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BRIEF REPORT

Childhood-onset autoimmune cytopenia as the presenting feature of biallelic *ACP5* mutations

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Abstract

Childhood-onset chronic and refractory cytopenias are rare and may be genetic in etiology. We report three pediatric cases of severe autoimmune thrombocytopenia or anemia associated with growth retardation and spastic diplegia with intracranial calcification. The identification of platyspondyly and metaphyseal lesions suggested a potential diagnosis of spondyloenchondrodysplasia (SPENCD), which was confirmed with the identification of biallelic *ACP5* mutations. Two patients demonstrated elevated serum interferon alpha levels. Our report highlights *ACP5*-associated disease as a cause of childhood-onset autoimmune cytopenia, particularly combined with growth retardation and/or spasticity. Furthermore, a role for type I interferon in the pathogenesis of autoimmune cytopenias is supported.

KEYWORDS

autoimmunity, cytopenia, interferon, spondyloenchondrodysplasia

1 | INTRODUCTION

Chronic and refractory cytopenias in pediatrics are rare and may be associated with inherited immunodeficiencies. The identification of a monogenic cause of cytopenia has implications for both family genetic counselling and treatment. Recently, biallelic mutations of the gene *ACP5* have been found to be causative of spondyloenchondrodysplasia (SPENCD) (OMIM 271550).^{1,2} SPENCD was initially described as a skeletal dysplasia manifesting as enchondromatous-like changes of the long bone metaphyses and platyspondyly, with resulting short stature.^{3,4} An associated neurological phenotype with intracranial calcification and spasticity⁵ and a predisposition to autoimmune disease, including autoimmune cytopenia is well recognized.^{6,7} Biallelic *ACP5* mutations result in a loss of activity of the encoded protein tartrate resistant acid phosphatase (TRAP) and are also associated

with an upregulation of interferon-alpha ($INF\alpha$) and type I interferon-stimulated genes.²

We describe three children, under the age of 10 years, with severe autoimmune cytopenia with a confirmed diagnosis of SPENCD and highlight the hematological characteristics, disease course, and treatment of their cytopenia.

2 | CASES

Three patients were referred to two pediatric hematology departments with features of early-onset autoimmune cytopenia (Table 1). Patient 1 presented at 5 years of age with an episode of intracranial bleeding and was shown to have profound thrombocytopenia, in addition to neutropenia. Autoimmune thrombocytopenia was subsequently diagnosed as an elevated number of megakaryocytes were identified following bone marrow smear examination (and alternative etiologies were excluded according to established guidelines).^{8,9} On examination, there was evident growth retardation and spastic diplegia, with normal psychomotor development. Intracranial calcification was detected on cranial imaging (Fig. 1) and antinuclear antibodies (ANAs) were identified. In view of the intracerebral

ABBREVIATIONS: AGS, Aicardi–Goutières syndrome; AIHA, autoimmune hemolytic anemia; ALPS, autoimmune lymphoproliferative syndrome; ANAs, antinuclear antibodies; CSF, cerebrospinal fluid; INF, interferon; ITP, primary immune thrombocytopenia; IVIG, intravenous immunoglobulin gamma; OPN, osteopontin; pDCs, plasmacytoid dendritic cells; TLR9, Toll-like receptor 9; TRAP, tartrate resistant acid phosphatase; SLE, systemic lupus erythematosus; SPENCD, spondyloenchondrodysplasia

TABLE 1 Demographic, clinical, biological, and radiological features in three patients with autoimmune cytopenia secondary to *ACP5* gene mutations

	Patient 1	Patient 2	Patient 3
Age of cytopenia (years)/sex/country of origin	5/female/Senegal	6/female/Turkey	3/male/Pakistan
Cytopenia(s)	AITP	AITP	AIHA
Family history	One sister deceased (unexplained anemia) A brother with mutation	No	Two late miscarriages (mother)
Consanguinity	No	Yes	Yes
Spastic diplegia	Yes	No	Yes
Height (SD) ^a	2	2.5	4
CBC at onset ^a :			
Hemoglobin value (10.5–14 g/dl)	6	12.2	4
Reticulocyte count ($<120 \times 10^9 \text{ l}^{-1}$)	$116 \times 10^9 \text{ l}^{-1}$	$60 \times 10^9 \text{ l}^{-1}$	$150 \times 10^9 \text{ l}^{-1}$
Neutrophil count ($\geq 1.5 \times 10^9 \text{ l}^{-1}$)	$0.7 \times 10^9 \text{ l}^{-1}$	$2.3 \times 10^9 \text{ l}^{-1}$	$1.5 \times 10^9 \text{ l}^{-1}$
Platelet count ($\geq 150 \times 10^9 \text{ l}^{-1}$)	$0.7 \times 10^9 \text{ l}^{-1}$	$4.0 \times 10^9 \text{ l}^{-1}$	$150 \times 10^9 \text{ l}^{-1}$
Autoantibodies "titer" (time elapsed from diagnosis, months)	<ul style="list-style-type: none"> - ANA "1:200" (0) - ds DNA negative - Antiplatelet abs: anti-IaIIa, anti-IbIIIa, and anti-IbIX^b (0) - Antineutrophils abs^c: anti-CD16 IgG (0) 	<ul style="list-style-type: none"> - ANA "1:5,000" (12) - ds DNA + - Antithyroglobulin abs (12) 	<ul style="list-style-type: none"> - ANA "1:400" (0) - ds DNA abs "1:1,280," Farr test: 29% (normal values <20%) (0) - IgA et IgG antiIgadin abs (16)
Skeletal radiographs:	Yes	Yes	Yes
Platyspondyly	Yes	Yes	Yes
Metaphyseal dysplasia			
Intracranial calcification	Yes	Na	Yes
Serum IFN α (N < 2 IU/l)	37	Na	55
CSF IFN α	Negative	Na	Negative
<i>ACP5</i> gene mutation	c.643G>A/p.Gly215Arg Homozygous	c.155A>C/p.Lys52Thr Homozygous	c.667C>T/p.Gln223X Homozygous

^aData at onset of autoimmune cytopenia.^bBy MAIPA enzyme immunoassay, both direct and indirect test.^cBy immunofluorescence assay, indirect test and MAIGA enzyme immunoassay, indirect test.

abs, antibodies; CBC, complete blood count; ds DNA, double-strain DNA; Na, not available.

calcification, autoimmunity and growth retardation, the diagnosis of SPENCD was considered and supported by skeletal x-rays (Table 1 and Fig. 1). An elevated level of IFN α was demonstrated in the serum but not in the cerebrospinal fluid (CSF). Subsequent genetic analysis, performed with informed consent, revealed homozygous mutations in *ACP5*. Complete hematological remission was obtained following treatment with corticosteroids with intravenous immunoglobulins (intravenous immunoglobulin gamma [IVIG]). Remission off treatment has now been sustained for 6 months.

Patient 2 presented with a petechial rash and bruising at 6 years of age, secondary to thrombocytopenia, which was autoimmune in etiology (assessed using the criteria noted above). Clinical examination demonstrated growth retardation, without neurologic impairment and both platyspondyly and metaphyseal dysplasia were shown on skeletal x-rays, in keeping with a clinical diagnosis of SPENCD (Table 1 and Fig. 1). Homozygous mutations in *ACP5* were subsequently identi-

fied. One year following initial presentation, ANAs were detected, while IFN α testing was not undertaken. Hematological remission was obtained following treatment with corticosteroids and IVIG, but a relapse of thrombocytopenia while tapering the corticosteroids required a further dose increase to achieve complete remission.

Patient 3 presented with pallor at 3 years of age and investigations revealed anemia. In view of the additional findings of a reticulocytosis and a positive direct antiglobulin test, a diagnosis of autoimmune hemolytic anemia (AIHA) was made. Growth retardation and spastic diplegia were noted on examination, with normal psychomotor development. Skeletal x-rays suggested a diagnosis of SPENCD, while cranial imaging revealed intracranial calcification (Table 1 and Fig. 1). Genetic testing demonstrated homozygous mutations in *ACP5*, identification of ANAs, and elevated level of IFN α levels in the serum but not in the CSF. Hematological remission was obtained following treatment with corticosteroids; however, tapering of the corticosteroids induced

FIGURE 1 (A) Calcification in left globus pallidus putamen and in the right temporo-occipital deep white matter (patient 1). (B) Bilateral globus pallidus calcification (patient 3). (C) Platyspondyly (patient 2). (D) Epiphyseal lesions of the proximal femur (patient 1). (E) Metaphyseal lesions of the distal femur, proximal tibia, and fibula (patient 3). (F) Metaphyseal lesions of the distal radius and ulna (patient 3).

relapse. Complete remission was achieved with the addition of two rituximab infusions (375 mg/m²) and has now been sustained for 9 years. However, a persistent hypogammaglobulinemia (immunoglobulin G <3 g/l; normal 5–11.5 g/l) and low memory-B lymphocyte count (2–4% of total CD19+ B cells; normal >10%) developed after rituximab, necessitating replacement IVIG therapy. Prior to treatment with rituximab, the patient had normal serum IgG, IgA, and IgM levels, but his lymphocyte phenotype was not studied.

3 | DISCUSSION

Herein, we reported three patients, all under 10 years of age at presentation, harboring homozygous *ACP5* mutations associated with severe autoimmune thrombocytopenia and/or hemolytic anemia.

Primary immune thrombocytopenia (ITP) is the most common cause of thrombocytopenia in childhood between 18 months and 8 years of age.⁹ Its diagnosis in children is one of exclusion, and a presumptive diagnosis is made after a careful consideration of history, physical examination, complete blood count, and examination of the peripheral blood smear.^{8,9} AIHA is the main cause of acquired extracorporeal hemolysis in children,¹⁰ characterized by the presence of autoantibodies that bind to the erythrocyte surface membrane and lead to premature red cell destruction.

The diagnosis of an inherited autoimmune cytopenia should be considered particularly in cases of parental consanguinity, where there is family history of autoimmunity and/or infections, and if there are additional physical anomalies.¹⁰ The commonest inherited condition associated with autoimmune cytopenia is autoimmune lymphoproliferative syndrome (ALPS).¹¹ Less frequently, hematological autoimmunity may occur in combined immunodeficiencies, such as Wiscott–Aldrich

syndrome or Schimke syndrome.¹² More recently, disorders in which autoimmune cytopenias are the predominant phenotype have been recognized, secondary to hypomorphic variants in genes previously reported in association with combined immunodeficiencies, including genes linked to SCID¹³ and also LRBA,¹⁴ STAT3,⁴ and CTLA4.¹⁵ Finally, monogenic interferonopathies, particularly Aicardi-Goutières syndrome (AGS) and SPENCD, have been identified as conditions predisposing to autoimmunity, with an upregulation of type I IFN, apparently central to their pathogenesis.¹⁶

Both AGS and SPENCD may present with spasticity, intracranial calcification, and an elevation of type I IFN α and interferon-stimulated genes with a preponderance to autoimmune disease, including systemic lupus erythematosus (SLE). Patients with AGS may present with severe early-onset encephalopathy with elevated IFN α in both the CSF and serum. While patients with biallelic *ACP5* mutations have spondyloenchondrodysplasia, manifesting as enchondromatous-like changes of the metaphyses in long bones and platyspondyly, associated with short stature. Growth failure and spasticity were the two cardinal features, which led us to sequence *ACP5* in our patients. Of note, both patients with neurological impairment demonstrated elevated levels of IFN α in the serum, while CSF levels were normal, in keeping with previous observation by Navarro et al.¹⁷ A marked predisposition to autoimmunity has been demonstrated in association with biallelic *ACP5* mutations^{6,18} including SLE, isolated autoimmune cytopenia, Sjögren syndrome, hypothyroidism, inflammatory myositis, and vasculitis.^{1,2,6,17,19,20} Autoimmune thrombocytopenia was the most frequent autoimmune manifestation reported (nine of 24 patients), while AIHA and autoimmune neutropenia were described in two and four patients, respectively.^{1,2} Thrombocytopenia was most frequently associated with SLE (six of nine cases).

Details regarding age of onset, severity, and treatment of thrombocytopenia have not been discussed in detail in the reports to date. In our patients, autoimmune cytopenia was of early onset and the presence of ANAs could imply a subsequent evolution toward SLE. It has been suggested that autoimmune cytopenia in SPENCD evolves more frequently toward corticosteroid dependence.¹ In two patients, complete and sustained hematological remission was only obtained following treatment adaptation. However, a marked hypogammaglobulinemia developed in patient 3 treated with rituximab, a phenomenon recognized in patients with ALPS.²¹ Although a conclusion cannot be drawn from a single patient, careful immunological evaluation is recommended before and after treatment with rituximab, which may be best avoided if alternatives are available.

The pathogenesis of autoimmunity in SPENCD is not yet completely understood. However, it is known that biallelic mutations in *ACP5* result in a marked decrease in TRAP expression and activity in the serum and leukocytes.^{1,2} It is postulated that this absence of phosphatase activity results in reduced dephosphorylation of the highly phosphorylated glycoprotein osteopontin (OPN). OPN is a multifunctional protein with an intracellular isoform, which, in mice, has been shown to form a signalsome with the innate pattern recognition receptor Toll-like receptor 9 (TLR9) and signal to IFN α production in plasmacytoid dendritic cells (pDCs).^{22,23} Thus, a functional excess of intracellular phospho-OPN might be the basis of the autoimmune disease in SPENCD through exaggerated TLR9 signaling and increased IFN α production by pDCs. This hypothesis may be supported by the demonstration of elevated levels of IFN α and a type I IFN expression signature in patients with biallelic *ACP5* mutations.² Interestingly, in AGS, thrombocytopenia is the most common autoimmune manifestation.¹⁶ Thus, the type I IFN pathway may have a key role in the pathogenesis of a subset of autoimmune thrombocytopenia cases, as it has in the pathogenesis of SLE autoimmunity.²⁴

In conclusion, *ACP5* mutations should be considered in childhood-onset autoimmune cytopenia, especially if growth retardation or spasticity are present. Furthermore, we postulate that the type I IFN pathway may play a key role in the pathogenesis of a subset of autoimmune cytopenias.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest. The authors have no financial relationships relevant to this article to disclose.

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