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CASE REPORT

A germline variant of *TP53* in paediatric diffuse leptomeningeal glioneuronal tumour

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Abstract

Purpose Diffuse leptomeningeal glioneuronal tumour (DLGNT) is an extremely rare tumour involving the neuroaxis. At present, its exact pathogenesis and associated factors remain incompletely characterised. Recent molecular investigations in a small cohort have offered some insights into this disease. However, the role of germline findings has not yet been fully explored in affected patients. The authors present a case of DLGNT, focusing on the clinical and molecular features with reference to current disease knowledge.

Methods A 4-year-old male presented with raised intracranial pressure symptoms secondary to extensive leptomeningeal disease of the brain and spine. Preliminary impression was that of an inflammatory lesion.

Results A lumbar biopsy of the lesion confirmed DLGNT on routine diagnostic examination. Further molecular analysis of his tumour and blood demonstrated a previously unreported *TP53* exon 5 (c.147V > I) germline variant. Based on the latest DLGNT molecular subtyping classification, his tumour falls into the group with better clinical outcome. However, his germline findings may add an extra layer of uncertainty to his overall prognosis.

Conclusion Given that much remains unknown in many rare paediatric tumours at this stage, isolated findings found in an individual may be of significance. Supplementary genetic information, together with tumour molecular analysis, add to our current understanding of this uncommon disease. This case highlights the benefit of combined clinical and molecular efforts, including germline testing, especially for children affected by such challenging neoplasms.

Keywords Glioma · Diffuse leptomeningeal glioneuronal tumour · TP53

Introduction

Diffuse leptomeningeal glioneuronal tumour (DLGNT) is an extremely rare entity. Previous publications have concluded that these lesions represent a unique class of low-grade neuroepithelial tumours that show variable neuronal/neurocytic and glial differentiation [1]. Owing to the infrequency of clinical cases, this recently described tumour has not yet been assigned a grade in the current WHO 2016 classification of central nervous system tumours [2]. It is reputed to be predominantly slowly progressive, and exhibits little, if any, parenchymal involvement [3]. Advancements in

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technology have provided recent insights into DLGNT [4–6]. Despite this, there remains significant knowledge gaps in the exact biological mechanisms of this neoplasm.

Case presentation

A previously well and developmentally normal 3-year-old male presented with symptoms of progressively worsening raised intracranial pressure, associated with gait disturbance. There was no complaint of back pain or radicular pain, and no voiding difficulty. Imaging of the neuro-axis showed evidence of hydrocephalus with leptomeningeal enhancement along the basal cisterns, posterior fossa structures and cranial nerves (Fig. 1). In addition, there was diffuse, thick and slightly nodular spinal enhancement with an intramedullary enhancing focus in the lower thoracic cord (Figs. 2 and 3). Positron emission tomography–computed tomography (PET-CT) did not demonstrate any fluorodeoxyglucose (FDG)-positive lesions. Ophthalmology review confirmed bilateral mild papilloedema. There was no report of abnormalities in visual acuity or convergence and no opthalmoplegia. Cerebrospinal fluid (CSF) sampling showed elevated protein levels (1.12 g/L) but normal glucose levels and no pleocytosis. There were no tumour cells present in the CSF. Laboratory investigations to search for infective causes including Tuberculosis and Cryptococcus were negative.

Decision was made for surgery to address two key issues: symptomatic hydrocephalus and tissue diagnosis to guide further treatment. Laminectomy at the level of L4/5 and biopsy of the abnormal leptomeninges was performed. Intraoperatively the dura was found to be normal in appearance; however, the arachnoid was seen to be thickened and slightly vascular, coating and encasing the cauda equina nerve roots (Fig. 4). Following that, a right frontal ventriculo-peritoneal shunt was inserted for treatment of his hydrocephalus. His symptoms of headache and vomiting improved with the intervention. Papilledema also resolved on subsequent ophthalmology review.

Histopathological features of the arachnoid samples were consistent with that of DLGNT. There were also rare small slender fascicles of Schwann-like cells. No high-grade nuclear atypia, mitoses, necrosis or microvascular proliferation was seen (Fig. 5a). Further immunohistochemistry (IHC) tests showed positivity for S100, GFAP and synaptophysin (Fig. 5b–d). However, IDH1 (R132H) and neurofilament stains were negative. The Ki67 labelling index was < 1%. Fluorescence in situ hybridization (FISH) showed 1p36 deletion but was negative for 19q13 deletion (Fig. 6a, b). *KIAA1549-BRAF* fusion via RT-PCR (real-time polymerase chain reaction) amplification was positive.

The tumour specimen was also submitted to Foundation Medicine, a clinical laboratory improvement amendments (CLIA)-certified lab for next-generation sequencing (NGS) [7]. The FoundationOne® panel was used, which is a hybrid-capture based NGS test. This assay has been described in previous studies [7, 8]. Here, significant genomic alterations included a *TP53* exon 5 variant (c.147V > I) single-nucleotide polymorphism (SNP) and *KIAA1549-BRAF* fusion (Fig. 7). The latter reaffirmed our previous diagnostic results. Furthermore, the 315-gene panel included the entire DNA coding sequence for the detection of base substitutions, insertions/ deletions and copy number alterations for the H3F3A gene (Supplementary Fig. A). This did not detect H3K27M which is associated with diffuse midline gliomas. In view of the *TP53* finding, the patient underwent further germline screening under the Cancer Genetics team. The same *TP53* exon 5 variant (c.147V > I) was found in his blood

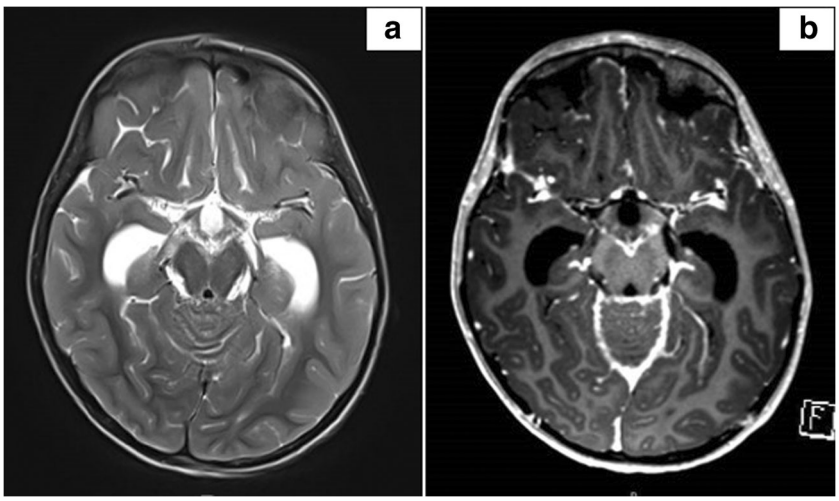

Fig. 1 **a**, **b** Representative T2-weighted and post-contrast T1-weighted sequence MRI brain images in axial view. Figure 1a demonstrates dilatation of both temporal horns and third ventricle secondary to obstructive hydrocephalus. Figure 1b shows extensive leptomeningeal enhancement without significant intraparenchymal disease [10]

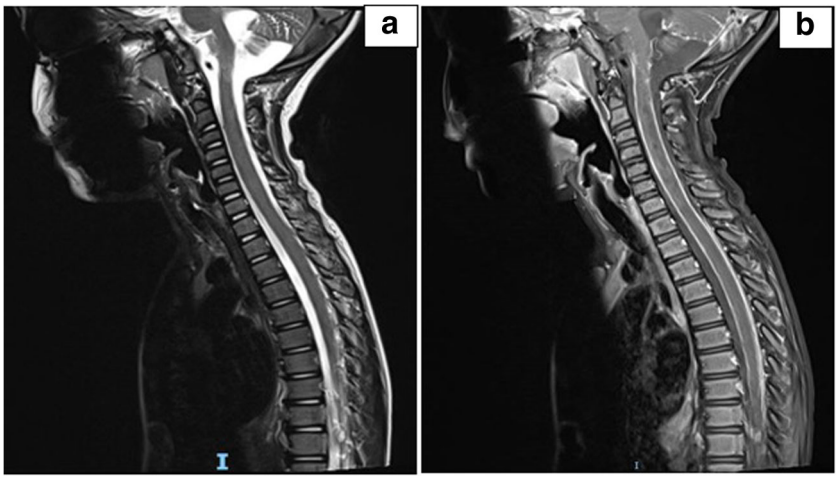

Fig. 2 **a**, **b** T2-weighted and post-contrast T1-weighted sequence MRI cervico-thoracic spine images in sagittal view, respectively. Figure 2b has evidence of enhancing and thickened leptomeningeal involvement throughout the length of the cervio-thoracic cord

sample, confirming that this variant was in his germline, and hence, not somatic in nature. In view of the germline *TP53* finding, his family members were referred for comprehensive genetic screening as well.

The patient was commenced on chemotherapy using a modified Children's Cancer Group protocol for low-grade glioma (COG A9952). The regimen consisted of a combination of vincristine and carboplatin [9]. He completed induction treatment uneventfully. However, during maintenance chemotherapy, he developed bilateral foot drop secondary to peripheral neuropathy. In view of his symptoms, decision was made to withhold vincristine and continue carboplatin monotherapy [10]. He was referred to the paediatric neurologist for management of his bilateral foot drop, which showed mild improvement after physiotherapy and discontinuation of vincristine. Post-adjuvant treatment, repeat MRI brain and spine imaging reported largely stable intracranial and spinal leptomeningeal enhancement, with less prominence of previous intramedullary disease. He continues to be on close surveillance under our multi-disciplinary neuro-oncology team.

Discussion

Diffuse leptomeningeal glioneuronal tumour is a novel entity under the neuronal and mixed neuronal-glial tumours in the WHO 2016 classification [11]. From clinical observations, DLGNT patients commonly present with signs and symptoms of progressive hydrocephalus—that is, headache, nausea, vomiting, ataxia and, or papilledema. Often, neuroimaging would reveal diffuse leptomeningeal enhancement (with or without a recognisable intraparenchymal component), and cerebrospinal fluid (CSF) sampling would show elevated protein but absent tumour cells. This constellation of clinical signs, together with neuroimaging, often cause affected patients to be misdiagnosed with meningitis or disseminated neoplasia [1]. Other neoplastic differentials for DLGNT include leptomeningeal dissemination of intraparenchymal diffuse astrocytoma, oligodendroglial glioma and pilocytic astrocytoma [12]. Broadly speaking, paediatric low-grade gliomas are a heterogeneous group. They include tumours of astrocytic, oligodendroglial and mixed glial-neuronal histology [13]. As a

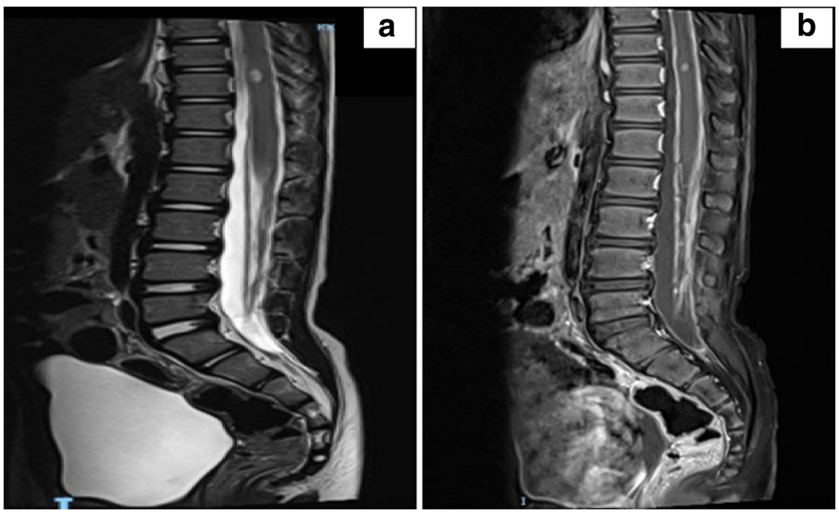

Fig. 3 **a**, **b** T2-weighted and post-contrast T1-weighted sequence MRI lower thoracic and lumbo-sacral spine images in sagittal view, respectively. There is a prominent intramedullary focus at the level of T10/T11 seen in both images. (Of interest, the bladder was noted to be full on the MRI which was performed under general anaesthesia. Subsequent clinical assessment when the patient was awake showed he had no voiding issues)

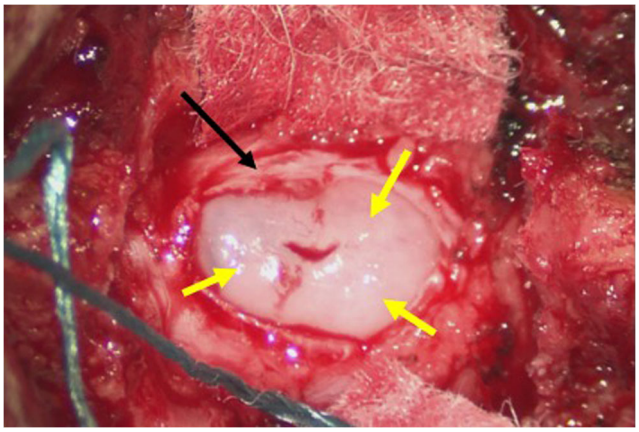

Fig. 4 An intraoperative photo demonstrating the lesion. Yellow arrows point to the pearly-white thickened layer involving the arachnoid that was biopsied during surgery. Black arrow points to the normal-looking dura that was adherent over the lesion

result, some of these may have similar radiological features and or clinical presentations. Therefore, the role of conscientious histopathological and ancillary molecular tests to ensure correct diagnosis is critical.

A case series by Rodrigeuz et al reports the existence of high frequency of concurrent *KIAA1549-BRAF* fusions and 1p deletions in these tumours [3]. The same study also demonstrated that most DLGNT tumours lack BRAF V600E mutations—a pertinent finding as the latter is commonly seen in pleomorphic xanthoastrocytoma and some gangliogliomas [5, 14]. Of note, similar findings were observed in a recent genome-wide DNA methylation screening of 30 DLGNT patients, placing them into two subgroups: DLGNT methylation class (MC)-1 and DLGNT methylation class (MC)-2 [6]. Interestingly, all cases had loss of chromosomal arm 1p. Next, co-deletion of 1p/19q was frequently observed in DLGNT-MC-1, and DLGNT-MC-2 cases displayed a gain of chromosomal arm 1q. Both subgroups had recurrent genetic alterations leading to an aberrant *MAPK/ERK* pathway, with *KIAA1549-BRAF* fusion being the most frequent event. The DLGNT-MC-1 group reported lower age at diagnosis (median 5 vs 14 years) and a clinically more favourable course [6].

At present, there is no established therapeutic guidelines for DLGNT patients. This is likely due to the rarity of this particular CNS tumour. Published reports cite the use of various chemotherapy regimens with or without radiation [5, 15–18]. Majority of these studies show relatively good clinical outcomes with adequate disease control. Nonetheless, these studies are mostly based on small cohorts; hence, large-scale, randomised prospective trials are needed to ascertain the best adjuvant treatment regimen for such patients. The MAPK/

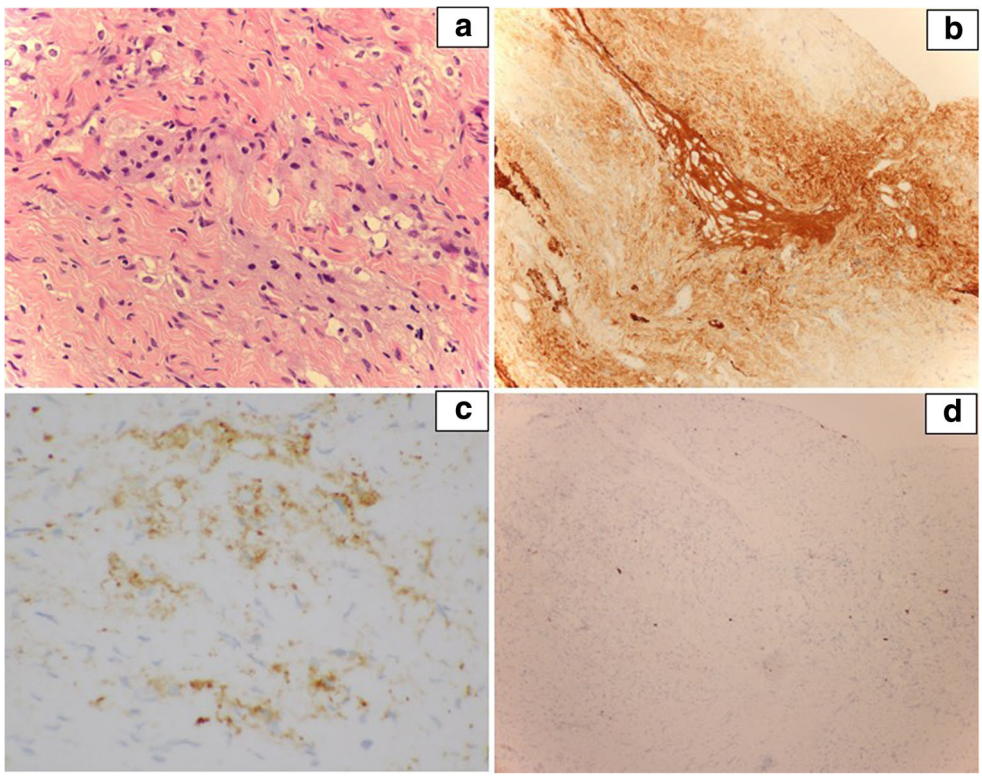

Fig. 5 **a** Haemotoxylin and eosin slide (×20). Morphology shows thickened, mildly inflamed, fibrotic arachnoid diffusely infiltrated by a low-grade neuroepithelial tumour. It comprises of haphazard pauci-cellular fascicles, islands of basophilic and focally myxoid neurophil interspersed amongst the collagen bundles, containing cells with monotonous small round-to-oval shaped nuclei, indistinct cell borders and focally granular cytoplasm. **b**–**d** IHC stains for glial fibrillary acidic protein (GFAP), syanptophysin and Ki67, respectively. As depicted, there is positivity for GFAP (**b**) and synaptophysin (**c**). The Ki67 index (**d**) was low, at < 1%. All IHC figures have a magnification of ×40, except for Fig. 5c which has a ×60 magnification

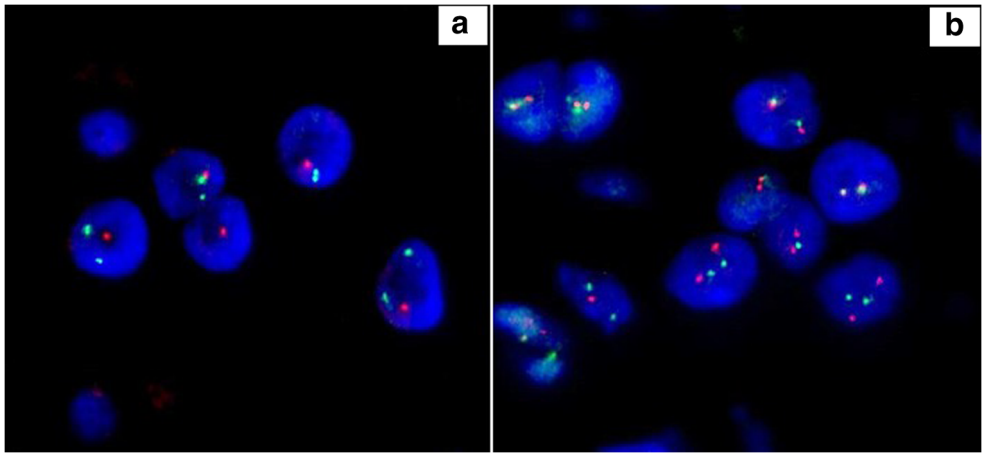

Fig. 6 **a**, **b** The 1p/1q FISH panel and 19p/19q FISH panel, respectively. Interphase FISH using locus specific identifier (LSI) 1p36/1q25 and 19q13/19p13 dual-colour probes were performed on the tumour. Of the 1p36, 83.3% was deleted and the ratio of 1p36/1q25 was 0.57. However, only 8.3% of 19q13 was deleted and the ratio of 19q13/19p13 was 0.96. Hence, the test was concluded to be positive for 1p36 deletion but negative for 19q13 deletion. (For references in this test, the cut-off ratio for a deletion is 0.80 and the normal cut-off level is 17.2% of 60 nuclei enumerated)

ERK alterations found in this tumour represent therapeutic targets and imply that respective BRAF or MEK inhibitors may be alternative treatment options.

Based on the above evidence, our patient has potential features of a more favourable DLGNT-MC-1 subtype. However, the additional finding of his germline variant has added another layer of uncertainty to his overall prognosis. In Dodgshun et al's study of 10 patients, 1 patient is reported to have *RAF*1 germline mutation concurrently with cardio-facio-cutaneous syndrome—a Noonan's-like syndrome [5]. At this point, we will like to highlight that germline findings in non-syndromic DLGNT patients, as demonstrated in our patient, have not yet been reported. Currently, modern molecular techniques enable the identification of unexpected cancer-

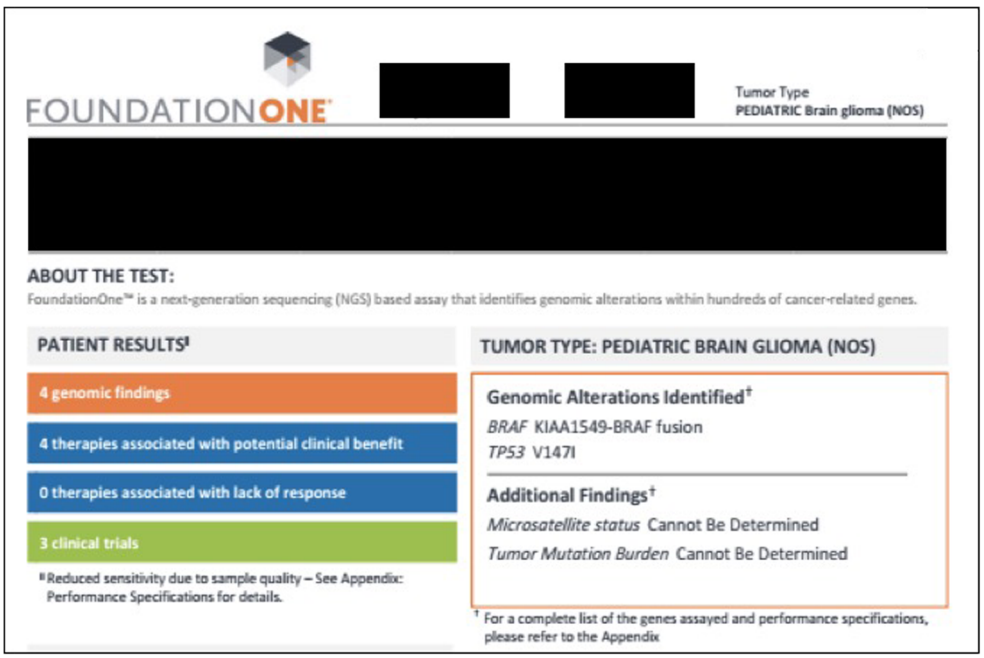

Fig. 7 Screenshot of the patient's anonymized FoundationOne® report

predisposing gene mutations, such as acquired de novo variations, as well as variations of unknown significance [19]. In the context of paediatric cancers, *TP53*-related findings may be clinically relevant for risk of future malignancies and, or response to treatment. Given that rare paediatric tumours remain poorly understood at this stage, an isolated finding found in a single patient may be of significance [20]. The authors advocate continued emphasis on combined clinical and molecular analyses to guide disease management, especially for children affected by such challenging neoplasms.

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Compliance with ethical standards

Conflict of interest We, the authors of this manuscript, report no funding, financial support or industrial affiliations received for the writing of this article. In addition, we report no conflict of interest concerning the material or methods used in this paper. This manuscript has not been published and is not being considered for publication elsewhere.

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