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Prevalence and Diagnostic Challenge of Hemophagocytic Lymphohistiocytosis Syndrome in Critically Ill Patients

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ABSTRACT

Background: Hemophagocytic lymphohistiocytosis (HLH) is a rare hyperinflammatory syndrome. It is a severe condition with a challenging diagnosis in the intensive care unit (ICU), for which current recommendations rely on fulfilling five of eight HLH-2004 criteria.

Methods: Our single-center retrospective cohort included adults admitted to the ICU in whom HLH was diagnosed, suspected, or excluded. The primary outcome was the prevalence of HLH in the ICU according to the HLH 2004 criteria. Additional factors analyzed included prognosis and diagnosis (determined by clinician judgment, HLH 2004 criteria with a four variable threshold and HScore).

Results: Among 123 included patients, 30 met HLH-2004 criteria, corresponding to a prevalence of 0.42% of ICU admissions, likely underestimated. Their 28-day mortality rate was 46.7%; their 1-year mortality rate was 75%. In the entire cohort, absence of infection as a trigger and higher SAPS II scores were independently associated with 28-day mortality in multivariate analysis. Diagnostic concordance was 69.1% with the HScore, 61.8% with clinician judgment, and 71.5% with the four-variable HLH-2004 threshold, yielding an overall agreement of 46.3%.

Conclusion: Our study describes a population of adults suspected of HLH in the ICU. Improved diagnostic strategies are needed to facilitate timely recognition and optimize patient outcomes.

1 | Background

Hemophagocytic lymphohistiocytosis (HLH) is a severe hyperinflammatory syndrome driven by dysregulated immune activation. It is characterized by a cytokine storm leading to clinical manifestations such as fever, cytopenias, organomegaly, and multi-organ dysfunction [1, 2]. HLH can be classified into primary (genetic) forms, typically occurring in pediatric populations, and secondary forms triggered by infections, malignancies, or autoimmune diseases [3, 4].

Early identification and intervention are crucial to prevent multi-organ failure and improve outcomes [5, 6]. However, diagnosing HLH in critically ill adults presents challenges due to the non-specific nature of its symptoms and significant overlap with other conditions, including sepsis, systemic inflammatory response syndrome, and multi-organ dysfunction syndrome [7–9].

Two principal diagnostic tools are currently employed to identify HLH: the HLH-2004 diagnostic criteria and the HScore.

Abbreviations: ECCO₂R, extracorporeal CO₂ removal; ECMO, extracorporeal membrane oxygenation; HLH, hemophagocytic lymphohistiocytosis; ICU, intensive care unit; NK, natural killer; SAPS, simplified acute physiology score; sCD25, soluble CD25; SOFA, sequential organ failure assessment.

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The HLH-2004 criteria were initially developed for pediatric populations and are currently recommended in HLH diagnosis [3, 10]. They require the presence of five out of eight clinical and laboratory findings, which include fever, splenomegaly, cytopenias affecting at least two cell lines, hypertriglyceridemia or hypofibrinogenemia, hyperferritinemia, hemophagocytosis in bone marrow or other tissues, elevated soluble CD25 (sCD25), and reduced natural killer (NK) cell activity. While these criteria provide a structured approach, they face several limitations in adult ICU populations. Key parameters such as NK cell activity [4, 11, 12] and sCD25 [13–15] are often unavailable due to technical and logistical challenges. Additionally, many of the criteria, such as fever and cytopenias, are non-specific and frequently observed in other critical conditions, leading to diagnostic uncertainty [16, 17].

The HScore, based on a probabilistic model, was developed to estimate the likelihood of secondary HLH in adults [18] outside of intensive care. It incorporates nine variables including immunosuppression, fever, organomegaly, cytopenias, hyperferritinemia, hypertriglyceridemia, hypofibrinogenemia, elevated transaminases, and hemophagocytosis. The score obtained is associated with a diagnostic probability of HLH which is usually retained for an HScore ≥ 169 . The HScore is widely recognized for its ease of use and accessibility, as it avoids reliance on complex assays like NK cell activity or sCD25 [19]. However, its retrospective validation limits its applicability in ICU settings. Moreover, as well as the HLH-2004 criteria, the specificity of the HScore diminishes in critically ill populations, where inflammatory markers are commonly elevated [16, 17, 20–22].

As described above, both the HLH-2004 criteria and the HScore face significant limitations when applied to ICU adult patients. The lack of specificity in clinical and laboratory findings frequently leads to diagnostic overlap with sepsis and other hyperinflammatory syndromes. Additionally, the thresholds for hyperferritinemia ($\geq 500 \mu\text{g/L}$) [7, 17, 23–25] and other biomarkers may inherently have low specificity in adult populations. Moreover, neither of these diagnostic tools has been validated in prospective studies involving critically ill adult patients. These limitations contribute to an imprecise estimation of the incidence of secondary HLH in the ICU [6, 10, 26, 27] and underscore the need for improved diagnostic tools, including novel biomarkers or validated scoring systems tailored to critically ill adults. Among these, the use of the HLH 2004 criteria with a four-variable threshold is proposed as it could improve the sensitivity and the precocity of the diagnosis [16, 28].

Given the indeterminate incidence and the diagnostic challenge, we performed a retrospective analysis of suspected, retained, or ruled-out HLH cases in a single-center ICU, focusing on the diagnostic methods used, their limitations, and the clinical and prognostic characteristics of patients.

2 | Methods

2.1 | Study Design and Setting

This retrospective and observational study was conducted in the medical intensive care department of Caen University Hospital, France, from January 2014 to December 2022. The study was

approved by the local ethics committee (ID 5306; March 19, 2024). Patient consent was waived due to the retrospective nature of the study.

2.2 | Population

Cases were identified through electronic medical record reviews using keywords such as “hemophagocytic lymphohistiocytosis” and “macrophage activation syndrome.” Included patients were adults (≥ 18 years) admitted to the ICU with a diagnosis of HLH suspected, retained, or refuted by the clinician during the hospitalization. Patients for whom the diagnosis of HLH syndrome was retained before their admission to intensive care but had led to their transfer to intensive care were also included in the same way.

2.3 | Data Collection

Data were extracted from medical records, including demographic information, comorbidities, presenting symptoms, laboratory findings, diagnostic evaluations, and treatments. Diagnostic criteria were assessed using the HLH-2004 framework and HScore calculations. Severity was assessed by the sequential organ failure assessment (SOFA) score [29] and the simplified acute physiology score (SAPS) II [30]. Confounding factors affecting diagnostic criteria were first identified through a literature review, and then systematically searched for in medical records. Outcomes included ICU length of stay, mortality rates in ICU at 28 days and 1 year.

2.4 | Diagnostic Definitions

HLH was *confirmed* using the HLH-2004 criteria when at least five out of eight criteria were met, as recommended (including ferritin, with the standard threshold of $500 \mu\text{g/L}$). For the other diagnostic tools evaluated, a diagnosis was established with an HScore ≥ 169 , or with at least four out of eight criteria for the HLH-2004 criteria using a four-variable threshold, or if the diagnosis was documented in the medical record based on the clinician's assessment (a medical team experienced in general intensive care, but not specific expertise in HLH).

Clinician-established diagnoses were classified as retained, possible, or refuted according to the medical records. The diagnostic process was not externally influenced and was left to the clinicians' independent judgment.

2.5 | Objectives

Our primary objective was to estimate the prevalence rate of HLH among ICU admissions. Secondary objectives were to describe diagnostic tools used, to describe patients suspected or diagnosed with HLH in terms of demographic and anamnestic characteristics, prognosis, triggering factors for HLH and initiation of specific treatment, and to characterize risk factors for mortality at 28 days of patients suspected or diagnosed with HLH admitted to the ICU.

2.6 | Statistical Analysis

Results are presented in median [interquartiles (IQ)] and number (percentage), as appropriate. The prevalence rate of confirmed HLH among adult patients' ICU admissions during ICU stay was calculated with a 95% confidence interval (CI).

Descriptive statistics were used for baseline characteristics. Comparisons between groups (confirmed HLH or unconfirmed HLH) were conducted using chi-square tests and Fisher exact tests for categorical variables or Mann–Whitney *U* tests for continuous variables. Diagnostic agreement and performance metrics, including sensitivity and specificity, were calculated relative to the HLH-2004 criteria with a five-variable threshold as the reference standard. Kaplan–Meier survival analysis was used to evaluate mortality. To identify independent predictors of mortality at 28 days among the entire cohort, a multivariate logistic regression was performed. Variables associated with 28-day mortality with $p < 0.2$ in the univariate analysis were incorporated into the multivariate analysis. The performance of the final model on the test set was assessed using the c-index and its 95% CI.

All tests were two-sided, and p values less than 0.05 were considered statistically significant. No correction of the multiplicity of tests was carried out.

All analyses were performed using R software (version 4.4.3, R foundation for statistical computing, Vienna, Austria; <https://www.r-project.org/>).

3 | Results

3.1 | Patient Characteristics and HLH Prevalence

Out of 7200 ICU admissions between January 1, 2014 and December 31, 2022, 140 eligible patients were identified (Figure 1). Of these, 123 patients were finally included, with a HLH retained by the clinician for 57 of them (46.3%), a HLH described as possible for 36 (29.3%), and a HLH refuted for 30 (24.4%). Among these 123

patients, the HLH-2004 criteria confirmed the diagnosis of HLH in 30 patients, leading to a prevalence of HLH among patients admitted to the ICU of 0.42% (95% CI, 0.27%–0.57%).

Patient characteristics are presented in Table 1. The median age was 62 years [51;69], and 65.9% were male. Common comorbidities included mostly immunosuppression (49.6%) and hematologic malignancies (34.2%). The main reason for ICU admission was respiratory failure (38.2%). ICU mortality was 39%, increasing to 45.1% at 28 days and 60.7% at 1 year. Patient characteristics and clinical outcomes did not differ significantly between those with confirmed and unconfirmed HLH diagnoses.

3.2 | Triggers and Alternative Diagnosis

Among patients with confirmed HLH, the most common triggers were infections (33.3%), hematologic malignancies (30%), and a combination of both (23.3%). Infectious causes were mainly viral (including Influenza, Epstein–Barr Virus, Metapneumovirus, Enterovirus/Rhinovirus, and Coronavirus 2 (SarsCov2)), while fungal and bacterial infections were less frequent. Hematologic malignancies were primarily newly diagnosed B-cell lymphomas, followed by T-cell lymphomas, lymphoma relapses, and other hematologic disorders, including acute myeloid leukemia (Figure S1).

Among patients with unconfirmed HLH, alternative explanations for cytopenias were most commonly drug toxicity (21.4%), infections (16.1%), or hematologic malignancies (12.5%). Less frequently, when the cause was not unknown (16.1%) or unrecoverable (16.1%), other identified aetiologies included disseminated intravascular coagulation (5.4%), vitamin deficiencies (5.4%), hemolysis (9%), hypersplenism (3.6%), or other causes (7.1%), such as hemorrhagic shock and systemic diseases (Figure S2).

3.3 | Diagnostic Criteria

HLH-2004 criteria are presented in Table 2. NK cell activity and sCD25 were not assessed in any patient due to lack of

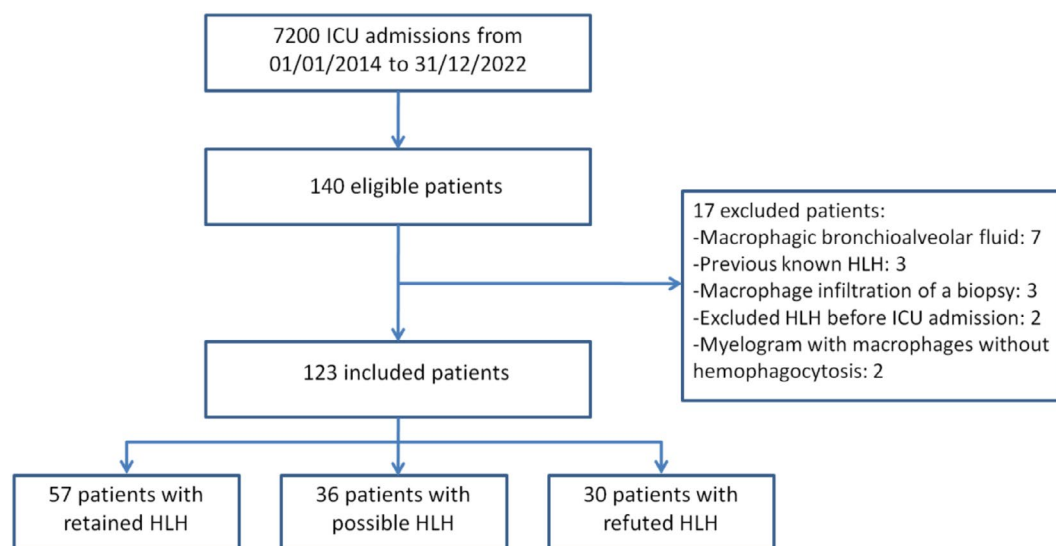


FIGURE 1 | Flowchart.

TABLE 1 | Patient characteristics.

Characteristics	Total (n = 123)	Confirmed HLH (n = 30)	Unconfirmed HLH (n = 93)	p
<i>Demographic variables</i>				
Age, median [IQ]	62 [51;69]	61 [44;68.5]	63 [53;70]	0.23
Male, n (%)	81 (65.9)	23 (76.7)	58 (62.4)	0.15
<i>Comorbidities</i>				
Solid neoplasia, n (%)	23 (18.7)	2 (6.7)	21 (22.6)	0.052
Hematologic malignancies, n (%)	42 (34.2)	13 (43.3)	29 (31.2)	0.22
Immunosuppression, n (%)	61 (49.6)	19 (63.3)	42 (45.2)	0.08
Systemic disease, n (%)	11 (8.9)	4 (13.3)	7 (7.5)	0.33
<i>Reason for ICU admission</i>				
Hemodynamic, n (%)	29 (23.6)	7 (23.3)	22 (23.7)	—
Respiratory, n (%)	47 (38.2)	7 (23.3)	40 (43.0)	—
Neurological, n (%)	9 (7.3)	3 (10.0)	6 (6.5)	—
Renal, n (%)	4 (3.3)	2 (6.7)	2 (2.2)	—
Hematologic, n (%)	5 (4.1)	1 (3.3)	4 (4.3)	—
Multi-organ, n (%)	29 (23.6)	10 (33.3)	19 (20.4)	—
<i>Prognostic scores</i>				
SOFA, median [IQ]	10 [8;13]	12 [9;15]	10 [7;12]	0.08
SAPS II, median [IQ]	54 [39.5;64.5]	55 [44.8;68.3]	54 [38.5;63.5]	0.33
<i>Organ support therapy</i>				
Extrarenal purification, n (%)	57 (46.3)	16 (53.3)	41 (44.1)	0.38
Vasopressor, n (%)	93 (75.6)	21 (70.0)	72 (77.4)	0.41
Inotrope, n (%)	15 (12.2)	4 (13.3)	11 (11.8)	0.83
<i>Ventilatory support</i>				
Oxygen therapy, n (%)	19 (15.5)	6 (20.0)	13 (14.0)	0.43
High-flow nasal oxygen, n (%)	8 (6.5)	3 (10.0)	5 (5.4)	—
Non invasive ventilation, n (%)	3 (2.4)	0 (0)	3 (3.2)	—
Invasive ventilation, n (%)	81 (65.9)	18 (60.0)	63 (67.7)	—
ECMO, n (%)	3 (2.4)	0 (0)	3 (3.23)	—
<i>Organ failure</i>				
Renal, n (%)	113 (91.9)	27 (90.0)	86 (92.5)	0.67
Liver, n (%)	89 (72.4)	23 (76.7)	66 (71.0)	0.60
Neurological, n (%)	72 (58.5)	21 (70.0)	51 (54.8)	0.18
<i>Outcome</i>				
Length of stay, median [IQ]	10 [3;18]	8.5 [4;16.8]	10 [3;18]	0.92
ICU mortality, n (%)	48 (39.0)	11 (36.7)	37 (39.8)	0.76
Mortality at 28 days, n (%)	55 (45.1)	14 (46.7)	41 (44.6)	0.84
Mortality at 1 year, n (%)	71 (60.7)	21 (75.0)	50 (56.2)	0.07

Abbreviations: ECMO, extracorporeal membrane oxygenation; ICU, intensive care unit; IQ, interquartiles; SAPS, simplified acute physiology score; SOFA, sequential organ failure assessment.

TABLE 2 | HLH-2004 criteria: research, presence, and confounding factors.

Criteria	Total (n = 123)	Retained diagnosis (n = 57)	Possible diagnosis (n = 36)	Refuted diagnosis (n = 30)	Confirmed HLH (n = 30)	Unconfirmed HLH (n = 93)	p (HLH or not)
<i>Fever, n (%)</i>							
Research	123 (100)	57 (100)	36 (100)	30 (100)	30 (100)	93 (100)	1
Presence	101 (82.1)	52 (91.2)	29 (80.6)	20 (66.7)	30 (100)	71 (76.3)	0.003
Confounding factor	118 (95.9)	55 (96.5)	35 (97.2)	28 (93.3)	27 (90)	91 (97.8)	0.09
<i>Organomegaly, n (%)</i>							
Ultrasound research	34 (27.6)	20 (35.1)	10 (27.8)	4 (13.3)	14 (46.7)	20 (21.5)	0.007
Scanner research	87 (70.7)	42 (73.7)	23 (63.9)	22 (73.3)	28 (93.3)	59 (63.4)	0.002
Splenomegaly presence	24 (19.5)	15 (26.3)	6 (16.7)	3 (10.0)	18 (60.0)	6 (6.5)	<0.001
Hepatomegaly presence	41 (33.3)	23 (40.4)	10 (27.8)	8 (26.7)	19 (63.3)	22 (23.7)	<0.001
Confounding factor	57 (46.3)	28 (49.1)	15 (41.7)	14 (46.7)	20 (66.7)	37 (39.8)	0.01
<i>Hyperferritinemia, n (%)</i>							
Research	114 (92.7)	56 (98.2)	31 (86.1)	27 (90.0)	30 (100)	84 (90.3)	0.11
Presence	101 (88.6)	52 (92.9)	28 (90.3)	21 (77.8)	29 (96.7)	72 (85.7)	0.18
Confounding factor	113 (99.1)	56 (100)	31 (100)	26 (96.3)	30 (100)	83 (98.8)	1
<i>Bicytopenia, n (%)</i>							
Research	123 (100)	57 (100)	36 (100)	30 (100)	30 (100)	93 (100)	1
Presence	92 (74.8)	44 (77.2)	25 (69.4)	23 (76.7)	28 (93.3)	64 (68.8)	0.007
Confounding factor	121 (98.4)	56 (98.2)	35 (97.2)	30 (100)	29 (96.7)	92 (98.9)	0.43
<i>Hypertriglyceridemia or hypofibrinogenemia, n (%)</i>							
Research	118 (95.9)	56 (98.2)	34 (94.4)	28 (93.3)	29 (96.7)	89 (95.7)	1
Presence	54 (45.8)	28 (50.0)	17 (50.0)	9 (32.1)	24 (82.8)	30 (33.7)	<0.001
Confounding factor	96 (81.4)	44 (78.6)	26 (76.5)	26 (92.9)	23 (79.3)	73 (82.0)	0.74
<i>Hemophagocytosis, n (%)</i>							
Research	87 (70.7)	47 (82.5)	22 (61.1)	18 (60.0)	29 (96.7)	58 (62.4)	<0.001
Presence	61 (70.1)	45 (95.7)	16 (72.7)	0 (0)	26 (86.7)	35 (60.3)	0.005
<i>Elevated sCD25, n (%)</i>							
Research	0 (0.0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1
<i>Reduced NK cell activity, n (%)</i>							
Research	0 (0.0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1

accessibility. Organomegaly and hemophagocytosis criteria were significantly more frequently documented in patients with a confirmed HLH diagnosis. Confounding factors were

identified in 82.1% of cases across all collected criteria, reaching up to 99.1% for hyperferritinemia. Fever was the most common presenting symptom, observed in 82.1% of the total cohort and in

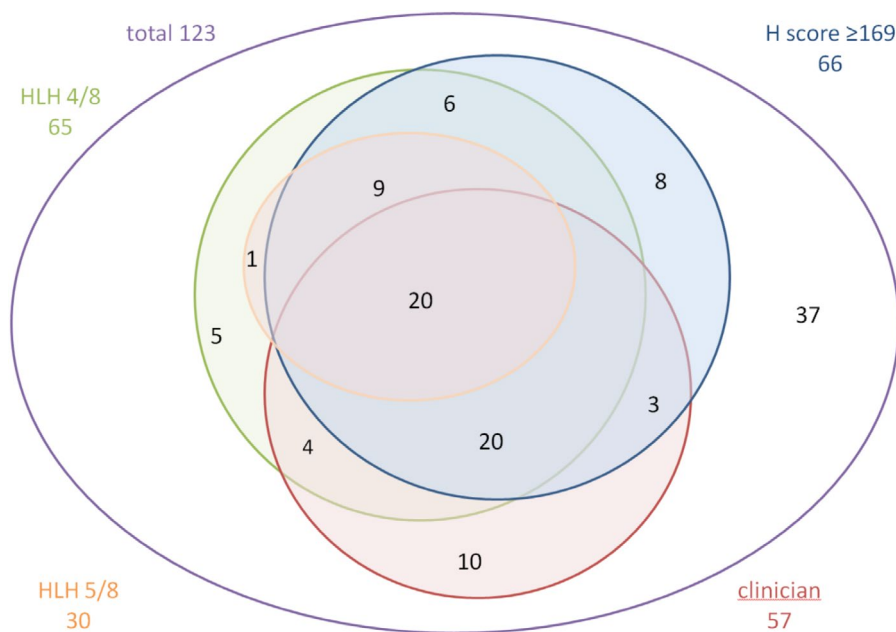


FIGURE 2 | Agreement between diagnostic methods.

100% of confirmed HLH cases. Hyperferritinemia ($\geq 500 \mu\text{g/L}$) was present in 96.7% of confirmed HLH and 85.7% of unconfirmed cases, with no significant difference between groups. The median peak ferritin levels were 4661 and 1447 ng/mL, respectively. Fever, organomegaly, bicytopenia, hemophagocytosis, and either hypertriglyceridemia or hypofibrinogenemia were significantly more prevalent among patients with a confirmed HLH diagnosis.

Using different diagnostic approaches (Figure 2 and Table S1), HLH was diagnosed in 30 patients (24.4% of the cohort) based on the HLH-2004 criteria with a five-variable threshold, in 65 patients (52.8%) using the HLH-2004 criteria with a four-variable threshold, in 66 patients (53.7%) using the HScore and in 57 patients (46.3%) based on clinician's judgment. Concordance of the HLH-2004 criteria (five-variable threshold) was 69.1% with the HScore, 61.8% with the clinician's diagnosis, and 71.5% with the HLH-2004 criteria with a four-variable threshold (Table S1). Overall agreement between diagnostic methods was moderate (46.3%).

The HScore and the HLH-2004 criteria with a four-variable threshold demonstrated high sensitivity (96.7% and 100%, respectively), but lower specificity (60.2% and 62.4%). The clinician's diagnosis showed a comparable specificity (60.2%) but had a lower sensitivity (66.7%) (Table S2).

3.4 | Outcome

Survival rates at 28 days and 1 year did not differ significantly between patients with confirmed and unconfirmed HLH diagnoses (Figure 3). It should be noted that most of the deaths observed occurred during the first 100 days of follow-up, as shown by the Kaplan–Meier curve. By univariate analysis, 28-day mortality was associated with comorbidity conditions

such as hematologic malignancies ($p < 0.001$) and immunosuppression ($p < 0.001$), higher SAPSII ($p = 0.004$), higher HScore ($p = 0.044$), hematologic malignancies with ($p = 0.001$) or without ($p = 0.04$) infections as HLH trigger, the use of vasopressors in ICU ($p = 0.02$), and the need for etoposide as HLH therapeutic management ($p = 0.019$) (Table S3). In the overall cohort, multivariate analysis identified higher SAPS II scores (OR, 1.30 [1.02–1.34]; $p = 0.031$) and non-infectious triggers (OR, 0.38 [0.17–0.88]; $p = 0.024$ for infectious triggers) as independent predictors of mortality at 28 days (Figure 4). The discrimination performance of the final model was moderate, with a c-index of 0.72 (95% CI, 0.61–0.81). However, multivariate analysis could not be conducted specifically in the confirmed HLH group due to the limited number of events.

4 | Discussion

In this retrospective analysis of suspected, retained or ruled-out HLH cases in a single-center ICU, we describe diagnostic methods used with their limitations, clinical and prognostic characteristics of patients. This is the first study to assess diagnostic concordance among the four commonly used HLH diagnostic tools in a general adult ICU population—HLH-2004 criteria with both five- and four-variable thresholds, the HScore, and clinician's diagnosis—demonstrating a moderate agreement (46.3%). A total of 30 HLH cases were confirmed, suggesting a likely underestimated prevalence of 0.42% among ICU admissions. No significant differences were observed between patients with confirmed and unconfirmed HLH diagnoses regarding baseline characteristics, ICU management, or prognosis. In the overall cohort, the absence of infection as a triggering factor and higher SAPS II scores were independently associated with 28-day mortality in multivariate analysis.

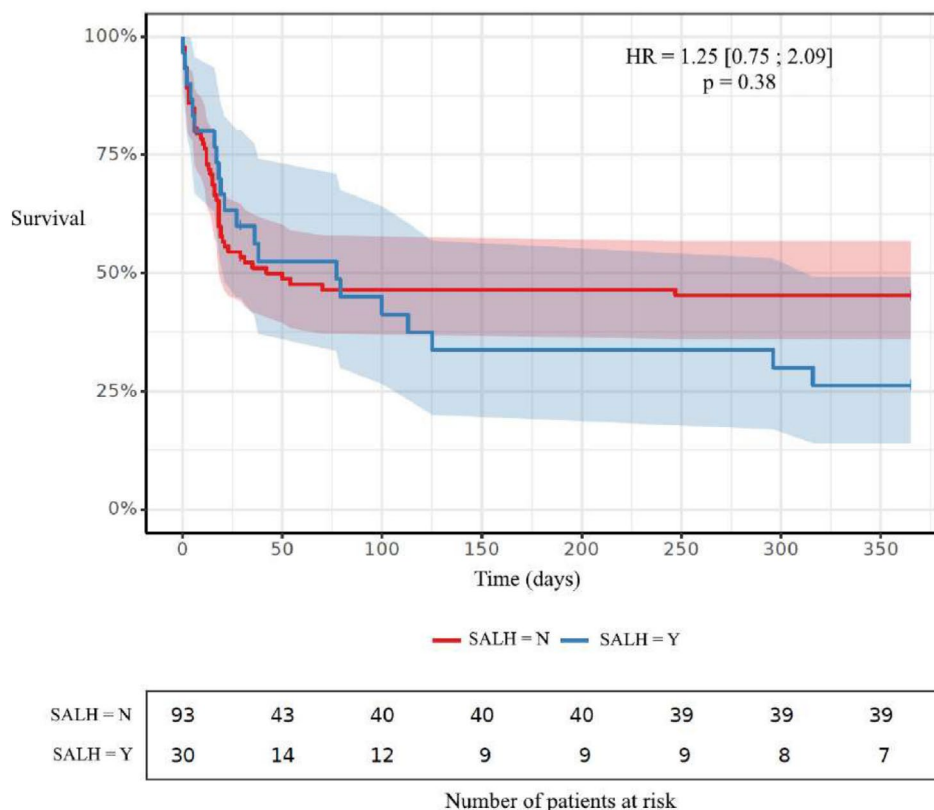


FIGURE 3 | Survival at 1 year.

4.1 | Diagnostic Challenges

Diagnosing HLH in critically ill adults remains exceptionally complex due to the significant overlap of its clinical and biological features with other hyperinflammatory and critical conditions. This diagnostic complexity contributes to substantial underdiagnosis, potentially affecting up to 75% of cases in intensive care [7]. Conditions such as sepsis, systemic inflammatory response syndrome, and multi-organ dysfunction syndrome frequently mimic HLH, complicating differential diagnoses [2, 25, 31, 32]. Fever, cytopenias, and hyperferritinemia, hallmark features of HLH, are common in ICU patients with various conditions, diminishing their diagnostic specificity. For example, while hyperferritinemia is a critical diagnostic marker, its utility may be limited in ICU settings where inflammatory markers are frequently elevated [23–25, 33]. Ferritin thresholds tailored to critically ill adults could improve specificity but require further validation.

The variability of bone marrow findings also complicates diagnosis. Hemophagocytosis, considered a hallmark feature of HLH, is not always present in medullar samples or may occur in non-HLH conditions such as severe infections or malignancies [27, 34–37]. This needs careful clinical correlation rather than sole reliance on histopathological findings. Thus, no isolated clinical or biological criterion is currently sufficiently effective [5, 27].

Additionally, another challenge lies in the delayed recognition of HLH. The syndrome often evolves over time, with early presentations being non-specific [2, 5, 10]. Diagnostic delays may also result from fragmented clinical assessments and the lack of

standardized protocols for HLH evaluation in ICUs. However, its late recognition can worsen its already severe prognosis [10, 38].

Finally, the reliance on the HLH-2004 criteria and the HScore introduces certain limitations. The HLH-2004 criteria, initially developed for pediatric populations and currently recommended, include parameters such as NK cell activity and sCD25 levels. These parameters are often unavailable or delayed in adult ICU settings due to technical or logistical challenges [4, 11, 13–15]. Additionally, there is a lack of prospective data supporting the use of these criteria, even more with only six of the criteria being collected in adult ICU populations. In contrast, although the prospective validation of the HScore also remains insufficient and it is not yet recommended in the most recent international intensive care guidelines [10], it is more practical, broadly applicable, and widely recommended outside ICU combined with an expert multidisciplinary team approach [19]. These advantages, together with evidence from several studies demonstrating its good diagnostic performance [16, 21], may support its use in the ICU setting—as is already increasingly observed in routine clinical practice. However, both tools may underperform in cases involving multiple triggers, such as combined infections and malignancies, which are common in ICU patients. The absence of prospective validation for current diagnostic criteria in ICU populations further emphasizes the need for the development of improved diagnostic tools and algorithms that integrate clinical, laboratory, and emerging biomarker data [39, 40].

In our study, the diagnostic performance of the HScore aligns with the literature regarding its good sensitivity. However, the specificity observed (60.2%) is lower than the range typically reported in the literature, which is between 70% and 99% [16–18,

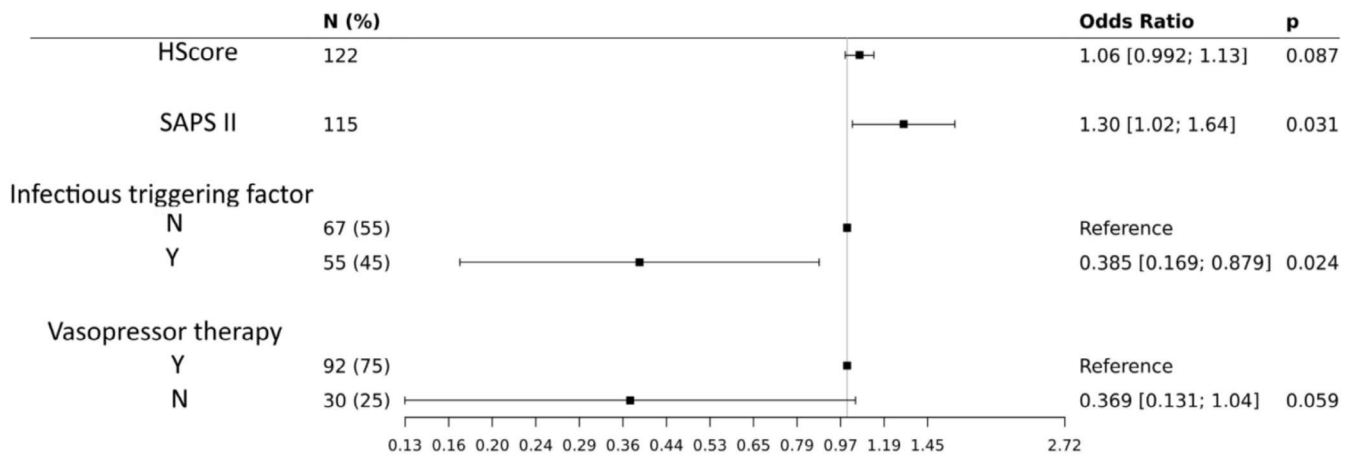


FIGURE 4 | Multivariate analysis of predictors of mortality at 28 days in the overall cohort.

20–22]. This discrepancy may be attributed to differences in the selection of control subjects. Specifically, all control subjects in our study were ICU patients, all of whom had at least a suspected diagnosis of HLH during hospitalization, often at an advanced stage of severity and disease progression. This contrasts with the broader variability of control groups in the literature, which include patients with hematologic malignancies not hospitalized in intensive care, who were neither suspected nor diagnosed with HLH. Furthermore, this study design does not allow to investigate a potential superiority of the HScore over the HLH-2004 criteria, currently regarded as the gold standard, which may result in an artificially lowered diagnostic performance of the HScore.

4.2 | Clinical Implications

Our study highlights the importance of heightened clinical suspicion and thorough diagnostic evaluation in suspected HLH cases [10]. The key challenge is improving the identification of patients with this hyperinflammatory syndrome to optimize management tailored to their individual heterogeneity, particularly with regard to severity, underlying triggers, and disease progression [5, 41–43]. Such tailored approaches are crucial for improving patient outcomes. The high mortality observed, especially among patients with hematologic malignancies [16, 37, 38], underscores the urgent need for personalized management strategies.

4.3 | Future Directions

Development of novel diagnostic tools holds promise for improving the accuracy and timeliness of HLH diagnosis. Prominent among these are immunophenotyping approaches that focus on lymphocyte and monocyte activation markers. For instance, lymphocyte HLA-DR and CD38 expression have emerged as potential indicators of excessive T-cell activation [44–46]. Elevated expression on CD8+ T cells correlates with T-cell hyperactivation, which may distinguish HLH from other hyperinflammatory states such as sepsis. Thus, CD38, a marker of T-cell activation, may provide additional specificity. Monocyte HLA-DR expression is another promising tool, reflecting the activation status

of antigen-presenting cells [47, 48]. While HLA-DR expression is typically reduced in sepsis, it is elevated in HLH. Combining these markers with traditional diagnostic criteria could enhance differentiation between HLH and mimicking conditions.

In addition, soluble markers such as soluble CD163 [49] and soluble CD14 [50] (reflecting monocyte/macrophage activation) and glycosylated ferritin [28] are being investigated as adjuncts to existing tools. Cytokine profiling [39], particularly interferon-gamma-induced chemokines like CXCL9, and other tools such as the OHI index, combining hyperferritinemia and elevation of sCD25 [51], may also offer valuable diagnostic and prognostic insights.

These tools require prospective validation in critically ill populations to assess their impact on diagnostic precision and clinical outcomes. Then, larger studies should aim to explore the integration of these novel biomarkers into existing diagnostic frameworks.

4.4 | Study Limitations

This study is subject to several limitations that may impact the interpretation of its findings. First, the retrospective design inherently introduces selection and information biases. Patients with milder forms of HLH or rapidly fatal courses may have been underrepresented, as their diagnoses may not have been captured in clinical records. Additionally, the reliance on medical records limited the identification of cases and the availability of some data for certain HLH-2004 criteria, although the number of missing data was small (Table S4), which may have affected diagnostic classifications. We had no available data on sCD25 levels or NK cell activity for any of the patients, which may have influenced the diagnostic performance of the HLH-2004 criteria and, consequently, other diagnostic tools. Although this limitation is common in studies on the topic and reflects real-world clinical practice, it may also have affected the estimation of HLH prevalence and other study endpoints. Moreover, the collected cases may have been subject to classification bias due to the absence of independent expert validation. While this differs from some previous studies, it accurately represents routine clinical practice. Classification bias may also have resulted from the use of HLH-2004 criteria with a five-variable threshold

to confirm HLH diagnosis. Although in line with current recommendations, the choice of this diagnostic method as the reference method and therefore as the supposed best diagnostic method may raise questions. All these factors have probably led to an underestimation of HLH prevalence.

The single-center nature of this study further restricts generalizability. As a tertiary referral center, our ICU manages a high proportion of patients with hematologic malignancies, potentially skewing the observed prevalence and outcomes of HLH. Differences in diagnostic and therapeutic practices across institutions may also limit the applicability of our findings to other settings.

Lastly, temporal changes over the nine-year study period represent another limitation. Advances in diagnostic tools and therapeutic approaches during this time may have influenced both case identification and patient outcomes. The heterogeneity of care practices and diagnostic thresholds over time introduces variability that could not be fully accounted for in the analysis.

5 | Conclusion

This retrospective study underscores the diagnostic challenges of HLH in ICU settings, the limitations of current diagnostic criteria, and the need for improved diagnostic tools. The HLH-2004 criteria and HScore are valuable tools and should be applied as soon as HLH is suspected, with appropriate contextual interpretation by the clinician. Infections were the most frequent triggers and are associated with better outcomes. Future research should focus on refining diagnostic criteria and exploring novel biomarkers to enhance early recognition and management of HLH in critically ill adults.

Author Contributions

C.Q. and D.C. contributed to manuscript writing, preparation, and creation of the published work. M.B., S.G., and S.C. participated in the initial draft, contributed to critical review and commentaries.

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Ethics Statement

The Local Health Research Ethics Committee of Caen University Hospital approved this retrospective observational study (ID 5306; March 19, 2024).

Consent

The authors have nothing to report.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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Supporting Information

Additional supporting information can be found online in the Supporting Information section. **Data S1:** Supporting Information.