HAEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS IN A GHANAIAN CHILD

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SUMMARY
We report a case of a previously well nine-month-old infant who presented with prolonged fever, hepatosplenomegaly and pancytopenia. A diagnosis of haemophagocytic lymphohistiocytosis (HLH) was made during the course of hospital admission. There was good initial response to dexamethasone but the patient died less than two months after diagnosis. This is the first report of HLH from Ghana. The disease has a significant mortality rate if untreated and a high index of suspicion is required in all severely ill children.

Key words: Haemophagocytic lymphohistiocytosis, children, hyperinflammation, pancytopenia, dexamethasone

INTRODUCTION
Haemophagocytic lymphohistiocytosis (HLH) is a rare but potentially fatal disease in which there is an exaggerated though ineffective immune response.[1] Although the pathogenesis of HLH is still not fully clear, the most commonly implicated pathway involves defects in perforin-mediated cell lysis, a key function of cytotoxic T cells and natural killer (NK) cells.[1,2] The clinical manifestations of HLH result from over activation of CD8+ T lymphocytes and macrophages, proliferation and infiltration of these cells into various organs and excessive cytokine production.[3]

Historically, two forms of HLH are recognized: primary (or genetic) HLH and secondary (or acquired) HLH.[1] Primary HLH can either be seen in association with inherited immunodeficiency disorders such as X-linked lymphoproliferative syndrome, Chediak-Higashi syndrome and Hermansky-Pudlak syndrome Type II, or may occur in isolation, as the sole manifestation of disease.[1]

Mutations involving the perforin gene, syntaxin 11, Munc 13-4 and Munc 18-2 have been identified in patients with primary HLH.[4] Secondary HLH is associated with malignancies, rheumatologic disorders and infections.[3]

Although various infectious agents (viruses, bacteria, fungi and parasites) can trigger HLH, Epstein Barr virus (EBV) is the most commonly associated infectious cause.[1,3]

Based on the Histiocyte Society HLH-2004 guidelines, a diagnosis of HLH can be established if there is either a molecular diagnosis consistent with HLH or if five out of the following eight criteria are fulfilled: fever; splenomegaly; cytopenias affecting ≥2 of 3 lineages in the peripheral blood: haemoglobin <9g/dl (in infants <4 weeks: haemoglobin <10g/dl), platelets <100 x 10^9/L and neutrophils <1.0 x 10^9/L; hypertriglyceridemia (fasting triglycerides ≥3.0 mmol/L) and/or hypofibrinogenemia (fibrinogen ≤ 1.5g/L); haemophagocytosis in bone marrow or spleen or lymph nodes, with no evidence of malignancy; low or absent NK-cell activity (according to local laboratory reference); ferritin ≥500µg/L; and soluble CD25 (i.e. soluble IL-2 receptor) ≥2400U/ml.[5] Supportive criteria include cerebrospinal fluid (CSF) pleocytosis (lymphocytes, macrophages) and/or elevated CSF protein, neurologic symptoms (e.g. lethargy, cranial nerve palsies, seizures), conjugated hyperbilirubinemia and transaminitis, skin rash, hypoalbuminemia, hyponatremia, elevated D-dimers and lactate dehydrogenase.[1,6]

Diagnosis of HLH is often delayed or missed due to several factors including physicians being unaware of the disease entity, non-specific clinical features, variable clinical course and unavailability of several diagnostic tests, particularly in resource-limited settings.[3,7] Both primary and secondary HLH can be rapidly fatal and must be recognized early to enable institution of appropriate therapy.[1]

CASE REPORT
A 9 month old previously healthy, female infant, born to non-consanguineous parents, presented with a two-week history of fever and diarrhoea.
There was no vomiting but she was feeding poorly and had cough and rhinorrhoea. Pregnancy and delivery

history were uneventful. Birth weight was 3.2kg and she had been exclusively breastfed for 6 months. Her immunizations were up to date and developmental milestones were appropriate for her age. Family history was non-contributory. On initial examination, she was pale and febrile (37.8^{o}C, axillary temperature) and looked acutely unwell.

She was fully conscious but lethargic. Abdominal examination revealed hepatomegaly and splenomegaly of 6cm and 5cm below the right and left costal margins, respectively. The rest of the systemic examination was normal. Provisional differential diagnoses included enteric septicaemia and sickle cell disease. She was started on intravenous ceftriaxone and intravenous fluids. Initial laboratory results were as follows: haemoglobin 5.2 g/dl, white blood cell count 13.1 x 10^9/L (with absolute neutrophil count of 0.3 x 10^9/L), platelets 17 x 10^9/L, ESR 11mmfall/hour and haemoglobin genotype AA.

Blood film for malaria parasites was negative. Serum electrolytes, urea and creatinine levels were normal for age. Liver function tests showed decreased serum albumin (24g/L), increased conjugated bilirubin (12µmol/L) and aspartate transaminase (213U/L), and normal alanine transaminase (36U/L). Over the next few days, she developed facial puffiness, diffuse hypopigmented skin lesions, gum bleeding and bleeding from phlebotomy sites. She was severely pale and had remained persistently febrile (up to 40°C). She received red cell and platelet transfusions.

Blood and urine cultures were negative for bacterial growth. HIV testing was also negative. The CSF contained 21 cells/µL (mostly lymphocytes) and CSF protein level was elevated - 0.9g/L (normal range: 0.15 – 0.45g/L). A bone marrow aspirate was performed five days after admission to exclude acute leukaemia and also look for evidence of haemophagocytosis. No leukaemic blasts were seen but numerous haemophagocytes were present in the marrow.

Based on clinical findings thus far, the patient was started empirically on IV dexamethasone 10mg/m^2 daily, with supportive therapy (ranitidine, co-trimoxazole and fluconazole). Subsequent laboratory investigations were as follows: serum ferritin 1374µg/L (normal 13-150µg/L) and triglycerides-2.68 mmol/L (normal - <1.69mmol/L). EBV testing was not done. Fibrinogen testing, NK-cell function and soluble IL-2 testing were unavailable.

The patient defervesced within 24 hours of starting dexamethasone and continued to show significant clinical improvement over the next few days. She was discharged home after a total of 12 days of hospitalization. On two subsequent follow up visits, she remained clinically well with resolution of hepatosplenomegaly, improvements in her blood counts and normalization of her serum ferritin levels (Table I). Her steroid doses were tapered from week 3 as per the HLH-2004 guidelines for de-escalating dexamethasone in the initial phase of therapy.[5]

Table I Full blood count and serum ferritin results, prior to and while on dexamethasone therapy

Timeline / Laboratory test	Baseline (initial presentation)	First week of admissi (prior to starting dexamethasone)	Week one of dexamethasone therapy	Week three of dexamethasone therapy
Haemoglobin (g/dL)	5.2	7.8*	9.1	11.4
Platelet count (x 10^9/L)	17	37**	42	89
Absolute neutrophil count (x 10^9/L)	0.3	0.7	0.7	2.1
Serum Ferritin (µg/L)		1374	Not done	113

**** Post-transfusion with whole blood***
*****Post-transfusion with platelet concentrate***

Five weeks after commencement of steroids (at which time her dexamethasone dose was at 2.5mg/m^2/day), she was rushed into the emergency room unconscious and pale, with a temperature of 37.6°C. Random blood sugar was 10.1 mmol/L. She had a hepatomegaly of 6cm and a markedly enlarged spleen, 10cm below the left costal margin. Laboratory tests done showed haemoglobin 8.0 g/dl, white blood cell count 5.4 x 10^9/l (neutrophils – 0.5), platelets 27 x 10^9/L and negative blood film for malaria parasites. Conjugated bilirubin was high (23 µmol/L), as were the liver transaminases (alanine aminotransferase, 67 IU/L and aspartate aminotransferase, 328 IU/L). Serum ferritin level was markedly elevated (7917µg/L).

The dexamethasone dose was immediately increased to 10mg/m^2/day for suspected relapsed HLH and she was started on broad spectrum intravenous antibiotics for presumed sepsis, but unfortunately passed away three hours after presentation.

DISCUSSION

The HLH diagnostic criteria that our patient met were fever, splenomegaly, cytopenias, elevated serum ferritin and the presence of haemophagocytes in the bone marrow. Supportive criteria that were present included conjugated hyperbilirubinemia, transaminitis, abnormal bleeding, skin rash, CSF pleocytosis and elevated CSF protein. Fasting triglyceride level, though elevated in our patient, did not meet the specified threshold of 3.0mmol/L.[5] Testing for fibrinogen level, soluble CD 25 and NK cell activity are not readily available in Ghana and so could not be determined.

Uncontrolled immune activation and hypercytokinaemia are responsible for most of the presenting features of HLH.[8] However, no single clinical feature is diagnostic and a high index of suspicion is required to make a diagnosis. Due to its non-specific clinical presentation, children with HLH may be diagnosed as having sepsis, systemic inflammatory response syndrome (SIRS), hepatitis, acute liver failure, multi-organ dysfunction or metabolic disorders.[3,4,7] To complicate matters further, sepsis and HLH may co-exist, as the former often triggers the latter.[4]

Failure of patients to respond to first line therapy and progressive clinical deterioration, despite maximal therapy, should alert the clinician to the possibility of HLH.[8] Not all the diagnostic criteria of HLH may be present when the patient is first seen; close clinical monitoring and repeated laboratory testing are thus required.[7] In our patient, the diagnosis was made after 9 days on admission, during which time there was worsening of her clinical condition.

We were unable to determine if our patient had primary or secondary HLH. Due to limited diagnostic capabilities, we could not perform any molecular testing. The family history was negative for HLH and the parents were non-consanguineous. Primary HLH is inherited as an autosomal recessive disorder, occurring more frequently when there is parental consanguinity.[8] Patients with primary HLH most often present in infancy, while secondary HLH is usually seen in older children in association with concurrent infections, malignancy or other medical conditions that serve as underlying triggers of HLH.[7] However, there are reports of primary HLH first presenting in adolescence and adulthood.[9,10]

The distinction between primary and secondary HLH often remains challenging in the acute setting, unless there is a known genetic defect or family history.[3] Early institution of appropriate therapy is crucial.

Although spontaneous remission of secondary HLH may occur, many patients require HLH-directed therapy in addition to treatment of any underlying illnesses.[4] The immediate aims in the treatment of any patient with HLH are to suppress the severe hyperinflammation and to kill the antigen-presenting cells, thus removing the stimulus for the ongoing but ineffective immune activation.[8]

In primary HLH, the ultimate goal is stem cell transplantation (SCT), which represents the only chance of cure.[3] For induction therapy, dexamethasone, etoposide and cyclosporine A are used, with intrathecal therapy given to patients with persistent active CNS disease or CNS reactivation.[7] Steroids alone have been used successfully as first line therapy in some patients although there should be rapid progression to full combination therapy if the disease does not respond.[1]

In EBV-associated HLH, patients who receive etoposide in the first four weeks of treatment have better outcomes.[4] One of the concerns with the use of etoposide is the potential risk of developing secondary malignancies and there are suggestions that etoposide should only be used in patients with severe or fulminant disease and in those who do not respond to non-etoposide containing treatment regimens.[3,4] With early active therapy and SCT, survival in primary HLH has improved from 0% to over 60%.[3] Patients with secondary HLH may qualify for SCT if there is refractory or relapsed disease.[3]

Persistence or recurrence of fever in patients being treated for HLH could either signify ongoing disease or opportunistic infection. Both bacterial and invasive fungal infections are important causes of mortality.[11] Patients should initially be evaluated for recurrent disease, started on broad-spectrum antibiotics, and dexamethasone/etoposide should be restarted or re-escalated.[7]

Salvage therapies that have been used in refractory HLH include antithymocyte globulin (ATG), anti-CD52 antibody (alemtuzumab), anti-CD25 antibody (daclizumab), anti-tumour necrosis factor antibody (infliximab), plasma exchange and high dose pulse corticosteroids.[3-5] It is important to exclude a secondary infectious process prior to maximizing immunosuppressive treatment with salvage therapies.[1]

Our patient had an excellent initial response to dexamethasone as monotherapy, but suffered early reactivation of the disease and passed away before any potential salvage therapies could be considered. She was also started on antibiotics for presumed infection but this was not confirmed by bacteriologic culture.

CONCLUSION

HLH should be suspected in any child who is severely ill and deteriorating clinically or who does not improve with conventional therapies. Even where an underlying aetiology like infection, malignancy or autoimmune disease is apparent, HLH-directed therapy is often required to quell the hyper-inflammatory response. SCT and salvage therapies may be used for refractory or recurrent disease.

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