CME/SAM

Performances of the H-Score for Diagnosis of Hemophagocytic Lymphohistiocytosis in Adult and Pediatric Patients

France Debaugnies, PharmD,[1,5] Bhavna Mahadeb, MD,[1] Alina Ferster, MD,[2] Nathalie Meuleman, MD, PhD,[3] Laurence Rozen, PharmD,[4] Anne Demulder, MD, PhD,[4] and Francis Corazza, MD, PhD[1]

From the [1]Laboratory of Immunology and [4]Laboratory of Hematology and Hemostasis, Université Libre de Bruxelles, CHU Brugmann, Brussels, Belgium; [2]Department of Hematology and Oncology, Hôpital Universitaire des Enfants Reine Fabiola, Brussels, Belgium; [3]Department of Hematology, Jules Bordet Institute, Brussel, Belgium; and [5]Laboratory of Biochemistry, Laboratoire National de Santé, Dudelange, Luxembourg.

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ABSTRACT

Objectives: *In this study, we compared the performances of adapted hemophagocytic lymphohistiocytosis (HLH)–2004 guidelines with those of the new diagnostic H-score to identify patients with HLH in a multicenter cohort consisting of adult and pediatric cases of suspected HLH.*

Methods: *The study sample consisted of 147 cases, including 20 adults and 16 children with HLH. Two sets of biological data were evaluated: at presentation and the maximal values reached during the episode.*

Results: *At presentation, for both children and adults, the H-score was more efficient than adapted HLH-2004 guidelines to identify HLH. The diagnostic sensitivity and specificity were respectively 100% and 80% for children and 90% and 79% for adults. However, for adults, performances became comparable between adapted HLH-2004 guidelines and H-score as patient clinical status worsened. The specificity decreased to 73% for the same sensitivity.*

Conclusions: *The adapted HLH-2004 guidelines seem less powerful and H-score seems to be more appropriate for children, which may be due to less significantly marked biological features. For adults, H-score performances are better when determined at presentation. The cutoff value of the H-score should be adapted depending on the target population to obtain optimal specificity.*

Upon completion of this activity you will be able to:
- list the most important clinical and biological diagnostic criteria for hemophagocytic lymphohistiocytosis (HLH).
- discuss the limitations of the available diagnostic scoring systems for HLH in adults and children.
- compare the H-score and HLH-2004 diagnostic criteria for HLH.

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Hemophagocytic lymphohistiocytosis (HLH) is a life-threatening hyperinflammatory syndrome resulting from a highly stimulated but ineffective immune response, which leads to overactivation of macrophages and lymphocytes, hemophagocytosis, severe systemic inflammation, and multiorgan damage. This syndrome may be the consequence of a genetic defect in cytolytic activity of natural killer (NK) cells and cytotoxic T lymphocytes or may be promoted by an acquired immunodeficiency. HLH is classified as primary (genetic) or secondary (acquired) depending on whether a genetic disorder predisposing to HLH has been identified. Most often, the trigger of HLH is an infectious agent, but acquired forms of HLH have been observed in association with a variety of underlying immune-stimulating diseases (namely, malignancies, metabolic disorders, and

autoinflammatory/autoimmune diseases). Furthermore, HLH can be facilitated by acquired immune deficiencies, iatrogenic immune suppression, and organ or stem cell transplantation.[1] Diagnosis of the acute syndrome relies on a set of nonspecific clinical and laboratory features, including fever, hepatosplenomegaly, hemophagocytosis, cytopenia, hypofibrinogenemia, and increased ferritin, triglycerides, soluble CD25 (sCD25), and liver enzyme levels. HLH represents the extreme upper end on a gradient of adequate inflammatory response to excessive hyperinflammation; therefore, only the magnitude of clinical and laboratory abnormalities and progressiveness of the symptoms are characteristic of the syndrome.[2] Early introduction of the most adequate therapy, considering the triggering condition, is crucial to block the cytokine storm before eventual organ failure and death. However, it is always challenging to diagnose an episode of HLH, especially at an early stage when the entire picture of the disease is still incomplete. Furthermore, several underlying conditions such as septicemia[3] or lymphoma[4] can mimic or underlie HLH. Consequently, HLH remains an underrecognized cause of death in patients with septic shock and multiorgan failure.[5]

Nowadays, the 2004 diagnostic guidelines for HLH proposed by the Histiocyte Society are still the most widely used criteria to define and diagnose HLH in clinical practice,[6] although these were poorly validated for most cases encountered. This is especially true for acquired forms of HLH and in the adult population, since those guidelines were initially developed to diagnose primary forms of HLH in a pediatric population. Recently, alternative parameters have been evaluated to define a new diagnostic score: the H-score.[7] This H-score corresponds to a set of weighted criteria, allowing more effective estimation of the individual's risk of having HLH. However, it has only been validated for the diagnosis of reactive forms of HLH in an adult cohort.[8]

The aim of the present study is to compare the performances of the HLH-2004 guidelines with those of the H-score to identify patients with HLH in a retrospective cohort, including both adult and pediatric patients for whom the diagnosis of HLH was suspected in the routine practice of four university hospitals.

Materials and Methods

Patient Selection

We retrospectively conducted a keyword search (including *hemophagocytosis* or *macrophage activation*) in the hematology laboratory database of four university-affiliated tertiary hospitals (one pediatric center, one oncology adult center, and two major general hospitals) in Brussels, Belgium. The search included all bone marrow aspirates requested between January 2011 and December 2014 and encompassed the final result field, allowing extraction of cases with evidence of hemophagocytosis and cases in which no evidence of hemophagocytosis had been detected. Only the first aspiration was considered for each patient. According to the laboratory's procedures, they correspond to all patients with signs and symptoms of suspected HLH, for whom a marrow puncture was requested, as well as those with reported findings of hemophagocytosis due to other causes.

We then retrospectively reviewed the medical records of these patients. The collected data included age, sex, highest recorded temperature, presence of hepatomegaly/splenomegaly, earlier medical history, known underlying immunosuppression (due to human immunodeficiency virus [HIV] or treatment with an immunosuppressive agent), diagnosis of hemophagocytosis retained or not by the physician, treatment prescribed, underlying disease, and duration of episode (defined as duration of fever or hospitalization). The following laboratory data were also collected: hemoglobin levels; platelet, leukocyte, neutrophil, and lymphocyte counts; liver enzymes; ferritin; triglycerides; fibrinogen levels; and the presence or absence of images of hemophagocytosis on bone marrow smears. First, we extracted the biological parameters concomitant to bone marrow aspiration (in a range of 2 days before or after) corresponding to the presentation data set. Second, we recorded the highest or lowest value of each biological parameter registered during the episode corresponding to the diagnostic confirmation data set, allowing the determination of the maximal value of the scores reached for each patient. The study was approved by the ethics committees of each institution.

Patient Classification

Patients were classified as having HLH based on the final diagnosis retained by the medical team caring for the patient. For doubtful cases, two of the authors (A.F. and N.M.), who have at least 10 years' experience in diagnosis of and care for patients with HLH, reclassified the patients as positive, negative, or undetermined cases based on their medical records. The control group consisted of patients for whom the diagnosis was not retained by the medical team but did not include undetermined cases. Patients with HLH and controls were then classified according to the HLH-2004 diagnostic guidelines and the H-score. As neither NK cell activity nor sCD25 levels were determined in most patients, the HLH-2004 diagnostic rule was amended to presume the diagnosis of HLH when at least either four or five among six of the remaining criteria were met. The adapted HLH-2004 guidelines and the H-score are detailed in

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Table 1. The H-score was determined as described by Fardet et al.[8] The best cutoff value proposed by the authors to diagnose HLH was 169.

Statistical Analysis

Categorical variables are presented as an absolute number expressed in terms of percentage. Comparison of categorical variables was done using the Fisher exact test, and diagnostic odds ratios were calculated for each criterion. To evaluate the diagnostic performances of the HLH-2004 guidelines and the H-score to discriminate patients with HLH from control patients, we determined their diagnostic sensitivity, specificity, and accurate classification percentage by taking as a reference the diagnosis retained by the medical team. For the H-score, we evaluated the cutoff proposed by Fardet et al[8] and an optimal cutoff value obtained by receiver operating characteristic (ROC) curve analysis. This optimal cutoff value corresponds to the highest value of the sum of sensitivity and specificity. Data were analyzed using GraphPad Prism5 (GraphPad Software, La Jolla, CA).

Results

Study Population

A query performed in our laboratory database identified 147 patients for whom a bone marrow puncture had been requested because of suspected HLH or revealed hemophagocytosis regardless of the indication. According to the final diagnosis retained by the referring physician and the review of the patients' medical records, 36 patients of these 147 cases were ultimately diagnosed with HLH, 16 of 73 children and 20 of 74 adults. For one child, the diagnosis of HLH could not be formally excluded. For six adults, the diagnosis remained undetermined. Since NK cell activity measurement data and sCD25 levels were available for only five of the 147 patients in our cohort, we chose to omit these criteria from our analysis. The characteristics of the study population are shown in **Table 2**. At presentation, 81% of children with HLH and 75% of adults with HLH met at least four of the six diagnostic criteria of the HLH-2004 guidelines. The median (interquartile range [IQR]) H-score was 187 (152-202) for children with HLH and 235 (152-274) for adults with HLH. At diagnostic confirmation, 88% of children and 90% of adults with HLH met at least four of the six diagnostic criteria of the HLH-2004 guidelines. The median (IQR) H-score was 205 (174-268) for children with HLH and 270 (193-300) for adults with HLH.

The frequency of fulfillment of H-score items is presented in **Table 3** (determined with the diagnostic confirmation data set). Sixty-nine percent of the patients with HLH (25 of 36) had evidence of hemophagocytosis in bone marrow aspirates compared with 71% (74 of 104) in the control group. In children with HLH, the prevalence of fever, hepatomegaly, splenomegaly, pancytopenia, and increased levels of aspartate aminotransferase was higher than in the control group. In adults with HLH, the prevalence of hepatosplenomegaly, pancytopenia, increased levels of ferritin above 6,000 ng/mL, triglycerides greater than 4 mmol/L, and decreased levels of fibrinogen was higher than in the control group.

The frequency of fulfillment of clinical and biological items of children with HLH was comparable to that seen in adults with HLH **Figure 1**. Only splenomegaly was significantly more frequent in adults.

Table 1
Parameters Included in the Adapted HLH-2004 Guidelines and H-Score and the Number of Points Associated With Each Criterion for Scoring[a]

Parameter	Adapted HLH-2004 Guidelines	H-Score
Fever (°C)	0 (<38.5) or 1 (≥38.5)	0 (<38.4), 33 (38.4–39.4), or 49 (>39.4)
Splenomegaly	0 (no) or 1 (yes)	
Organomegaly		0 (no), 23 (hepatomegaly or splenomegaly), or 38 (hepatomegaly and splenomegaly)
Cytopenia	0 (one lineage) or 1 (two or three lineages)[b]	0 (one lineage), 24 (two lineages), or 34 (three lineages)[c]
Ferritin (ng/mL)	0 (<500) or 1 (≥500)	0 (<2,000), 35 (2,000-6,000), or 50 (>6,000)
Triglycerides (mmol/L)	0 (<3) or 1 (≥3)	0 (<1.5), 44 (1.5-4), or 64 (>4)
Fibrinogen (g/L)	0 (>1.5) or 1 (≤1.5)[d]	0 (>2.5) or 30 (≤2.5)
Hemophagocytosis in bone marrow	0 (no) or 1 (yes)	0 (no) or 35 (yes)
Aspartate aminotransferase (IU/L)		0 (<30) or 19 (≥30)
Known underlying immunosuppression		0 (no) or 18 (yes)

HLH, hemophagocytic lymphohistiocytosis.
[a]Data are presented as number of points, with values in parentheses.
[b]Defined as hemoglobin less than 90 g/L, platelets less than 100×10^9/L, and neutrophils less than 1.0×10^9/L.
[c]Defined as hemoglobin 92 g/L or less, platelets 110×10^9/L or less, and leukocytes 5×10^9/L or less.
[d]The point is not added if there is already one point for triglycerides.

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Table 2
Characteristics of the Study Population Classified by the Medical Team Caring for the Patient as Being Negative, Undetermined, or Positive for the Diagnosis of HLH[a]

Characteristic	Children (n = 73)			Adults (n = 74)		
	HLH (n = 16)	Undetermined (n = 1)	Controls (n = 56)	HLH (n = 20)	Undetermined (n = 6)	Controls (n = 48)
Male/female sex	5 (31)/11 (69)	0/1 (100)	37 (66)/19 (34)	11 (55)/9(45)	5 (83)/1 (17)	29 (60)/19 (40)
Age, median (interquartile range), y	9 (3-12)	0	6 (3-10)	59 (45-66)	49 (38-56)	57 (39-69)
Underlying disease						
Infection	8 (50)	0 (0)	2 (4)	7 (35)	1 (17)	13 (27)
Hematologic malignancy	2 (13)	0 (0)	22 (39)	3 (15)	2 (33)	15 (31)
Hematologic malignancy + infection	1 (6)	0 (0)	2 (4)	6 (30)	2 (33)	2 (4)
Solid cancer	0 (0)	0 (0)	9 (16)	1 (5)	0 (0)	4 (8)
Autoimmune/inflammatory disease	4 (25)	0 (0)	2 (4)	0 (0)	1 (17)	3 (6)
Autoimmune/inflammatory disease + infection	0 (0)	0 (0)	1 (1)	2 (10)	0 (0)	3 (6)
Familial lymphohistiocytosis	1 (6)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Other/unknown	0 (0)	1 (100)	18 (32)	1 (5)	0 (0)	8 (17)
H-score, median (interquartile range)[b]	205 (174-268)	199	75 (53-107)	270 (193-300)	174 (160-224)	136 (87-173)
Adapted HLH-2004 guidelines						
≥4 of 6 criteria[b]	14 (88)	1 (100)	4 (7)	18 (90)	3 (50)	13 (27)

HLH, hemophagocytic lymphohistiocytosis.
[a]Values are presented as number (%) unless otherwise indicated.
[b]Scores determined with the diagnostic confirmation data set.

Table 3
Frequency of Fulfillment of Individual Items of the H-Score in Control Groups and Patients With HLH Determined With the Diagnostic Confirmation Data Set[a]

Characteristic	Children (n = 72)				Adults (n = 68)			
	HLH (n = 16), No. (%)	Controls (n = 56), No. (%)	Odds Ratio	*P* Value	HLH (n = 20), No. (%)	Controls (n = 48), No. (%)	Odds Ratio	*P* Value
Known underlying immunosuppression	5/16 (31)	28/56 (50)	0.5	.3	11/20 (55)	25/48 (52)	1.1	1
Fever								
38.4-39.4 °C	8/16 (50)	8/56 (14)	6	**.005**	7/18 (39)	9/42 (21)	2.3	.2
≥39.5 °C	5/16 (31)	2/56 (4)	12.3	**.005**	6/18 (33)	4/42 (10)	4.8	.05
Hepatomegaly or splenomegaly	5/16 (31)	5/56 (9)	4.6	**.04**	10/20 (50)	12/47 (26)	2.9	.08
Hepatomegaly and splenomegaly	7/16 (44)	4/56 (7)	10.1	**.002**	8/19 (42)	5/45 (11)	5.8	**.01**
Cytopenia of two lineages	1/16 (6)	9/56 (16)	0.35	.4	1/20 (5)	9/48 (19)	0.2	.3
Cytopenia of three lineages	13/16 (81)	13/56 (23)	14.3	**<.0001**	19/20 (95)	27/48 (56)	14.8	**.002**
Ferritin								
2,000-6,000 ng/mL	6/16 (38)	2/20 (10)	5.4	.1	5/20 (25)	13/44 (30)	0.8	.8
>6,000 ng/mL	6/16 (38)	2/20 (10)	5.4	.1	12/20 (60)	9/44 (20)	5.8	**.004**
Triglycerides								
1.5-4 mmol/L	9/15 (60)	9/14 (64)	0.8	1	7/19 (37)	19/32 (59)	0.4	.2
>4 mmol/L	2/15 (13)	1/14 (7)	2	1	11/19 (58)	4/32 (13)	9.6	**.001**
Fibrinogen ≤2.5 g/L	10/16 (63)	10/29 (34)	3.2	.1	16/20 (80)	8/35 (23)	13.5	**<.0001**
AST ≥30 IU/L	15/16 (94)	30/53 (57)	11.5	**.007**	20/20 (100)	38/45 (84)	8	.09
Hemophagocytosis in bone marrow	11/16 (69)	50/56 (89)	0.3	.06	14/20 (70)	24/48 (50)	2.3	.2

AST, aspartate aminotransferase; HLH, hemophagocytic lymphohistiocytosis.
[a]*P* value determined using the Fisher exact test. Significant *P* values (<.05) are indicated in bold type. Missing data concerned only the control group: triglycerides (data missing for 33% and 75% of adults and children, respectively), fibrinogen (data missing for 27% and 48% of adults and children, respectively), and ferritin (data missing for 64% of children).

Performances of the H-Score Compared With HLH-2004 Guidelines in the Diagnosis of HLH

The diagnostic sensitivity and specificity of the H-score and the adapted HLH-2004 guidelines are presented in **Table 4**. Considering the ROC curve analysis **Figure 2**, the optimal cutoff value of the H-score was 120 for children when determined with the presentation data set and 141 with the diagnostic confirmation data set, based on extreme values registered during the episode. These values were respectively 138 and 185 for adults.

In children, the highest sensitivity (100%) was achieved for the H-score with an adapted cutoff while HLH-2004 guidelines had maximum specificity (up to 100%). When the maximal score values were considered, the best compromise

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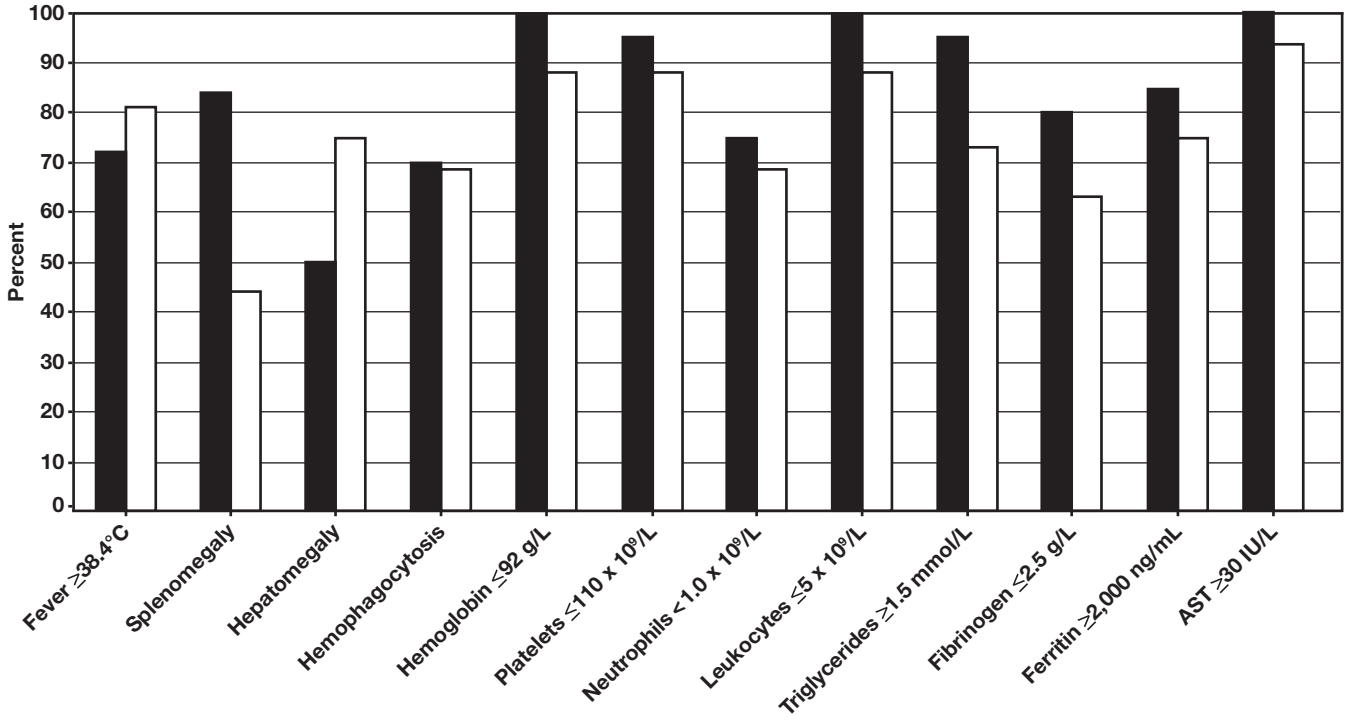

Figure 1 Percentage of fulfillment of clinical and biological items in children (white bars) and adults (black bars) with hemophagocytic lymphohistiocytosis (HLH) determined with the diagnostic confirmation data set. Splenomegaly was significantly more frequent in adults compared with children with HLH ($P = .03$). *P* value determined using the Fisher exact test. AST, aspartate aminotransferase.

Table 4
Sensitivity, Specificity, and Percentage of Accurate Classification for the Adapted HLH-2004 Guidelines and the H-Score With Adapted Cutoff in the Discrimination of Patients With HLH From Control Groups[a]

	HLH Children vs Control Group, %			HLH Adults vs Control Group, %		
Characteristic	**Sensitivity**	**Specificity**	**Accurate Classification**	**Sensitivity**	**Specificity**	**Accurate Classification**
At initial presentation						
Adapted HLH-2004 guidelines (at least four of six criteria)	**81**	**100**	95	75	92	80
Adapted HLH-2004 guidelines (at least five of six criteria)	25	100	82	55	100	80
H-score >169	63	91	84	65	90	76
H-score >120	**100**	**80**	64	95	73	73
H-score >138	65	89	86	**90**	**79**	86
Maximal score value[b]						
Adapted HLH-2004 guidelines (at least four of six criteria)	88	93	92	90	73	78
Adapted HLH-2004 guidelines (at least five of six criteria)	44	100	88	**80**	**96**	91
H-score >169	81	89	88	90	73	78
H-score >141	**100**	**88**	90	95	56	69
H-score >185	69	91	86	**85**	**88**	87

HLH, hemophagocytic lymphohistiocytosis.
[a]Bold values represent the highest value of the sum of sensitivity and specificity.
[b]Maximal score value was determined using the diagnostic confirmation data set consisting of extreme values reached during the episode for each parameter.

between sensitivity and specificity was provided by the H-score with a cutoff of 141 (ie, 100% of sensitivity and 88% of specificity).

In adults at initial presentation, we observed that the best trade-off between sensitivity and specificity was provided by the H-score with an adapted cutoff. However, when the H-score was determined with the diagnostic confirmation data set, for a same sensitivity of 90%, the specificity decreased from 79% to 73%. With this data set, the best trade-off between sensitivity and specificity was comparable between five of six criteria of the adapted HLH-2004 guidelines and H-score. Five of six criteria of the adapted

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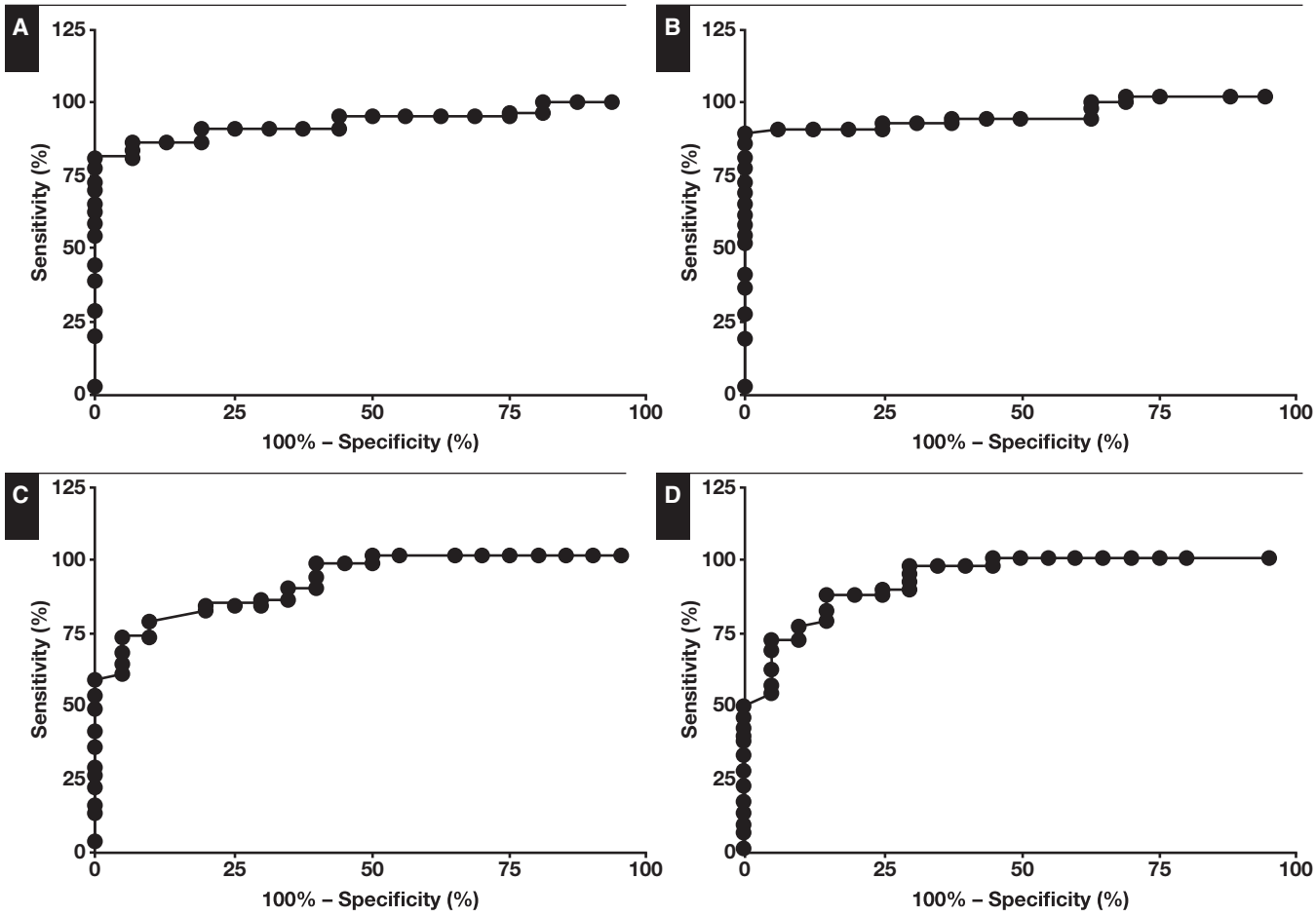

Figure 2 Receiver operating characteristic (ROC) curve analysis for diagnosis of hemophagocytic lymphohistiocytosis in children (**A**, **B**) and adults (**C**, **D**) with H-score computed with the presentation data set (**A**, **C**) or maximal H-score value obtained with the diagnostic confirmation data set (**B**, **D**). The optimal cutoff was selected as the threshold giving the highest value for the sum of sensitivity and specificity. **A**, Area under the curve (AUC) = 0.9286; **B**, AUC = 0.9425; **C**, AUC = 0.9151; **D**, AUC = 0.9307.

HLH-2004 guidelines were the most specific, reaching a specificity of 96%, whereas H-score was the most sensitive with a sensitivity of 85%.

Discrepant Results

Of 74 adult cases, nine yielded discrepant results, including six false positives and three false negatives. The false positives included four cases of hematologic malignancies and two cases of miliary tuberculosis. All six had an H-score above 185, and two met five of six criteria of the adapted HLH-2004 guidelines. Their scores and diagnosis are described in **Figure 3A**. The three adults with HLH having an H-score of less than 185 had an underlying bacterial infection; two were also HIV positive, and one received a specific treatment for HLH.

Seven pediatric controls had an H-score above 141 **Figure 3B**, of whom four met four of six criteria of the adapted HLH-2004 guidelines. The diagnoses retained for these patients were graft vs host disease, thiamine-responsive megaloblastic anemia, systemic Langerhans cell histiocytosis, and progressive familial cholestasis type 2.

Agreement Between H-Score and HLH-2004 Guidelines in Undetermined Cases

The baby with an undetermined diagnosis, for whom HLH could not be formally excluded, had an H-score of 199 and met five of the six criteria of the adapted HLH-2004 guidelines. She was a preterm neonate with multisystem organ failure who died 1 week after birth.

Among adults with an undetermined diagnosis (n = 6), two had an H-score higher than 185 and met four of the six criteria of the adapted HLH-2004 guidelines. They included a patient with systemic lupus erythematosus suspected of having either HLH or catastrophic antiphospholipid syndrome who rapidly died after admission and another patient with a posttransplantation reactivation of Epstein-Barr virus.

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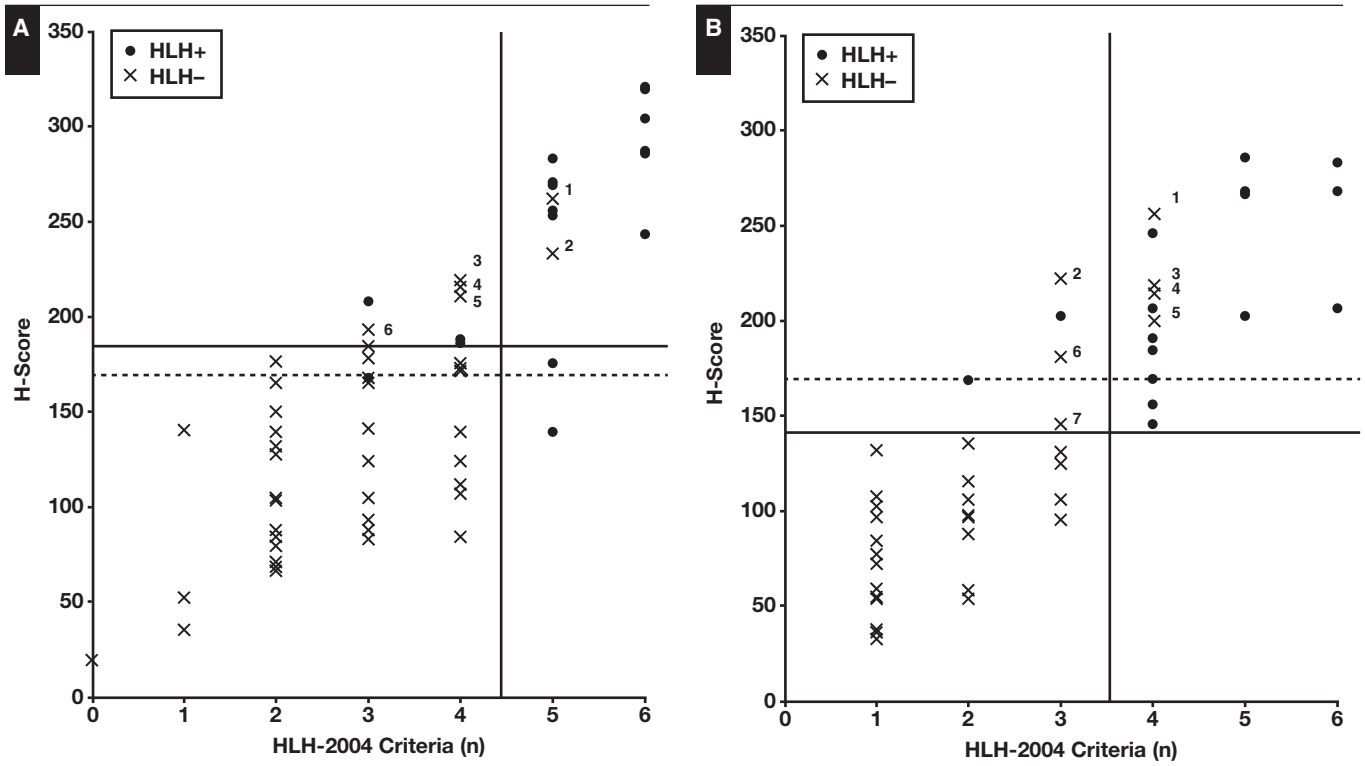

Figure 3 Discrepant results between the H-score and hemophagocytic lymphohistiocytosis (HLH)–2004 guidelines (observed with the diagnostic confirmation data set): adult cohort (**A**) and pediatric cohort (**B**). For the H-score, the broken lines indicate the cutoff of Fardet et al,[8] and the solid lines indicate the optimal cutoff. HLH+, patients with HLH; HLH–, controls. Diagnosis of discrepant results for the controls are as follows. **A**, In adults: 1, miliary tuberculosis; 2, chronic lymphocytic leukemia with chlorambucil intoxication; 3, chronic lymphocytic leukemia in progression to lymphoma; 4, miliary tuberculosis; 5, anaplastic lymphoma; and 6, chronic myelomonocytic leukemia with severe sepsis. **B**, In children: 1, graft vs host disease; 2, Epstein-Barr virus reactivation following transplantation; 3, thiamine-responsive megaloblastic anemia; 4, systemic Langerhans cell histiocytosis; 5, progressive familial cholestasis type 2; 6, hemolytic anemia due to G6PD deficiency with influenza A infection; and 7, idiopathic aplastic anemia.

Of the remaining four patients, one had an H-score higher than 169 but less than 185 and met three of the six criteria of the adapted HLH-2004 guidelines. He was admitted for acute myeloid leukemia with sepsis. Two patients had an H-score less than 169 and met three of the six criteria of the adapted HLH-2004 guidelines. The first patient had a pulmonary infection treated empirically, and the second patient was admitted for an acute leukemia and died rapidly after admission. The last case had an H-score less than 169 and met five of the six criteria of the adapted HLH-2004 guidelines. He was admitted for an acute monoblastic leukemia with pulmonary embolism and disseminated intravascular coagulation.

Discussion

In this study, we retrospectively assessed the performances of the H-score to diagnose HLH in two distinct cohorts of pediatric and adult patients, and we compared them with the adapted HLH-2004 guidelines. The patients were selected on the basis of a bone marrow morphologic examination explicitly reporting the absence or presence of hemophagocytosis. According to our laboratory's procedures, a systematic screening for hemophagocytosis is performed and reported for patients with signs and symptoms of suspected HLH. The term *hemophagocytosis* is also mentioned if hemophagocytosis is incidentally observed regardless of the clinical information received. Even if the presence of hemophagocytosis has a very low predictive value[9] for the diagnosis of HLH, bone marrow examination is still recommended for HLH suspicion mainly to rule out or identify other confounding or underlying diseases.[10] We noted that the frequency of hemophagocytosis was not different between patients with HLH and control patients in our cohort, with very low odd ratios (Table 3), in accordance with the observations of Ho et al.[9]

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Our search strategy restricted our evaluation to patients suspected of having HLH who underwent bone marrow aspiration. Despite the possibility of this selection bias, the different etiologies underlying reactive hemophagocytic syndrome encountered in our population are representative of the heterogeneity of the causes of the disease and in agreement with recently published data for adult cases.[11,12] In our adult cohort, hematologic malignancy (combined or not with infection) was the predominant cause of HLH (45%), followed by infection (35%) and autoimmune or autoinflammatory disease (10%). Fifty-five percent of the patients with HLH were immunocompromised (HIV positive or undergoing immunosuppressive therapy). In children, infection was the predominant cause (50%), followed by autoimmune or autoinflammatory disease (25%) and hematologic malignancy (19%). The prevalence of hematologic malignancy was greater in our cohort than the 8% value reported for children and adolescents by Lehmberg et al.[13] This could be explained by the recruitment of children mostly in the hemato-oncology unit. Furthermore, patients who developed HLH following stem cell transplantation were also included in this group. Thirty-one percent of the children with HLH were immunocompromised.

Two sets of biological data were analyzed to assess the H-score in the same condition than in the validation study[8] and to determine the maximal value of the scores reached for each control and patient with HLH during the episode. Hemoglobin, neutrophil counts, and fibrinogen were the parameters that changed the most during the HLH episode. It resulted in a 25% to 35% increase in patients with HLH who met the criteria for anemia, neutropenia, and hypofibrinogenemia (difference being significant only for hemoglobin in adults, $P = .008$, data not shown).

In their validation study, Fardet et al[8] reported an optimal cutoff value of 169 for the H-score. This cutoff resulted in a 93% diagnostic sensitivity and a 86% diagnostic specificity for acquired HLH in adults. In our study, to obtain the same specificity with the presentation data set, the cutoff of the H-score could be decreased to 162 in adults achieving a 65% sensitivity and to 131 in children achieving a 94% sensitivity.

In adults with HLH, an H-score of 138 seems to be more predictive with the presentation data set than the four of six or five of six criteria of the adapted HLH-2004 guidelines (Table 4), achieving a 90% sensitivity and 79% specificity. This difference is less marked with the diagnostic confirmation data set. For the same sensitivity, we observed a decrease of specificity of the H-score between these two data sets. The performances become comparable between five of six criteria of the adapted HLH-2004 guidelines and an H-score of 185. Therefore, it is more difficult to discriminate adults with HLH from our controls when the clinical condition of the patient deteriorates.

In children with HLH, the clinical characteristics are less marked than in adults with HLH. The addition of one criterion for the HLH-2004 guidelines (compared with the five of six criteria of the adapted HLH-2004 guidelines) reduces by 50% the number of children with HLH with a positive score. This suggests that five of six criteria of the adapted HLH-2004 guidelines could be too restrictive in many cases, especially in children. The value of the cutoff for H-score should be adjusted accordingly.

Another limitation of our study is that data were collected retrospectively from clinical charts with missing and possibly erroneous information (especially in the control groups). However, available data could be assumed to reflect routine clinical management of suspected cases, with the availability of data being related to the clinician's degree of HLH suspicion. Our control group may indeed include patients with hemophagocytosis who have a low suspicion of HLH, which could overestimate the diagnostic specificity of the scores. However, this bias should have the same impact on both scores. Moreover, patients should ideally have been categorized according to underlying conditions to assess an optimal cutoff for the H-score. However, in practice, the underlying disease is not always identified before HLH occurs, making it difficult to choose the best cutoff for each category of patient.

We noted that four of six adult patients misclassified by the H-score (>185) had lymphoma at diagnosis or miliary tuberculosis. As already reported, these diseases present many confounding factors with HLH, which complicates differential diagnosis before the identification of the underlying cause. Concerning the treatment of patients with HLH, 14 (70%) adults and eight (50%) children received a specific HLH-directed treatment. HLH is frequently lethal if not quickly treated and may explain the rapid death of two of the three deceased undetermined cases. Overall, three of the seven undetermined cases had an H-score above the optimal cutoff.

In conclusion, this multicenter study has the advantage to comparatively evaluate the performances of H-score and HLH-2004 criteria to diagnose HLH in adults and children, in the context of daily clinical practice. The H-score seems to be less restrictive than the HLH-2004 criteria. In children, H-score presents a better sensitivity than the HLH-2004 guidelines, with an adequate specificity with both data sets. In adults, H-score performances are better when determined at initial presentation than when the laboratory parameters reach their extreme values. The cutoff value of the H-score should be adapted based on the target population. This study also confirms that the main causes of reactive hemophagocytic syndrome are malignancies in adults and infections in children. For severely ill patients with deteriorating health, if the calculated score is borderline or negative, high clinical suspicion of HLH should be sufficient to justify early

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initiation of the therapy, given the fatal evolution of the disease with delayed treatment.

Corresponding author: France Debaugnies, PharmD, 4 Place Van Gehuchten, 1020 Brussels, Belgium; fdebaugn@ulb.ac.be.

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