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Molecular Case Studies

RAPID CANCER COMMUNICATION

A germline *PALB2* pathogenic variant identified in a pediatric high-grade glioma

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Abstract *PALB2* (partner and localizer of BRCA2) gene encodes a protein that colocalizes with *BRCA2* in nuclear foci and likely permits the stable intranuclear localization and accumulation of *BRCA2*. *PALB2* plays a critical role in maintaining genome integrity through its role in the Fanconi anemia and homologous recombination DNA repair pathways. It has a known loss-of-function disease mechanism. Biallelic *PALB2* pathogenic variants have been described in autosomal recessive Fanconi anemia. Heterozygous pathogenic variants in *PALB2* are associated with increased risk for female and male breast cancer and pancreatic cancer (*Science* 324: 217; *Cancer Res* 71: 2222–2229; *N Engl J Med* 371: 497–506). Heterozygous germline *PALB2* mutations have also been observed in patients with medulloblastoma (*Lancet Oncol* 19: 785–798). However, *PALB2*-related cancer predisposition to high-grade gliomas has not been reported. Here we report a germline *PALB2* pathogenic variant (c.509_510delGA, p.Arg170Ilefs*14, NM_024675.3) found in a pediatric patient with high-grade glioma. This variant was first identified by tumor sequencing using the Children's Hospital of Philadelphia (CHOP) Comprehensive Solid Tumor Panel and then confirmed to be a germline change using the CHOP Comprehensive Hereditary Cancer Panel on DNA from a blood sample of this patient. Parental studies showed that this variant was paternally inherited. Further studies are needed to illustrate if pathogenic variants in *PALB2* convey increased risk to developing brain tumor. This case also highlights the potential of identifying germline mutation through tumor sequencing.

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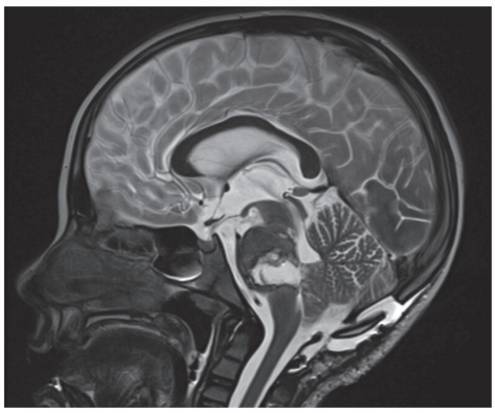
CASE PRESENTATION

The patient was a 7-yr-old female who was diagnosed with anaplastic astrocytoma, isocitrate dehydrogenase (IDH)-wild type, World Health Organization (WHO) grade III involving brainstem, thalamus, cerebellum, and cervical spinal cord (Fig. 1A). The patient's family history was notable for a glioblastoma in maternal grandfather, lung cancer in maternal great grandfather, and gastrointestinal cancers in multiple paternal fourth-degree relatives.

A



B

Chromosome	Abnormality	Band	Genes
X	LOSS	whole chromosome	
2	AMP	partial 2p	*MYCN*
9	LOSS	9p	*CDKN2A*
9	LOSS	partial 9q	*GNAQ, NTRK2, FANCC, PTCH1* (homozygous loss of exons 3-18)
13	LOSS	partial 13q	*RB1* (Homozygous)
17	LOSS	17p	*TP53*
	LOSS	partial 17q	*NF1...PPM1D*
	cnLOH	partial 17q	*BRIP...RPTOR*
19	LOSS	whole chromosome	
21	GAIN	21q	*RUNX1* AMP

Figure 1. (*A*) T2-weighted imaging sagittal plane image showing pontine mass with evidence of intratumoral hemorrhage. (*B*) Copy-number variations (CNVs) identified in the tumor.

TECHNICAL ANALYSIS

Next-Generation Sequencing (NGS)

Two-hundred and thirty-eight genes associated with pediatric solid tumors were selected for the Children's Hospital of Philadelphia (CHOP) Comprehensive Solid Tumor Panel (CSTP) (Surrey et al. 2019). Custom DNA probes were designed using SureDesign (Agilent Technologies) to cover all exons, at least 10 bp of intronic sequences at exon/intron boundaries, and selected known intronic mutations. The CHOP Comprehensive Hereditary Cancer Panel (CHCP) interrogates 130 genes associated with cancer predisposition and covers all coding exons, at least 20 bp of intronic sequences at exon/intron boundaries, and known intronic mutations. Additional common single-nucleotide polymorphisms (SNPs) were added to both CSTP and CHCP to mimic a low-density SNP array for copy-number variation (CNV) analysis (Surrey et al. 2019). All custom DNA probes were synthesized and biotinylated to allow for target enrichment using streptavidin-conjugated beads (Agilent Technologies). For target RNA sequencing, 110 major fusion partner genes associated with cancer-related fusions were selected for the CHOP fusion panel (Chang et al. 2019). Multiplex polymerase chain reaction (PCR) technology, powered by unidirectional gene-specific primers, sample indexes, and molecular barcodes for multiplex targeted RNA sequencing (ArcherDX, Inc.), were used. Target-specific primers covering 673 exons were custom-designed to identify known fusions and potential novel fusions associated with 110 cancer genes.

Multiple clinically significant alterations were detected in the tumor, including *TP53* (c.724T > A, p.Cys242Ser), *NF1* (c.6854dup, p.Tyr2285*), *PALB2* (c.509_510delGA, p.Arg170Ilefs*14) (Table 1), *MYCN* amplification, and multiple other CNVs including loss of Chromosome 17 (Fig. 1B). No known or novel fusion genes were detected. The variant allele fractions (VAFs) were 0.75, 0.79, and 0.43 for the *TP53*, *NF1*, and *PALB2* variants, respectively. Further NGS studies using the CHCP on DNA from a blood sample of this patient was performed and identified the same *PALB2* c.509_510delGA pathogenic variant, demonstrating a germline change, along with pre- and post-testing genetic counseling. No other somatically identified variant was detected in the blood. Targeted sequencing of parental samples showed that this variant was paternally inherited.

Table 1. Sequence variants identified in the tumor and blood

Gene	Chr	HGVS DNA ref (if genic)	HGVS protein ref	Variant type	Predicted effect	Allele frequency	Target coverage	Present in	Germline/ somatic
PALB2	16	c.509_510delGA	p.Arg170Ilefs*14	Deletion	Frameshift	43%	2303×	Tumor/blood	Germline
TP53	17	c.724T>A	p.Cys242Ser	SNV	Deleterious	75%	842×	Tumor	Somatic
NF1	17	c.6854dup	p.Tyr2285*	Duplication	Nonsense	79%	220×	Tumor	Somatic

(HGVS) Human Genome Variation Society, (SNV) single-nucleotide variant.

VARIANT INTERPRETATION

The *PALB2* c.509_510delGA variant creates a frameshift starting at codon Arg170 in exon 4, which results in a premature stop codon 14 amino acids downstream. The variant is observed in the genome Aggregation Database (gnomAD) with an allele frequency of 0.003579% (9/251446, 0 homozygotes) and it has been identified as a germline variant in patients with ovarian cancer, breast cancer, and pancreatic ductal adenocarcinoma (Dansonka-Mieszkowska et al. 2010; Noskowicz et al. 2014; Borecka et al. 2016). The variant is reported in ClinVar (Variation ID 126757) and listed as pathogenic by 15 submissions. It is not reported in the Catalogue of Somatic Mutations in Cancer (COSMIC).

SUMMARY

A germline *PALB2* pathogenic variant (c.509_510delGA) was confirmed in a patient with a pediatric high-grade glioma after finding the variant in somatic tumor sequencing. Although it is not clear at the present time if the *PALB2* c.509_510delGA variant is associated with the brain tumor development in this patient, heterozygous germline *BRCA1* or *BRCA2* pathogenic variants have been reported in patients with brain tumors (Wilson et al. 2010; Shoua et al. 2018). Additionally, two somatic *PALB2* variants have been previously observed in high-grade gliomas (Mackay et al. 2017). Further functional studies are needed to explore if *PALB2* pathogenic variants predispose mutation carriers to central nervous system (CNS) tumors.

ADDITIONAL INFORMATION

Data Deposition and Access

The interpreted variant has been deposited in ClinVar (https://www.ncbi.nlm.nih.gov/clinvar/) under accession number VCV000126757.11. Other variants identified in the 238 gene in the CHOP Comprehensive Solid Tumor Panel and 130 genes in the Comprehensive Hereditary Cancer Panel are reported in the body of the manuscript. The patient did not provide consent for public deposition of all raw sequencing data.

Ethics Statement

A case report does not constitute human subjects research. Per CHOP policy, it therefore does not require IRB review.

Author Contributions

M.M.L. and Y.Z. designed the study. M.M.L., Y.Z., J.S., J.W., F.X., F.L., K.C., K.Z., M.L., and S.P.M. collected and analyzed the data. M.M.L., Y.Z., J.S., J.W., and F.X. wrote the manuscript. J.B.F., K.A.C., and P.B.S. provided clinical data. M.M.L., Y.Z., J.S., J.W., F.X., F.L., K.C., K.Z., M.L., J.B.F., K.A.C., S.P.M., A.C.R., and P.B.S. reviewed the manuscript.

Competing Interest Statement

The authors have declared no competing interest.

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