Case Report

Familial CNS-Isolated Hemophagocytic Lymphohistiocytosis Due to a Novel PRF1 Mutation Triggered by SARS-CoV2

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Abstract

Background: Hemophagocytic lymphohistiocytosis (HLH) is a potentially fatal hyperinflammatory condition that presents with fever, hepatosplenomegaly, and characteristic laboratory findings. Mutations in the perforin gene PRF1 have been implicated in cases of familial HLH (fHLH) and can cause isolated CNS-HLH in the absence of systemic HLH. **Results:** A five year-old boy presented with three weeks of headache, blurry vision, and emesis. He was diagnosed with acute disseminated encephalomyelitis (ADEM), thought to be triggered by SARS-CoV-2 given positive nasopharyngeal testing. He completed a five day course of high dose IV methylprednisolone and plasma exchange. In the subsequent months, he was admitted twice due to worsening clinical and radiological activity and after several courses of IV pulse steroids, plasmapheresis, and IV immunoglobulin (IVIG), his condition stabilized with rituximab and monthly IVIG. A few months later, his younger brother presented with a similar syndrome. It was discovered that his parents were second cousins, leading to concern for a genetic disorder. Genetic testing revealed a homozygous mutation for PRF1 in both siblings (variant c.4422G>A). **Conclusions:** This is the first presentation of CNS-isolated familial HLH triggered by SARS-CoV-2 in the pediatric population. Furthermore, this is the first report of this specific PRF1 mutation, the variant c.4422G>A, as pathogenic. It highlights the relevance of genetic testing in pediatric neuroinflammatory disorders that do not respond adequately to conventional treatments. It is possible that as our knowledge in neurogenetics develops, certain genes will be identified as predisposing factors to syndromes such as ADEM.

Keywords: ADEM, autoimmune, hemophagocytic lymphohistiocytosis, PRF1 mutation

Introduction

Hemophagocytic lymphohistiocytosis (HLH) is a potentially fatal hyperinflammatory condition that presents with fever, hepatosplenomegaly and characteristic laboratory findings.[1] HLH can be primary/familial (fHLH) in the setting of genetically inherited mutations or secondary/acquired (sHLH) when triggered by an infection, malignancy, or rheumatological disorder.[2]

HLH is caused by a defect in the cytotoxic function of natural killer (NK) and T cells which results in uncontrolled hypercytokinemia and widespread activation of T cells and macrophages, some of them which engage in hemophagocytosis.[3] The cytokine storm leads to systemic inflammation and multiorgan dysfunction, including neuroinflammation.[4] Usually, neuroinflammation is seen in the setting of a systemic disease but in rare cases, it presents as isolated CNS disease (isolated CNS-HLH).[4,5]

FHLH is an autosomal recessive genetically heterogeneous disorder caused by various mutations in the NK/T-cell cytotoxic pathway. Numerous gene defects have been described in fHLH, with PRF1 and UNC13D mutations representing the most commonly identified ones.[6] Specific mutations in the perforin gene PRF1 have been identified in isolated CNS-HLH.[5]

In fHLH, infections, usually viral, can trigger an inappropriate cytotoxic immune response.[1] Here we present a five year-old boy who was initially thought to have refractory acute disseminated encephalomyelitis (ADEM) after SARS-CoV-2 infection who was later found to have CNS-isolated HLH with PRF1 mutation (variant c.4422G>A), not previously known to be pathogenic.

Case Report

A five year-old previously healthy Hispanic boy presented to the emergency department (ED) with persistent headaches, blurry vision, and emesis. In the ED, a routine physical examination and workup, including basic metabolic and infectious studies, was unremarkable except for a positive SARS-CoV-2 RNA PCR nasopharyngeal swab. Therefore, his presentation was

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thought to be due to the viral illness, and the patient was discharged to home with symptomatic management. However, symptoms persisted, and three weeks later he returned to the ED. He redemonstrated positive PCR for SARS-CoV-2 RNA (Biofire Respiratory Panel) and as he was still endorsing neurological complaints, including headache, right eye blurry vision, and mild bilateral arm weakness, neuroimaging of the brain and spinal cord imaging was obtained.

MRI brain revealed multifocal lesions on T2-fluid attenuated inversion recovery with intense enhancement and diffusion restriction, most prominent in the right frontal lobe, basal ganglia, and cerebellum, with vasogenic edema and punctate foci of hemorrhage [Figure 1A]. Bilateral swelling of the optic nerves and optic chiasm was also evident. MRI spine revealed longitudinally extensive myelitis with an intensely enhancing, expansile intramedullary lesion from T1 to T3 [Figure 1B]. Neurology examination, although limited by irritability and poor cooperation, was most significant for bilateral upper extremity weakness (Medical research council (MRC) scale of 3/5, left worse than right), bilateral lower extremity strength appeared grossly intact (he was able to ambulate), and normal gait. Due to poor cooperation, visual acuity and reflexes could not be assessed.

Based on his age, and clinical and imaging findings, in the setting of a recent infection, ADEM was suspected. Neuromyelitis Optica was considered as well, awaiting aquaporin-4 antibodies testing. He was started on high-dose IV methylprednisolone at 30 mg/kg/day for five days. A lumbar puncture was performed the next day under sedation and cerebrospinal fluid (CSF) showed nine nucleated cells/mm^3 (97% lymphocytes), 44 mg/dL protein, and 77 mg/dL glucose. Oligoclonal bands were negative in CSF. D-dimer was mildly elevated (0.56 mcg/mL) as well as the erythrocyte sedimentation rate (17 mmol/hour). Fibrinogen was mildly decreased (139 mg/dL) and ferritin levels were not available. Lactate dehydrogenase was within normal limits (1.7 mmol/L) as well as vitamin D levels (22 mg/mL). Extensive infectious workup including Enterovirus PCR, Herpes Simplex Virus PCR, and West Nile virus IgG was negative. Table 1 summarizes the initial workup completed. Unfortunately, SARS-CoV-2 PCR testing in CSF was not available. Notably, myelin oligodendrocyte glycoprotein (MOG) and Aquaporin 4 antibodies, as well as a comprehensive autoimmune

Table 1: Initial investigations completed

Infectious evaluation	
Serum	Herpes Simplex Virus IgG antibody: positive
	Varicella-Zoster IgG antibody: positive
	Epstein-Barr Virus IgG antibody: positive
Cerebrospinal fluid	Herpes Simplex Virus PCR: negative
	Enterovirus PCR: negative
	West Nile IgM/IgG: negative
	Cytomegalovirus PCR: negative
	Toxoplasma Gondii PCR: negative
Autoimmune, inflammatory & additional evaluation	
Serum	Anti-MOG antibody: negative
	Aquaporin-4 antibody: negative
	Autoimmune paraneoplastic panel:[7] negative
	Erythrocyte sedimentation rate: 17 mmol/hour
	D-dimer: 0.56 mcg/mL
	Fibrinogen: 139 mg/dL
	Lactate dehydrogenase: 1.7 mmol/L
	Antinuclear antibodies: negative, <1:80
	Complete blood count: within normal range
	Comprehensive metabolic panel: within normal range
Cerebrospinal fluid	Oligoclonal bands: negative
	IgG index[1]: 0.74
	Cytology: negative
	Flow cytometry: negative

MOG=myelin oligodendrocyte glycoprotein, Ig=immunoglobulin [1]IgG index is an indicator of local IgG production. It is calculated with the following formula. Cerebrospinal fluid (CSF) IgG index = (CSF IgG x serum albumin)/(CSF albumin x serum IgG)

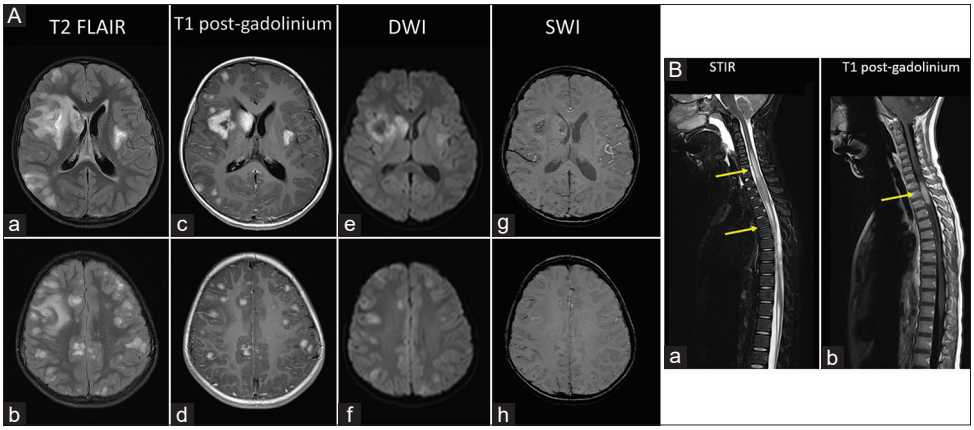

Figure 1: (A) Brain T2 FLAIR demonstrating multifocal hyperintense bilateral, patchy lesions (a and b) with contrast enhancement (c and d). Restricted diffusion is noted in some of these lesions (e and f) as well as hemorrhagic changes are noted in SWI (g and h). FLAIR: fluid-attenuated inversion recovery; DWI: diffusion-weighted magnetic resonance imaging; SWI: susceptibility weighted imaging. (B) Large confluent lesions extending the entire spinal cord and revealing longitudinally extensive myelitis (a) with an intensely enhancing, expansile intramedullary lesion from T1 to T3 (b). STIR: short-tau inversion recovery

encephalopathy panel, were negative. Given limited clinical and radiological improvement with steroids, five sessions of plasma exchange were completed. After completion of this therapy, the patient's encephalopathy and bilateral upper extremity strength improved, from 3/5 to 4+/5 on the MRC scale. The patient was discharged with plans to complete outpatient therapies.

Unfortunately, he returned to the ED a week later due to a recurrent headache. A repeat brain MRI revealed an increased periventricular T2 FLAIR signal compatible with mild hydrocephalus [Figure 2]. A repeat spine MRI also did not reveal any significant changes. Subsequently, he was placed on a five day course of high-dose IV steroids followed by a 6-week oral steroid taper and a 2 g/kg IV immunoglobulin (IVIG) course. His headache resolved and neuroimaging post-therapy revealed a resolution of the hydrocephalus. The patient remained clinically stable and was asymptomatic, however, on a follow-up brain MRI 2 months later, he again was noted to have worsening edema in the bilateral cerebellar hemispheres causing mild infratentorial herniation of the cerebellar tonsils and development of non-communicating hydrocephalus with mildly increased periventricular T2 FLAIR signal [Figure 2]. Despite the lack of overt clinical symptoms, given the concerning findings on imaging, the patient was given another five day course of high-dose intravenous (IV) steroids with a 2 g/kg IV immunoglobulin course and was discharged. Unfortunately, a month later, he had worsening headaches, and repeat neuroimaging redemonstrated recurrence and worsening of cerebellar edema, so he was admitted to the hospital for the third course of five days of high-dose IV steroids. This time, given the refractory inflammation, the patient received a rituximab infusion (approximately 500 mg/m^2). Repeat imaging one month later showed no significant change with no new lesions. The patient had no new clinical symptoms and was advised to initiate monthly IVIG infusions as adjunctive therapy with close follow-up.

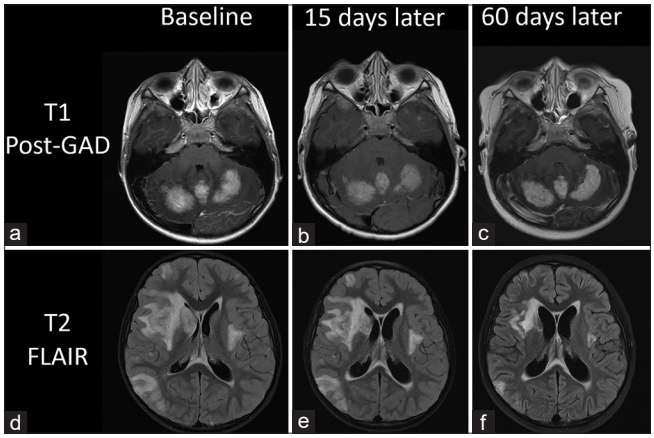

Figure 2: Longitudinal changes on brain MRI at 15 and 60 days since diagnosis. (a-c) reveal post-GAD enhancement in the cerebellum, demonstrating persistent and worsening edema at 60 days. (d-f) show T2-FLAIR lesions evolution over time. Right frontal hemisphere lesion is improved (f) when compared to baseline (d). However, there is increase periventricular signal as well as the size of the ventricles concerning for development of noncommunicating hydrocephalus which is consistent with the worsening inflammation of the cerebellum inflammation. Although not displayed in the figure, there is mild infratentorial herniation of the cerebellar tonsils through the foramen magnum. FLAIR: fluid-attenuated inversion recovery; GAD: gadolinium

He remained clinically stable on follow-up visits, but interestingly, almost one year after his presentation, his two year-old brother presented to the ED with ataxia and emesis and had similar MRI findings. On further history it was found that his parents were second cousins, leading to concerns about a genetic disorder. Genetic testing for leukodystrophies (Invitae leukodystrophy panel) was sent. Both the patient and his brother were found to have a homozygous mutation for the PRF1 gene (variant c.4422G>A). Immunological studies showed he had absent perforin expression and profoundly depressed NK cell function. The patient underwent an allogenic bone marrow transplant (BMT) with full engraftment and remains on graft versus host disease prophylaxis with tacrolimus and mycophenolate mofetil. IVIG and rituximab were discontinued post-BMT.

Discussion

This case describes CNS-isolated HLH triggered by SARS-CoV-2 in a pediatric patient. Furthermore, this is the first report of the PRF1 gene variant c.4422G>A to be a pathogenic mutation. Notably, until a genetic diagnosis was made, he was formulated as having aggressive ADEM and treated with conventional immunotherapies with transient improvement but lack of clinical and radiological remission which is highly atypical for ADEM. He had an unrevealing workup for recurrent demyelinating syndromes with negative AQP-4 and MOG antibodies. Given no better unifying diagnosis, the refractory nature of his disease was attributed to incompletely understood hyperinflammatory and vascular effects of SARS-CoV2 infection. His exceedingly refractory clinical course required escalation in immunosuppressive therapy to rituximab and IVIG. Only when his younger brother presented with a similar presentation did an investigation for an inherited process begin confirming of a homozygous PRF1 mutation, and the diagnosis of CNS-isolated fHLH.

Immune-mediated and inflammatory disorders of the CNS in the pediatric population are heterogeneous though presumably derives from immunological dysregulation affecting the CNS.[8] They can be primaries, such as ADEM, vasculitis, or antibody-mediated disorders, or secondary in the setting of a systemic process or an infection. Prompt recognition and a correct diagnosis are crucial steps to control inflammation and improve prognosis.[9] ADEM is an inflammatory demyelinating CNS disorder that usually presents with encephalopathy in children and it is thought to be triggered by infections or vaccinations. ADEM is usually monophasic but in some cases, it can be recurrent, especially when patients are anti-MOG antibody

positive (MOG-associated disease or MOGAD).[10] HLH is a rare disorder and although CNS manifestations are common, they usually occur in the setting of systemic inflammation.[5] Therefore, diagnosing CNS-isolated HLH requires a high index of suspicion. Key findings that may indicate this diagnosis are young age at presentation, consanguinity of parents, and affected siblings.[8] Interestingly, the patient in this clinical case met all three characteristics. In addition to HLH, there are other genetic disorders presenting with neuroinflammation such as interferonopathies which are a group of disorders characterized by defects in Interferon (IFN) - mediated responses. One of them is Aicardi–Goutières syndrome (AGS) which presents with early-onset encephalopathy and basal ganglia calcifications. In some cases, its clinical manifestations overlap with systemic lupus erythematosus (SLE), increasing its complexity.[11] There are other inherited leukodystrophies that can present with contrast enhancement lesions and can be misdiagnosed as primarily autoimmune such as Adrenoleukodystrophy, Alexander disease, and Krabbe disease among others.[12]

Isolated CNS-HLH as a cause of treatment-refractory neuroinflammation is an important disorder to include in patients with atypical ADEM or refractory MOGAD. It is possible that as our knowledge of neurogenetics develops, certain genes will be identified as predisposing factors to clinically recognizable syndromes such as ADEM.

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Conflicts of interest

There are no conflicts of interest.

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