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Case report

Hemophagocytic lymphohistiocytosis: a case series of a Brazilian institution



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ABSTRACT

Objective: To describe the clinical and laboratory presentation of hemophagocytic lymphohistiocytosis in children treated at a referral institution.

Methods: A retrospective descriptive study was carried out of seven children diagnosed with hemophagocytic lymphohistiocytosis between 2010 and 2012. The criteria for diagnosis were those proposed by the Histiocyte Society. When indicated, immunochemotherapy was prescribed according to the HLH94 and HLH2004 protocols of the Histiocyte Society.

Results: The patients' ages at diagnosis ranged from one month to nine years. All patients had splenomegaly, fever, anemia, thrombocytopenia, hyperferritinemia and hypertriglyceridemia. Bone marrow hemophagocytosis was detected in six patients. In six cases, infectious diseases triggered the syndrome. In two cases, associated with visceral leishmaniasis, remission was achieved after treatment of the underlying infection. Three patients, who had Epstein-Barr-related hemophagocytic lymphohistiocytosis, required treatment with immunochemotherapy. They are alive and in remission; one patient had symptoms of juvenile rheumatoid arthritis and another, who was suspected of having primary hemophagocytic lymphohistiocytosis, entered into remission after bone marrow transplantation. Two deaths (28.6%) occurred in patients with suspected primary hemophagocytic lymphohistiocytosis; one whose clinical picture was triggered by cytomegalovirus infection did not respond to immunochemotherapy and the other died before any specific treatment was provided.

Conclusion: As reported before, hemophagocytic lymphohistiocytosis has a multifaceted presentation with nonspecific signs and symptoms. In secondary forms, remission may be achieved by treating the underlying disease. In the primary forms, remission may be achieved with immunochemotherapy, but bone marrow transplantation is required for cure.

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Introduction

Hemophagocytic lymphohistiocytosis (HLH) is an immune hyperactivation syndrome characterized by clinical signs and symptoms of severe uncontrolled inflammation.¹

Diagnosis is based on combined clinical, laboratory, genetic, and morphological criteria. According to the Histiocyte Society,² in the absence of family history or of specific genetic tests, five out of eight criteria must be present in order to establish a diagnosis and initiate treatment (Table 1).

Common findings in HLH include persistent fever, hepatosplenomegaly, cytopenia, decreased activity of cytotoxic T lymphocytes and natural killer cells (NK), as well as widespread accumulation of lymphocytes and macrophages that carry out hemophagocytosis.³

HLH is included in the differential diagnosis of a number of diseases commonly found in children, such as autoimmune diseases, primary immunodeficiencies, malignancies, and infectious diseases.³

There are two main forms, primary or familial HLH and secondary HLH. Secondary HLH can be associated with infections, autoimmune disorders or malignancies. Despite attempts to differentiate between primary and secondary forms of HLH, their clinical features are very similar.^{4,5}

Remission may be achieved in primary HLH with immunochemotherapy and in cases of secondary HLH by treating the underlying disease.⁶⁻⁸ However, all patients with the familial form of HLH relapsed or died until treatment using allogeneic bone marrow transplantation (BMT) started to be used.^{2,6}

Given its rarity and the scarcity of data in the Brazilian literature, this retrospective study was carried out to describe the clinical and laboratory presentation of HLH in children followed at the Hematology Service of the Hospital das Clínicas, Universidade Federal de Minas Gerais (HC-UFMG) between January 2010 and July 2012.

The study was approved by the Institution's Research Ethics Committee (protocol no. 05683012.9.0000.5149), and patients and guardians were asked to sign informed consent forms.

Case report

Seven children were included. Age at diagnosis ranged from one month to nine years, with a median of 13 months. All patients presented with splenomegaly and fever. Other clinical manifestations are shown in Table 2. Only one case (#7) had a family history suggesting HLH.

All children had anemia, thrombocytopenia, hypertriglyceridemia, and hyperferritinemia at the time of diagnosis. Hypofibrinogenemia was observed in six cases (85.7%). Hemophagocytosis features were visible in the bone marrows of six patients (85.7%) at diagnosis (Table 2). Lumbar puncture with cerebrospinal fluid (CSF) analysis was performed in two cases; both had pleocytosis. Cranial computed tomography was performed in one patient (#4) which revealed diffuse cortical atrophy.

The estimated time between the onset of symptoms and HLH diagnosis ranged from 10 to 60 days (median of 50 days).

The initial investigation showed that in six cases (85.7%) the clinical picture was prompted by infectious diseases, two of which (33.3%) were secondary to visceral leishmaniasis, three (50%) after infections with Epstein-Barr virus (EBV) and one (16.7%) triggered by cytomegalovirus (Table 2).

Four cases were considered secondary forms: two had visceral leishmaniasis (#1 and #3) and entered into remission after the infection was treated with liposomal amphotericin B but without specific therapy for HLH. The initial clinical picture of the patient with positive serology (IgM) for EBV was severe; he received induction therapy following the HLH94 protocol, and entered into remission after eight weeks (#5). The last patient (#6) had a prior diagnosis of juvenile rheumatoid arthritis and developed HLH secondary to the underlying disease (macrophage activation syndrome) triggered by EBV infection. The child received induction therapy following the HLH2004 protocol and entered into remission after eight weeks. The secondary form cases are currently being followed as outpatients and continue in remission.

After a retrospective review, the other three cases were considered likely to have been primary forms of HLH. Data that contributed to this suspicion were: age at diagnosis (all were infants), no response to immunochemotherapy (in two cases), and in the third case, history of siblings dying with similar clinical presentations.

Two patients (#2 and #4) received immunochemotherapy (Histiocyte Society HLH2004 and HLH94 protocols)^{2,7,8} and failed to achieve remission after the induction and maintenance phases. The first patient (#2) underwent unrelated allogeneic bone marrow transplantation and responded well. He is currently being followed in the outpatient clinic. After a seven-month follow-up, the second patient (#4) died from severe sepsis of a pulmonary focus with active disease.

The third patient (#7) aged one month and with a family history of death of siblings from rapid infectious complications, died 48 h after admission, presenting with severe sepsis of an unknown etiologic agent before there was enough time

Table 1 – Criteria for the diagnosis of hemophagocytic lymphohistiocytosis.

- A. Molecular diagnosis compatible with hemophagocytic lymphohistiocytosis
OR
- B. At least five out of the eight following criteria:
1. Fever
 2. Splenomegaly
 3. Cytopenias (two or more lineages)
 - Hemoglobin <9 g/dL or <10 g/dL in newborn babies
 - Platelets <100 × 10⁹/L
 - Neutrophils <1.0 × 10⁹/L
 4. Hypertriglyceridemia (>265 mg/dL) or hypofibrinogenemia (<150 mg/dL)
 5. Hemophagocytosis in bone marrow, spleen, lymph nodes or liver without any evidence of malignancy
 6. Decreased or absent activity of natural killer cells
 7. Serum ferritin >500 µg/L
 8. Increased soluble CD25 (>2400 U/mL)

Modified from Henter et al.²

Table 2 – Clinical and laboratorial characteristics of seven children with hemophagocytic lymphohistiocytosis (HLH).

#	Gender	Age at diagnosis	Enlarged lymph nodes	Cutaneous rash	Jaundice	Fibrinogen (mg/dL)	Hemophagocytosis in bone marrow	Spinal fluid exam	Infections and associated diseases	Type of immunochemotherapy/present clinical status
1	F	5 months	No	No	Yes	89	No	ND	Visceral leishmaniasis	ND/Alive in remission
2	M	13 months	No	No	No	>400	Yes	Pleocytosis	Epstein-Barr virus	HLH 2004/Bone marrow transplantation in July 2012; alive in remission
3	M	5 months	No	No	No	97	Yes	ND	Visceral leishmaniasis	ND/Alive in remission
4	M	16 months	Yes	Yes	Yes	131	Yes	ND	Cytomegalovirus	HLH 1994/No remission. Death after sepsis
5	M	3 years	Yes	No	Yes	59	Yes	ND	Epstein-Barr virus	HLH 1994/Alive in remission
6	F	9 years	No	Yes	Yes	94	Yes	Pleocytosis	Epstein-Barr virus/Juvenile Rheumatoid Arthritis	HLH 2004/Alive in remission
7	M	1 month	No	No	No	35	Yes	ND	None proven	ND/Death after 48 h from hospital admission

F: female; M: male; ND: not done.

to start specific treatment. Results from the autopsy suggested HLH.

Mortality among patients in the study was 28.6% (2/7) and both deaths occurred in patients suspected of having primary HLH.

Discussion

HLH has multifaceted presentations with nonspecific signs and symptoms that are found in other clinical conditions. In the present study the most frequent clinical and laboratory findings at diagnosis were prolonged fever, splenomegaly, and cytopenias, particularly anemia and thrombocytopenia, as has already been described in the literature.³

Hemophagocytosis, the phenomenon that lends its name to the nosological entity, is not a prerequisite for the clinical diagnosis of HLH.⁶ This phenomenon was observable at some point during the follow-up in all but one case of this series. Nevertheless, absence of hemophagocytosis should not delay the diagnosis when the initial clinical picture fulfills the other criteria.³ This morphological phenomenon can also be induced by other events such as blood transfusion, infection, autoimmune diseases, and some types of bone marrow failure.⁶

Despite the limited access to more complex laboratory tests, common to many institutions where molecular genetic studies and assessment of NK cell activity and soluble CD25⁶ are not available, HLH diagnosis was possible in the seven reported cases because each fulfilled at least five of the eight criteria described by the Histiocyte Society.² The absence of molecular studies, however, prevented confirmation of the diagnosis of primary HLH cases, essential for predicting the risk of recurrence and for defining disease predisposition in asymptomatic family members.⁶

In this case series, only three patients underwent investigation for changes in the central nervous system (CNS). In two of these cases, the CSF was examined, and pleocytosis was detected in both, with atypical cells present in one. Neurological abnormalities are a prominent feature of this disease. CNS involvement can cause severe and irreversible damage. It is recommended that all patients should be submitted to neurological evaluations with lumbar puncture even when asymptomatic.^{6,9}

Familial HLH is considered an autosomal recessive genetic disorder with an estimated prevalence of 1/50,000 live births. Approximately 70–80% of patients with familial HLH have symptoms in the first year of life.³ Although cases with documented genetic defects are, by definition, primary, infectious processes appear as the trigger in the majority of these children. Because it is a recessive disorder, negative family history is common and results in such cases are wrongly defined as secondary, especially when initial treatment induces satisfactory clinical remission.⁵ Thus, the use of the terms primary and secondary HLH is not ideal.

In the present study, patients with suspected familial HLH manifested symptoms within the first two years of life. In two cases, a related infectious disease was identified. The patient whose diagnosis was established in the second month of life was the most severe case and rapidly progressed to death.

As described in the literature, the younger the patient is at diagnosis, the greater the severity of the presentation.^{2,6,8}

Six cases were related to infections, which also concurs with descriptions in several reviews on the topic.^{2,4,6,7} According to the Histiocyte Society study HLH94 and many other studies, the most common infectious trigger is the EBV; in these cases, the initial clinical picture is typically severe.^{6,7,10} However, there are reports that clinical presentation of HLH secondary to EBV infection is very variable, ranging from spontaneously resolved inflammation to extremely severe cases that require immunochemotherapy¹⁰ followed by stem cell transplantation.² Among the patients in this sample who had EBV-related HLH, all had very severe initial clinical presentations and required immunochemotherapy to control signs and symptoms.

In this series, two cases were associated with visceral leishmaniasis. Both achieved HLH remission after the underlying infection was treated and required no anti-inflammatory or cytotoxic therapies, also in agreement with the literature. It is important to note, however, that without treating the underlying infection, the mortality rate is very high.³

Macrophage activation syndrome occurred in one patient previously diagnosed with juvenile rheumatoid arthritis, the autoimmune disease most commonly associated with HLH. The patient initially presented with severe hepatitis and coagulopathy, very common findings in this specific group of patients.³

The immediate goal of treatment of patients with HLH is to suppress the hyperinflammation, responsible for the symptoms that put the patient's life at risk. Thus, chemotherapeutic and/or immunosuppressive agents such as corticosteroids, cyclosporin, and immunoglobulin are used. In familial HLH, the ultimate goal should be stem cell transplantation to substitute the defective immune system with properly functioning effector cells. Without treatment, uncontrolled hyperinflammation leads to persistent neutropenia and death from repeated fungal and bacterial infections or multiple organ failure.^{4,8}

A retrospective review of this series revealed that complete response after immunochemotherapeutic treatment was achieved only in patients with a diagnosis of secondary HLH. Although effective in prolonging the survival of patients with familial HLH, it is known that immunochemotherapy alone cannot cure this form of the disease.⁶

The mortality rate in our study was 28.6%, lower than the death rate described by the Histiocyte Society, which is around 50%.^{2,6,7} The number of cases in our series, however, is very small. If we consider that all deaths occurred in patients with primary HLH and of those, only the one who underwent BMT is alive, the data from this study is in agreement with the literature, which gives a 100% mortality rate for patients with primary HLH who are not submitted to BMT.^{3,6,10}

Despite the small sample size, test limitations, and the retrospective nature of this study, all of which are factors that prevent more definitive conclusions, the cases presented here demonstrate the importance of raising awareness about HLH among professionals involved in the care of pediatric patients. Access to diagnostic procedures that allow a more definite diagnosis of suspected cases of familial HLH are evidently needed, both for the patient and for conducting family

counseling. We should emphasize the need for prospective and cooperative studies in order to enhance understanding about HLH in Brazil, so as to improve its clinical management.

Conflicts of interest

The authors declare no conflicts of interest.

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