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Pediatric Oncology

Targetable *BRAF* and *RAF1* Alterations in Advanced Pediatric Cancers

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Abstract

RAF family protein kinases signal through the MAPK pathway to orchestrate cellular proliferation, survival, and transformation. Identifying *BRAF* alterations in pediatric cancers is critically important as therapeutic agents targeting BRAF or MEK may be incorporated into the clinical management of these patients. In this study, we performed comprehensive genomic profiling on 3,633 pediatric cancer samples and identified a cohort of 221 (6.1%) cases with known or novel alterations in *BRAF* or *RAF1* detected in extracranial solid tumors, brain tumors, or hematological malignancies. Eighty percent (176/221) of these tumors had a known-activating short variant (98, 55.7%), fusion (72, 40.9%), or insertion/deletion (6, 3.4%). Among *BRAF* altered cancers, the most common tumor types were brain tumors (74.4%), solid tumors (10.8%), hematological malignancies (9.1%), sarcomas (3.4%), and extracranial embryonal tumors (2.3%). *RAF1* fusions containing intact RAF1 kinase domain (encoded by exons 10–17) were identified in seven tumors, including two novel fusions *TMF1-RAF1* and *SOX6-RAF1*. Additionally, we highlight a subset of patients with brain tumor with positive clinical response to BRAF inhibitors, demonstrating the rationale for incorporating precision medicine into pediatric oncology. ***The Oncologist*** 2021;26: e153–e163

Implications for Practice: Precision medicine has not yet gained a strong foothold in pediatric cancers. This study describes the landscape of *BRAF* and *RAF1* genomic alterations across a diverse spectrum of pediatric cancers, primarily brain tumors, but also encompassing melanoma, sarcoma, several types of hematologic malignancy, and others. Given the availability of multiple U.S. Food and Drug Administration-approved BRAF inhibitors, identification of these alterations may assist with treatment decision making, as described here in three cases of pediatric cancer.

Introduction

Pediatric cancers are a leading cause of death in the U.S. among children aged 1 to 14 years [1]. Despite significant improvements in 5-year overall survival for this population, outcomes vary considerably depending on cancer type, with cure rates not exceeding 20% in patients with recurrent disease [2, 3].

Precision medicine, defined as biomarker-informed treatment, accounts for significant advances in management of patients with cancer during the past two decades, including trastuzumab for HER2-positive breast cancer [4], imatinib for BCR-ABL–driven chronic myeloid leukemia [5], crizotinib targeting *ALK*-rearranged non-small cell lung

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cancer [6], and *BRAF* V600E–targeting agents in melanoma [7, 8].

With certain exceptions, such as the use of tyrosine kinase inhibitors in Philadelphia chromosome–positive acute lymphoblastic leukemia, the targeted therapy paradigm has not been fully realized for pediatric patients with cancer. Improvements in cytotoxic chemotherapy and radiation therapy techniques have dramatically improved survival rates in many pediatric cancers over the past 50 years; however, certain tumor types continue to be resistant to standard therapeutic approaches [9], and when therapy is effective, long-term toxicities in survivors remain problematic [10–12].

Comprehensive genomic profiling (CGP) with next-generation sequencing is an effective tool for identifying clinically relevant genomic alterations (GAs) across diverse types of pediatric cancers, including low grade glioma (LGG) and high grade glioma (HGG) [13–15], osteosarcoma [16], neuroblastoma [17], medulloblastoma [18], thyroid carcinoma [19], acute myeloid leukemia (AML) [20], T-lineage acute lymphoblastic leukemia [21], gonadal tumors [22], and histiocytic neoplasms [23], with implications for more precise diagnoses, prognoses, and personalized therapeutic decision making.

BRAF encodes a member of the RAF family of protein kinases, which includes ARAF, BRAF, and CRAF (RAF1). These kinases function downstream of RAS as part of the MAPK (RAF-MEK-ERK) signaling cascade that facilitates cell proliferation, survival, and transformation [24, 25]. *BRAF* mutations have been reported in up to 20% of all cancers, with a majority occurring at the V600 position [26, 27]. *BRAF* fusions, which activate the MAPK pathway, have been reported in multiple tumor types [28] and are the most common genomic alteration in juvenile pilocytic astrocytoma (PA), a type of LGG [29]. *RAF1* fusions, which are functionally similar to *BRAF* fusions, are recurrent in adult solid tumors [30–32] and juvenile PA [15, 33–35]. Among LGGs, *BRAF* V600E may predict poorer long-term outcome after chemotherapy and radiation therapies compared with non–*BRAF* V600E tumors and those harboring *BRAF* fusions (*KIAA1549-BRAF*), although further study is needed [36–38]. Furthermore, *BRAF* V600E has been observed concurrent with *CDKN2A* loss in patients with ganglioglioma, although no significant difference in prognosis was identified compared with patients with *BRAF* V600E and intact *CDKN2A* [39].

Therapeutic strategies targeting BRAF-driven tumors rely mostly on U.S. Food and Drug Administration (FDA)-approved small molecule tyrosine kinase inhibitors (e.g., dabrafenib), approved in metastatic melanoma and non-small cell lung cancer, and vemurafenib, approved in metastatic melanoma and Erdheim-Chester disease. These and additional investigational *BRAF* V600E–targeting agents [40, 41] are under clinical evaluation for pediatric indications in multiple early phase trials. *BRAF*-altered pediatric malignancies have derived clinical benefit from *BRAF* V600E–targeting agents in central nervous system disease [2, 42–49] and histiocytic neoplasms [50, 51]. Targeting BRAF fusions remains a challenge in pediatric brain tumors, although reports demonstrating clinical benefit with MEK inhibitors are increasing [52–56]. Therapeutic modalities targeting *RAF1* fusion–positive tumors are even rarer, with no clinical studies (pediatric or adult) available. However, three reports demonstrated clinical benefit from trametinib in two adult patients with melanoma [30, 57] and a pediatric patient with LGG, respectively [58].

Despite gains in clinical success of biomarker-informed targeted therapy in children with cancer, access to relevant targeted therapy is limited [2, 5–38, 40–54, 58–61]. In this study, we sequenced tumors from 3,633 patients with pediatric cancer and identified a cohort of 221 cases with known and novel *BRAF* or *RAF1* alterations in extracranial solid tumors, brain tumors, or hematological malignancies. Furthermore, we highlight a subset of patients with brain tumors with positive clinical response to BRAF inhibitors, demonstrating the rationale for incorporating precision medicine into pediatric oncology.

Materials and Methods

CGP was performed in a Clinical Laboratory Improvement Amendments–certified, College of American Pathologists–accredited laboratory (Foundation Medicine, Inc., Cambridge, MA). Approval for this study, including a waiver of informed consent and a Health Insurance Portability and Accountability Act waiver of authorization, was obtained from the Western Institutional Review Board (protocol no. 20152817). The pathologic diagnosis of each case was confirmed on routine hematoxylin and eosin–stained slides, and all samples forwarded for DNA extraction contained a minimum of 20% tumor nuclear area as a proportion of all nucleated cells. In brief, ≥50 ng DNA was extracted from 40 microns of specimen from formalin-fixed, paraffin-embedded tissue blocks or unstained slides. The samples were assayed by CGP using adaptor ligation, and hybrid capture was performed for all coding exons from 287 (version 1) to 315 (version 2) cancer-related genes plus select introns from 19 (version 1) to 28 (version 2) genes frequently rearranged in cancer. Sequencing of captured libraries was performed using the Illumina HiSeq technology (Illumina, San Diego, CA) to a median exon coverage depth of at least 500× and analyzed for GAs, including short variant alterations (base substitutions, insertions, and deletions), copy number alterations (focal amplifications and homozygous deletions), and select gene fusions or rearrangements, as previously described [62]. Benign germline variants documented in publicly accessible population databases or recurrent variants of unknown significance that were predicted by an internally developed algorithm to be germline were removed, with the exception of known driver germline events (e.g., documented hereditary and deleterious *BRCA1/2* or *TP53* mutations) [63]. Somatic alterations present in the Catalog of Somatic Mutations in Cancer were highlighted as biologically significant [64]. Tumor mutational burden (TMB) was determined on 1.1 megabases of sequenced DNA for each case and categorized as low (0–5 mutations per megabase [mut/Mb]), intermediate (6–19 mut/Mb), or high (≥20 mut/Mb) as previously described [65]. Clinical histories, disease stage, and primary versus recurrent disease status of samples tested were not available.

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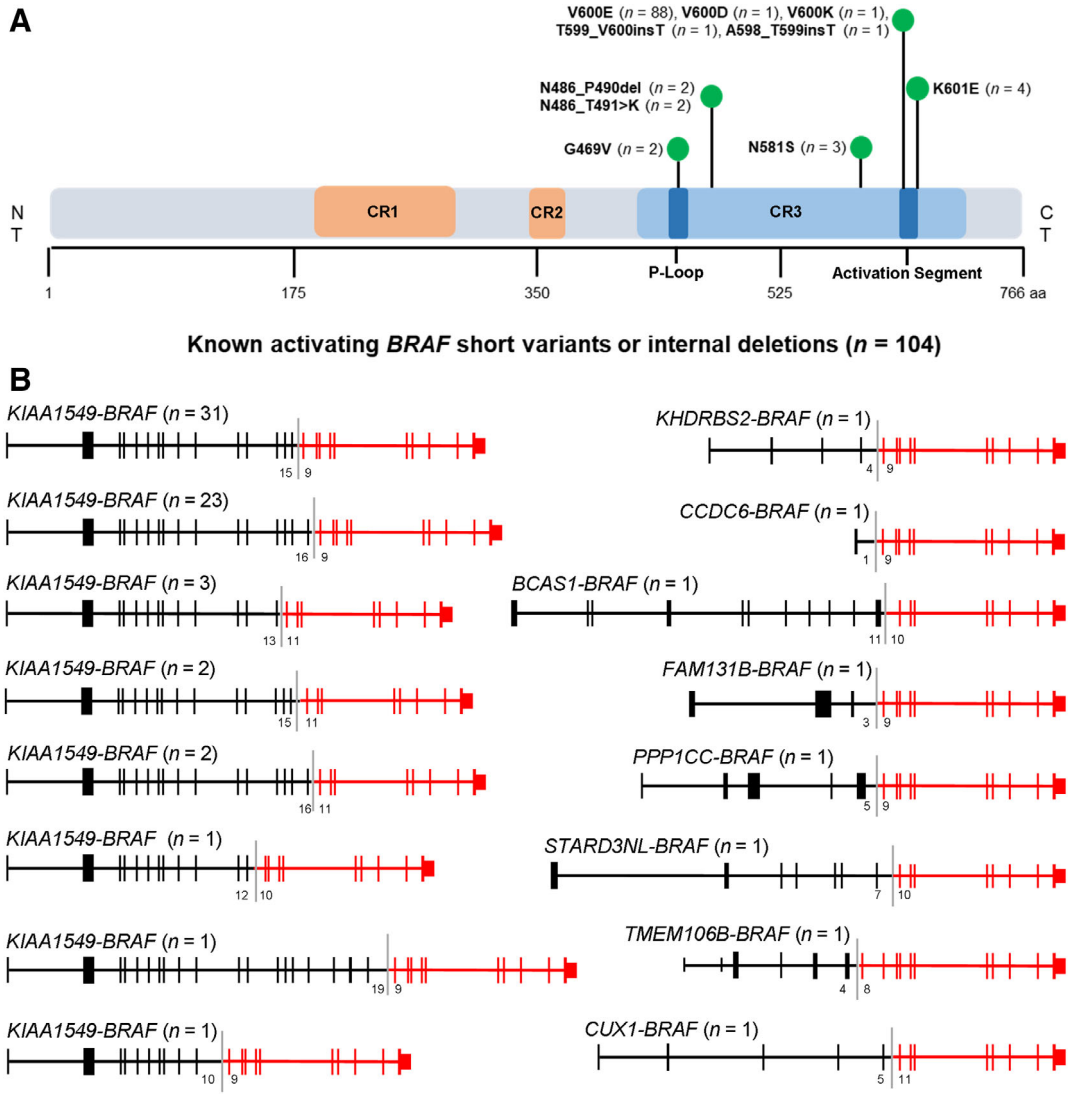

Figure 1. Landscape of known-activating *BRAF* alterations. **(A):** Schematic diagram of domains and alterations of *BRAF*. **(B):** Schematic of known-activating *BRAF* fusions, including those identified in this study (*STARD3NL-BRAF* and *KHDRBS2-BRAF*). Exon numbers at the fusion boundary are depicted below each fusion diagram.
Abbreviation: CR, conserved region.

Results

Characteristics of the Pediatric Cohort

CGP was performed on 3,633 pediatric (median 10.5 years, range < 1–21 years) cancer samples and revealed 221 (6.1%) unique samples that harbored alterations in *BRAF*. Alterations were classified as "known-activating" or "functionally impairing" if supported by publicly available, peer-reviewed biochemical data. An alteration was classified as "uncharacterized" if biochemical data supporting a specific functional status were not available at the time of this study's publication.

Of the *BRAF* mutation–positive cohort, 176 (79.6%) samples harbored a known-activating short variants (SVs), insertions/deletions (indels), or fusion; 34 (15.4%) harbored an uncharacterized SV, indel, or nonfusion rearrangement; 8 (3.6%) harbored a SV known to result in decreased protein function (i.e., functionally impairing); and 3 (1.4%) contained multiple uncharacterized or functionally impairing SVs (supplemental online Fig. 1A). Of the 176 samples bearing a *BRAF* known-activating alteration, 98 (55.7%) encompassed SVs, 72 (40.9%) fusions, and 6 (3.4%) indels (supplemental online Fig. 1B).

Known-activating *BRAF* alterations were identified in samples encompassing six primary histological categories: brain tumors (74.4%; 18 subtypes), other solid tumors (10.8%; 6 subtypes), hematological malignancies (9.1%; 5 subtypes), sarcomas (3.4%; 3 subtypes), and extracranial embryonal tumors (2.3% 2 subtypes) (supplemental online Fig. 2A). Brain tumors (n = 131) included pilocytic astrocytoma (PA), grade I (45; 34.4%); low grade glioma (LGG) not otherwise specified (NOS) (19; 14.5%); glioblastoma (GBM) (13; 9.9%); pilomyxoid astrocytoma, grade 2 (13; 9.9%); ganglioglioma, grade 1 (10; 7.6%); and 13 additional subtypes of varying frequency (supplemental online Fig. 2B).

Sarcomas (n = 6) included rhabdomyosarcoma (NOS) (2; 33.3%); sarcoma (NOS) (3; 50%); and rhabdomyosarcoma, embryonal (1; 16.7%) (supplemental online Fig. 2C).

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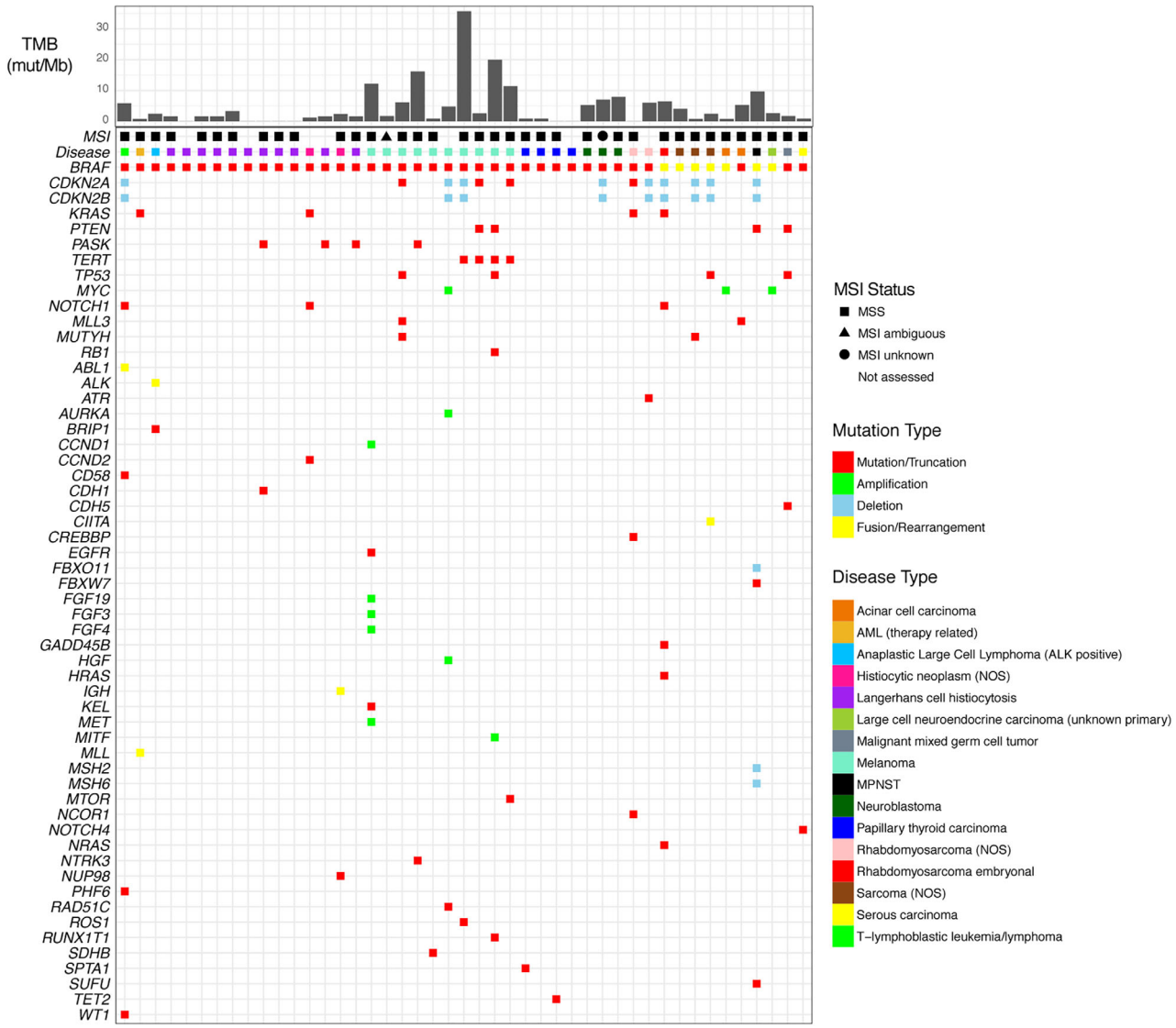

Figure 2. Genomic landscape of hematological malignancies and extracranial solid tumors with a known-activating *BRAF* alteration. Specimens are arranged from young to old (left to right) within each cancer type.
Abbreviations: AML, acute myeloid leukemia; MPNST, malignant peripheral nerve sheath tumor; MSI, microsatellite instability; MSS, microsatellite stable; mut/Mb, mutations per megabase; NOS, not otherwise specified; TMB, tumor mutational burden.

Embryonal tumors (n = 4) included neuroblastoma (3; 75%) and malignant mixed germ cell tumor (1; 25%) (supplemental online Fig. 2D). Hematological tumors (n = 16) included Langerhans cell histiocytosis (11; 68.8%), histiocytic cell neoplasm (NOS) (2; 12.5%), anaplastic large cell lymphoma (ALK-positive) (1; 6.3%), AML (treatment-related) (1; 6.3%), and T-lymphoblastic leukemia/lymphoma (1; 6.3%) (supplemental online Fig. 2E). Other solid tumors (n = 19) included melanoma (10; 52.6%), papillary thyroid carcinoma (4; 21.1%), acinar cell carcinoma (2; 10.5%), and three additional subtypes (supplemental online Fig. 2F).

Landscape of *BRAF* Known-Activating Variants

BRAF V600E accounted for 50% (88/176) of all identified known-activating variants, followed by K601E (2.3%; n = 4) and N581S (1.7%; n = 3). Less common known-activating SVs included G469V (1.1%; n = 2), V600D (0.6%; n = 1), and V600K (0.6%; n = 1). Known-activating indels identified included N486_T491 > K and N486_P490del (each at 1.1%, n = 2) and A598_T599insT and T599_V600insT (each at 0.6%; n = 1) (Fig. 1A). A known-activating *BRAF* fusion was identified in 72 cases (32.6% of the *BRAF* mutation–positive cohort), all of which contained an intact *BRAF* kinase domain (encoded by exons 11–18) and breakpoints in *BRAF* introns 7, 8, 9, or 10. Sixty-four (88.9%) of these included the *KIAA1549* fusion partner, with 8 distinct *KIAA1549-BRAF* fusion variants identified. *BRAF* fusions with unique fusion partners were identified in eight cases, with two involving the novel fusion partners *STARD3NL* and *KHDRBS2* (Fig. 1B).

Genomic Landscape of Hematologic Malignancies and Extracranial Solid Tumors with Known-Activating *BRAF* Alteration

Among 45 patients with extracranial solid tumors or hematologic malignancies harboring a known- activating *BRAF*

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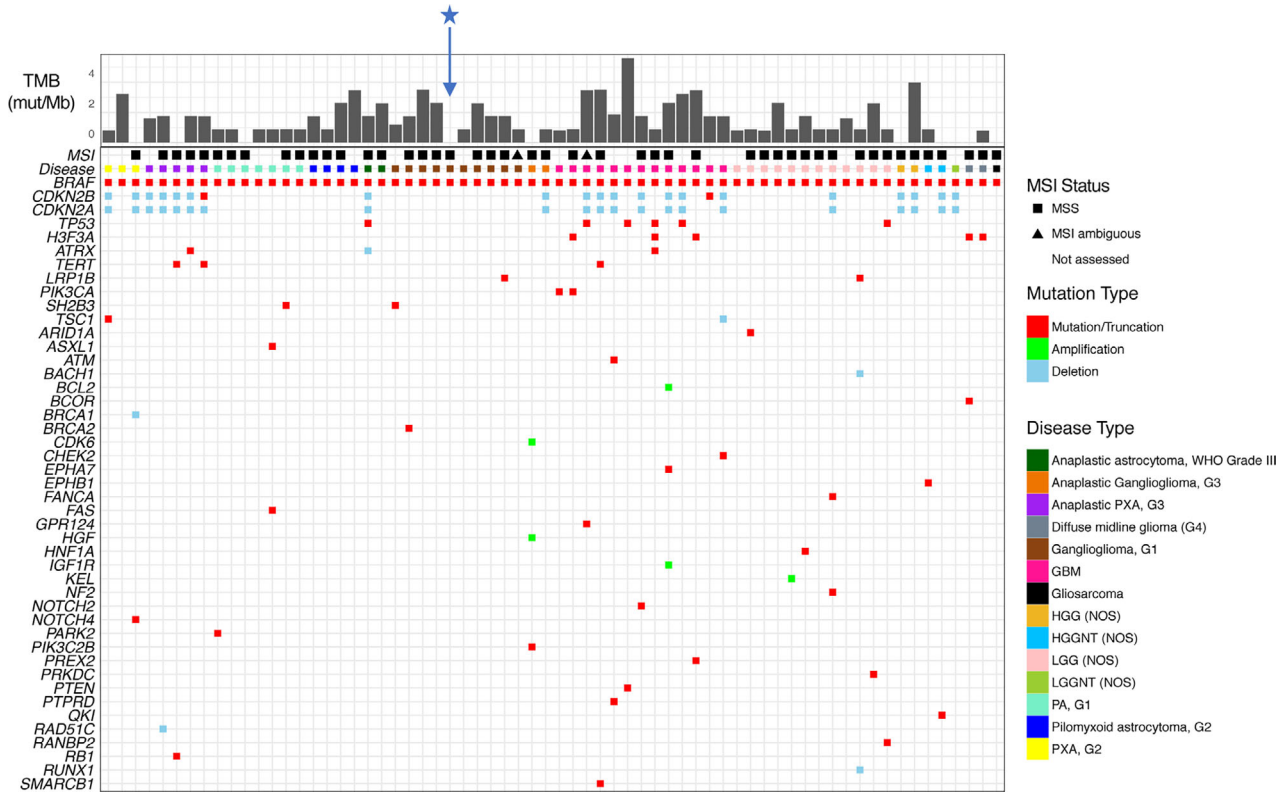

Figure 3. Genomic landscape of primary brain tumors bearing a known-activating *BRAF* short variant or indel. Specimens are arranged from young to old (left to right) within each cancer type. Blue star indicates the genomic profile associated with Index Case 1.

Abbreviations: G, grade; GBM, glioblastoma; HGG, high grade glioma; HGGNT, high grade glioneuronal tumor; LGG, low-grade glioma; LGGNT, low grade glioneuronal tumor; MSI, microsatellite instability; MSS, microsatellite stable; mut/Mb, mutations per megabase; NOS, not otherwise specified; PA, pilocytic astrocytoma; PXA, pleomorphic xanthoastrocytoma; TMB, tumor mutational burden; WHO, World Health Organization.

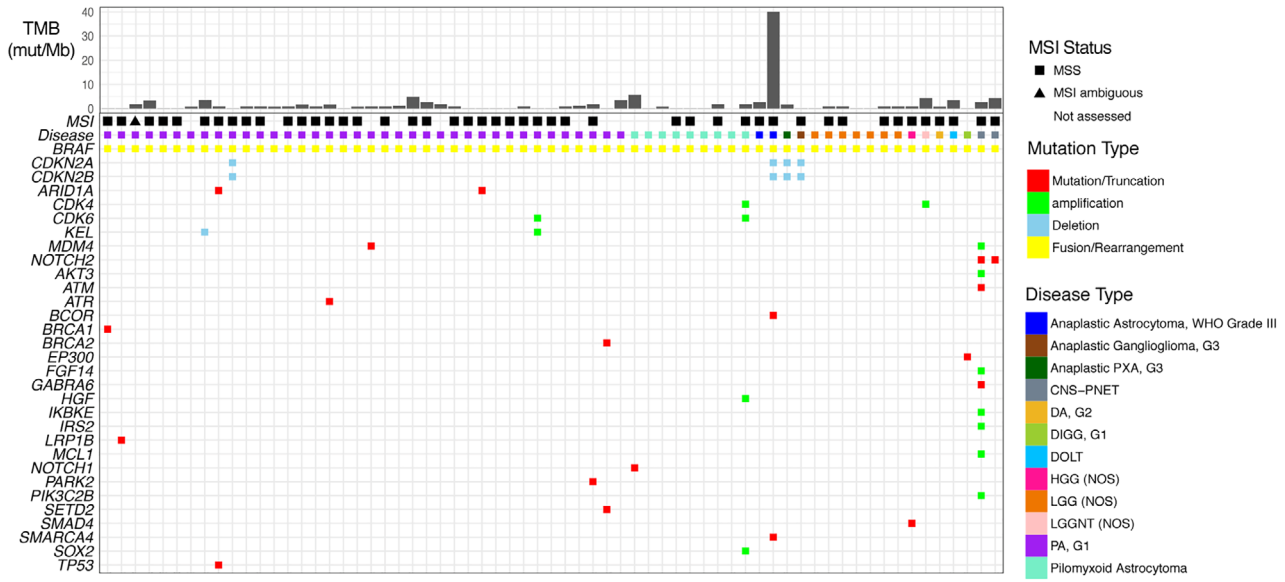

Figure 4. Genomic landscape of primary brain tumors bearing a known-activating *BRAF* fusion. Specimens are arranged from young to old (left to right) within each cancer type.

Abbreviations: CNS-PNET, central nervous system–primitive neuroectodermal tumor; DA, diffuse astrocytoma; DIGG, desmoplastic infantile ganglioglioma; DOLT, disseminated oligodendroglial-like leptomeningeal tumor; G, grade; HGG, high grade glioma; LGG, low-grade glioma; LGGNT, low grade glioneuronal tumor; MSI, microsatellite instability; MSS, microsatellite stable; mut/Mb, mutations per megabase; NOS, not otherwise specified; PA, pilocytic astrocytoma; PXA, pleomorphic xanthoastrocytoma; TMB, tumor mutational burden; WHO, World Health Organization.

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A

Diagnosis	Age (yrs)	*BRAF* Genomic Alteration
PA, G1	4	duplication of exons 9-18
LGG (NOS)	**10**	**deletion of exons 1-7**
Osteosarcoma	21	deletion of exons 1-7
Neuroblastoma	**4**	**translocation of exons 8-18 to chrX**
Neuroblastoma	7	duplication of exons 9-18

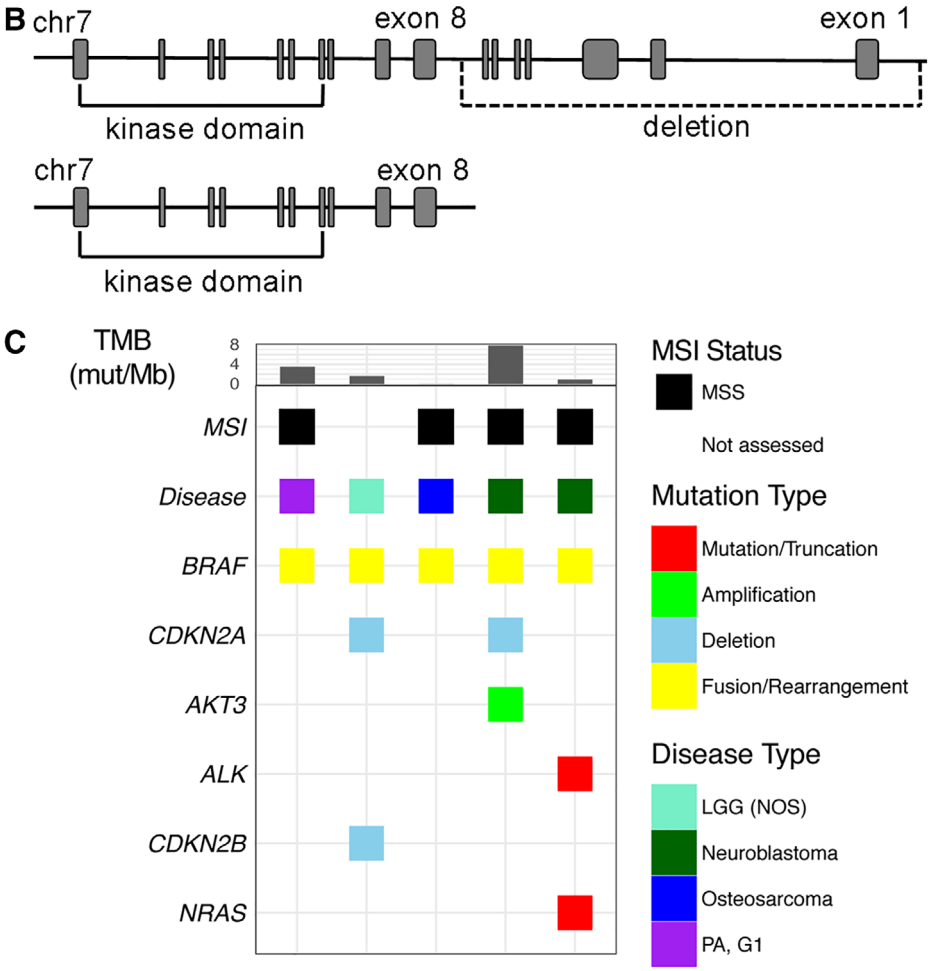

Figure 5. Landscape of *BRAF* nonfusion rearrangements. **(A):** Description of *BRAF* rearrangements in pediatric malignancy. **(B):** Schematic representing loss of *BRAF* exons 1–7. **(C):** Genomic landscape of pediatric cancers bearing a *BRAF* nonfusion rearrangement. Abbreviations: G, grade; LGG, low-grade glioma; MSI, microsatellite instability; MSS, microsatellite stable; mut/Mb, mutations per megabase; NOS, not otherwise specified; PA, pilocytic astrocytoma; TMB, tumor mutational burden.

alteration, *BRAF* V600E was the most common SV (n = 25/45, 55.6%) and was observed in AML (therapy-related) (n = 1); histiocytic neoplasms (n = 9); melanoma (n = 8); papillary thyroid carcinoma (n = 4); and rhabdomyosarcoma, acinar cell carcinoma, and serous carcinoma (n = 1 each). A known-activating *BRAF* fusion was identified in seven samples: *KIAA1549-BRAF* in embryonal rhabdomyosarcoma (n = 1), sarcoma (NOS) (n = 1), and malignant peripheral nerve sheath tumor (n = 1); *CUX1-BRAF* in sarcoma (NOS) (n = 1); *STARD3NL-BRAF* in sarcoma (NOS) (n = 1); *PPP1CC-BRAF* in acinar cell carcinoma (n = 1); and *KHDRBS2-BRAF* in large cell neuroendocrine carcinoma (unknown primary) (n = 1) (supplemental online Table 1).

Few co-occurring genomic driver alterations or signatures were identified in this cohort with the notable exception of the hematological and melanoma sample subsets. Thirteen of 45 samples contained either *CDKN2A/B* deletion (n = 9) or a *CDKN2A* truncating alteration (n = 4). Co-occurring known-activating *KRAS* SVs were found in four samples. The majority of cases contained low or intermediate TMB with the exception of melanoma samples, of which 50% (5/10) displayed high TMB. All cases that could be assessed for microsatellite instability demonstrated a microsatellite stable status (Fig. 2).

Genomic Landscape of Primary Brain Tumors with Known-Activating *BRAF* Alterations

Analysis of 131 primary brain tumors revealed 66 samples with known-activating *BRAF* SV or indels and 65 with a known-activating gene fusion. *BRAF* V600E was the most common (62/66 cases) SV or indel observed in 14 distinct histological subtypes. Less common, known-activating *BRAF* variants were identified in one patient with high grade

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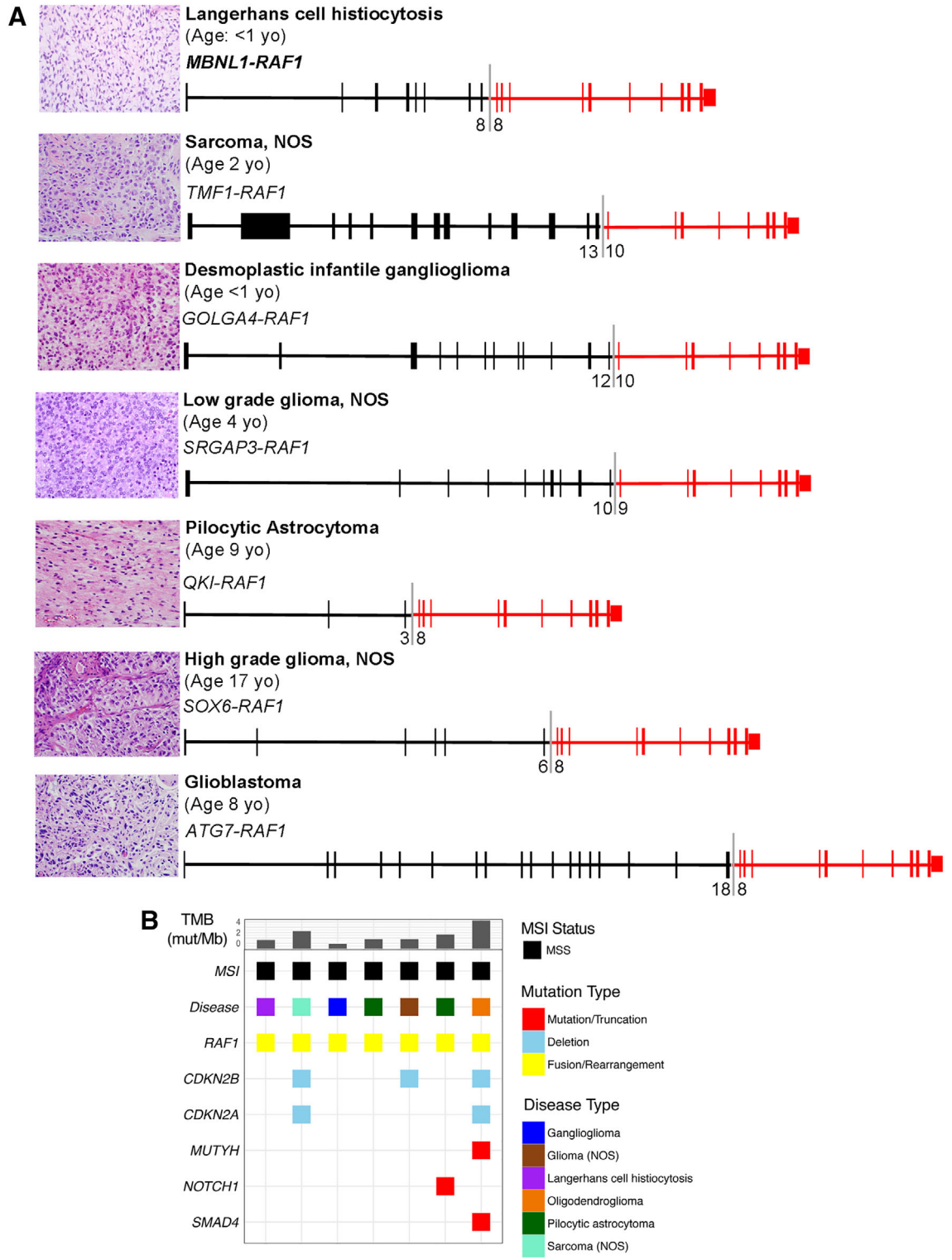

Figure 6. Landscape of *RAF* fusions. **(A):** Hematoxylin and eosin staining and corresponding schematics representing recurrent and novel *RAF1* kinase fusions identified in pediatric cancer. Exon numbers at the fusion boundary are depicted below each fusion diagram. **(B):** Genomic landscape of pediatric cancers bearing a known-activating *RAF1* fusion. Specimens are arranged from young to old (left to right) within each cancer type.
Abbreviations: MSI, microsatellite instability; MSS, microsatellite stable; mut/Mb, mutation; mutations per megabase; NOS, not otherwise specified; TMB, tumor mutational burden.

glioneuronal tumor (NOS) (N581S), one patient with GBM (N486_T491 > K), and two patients with LGG (NOS) (T599_V600insT, A598_T599insT, respectively) (supplemental online Table 2). All cases had low TMB, and those that could be assessed for microsatellite instability all demonstrated a microsatellite stable status (Fig. 3).

KIAA1549-BRAF was the most common fusion, identified in 61 of 65 fusion-positive cases represented by 16 tumor

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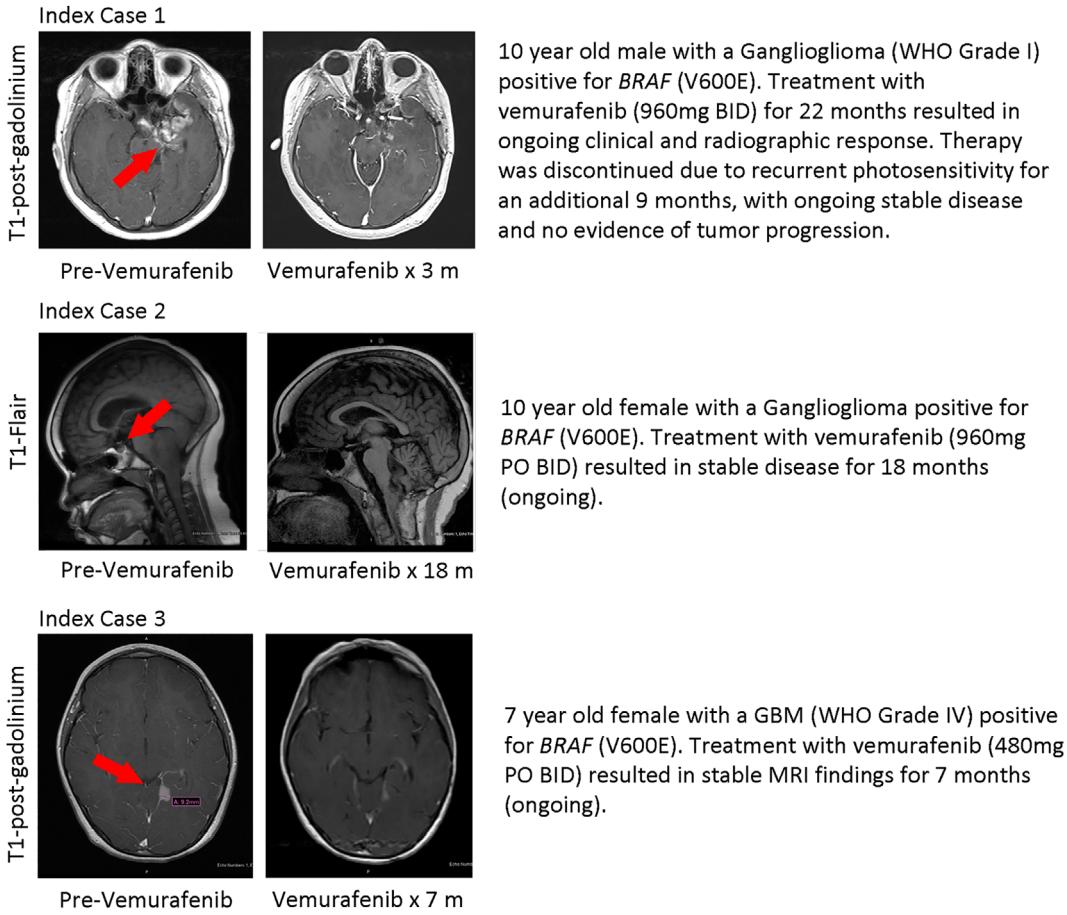

Figure 7. Postcontrast magnetic resonance imaging images showing decrease in tumor size in three separate *BRAF* V600E–positive pediatric patients after treatment with vemurafenib.
Abbreviations: BID, bis in die (twice daily); GBM, glioblastoma; MRI, magnetic resonance imaging; PO, per os (by mouth); WHO, World Health Organization.

subtypes, with highest frequency observed in PA, grade 1 (55% of fusion-positive specimens). Noncanonical *BRAF* fusions were identified in four other tumors: PA, grade 1 (*FAM131B-BRAF*); PA, grade 1 (*BCAS1-BRAF*); anaplastic pleomorphic xanthoastrocytoma, grade 3 (*CCDC6-BRAF*); and anaplastic ganglioglioma, grade 3 (*TMEM106B-BRAF*) (supplemental online Table 3). All the *BRAF* fusion–positive cases except for one contained low or intermediate TMB, the exception being a single tumor from a 20-year-old patient diagnosed with anaplastic astrocytoma World Health Organization (WHO) grade III that contained a TMB of >40 mut/Mb. All cases that were able to be assessed for microsatellite instability demonstrated microsatellite stable status (Fig. 4).

BRAF Nonfusion Rearrangements

Nonfusion *BRAF* rearrangements were identified in five patients, including one patient with PA, grade 1; one patient with LGG (NOS); two patients with neuroblastoma; and one patient with osteosarcoma (Fig. 5A). These noncanonical *BRAF* alterations manifested from one of three distinct chromosomal rearrangements, and all resulted in genomic loss or disruption of the *BRAF* N-terminal autoinhibitory domain with breakpoints in intron 7, 8, or 9, which has been shown to result in constitutive kinase activation in a RAS-independent manner [66] (Fig. 5B). All five specimens demonstrated low or intermediate TMB, and none demonstrated microsatellite instability (Fig. 5C).

RAF1 Known-Activating Fusions in Solid Tumors

Known-activating *RAF1* fusions (n = 7) were identified in five distinct brain tumor subtypes, one sarcoma, and one histiocytic neoplasm. All fusions contained an intact *RAF1* kinase domain (encoded by exons 10–17) with unique fusion partners and breakpoints in *RAF1* introns 7 or 9. Two involved the novel fusion partners *TMF1* (sarcoma [NOS]) and *SOX6* (HGG [NOS]) (Fig. 6A). All seven specimens demonstrated low TMB, and none demonstrated microsatellite instability (Fig. 6B).

Index Cases

Three patients with *BRAF* V600E–mutated brain tumors, including a 10-year-old boy and 10-year-old girl, each with ganglioglioma (WHO grade I) (Index Cases 1 and 2), and a 7-year-old girl with a GBM (WHO grade IV) (Index Case 3), who each experienced progression after conventional treatment, were independently treated with the BRAF inhibitor vemurafenib on a compassionate basis. Index Cases 1 and

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2 showed clinical and radiological response to the targeted therapy (960 mg b.i.d.) and remained on treatment 22 months and > 18 months, respectively, with ongoing response. After treatment with vemurafenib for 22 months, therapy was discontinued in Index Case 1 because of recurrent photosensitivity, and this patient has remained off treatment for >9 months with no radiologic or clinical evidence of tumor progression. The patient described in Index Case 3 was treated with 480 mg p.o. b.i.d. and showed stable magnetic resonance imaging findings >7 months with ongoing sustained response (Fig. 7).

Prior Molecular Testing

To better understand the extent to which prior molecular testing was used in this data set of tumors that harbored *BRAF* or *RAF1* known-activating alterations, we assessed cases (n = 35) with available clinical histories. Of those with prior *BRAF* molecular testing results, 19 (54.3%) reported results from prior testing methodologies inconsistent with the respective *BRAF* alteration type later identified with CGP (supplemental online Fig. 3A). Specifically, of eight PA cases with either *KIAA1549-BRAF* or *QKI-RAF1* fusion detected by CGP, six were previously tested for *BRAF* V600E by immunohistochemistry or polymerase chain reaction, and therefore the underlying *BRAF* fusion was not detected (supplemental online Fig. 3B).

Discussion

In this study we highlight the diverse landscape of pediatric cancer types that harbor genomic alterations in *BRAF* or *RAF1* and describe three index cases with durable benefit with RAF inhibitors. In our data set, alterations in *BRAF* likely to represent driver events were identified in approximately 6% of all pediatric tumors screened with CGP during routine clinical care. Key among these findings is that 25% of the tumor samples represent extracranial solid or hematologic tumor types for which single gene or broad panel testing for druggable biomarkers are unlikely to be employed routinely in a clinical setting. For example, *KIAA1549-BRAF*, *CUX1-BRAF*, *STARD3NL-BRAF*, or *TMF1-RAF1* fusions, which were identified in four separate patients with sarcoma in our study, would have likely gone unrecognized with standard of care molecular testing.

Multiple biomarker-informed targeted therapies have been developed for adult patients with cancer, but there continues to be significant lag time for similar development for pediatric cancers. Notable exceptions are recent age-agnostic therapy approvals, including larotrectinib and entrectinib for *NTRK* fusion–positive patients and the emergence of umbrella protocols, including the Children's Oncology Group–National Cancer Institute Pediatric Molecular Analysis for Therapeutic Choice (Pediatric MATCH) protocol [67]. To address this disparity, one potential strategy is the repurposing of off-label FDA-approved targeted therapies for pediatric patients with cancer with malignancies harboring relevant predictive biomarkers. Notably, a key challenge in implementing such a strategy is the ability to identify patients likely to benefit from a given targeted therapy. Per patient, single gene tests or other protein expression–based diagnostics suffer the limitations of requiring significant tissue and/or a limited range of biomarker detection. Sequential testing of individual biomarkers via multiple molecular diagnostic tests can result in significant loss of treatment time or in unnecessary toxicity because of use of conventional therapy. Our data are consistent with this; of cases with prior *BRAF* molecular testing results available, more than half of reported results were inconsistent with the respective *BRAF* alteration later identified by CGP. Moreover, even within one indication, diverse and druggable biomarkers are potentially discoverable. For example, at least 60% of PAs harbor *KIAA1549-BRAF* fusion. However, tumors found to be *BRAF* fusion–negative by standard molecular testing (e.g., fluorescence in situ hybridization) may instead harbor alternative activating variants in diverse genes including *BRAF*, *NTRK1–3*, *FGFR1*, *NF1*, or *KRAS* [14, 15, 68], all of which are directly or indirectly druggable with currently approved targeted therapies [69]. Large gene panel–based molecular profiling is currently the most efficient means of identifying the breadth of potentially clinically relevant variants in pediatric cancers.

Conclusion

There remains wide disparity in survival depending on cancer type in pediatric cancers. Improved therapeutic strategies are therefore urgently needed. Broad panel-based molecular profiling can efficiently identify multiple key genomic drivers and should therefore be considered a component of standard molecular testing in advanced or recurrent pediatric cancer types, regardless of disease indication.

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Disclosures

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