Mutation in the proline-serine-threonine phosphatase-interacting protein 1 (PSTPIP1) gene in a patient with acute lymphoblastic leukemia

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Abstract

Autoinflammatory syndromes are disorders characterized by recurrent or chronic inflammation caused by the dysregulation of the innate immune system. Hemophagocytic lymphohistiocytosis (HLH) is an aggressive and life-threatening syndrome of overactivation of the immune system.

We present a case of a 20-month-old boy who was referred to an oncology clinic because of HLH suspicion. In the preceding time, our patient suffered from a severe form of chickenpox with prolonged fever. Tests including myelogram, cerebrospinal fluid, and magnetic resonance (MR) of the brain gave a diagnosis of acute lymphoblastic leukemia from B lymphocyte precursors, without occupying the central nervous system. To exclude inherited HLH in our patient, next-generation sequencing was performed, which revealed a heterozygous missense mutation in exon 15 of the PSTPIP1 gene (c.1213C>T, R405C). No mutations of genes associated with familial HLH syndrome were found.

Our patient may be evidence that autoinflammatory diseases caused by PSTPIP1 gene mutations are not limited to the classical pyogenic arthritis, pyoderma gangrenosum, and acne (PAPA) phenotype but may have a different clinical presentation, and the spectrum of the PSTPIP1-associated inflammatory diseases (PAID) syndrome is more extensive than previously thought.

Key words: hemophagocytic lymphohistiocytosis, lymphoblastic leukemia, autoinflammatory syndrome.

Introduction

Autoinflammatory syndromes are a heterogeneous group of disorders characterized by recurrent or chronic inflammation caused by dysregulation of the innate immune system. Most of these diseases are very rare. Autoinflammatory syndromes are caused by genetic mutations in molecules that take part in regulation of the innate immune response. Monogenic inheritance is common [1]. In such cases, family history is relevant.

Immune dysregulation should be suspected in patients with an unusual presentation of infections — lasting longer, more severe than usual, atypical, with prolonged fever. The occurrence of a variety of inflammatory diseases in the family is a risk factor for autoinflammatory disorders as well. The main symptoms of autoinflammatory syndromes are prolonged fever, skin inflammation, and joint involvement, and patients develop them gradually through life. Inflammatory markers are usually elevated without a source of infection [1, 2].

Hemophagocytic lymphohistiocytosis (HLH) is an aggressive and life-threatening syndrome of excessive immune activation [3, 4]. It affects mostly children but can also occur in adults. Primary HLH has an underlying genetic disorder, and secondarily occurs as a response to another condition — cancer, infection, chemotherapy. Both primary and secondary HLH can be triggered by infections. HLH should be regarded as a typical final phenotype secondary to a range of diverse underlying molecular diagnoses rather than as a distinct disease. Many other autoimmune syndromes can have a similar phenotype [5, 6].

According to guidelines of the International Histiocyte Society, a diagnosis of HLH requires at least five of the following eight criteria: fever, splenomegaly, cytopenia, hypertriglyceridermia or hypofibrinogenemia, hyperferritinema, elevated soluble interleukin-2 receptor α (IL-2Rα),
decreased natural killer (NK) cell activity, and hemophagocytosis in the bone marrow [3, 4, 6]. Details are presented in Table 1. Proper diagnosis of autoinflammatory syndromes (including HLH) is necessary not only for treatment but also to establish the prognosis. Untreated HLH is always 100% fatal. Definite diagnosis of autoinflammatory syndromes is based on genetic testing, which is not commonly available.

Case description

A 20-month-old male patient was referred to the oncology clinic for evaluation of a possible HLH based on his laboratory test results. The patient was born at 40 weeks gestation to a healthy 34-year-old G3P3 mother, delivery by cesarean section, birth mass 3.8 kg, Apgar score of 10. The boy was vaccinated without complications until the age of 14 months according to the Polish schedule (https://szczepienia.pzh.gov.pl/en/immunization-schedule/) – BCG and hepatitis B – 24 hours after birth, 2 and 7 months, DTP 2, 3, 5 months, Hib 2, 4, and 6 months, PVC 2, 4 and 13 months, IPV 3, and 6 months, MMR 14 months. The boy was not vaccinated against VZV (it is not obligatory in Poland). He was previously diagnosed with atopic dermatitis and cows’ milk allergy. The parents are unrelated, and both are healthy. The family history concerned in particular the oldest brother who is an asymptomatic carrier of the heterozygous CFTR gene variant (F508del, and L467F) (diagnosed through newborn screening), which he inherited one from his mother and one from the father. Our patient’s mother’s sister was diagnosed with Wilson’s disease at the age of 25 (carrier of heterozygous H1069Q and Q1351X mutations in ATP7B gene), and additionally at the age of 41 with Crohn’s disease. Of the five siblings of the patient’s mother, Wilson’s disease was also confirmed in the mother’s youngest brother. Our patient and his mother are asymptomatic carriers of the H1069Q variant in the ATP7B gene. The boy is also an asymptomatic carrier of heterozygous variants in the CFTR gene, i.e., F508del and L467F. The rest of the family members, including both siblings, are healthy.

The current disease began a month earlier with enterocolitis. Five days before chickenpox, he was diagnosed with pharyngitis – treated with first-generation cephalosporin. The first hospitalization was on day 5 of fever in the course of chickenpox. The source of infection was unknown – family members were healthy, siblings and the patient himself did not attend kindergarten. On admission, physical examination revealed a rash on the trunk and swelling of hands and feet. Inflammatory markers were not clearly elevated (details are presented in Table 1). Chickenpox was severe, with many lesions and prolonged fever. VZV-DNA by PCR was found in the skin lesion scraping. He was treated with antibiotics and acyclovir for seven days. He had a fever for ten days in total. After four days without a fever, he was sent home. The day after discharge, he returned with the fever without any other symptoms. Laboratory tests on readmission are shown in Table 2. Seasonal infections such as influenza and respiratory syncytial virus (RSV) were excluded. He was empirically treated with 3rd generation cephalosporin. During the next four days, enlargement of the liver and spleen was increasing, anemia and thrombocytopenia appeared, and the level of ferritin and triglycerides had increased. HLH was suspected, and the child was referred to the oncology department.

On admission to the oncology ward, the boy was in a bad condition, he had a fever above 39°C, a rash on the trunk, swelling of the hands and feet, generalized lymphadenopathy, liver enlargement about 2 cm below the navel and spleen about 2 cm below the rib arch. Slight swelling of the upper eyelids was also found. Findings of bone marrow and lumbar puncture were not consistent with HLH. Myelogram showed 91% of cells with L1 morphology. Flow cytometric analysis revealed 86% blast CD19+, CD34+, cCD79α+, CD10+, TdT+, CD58+, CD38+, cIgM–, sIgM–, CD22+/-dim, CD20+/-.. The diagnosis of acute lymphoblastic leukemia from precursors of B lymphocytes, SR groups, without the involvement of the central nervous system, was made. Hemophagocytosis in bone marrow, count per 1.000 nucleated cells, was low – 0.01%. TEL-AML1, BCR-ABL, SIL-TAL1, KMT2A rearrangements have not been observed in leukemic cells. The study with CytoScan HD Affymetrix did not confirm the deletion of the IKZF1, PAX5, PAR1, and ERG gene. However, the bi-allelic deletion of CDKN2A and CDKN2B genes was found. 24-hour leukemic cell culture showed abnormal male karyotype – 46,XY, inv(1)(p13p36.3),del(9)(p21),del(13)(q32).

Treatment was consistent with the AIEOP-BFM pB ALL 2017 scheme. Chemotherapy was complicated by

| Table 1. Hemophagocytic lymphohistiocytosis (HLH) diagnostic criteria [6] |
|-----------------------------------------|
| **Five of the following eight findings** |
| Fever ≥ 38.5°C                          |
| Splenomegaly                            |
| Bicytopenia                             |
| Hemoglobin < 9 g/dl                     |
| platelets < 100,000/µl                  |
| absolute neutrophil count < 1000/µl    |
| Hypertriglyceridemia or hypofibrinogenemia (fasting triglycerides > 265 mg/dl, fibrinogen < 150 mg/dl) |
| Ferritin > 500 mcg/l                    |
| Low/absent NK cell activity             |
| Soluble CD25 elevation > 2,400 U/ml    |
| Hemophagocytosis in bone marrow, spleen, lymph node, or liver, or a molecular diagnosis consistent with HLH |

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infection (molluscum contagiosum), hepatotoxicity, neurotoxicity, cardiotoxicity (sinus bradycardia), and endovascular clotting (Leiden mutation, as well as mutations in prothrombin, and MTHFR genes were excluded). On day 15, hematological remission was obtained, and on day 33, remission was confirmed by flow cytometry and molecular tests. The patient remains in full remission of leukemia, continues treatment, molluscum contagiosum lesions are still observed but to a lesser extent. To exclude congenital HLH, a genetic test (by next-generation sequencing in a range of 5227 genes included in the SureSelect Custom Constitutional Panel, Agilent Technologies) was performed. The presence of a heterozygous missense mutation in exon 15 of the PSTPIP1 gene (c.1213C>T, R405C) and two heterozygous variants in the CFTR gene, i.e., F508del and L467F, was identified. Gene mutations associated with inherited HLH syndrome were not found. Neither parent carries this PSTPIP1 variant.

Discussion

The course of the disease in our patient and the results of the tests – recurrent and prolonged fever, enlarged liver and spleen, increased anemia and thrombocytopenia, high levels of ferritin and triglycerides – suggested HLH. The patient fulfilled four out of six tested HLH-2004 criteria. However, further studies have shown leukemia and a heterozygous missense mutation in the PSTPIP1 gene while excluding gene mutations associated with congenital HLH.

Proline-serine-threonine phosphatase-interacting protein 1 (PSTPIP1; also known as CD2 binding protein 1-CD2BP1) is a cytoskeletal protein within hematopoietic cells that serves as a scaffold for the binding of other cellular proteins, such as pyrin, protein tyrosine phosphatases, c-Abl, CD2, and WASP (Wiskott-Aldrich syndrome protein) [5, 6]. PSTPIP1 regulates several cellular functions, including IL-1β release, cytoskeleton organization, cell migration, and T-cell activation cytoskeleton-associated adaptor protein that modulates T-cell activation, cytoskeletal organization, and IL-1β release [7-9].

Table 2. Laboratory tests results during hospitalizations. The results in bold are outside the limits of the normal value

| Parameter | Value on 1st pediatric admission | Value on 2nd pediatric admission | Value on oncology admission | Normal values |
|-----------|----------------------------------|----------------------------------|-----------------------------|---------------|
| Hemoglobin (g/dl) | 10.4 | 9.0 | 8.8 | 10.1-13.0 |
| Erythrocytes (10⁹/µl) | 4.03 | 3.48 | 3.21 | 3.9-5.10 |
| Platelets (10⁹/µl) | 223 | 156 | 55 | 200-550 |
| Leukocytes 10⁹/µl | 14.3 | 28.1 | 6.4 | 3.80-10.0 |
| Neutrophils (10⁹/µl) | 13.2 | 13.2 | 2.1 | 2.0-7.0 |
| Neutrophils (%) | 92.31 | 46.98 | 32.81 | 35-55 |
| Albumin (g/l) | 36 | 34 | 21 | 35-52 |
| CRP (mg/l) | 10.64 | 8.5 | 3.25 | 0-5 |
| Procalcitonin (ng/ml) | 0.37 | 9.96 | 4.52 | < 0.05 |
| IgG (g/l) | 5.73 | ND | 5.12 | 4.53-9.16 |
| IgM (g/l) | 1.68 | ND | 1.71 | 0.19-1.46 |
| IgA (g/l) | 0.66 | ND | 1.24 | 0.2-1.00 |
| IgE (EU/ml) | 276.6 | ND | 281.4 | 0-60 |
| AST (U/l) | 120 | 313 | 385 | 0-32 |
| ALT (U/l) | 51 | 58 | 59 | 0-33 |
| CPK (U/l) | 39 | 51 | 47 | 29-168 |
| LDH (U/l) | 247 | 2583 | 3284 | 125-243 |
| Ferritin (ng/ml) | 210 | 18444 | 18284 | 20-150 |
| Fibrinogen (g/l) | 3.1 | 4.1 | 1.8 | 1.80-3.05 |
| D-dimers (mcg/ml) | 0.3 | 0.5 | 2.7 | < 0.4 |
| Triglycerides (mg/dl) | 90 | 251 | 201 | 0-150 |

CRP – C-reactive protein, AST – asparagine aminotransferase, ALT – alanine aminotransferase, CPK – creatine phosphokinase, LDH – lactate dehydrogenase.
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