofibromatosis type 1 (NF1) are brain tumor predisposing syndromes associated with cafe-au-lait macules (CALM). Due to this overlap, establishing the differential diagnosis can be difficult, but remains crucial as treatments and surveillance protocols clearly differ for affected patient according to the right hemisphere (temporal-frontal-parietal lobes). No specific enhancing lesion or restricted diffusion was noted. Histologically, the tissue had findings of diffuse low-grade astrocytoma. Molecular genetic testing was notable for fusions of ETV6 and NTRK3, a genotype concordant with a type high-grade astrocytic tumors. MGMT promoter methylation array was negative. Final diagnosis was glioblastoma, IDH and H3 wildtype. All the molecular features of this tumor were high-grade with an average progression of 2.5 years post-diagnosis. Targeted agents were not used in upfront treatment for this patient. This is a rare case of stabilization of disease with conventional treatment in a very aggressive tumor. This case is a rare constellation of an adult-type molecular profile in a pediatric patient with a low grade, less aggressive behavior profile, and highlights the need to better understand this rare subtype of tumors in children.
Preliminary results revealed a spectrum of genetic alterations in tumor aggressiveness suggesting that PDHGG are a diverse group of childhood brain tumours comprising multiple subgroups carrying distinct molecular drivers. Patient-derived models accurately recapitulating this underlying biology are critical for mechanistic/preclinical studies aimed at improving patient outcome, however their behaviour over time in the environments in which they are propagated, and how this relates to the human disease, is largely unknown. To explore this, we collected 94 models of PDHGG established as 2D/3D stem cell cultures in vitro, and generated patient-derived xenografts (PDX) in 38/62 specimens implanted orthotopically in vivo. We carried out exome/targeted sequencing, mRNA profiling and RNAseq to profile cultures through their first 25 passages in culture, and sequential implantation from p0-p2 in mice. In 15/33 cultures, we observed enrichment of gene expression signatures of non-malignant cells over the first 3 passages, with concurrent depletion of somatic mutations/ CNAs, excluding them from further study. The best strategy for de novo induction of GBM serves as a strong argument for a combination of MEK-inhibitors with immunotherapy.