석 사 학 위 논 문

Analysis of causes and survival of patients with hemophagocytic lymphohistiocytosis at tertiary medical centers in Korea

계 명 대 학 교 대 학 원

의 학 과

곽 아 브 라 함

지도교수 심 예 지

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이 논문을 석사학위 논문으로 제출함

2020년 8월

계명대학교대학원

의학과 소아청소년과 전공

곽 아 브 라 함

곽아브라함의 석사학위 논문을 인준함

주 심 도 영 록

부 심 심 예 지

부 심 허 미 화

계 명 대 학 교 대 학 원

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1. Introduction

Hemophagocytic lymphohistiocytosis (HLH) is a rare, rapidly progressive, life-threatening hematologic disorder with excessive immune activation.[1] HLH is a group of disorders characterized by 'hemophagocytosis', and accumulation of lymphocytes and macrophages, and often the phagocytosis of the hematologic cells by macrophages.[2] If HLH is left untreated, the patients survive for only a few months with severe progressive multi-organ failure.[1] In 1983, the long-term survival in patients with HLH was reported as only 4%.[3] HLH can be divided into two major categories: primary and secondary.[4] Primary HLH includes familial HLH (FHL), X-linked lymphoproliferative disease type 1 (XLP1), and other primary immunodeficiencies associated with HLH and partial albinism.[5,6] However, secondary HLH can develop because of various triggers, including infections, neoplasm, and autoimmune conditions.[4] Generally, HLH is frequently prevalent in infants from 0 to 18 months of age; however, this disorder is also observed in children, adolescents, and adults of all ages.[7]

In 1991, the Histiocyte Society firstly presented the diagnostic guidelines for HLH based on the common clinical, laboratory, and histopathological findings of HLH.[8] Subsequently, the prospective international treatment protocol, HLH-94 was introduced in 1994.[9] It is based on etoposide, corticosteroids, and cyclosporine A (CSA).[9] The HLH-94 protocol dramatically increased the survival rate of HLH patients to 54% with a median 6-year follow-up.[1,10] Thereafter, a modified treatment protocol, HLH-2004 with revised diagnostic criteria was introduced in 2004.[11] In HLH-2004, CSA is incorporated in the induction therapy; however, the survival outcomes in patients with HLH

was not different between two studies.[11,12] The HLH-94 and HLH-2004 protocols was mainly focused on pediatric patients with primary HLH, but secondary HLH is more common in Korea. Therefore, implementing the same treatment regimen in all HLH subtypes was questioned.[13]

Although primary and secondary HLHs exhibit common clinical features in the beginning, the distribution of each subtypes and respective prognoses are various among countries.[14,15] According to a nationwide study on pediatric patients with HLH by the Korea Histiocytosis Working Party from the Korean Society of Hematology, the most common genetic cause of primary HLH was FHL type 3 with *UNC13D* variants.[13] In Korean children, secondary HLH, the Epstein - Barr Virus (EBV) infection-induced type had a relatively high incidence in Korean children.[13] Moreover, the 5-year overall survival (OS) rate in 251 Korean children with HLH was 68%.[13]. In reported adult patients with HLH, neoplasms were a more common trigger factor in adults than in children.[16] Additionally, secondary HLH is the main cause of HLH subtype in adults.[17] However, until now, there are no data regarding adult patients with HLH or data that has compared children and adults with HLH in Korea. Therefore, we aimed to retrospectively review the causes and characteristics of HLH in both children and adult patients with different HLH subtypes in this study.

2. Materials and Methods

2.1. Subjects and ethical statement:

In this study, patients diagnosed with HLH at the Keimyung University Dongsan Hospital and Yeungnam University Medical Center from January 2001 to December 2019 were investigated. The medical records of the HLH patients were retrospectively reviewed for the age at diagnosis, sex, HLH etiology, treatment regimen, and death. This study was approved by the Institutional Review Board of Keimyung University Dongsan Hospital (IRB No. 2019-11-006), and the requirement for informed consent was waived.

2.2. Definition:

HLH was diagnosed according to the diagnostic criteria presented by the Histiocyte Society in 1991, and was updated in 2004.[11,18] According to the HLH-2004 guideline, at least five of the eight enlisted items must be met for the diagnosis of HLH: (1) fever ≥38.5℃, (2) splenomegaly, (3) bicytopenia affecting ≥2 cell lines (hemoglobin <90 g/L [hemoglobin <100 g/L in infants <4 weeks]; platelet $<100\times10^9$/L; neutrophil $<1.0\times10^9$/L), (4) hyperferritinemia (ferritin >500 μg/L), (5) hypertriglyceridemia (fasting triglyceride >3.0 mmol/L [265 mg/dL]) and/or hyperfibrinogenemia (fibrinogen, <1.5 g/L), (6) hemophagocytosis in the bone marrow, lymph node, spleen, or liver without evidence of malignancy, (7) elevated level of soluble CD25 (also called as interleukin-2 receptor) >2,400 U/mL, and (8) low or absent natural killer

(NK) cell activity.[5,11] Additionally, mutations in the genes of typical FHL (*PRF1, UNC13D, STX11, STXBP2*), XLP1 (*SH2D1A*) and X-linked lymphoproliferative syndrome type 2 (*BIRC4*) are sufficient to establish a diagnosis of HLH regardless of the number of fulfilled criteria of HLH-2004.[5,6,11]

2.3. Statistical analysis:

The variables are described as median values and range. The Mann - Whitney U test was utilized for comparing the variables between the two groups. To compare the ratio between the two groups, χ^2 test was used. The 5-year OS of the patients with HLH between the groups was compared by the Kaplan - Meier method with log-rank test and post hoc pairwise comparison. The 95% confidence intervals (CIs) were estimated using the means and standard errors. The Cox proportional hazards model was used for the multivariate analysis. *P* values <0.05 were considered significant. For all statistical analysis, we used SPSS software v23.0 (IBM Corp., Armonk, NY, USA).

3. Results

3.1. Baseline characteristics of the patients with HLH:

For the determined study period, 60 patients (male:female sex, 35:25) fulfilled the criteria for HLH. The median age at diagnosis of HLH was 7.0 years (range, 0.1 - 83 years). In the study population, 35 patients were children (age <18 years) and 25 patients were adults (age ≥18 years). Although HLH occurred in all age groups, more than 50% of the patients (56.7%, 34/60) developed HLH at below 10 years of age (Figure 1). The median follow-up duration of the HLH patients was 8.5 months (range, 0 - 204 months). When divided by patient age, the number of adult patients with HLH showed an increase with each year from 2001 to 2019 (Figure 2).

Upon classification, four patients had primary HLH comprising three and one patients with FLH and XLP1, respectively. Forty-eight patients had secondary HLH namely 20, 18, and 10 were infection-associated, neoplasm-associated, and autoimmune-associated HLH, respectively. In the remaining eight patients, the cause of HLH was unknown. In secondary HLH, EBV was the most common of the infection-associated HLH (6/20, 30%), lymphoma was the most common of the neoplasm-associated HLH (10/18, 55.6%), and Kawasaki disease was the most common of the autoimmune-associated HLH (5/10, 50%). With respect to the treatment regimen, 28 patients were treated with HLH-2004- or HLH-94-based immunochemotherapy and the remaining 32 patients were treated with other regimen; corticosteroid was the most commonly used medication (20/32, 62.5%). The baseline characteristics of

the HLH patients are presented in Table 1.

3.2. Comparisons of the course of HLH according to the age:

The underlying pathophysiology of HLH according to the age group is shown in Figure 3. In children with HLH, infection was the most common cause (14/35, 40%), followed by autoimmune diseases (6/35, 17.1%), uncertain causes (7/35, 20%), neoplasm (5/35, 14.3%), and primary HLH (3/35, 8.6%). In adults with HLH, neoplasm was the most common cause (13/25, 52%), followed by infection (6/25, 24%), autoimmune diseases (4/25, 16%), and primary HLH or uncertain cause (both, 1/25, 4%). The ratio of infection, as the cause of HLH, was higher in children (40%) than in adults (24%) (P=0.029). Moreover, the ratio of neoplasm was higher in adults (52%) than in children (14.3%) (P=0.004). Conversely, difference in the ratio of autoimmune diseases, uncertain causes, and primary HLH was not significant between children and adults (P=1.000, P=0.123, and P=0.634, respectively).

3.3. Comparisons of the variables of HLH patients between children and adults:

The comparison of the variables of clinical manifestations, laboratory or imaging tests, and treatment regimen in HLH patients between children and adults are presented in Table 2. In case of clinical manifestations,

the ratio of splenomegaly in adults (24/25, 96%) was higher than that in the children (24/35, 68.6%) (P=0.010). Conversely, the ratio of hepatomegaly in children (28/35, 80%) was higher than that in adults (10/25, 40%) (P=0.003). The liver enzyme, alanine aminotransferase (ALT), was also higher in children (median ALT 232 U/L, range 14 - 1,230 U/L) than that in adults (median ALT 120 U/L, range 8 - 2,019 U/L) (P=0.024). With respect to the treatment regimen, the ratio of treatment with HLH-2004/94 protocol was higher in children (28/35, 80%) than that in adults (0/25, 0%) (P<0.001). Differences in the variables for the rest of the clinical features and test findings between children and adults was not significant.

3.4. Survival of HLH patients:

The 5-year OS rate of all patients with HLH (N=60) was 59.9% (95% CI: 46.6 - 73.2) (Figure 4). Difference in the 5-year OS was not significant between the male (56.5%, 95% CI: 38.9 - 74.1) and female (65.6%, 95% CI: 45.8 - 85.4) sexes. According to the age, the difference in 5-year OS between children (72.1%, 95% CI: 56.4 - 87.8) and adults (37.4%, 95% CI: 11.3-63.5) was significant (Figure 5). Based on the HLH classification, the 5-year OS rates in patients with primary, infection-associated, neoplasm-associated, autoimmune-associated, and uncertain cause of HLH were 25% (95% CI: 0 - 67.5), 85% (95% CI: 69.3 - 100), 26.7% (95% CI: 3.6 - 49.8), 87.5% (95% CI: 64.6 - 100), and 62.5% (95% CI: 29 - 96), respectively (Fig. 6). Comparatively, the difference in the 5-year OS between the primary and infection-associated HLHs was significant (P=0.024). The difference in the 5-year OS between the neoplasm-associated and infection-associated

HLHs was significant (P=0.001). Additionally, the difference in the 5-year OS between the neoplasm-associated and autoimmune-associated HLHs was significant (P=0.010).

In terms of the treatment regimen, the 5-year OS rate in the patients treated with HLH-2004 protocol was 74.5% (95% CI: 54.9 - 94.1) and in patients treated with HLH-94 protocol was 68.6% (95% CI: 32.1 - 100), and that in patients who received other treatments was 45.8% (95% CI: 25 - 66.6) (Fig. 7). Difference in the 5-year OS between HLH-2004 and HLH-94 protocol was not significant (P=0.904). Difference in the 5-year OS in patients treated with HLH-2004/94 (73.1%, 95% CI: 55.9 - 90.3) and others (45.8%, 95% CI: 25 - 66.6) was significant (P=0.017).

3.5. Comparisons of the variables of HLH patients between the survival and death groups

The comparison of the variables of patients with HLH between the survival and death groups are presented in Table 3. The age at the time of diagnosis in the survival group (median 5.3 years, range 0.1 - 76.5 years) was lower than that in the death group (median 39.3 years, range 0.1 - 89 years) (P=0.007). The ratio of primary or neoplasm-induced HLHs in the survival group (5/37, 13.5%) was lower than that of the death group (16/23, 69.6%) (P<0.001). The hemoglobin level in the survival group (median 99 g/L, range 51 - 157 g/L) was higher than that in the death group (median 86 g/L, range 64 - 116 g/L) (P=0.006). The platelet level in the survival group (median 67×10^9/L, range 25 - 384×10^9/L) was higher than that in the death group (median 51×10^9/L, range 15 - 117×10^9/L) (P=0.030). The ratio of the patients with

abnormality on brain magnetic resonance imaging (MRI) in the survival group (1/12, 8.3%) was lower than that in the death group (4/6, 66.7%) (P=0.022). However, primary or neoplasm-induced HLH was the only dependent risk factor of death in patients with HLH as determined by multivariate analysis using the Cox proportional hazards model (P=0.036, hazard ratio 10.171, 95% CI: 1.163 - 88.939). Differences in the variables for the rest of the clinical features and test findings between children and adults were not significant.

Table 1A. Baseline characteristics of the patients with hemophagocytic lymphohistiocytosis.

Baseline characteristics of HLH patients (N=60)	N (%)
Age at diagnosis of HLH (years) (median, range)	7 (0.1 - 83)
Age < 18 years (n, %)	35 (58.3)
Age ≥ 18 years (n, %)	25 (41.7)
Sex (Male : Female)	35 : 25
Classification	
Primary HLH (n, %)	4 (6.7)
Familial HLH type 3 (*UNC13D*)	2
Familial HLH type 2 (*PRF1*)	1
X-linked lymphoproliferative disease 1 (*SH2D1A*)	1
Secondary HLH (n, %)	48 (80.0)
Infection-associated	20
Epstein - Barr virus	6
Cytomegalovirus	3
Severe fever with thrombocytopenia syndrome virus	3
Mycoplasma pneumoniae	2
Enterococcus (urinary tract infection)	2
Adenovirus	1
Streptococcus galactiae (meningitis)	1
Achromobacter xylosoxidans	1
Unknown organism (infectious colitis)	1
Neoplasm-associated	18
Lymphoma	10
Acute leukemia	3
Castleman disease	2
Myelodysplastic syndrome	1
Hepatocellular carcinoma	1
Pancreatic cancer	1
Autoimmune-associated	10
Kawasaki disease	5
Kikuchi disease	2
Rheumatoid arthritis	1
Systemic lupus erythematosus	1
Steven-Johnson syndrome	1
Uncertain cause (n, %)	8 (13.3)

Table 1B. Baseline characteristics of the patients with hemophagocytic lymphohistiocytosis (continued).

Treatment regimen (n, %)	
HLH-2004	21 (35)
HLH-94	7 (11.7)
Others	32 (53.3)
Corticosteroid	7
Antibiotics	5
Chemotherapy for underlying neoplasm	3
Corticosteroid + antibiotics	4
Corticosteroid + chemotherapy	3
Corticosteroid + Immunoglobulin G + antibiotics	2
Immunoglobulin G + antibiotics	2
Corticosteroid + Immunoglobulin G	1
Corticosteroid + Immunoglobulin G + Cyclosporin A + antibiotics	1
Corticosteroid + Immunoglobulin G + Cyclosporin A	1
Corticosteroid + plasma exchange	1
No treatment	2

HLH, hemophagocytic lymphohistiocytosis

Table 2A. Comparison of the variable between children and adults with hemophagocytic lymphohistiocytosis.

	Children (N=35)	Adults (N=25)	*P*
Sex (M:F)	21:14	14:11	0.481
EBV (%, n/n)	20.6 (7/34)	20 (4/20)	1.000
Fever (%, n/n)	100 (35/35)	100 (25/25)	-
Splenomegaly	68.6 (24/35)	96 (24/25)	0.010*
Hepatomegaly	80 (28/35)	40 (10/25)	0.003*
Neurologic Symptom or sign	11.4 (4/35)	16 (4/25)	0.708
Rash	37.1 (13/35)	20 (5/25)	0.253
Hemoglobin (g/L) (median, range)	90 (51 - 121)	89 (64 - 157)	0.685
Neutrophil count ($\times 10^9$/L) (median, range)	0.97 (0.06 - 14.1)	1.0 (0.18 - 10.2)	0.589
Platelet count ($\times 10^9$/L) (median, range)	67 (25 - 384)	52 (15 - 158)	0.184
Triglyceride (mg/dL) (median, range)	219 (47 - 742)	244 (128 - 274) (N=5)	0.988
Fibrinogen (g/L) (median, range)	1.32 (0.70 - 4.22)	2.18 (0.45 - 6.18)	0.051
Ferritin (μg/L) (median, range)	3,225 (185 - 204,887)	2,450 (764 - 19,640) (N=23)	0.765
sCD25 (U/mL) (median, range)	4,745 (825 - 27,060) (N=13)	2817 (N=1)	0.714
NK cell activity (%) (median, range)	94 (40 - 2,000) (N=6)	2000 (N=1)	0.286
Hemophagocytosis (%, n/n)	91.4 (32/35)	96 (24/25)	0.634
AST (U/L) (median, range)	450 (22 - 5,137)	221 (11 - 2,237)	0.063
ALT (U/L) (median, range)	232 (14 - 1,230)	120 (8 - 2,019)	0.024*
LDH (U/L) (median, range)	1,565 (266 - 5,8611) (N=32)	1,660 (262 - 6,369) (N=23)	0.666
Total bilirubin (mg/dL) (median, range)	1.2 (0.1 - 11.8)	1.1 (0.3 - 8.2)	0.584
Direct bilirubin (mg/dL) (median, range)	0.6 (0 - 8.8)	1.3 (0.2 - 6.2) (N=12)	0.617
PT (sec)	12.9 (11.7 - 31.6) (N=34)	13.7 (11 - 19.4)	0.758
INR	1.19 (1 - 2.85) (N=34)	1.18 (0.93 - 1.85)	0.515

Table 2B. Comparison of the variable between children and adults with hemophagocytic lymphohistiocytosis (continued).

aPTT (sec)	40.1 (25.5 - 200) (N=33)	39.3 (21.6 - 68.8)	0.921
CSF pleocytosis	14.8 (4/27)	0 (0/1)	1.000
CSF proteinosis	28 (7/25)	0 (0/1)	1.000
Abnormality on brain MRI (%, n/n)	31.3 (5/16)	0 (0/2)	1.000
HLH-2004/94 Treatment	80 (28/35)	0 (0/25)	< 0.001*

EBV, Epstein - Barr virus; sCD25, soluble CD25; AST, Aspartate transaminase; ALT, Alanine transaminase; LDH, lactate dehydrogenase; PT, Prothrombin time; INR, International normalized ratio; aPTT, activated partial thromboplastin time; CSF, Cerebrospinal fluid; MRI, Magnetic resonance imaging; HLH, hemophagocytic lymphohistiocytosis

Table 3A. Comparison of the variables between the survival group and death group.

	Survival group (N=37)	Death group (N=23)	*P*
Sex (M:F)	20:17	15:8	0.432
Age (years) (median, range)	5.3 (0.1 - 76.5)	39.3 (0.1 - 89)	0.007*
Primary or neoplasm-induced HLH (%, n/n)	13.5 (5/37)	69.6 (16/23)	<0.001*
EBV (%, n/n)	18.8 (6/32)	22.7 5(5/22)	0.743
Fever (%, n/n)	100 (37/37)	100 (23/23)	-
Splenomegaly	73 (27/37)	91.3 (21/23)	0.107
Hepatomegaly	67.6 (25/37)	56.5 (13/23)	0.421
Neurologic symptom or sign	10.8 (4/37)	17.4 (4/23)	0.468
Rash	35.1 (13/37)	21.7 (5/23)	0.387
Hemoglobin (g/L) (median, range)	99 (51 - 157)	86 (64 - 116)	0.006*
Neutrophil count ($\times 10^9$/L) (median, range)	0.90 (0.13 - 14.1)	1.0 (0.06 - 10.2)	0.638
Platelet count ($\times 10^9$/L) (median, range)	67 (25 - 384)	51 (15 - 117)	0.030*
Triglyceride (mg/dL) (median, range)	228 (47 - 742)	206 (48 - 347)	0.335
Fibrinogen (g/L) (median, range)	1.48 (0.70 - 5.0)	1.40 (0.45 - 6.18)	0.893
Ferritin (μg/L) (median, range)	3,200 (528 - 204,887)	2,935 (185 - 57,186)	0.850
sCD25 (U/mL) (median, range)	4,285 (825 - 10,013) (N=10)	17,647 (1,139 - 27,060) (N=4)	0.142
NK cell activity (%) (median, range)	40 (40 - 2,000) (N=5)	1,074 (148 - 2,000) (N=2)	0.381
Hemophagocytosis (%, n/n)	89.2 (33/37)	100 (23/23)	0.288
AST (U/L) (median, range)	307 (11 - 5137)	285 (22 - 4829)	0.238
ALT (U/L) (median, range)	178 (8 - 2019)	120 (14 - 1043)	0.110
LDH (U/L) (median, range)	1,749 (262 - 5,8611) (N=35)	1164 (437 - 15,062) (N=20)	0.110

Table 3B. Comparison of the variables between the survival group and death group (continued).

T-bil (mg/dL) (median, range)	1 (0.3 - 11.8)	1.2 (0.1 - 8.2)	0.171
D-bil (mg/dL) (median, range)	0.6 (0.1 - 8.8)	1.4 (0 - 6.2) (N=16)	0.710
PT (sec)	12.9 (11 - 22.3) (N=36)	13.8 (12.9 - 16.5)	0.229
INR	1.17 (0.99 - 2.07) (N=36)	1.27 (0.93 - 2.85)	0.368
aPTT (sec)	40.1 (26 - 79.6) (N=35)	38.6 (21.6 - 200)	0.681
CSF pleocytosis	25 (2/8)	10 (2/20)	0.555
CSF proteinosis	27.8 (5/18)	25 (2/8)	1.000
Abnormality on brain MRI (%, n/n)	8.3 (1/12)	66.7 (4/6)	0.022*
HLH-2004/94 Treatment	56.8 (21/37)	30.4 (7/23)	0.064

EBV, Epstein - Barr virus; sCD25, soluble CD25; AST, Aspartate transaminase; ALT, Alanine transaminase; LDH, lactate dehydrogenase; PT, Prothrombin time; INR, International normalized ratio; aPTT, activated partial thromboplastin time; CSF, Cerebrospinal fluid; MRI, Magnetic resonance imaging; HLH, hemophagocytic lymphohistiocytosis

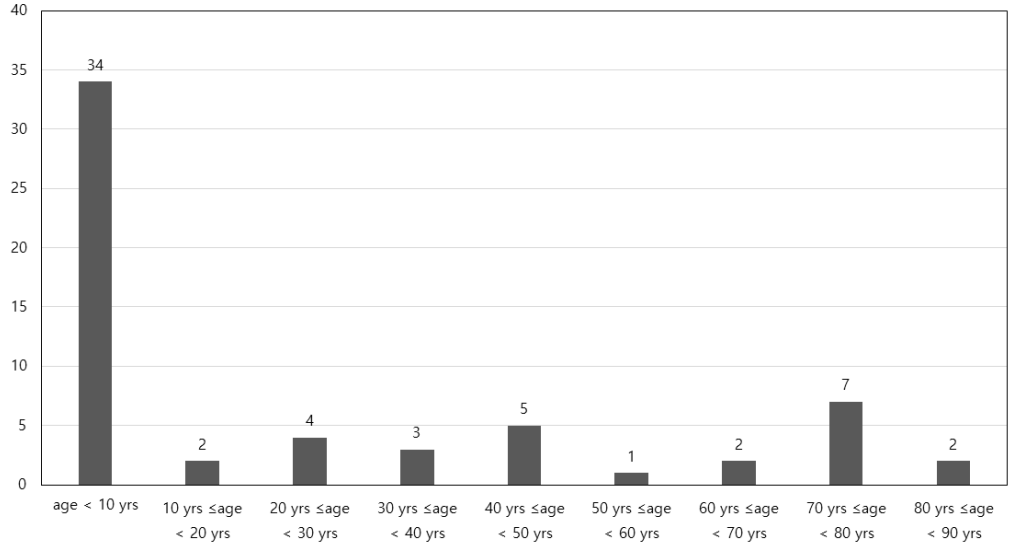
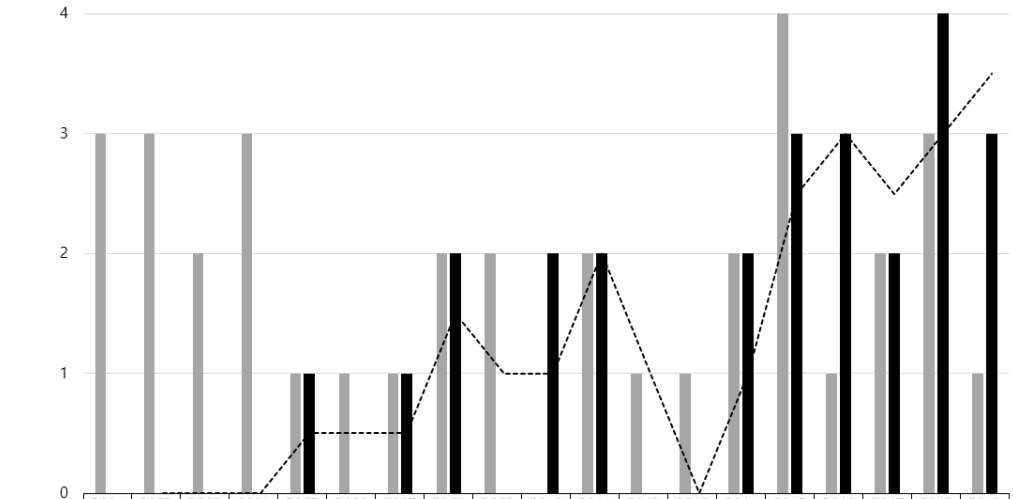


Figure 1. Age distribution of the patients with hemophagocytic lymphohistiocytosis (HLH) (N=60). Although HLH occurred in all age groups, more than half of the patients (56.7%, 34/60) developed the disease at below 10 years of age.



	2001	2002	2003	3004	2005	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019
Children	3	3	2	3	1	1	1	2	2	0	2	1	1	2	4	1	2	3	1
Adults	0	0	0	0	1	0	1	2	0	2	2	0	0	2	3	3	2	4	3

Figure 2. Number of patients diagnosed with hemophagocytic lymphohistiocytosis (HLH) per year. On dividing by the patients' age, the number of adult patients with HLH have been noted to increase each year over time from 2001 to 2019.

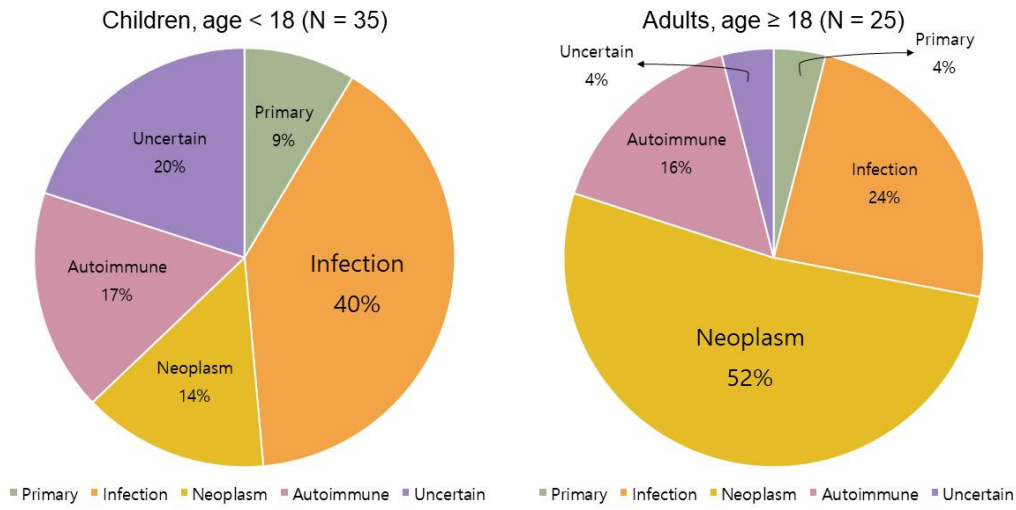


Figure 3. Underlying causes of hemophagocytic lymphohistiocytosis (HLH) according to the age groups. In case of children, infection was the most common cause of HLH (14/35, 40%). However, in adults, neoplasm was the most common cause of HLH (13/25, 52%). The ratio of infection was higher in children (40%) than in adults (24%) (P=0.029). Further, the ratio of neoplasm was higher in adults (52%) than in children (14.3%) (P=0.004).

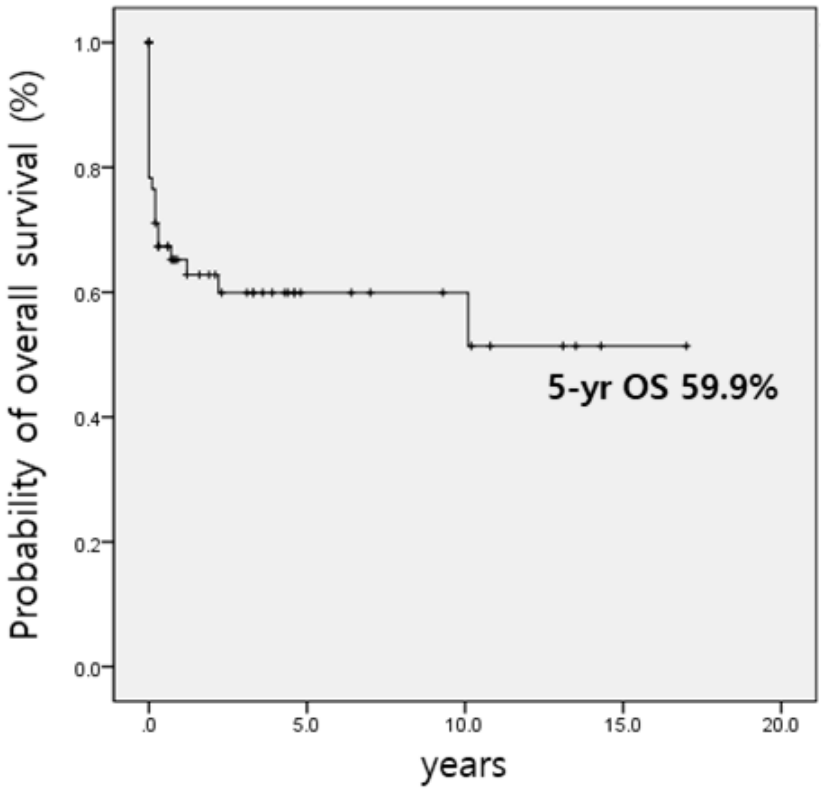


Figure 4. In all the patients with hemophagocytic lymphohistiocytosis (N=60), the 5-year overall survival rate was 59.9% (95% CI: 46.6 - 73.2).

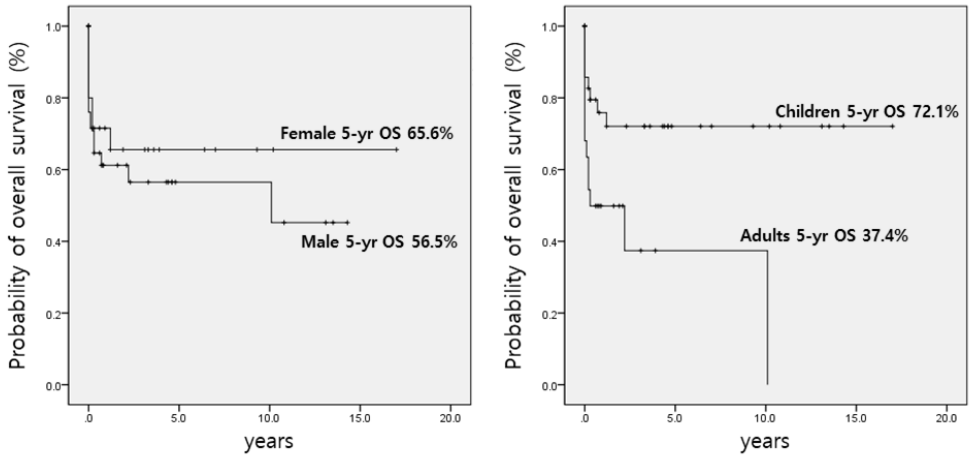


Figure 5. The 5-year overall survival (OS) rate was 56.5% (95% CI: 38.9 - 74.1) in men (N=35) and 65.6% (95% CI: 45.8 - 85.4) in women (N=25). Difference in the OS between men and women was not significant (P=0.557). The 5-year OS rate was 72.1% (95% CI: 56.4 - 87.8) in children (N=35) and 37.4% (95% CI: 11.3-63.5) in adults (N=25). The difference in the OS between children and adults was significant (P=0.007).

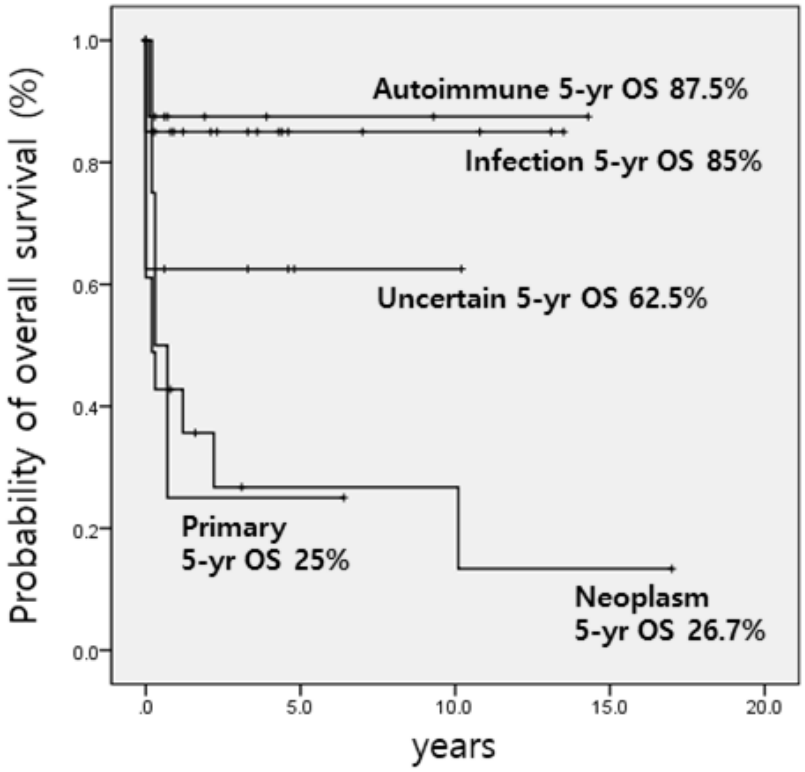


Figure 6. The 5-year overall survival (OS) rate according to the classification of hemophagocytic lymphohistiocytosis (HLH). Difference in the 5-year OS between patients with primary HLH and those with infection-associated HLH was significant (P=0.024). Difference in the 5-year OS between those with neoplasm-associated HLH and those with infection-associated HLH was significant (P=0.001). Difference in the 5-year OS between patients with neoplasm-associated HLH and those with autoimmune-associated HLH was significant (P=0.010).

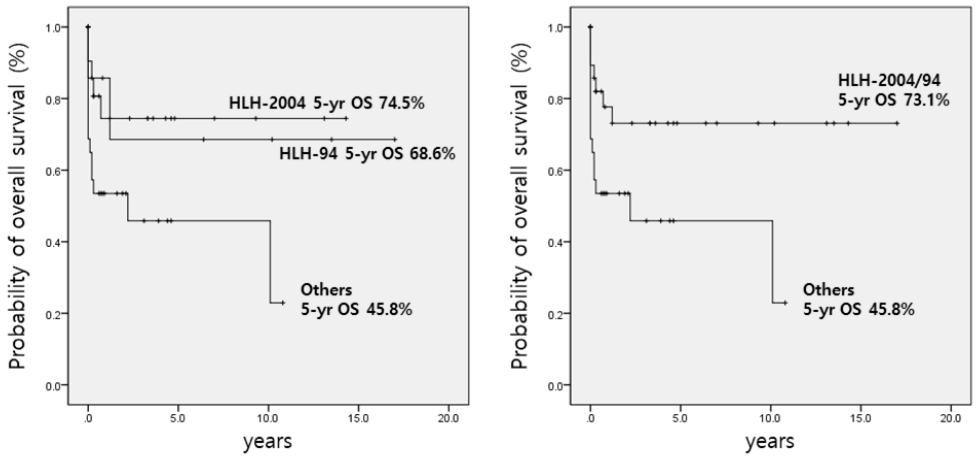


Figure 7. The 5-year overall survival (OS) rate in the patients with hemophagocytic lymphohistiocytosis (HLH) according to the treatment regimen. There was statistical difference in the 5-year OS between HLH-2004/94 (73.1%, 95% CI: 55.9 - 90.3) and others (45.8%, 95% CI: 25 - 66.6) (P=0.017). Additionally, the difference in the 5-year OS between HLH-2004 and HLH-94 protocol was not significant (P=0.904).

4. Discussion

In this study, we retrospectively reviewed the causes and survival rates in both children and adults with different HLH subtypes. Moreover, we analyzed the difference in the clinical characteristics between children and adults with HLH. Based on the statistical analyses of this study, HLH occurrence is observed primarily in children (median age 7 years); however, from the analysis of the duration from 2001 to 2019, the number of adult patients diagnosed with HLH has recently increased. During the review of the medical data for this retrospective study, several patients suspected to have HLH and treated with corticosteroid or immunoglobulin were not included from this study, because they had not performed all tests corresponding to the diagnostic criteria of HLH. Therefore, the increase in the number of diagnoses of adult patients with HLH during the study period is not exactly an increase in HLH cases, but the result of active examinations with an increasing awareness among the medical staff. The awareness of the symptoms of HLH as well as the diagnostic criteria is crucial for physicians, because early diagnosis and prompt therapy are crucial for the outcome given the life-threatening nature of HLH.

As with the previous Korean nationwide pediatric HLH data from the Korea Histiocytosis Working Party,[13] EBV was the main cause of secondary HLH in this study. This is an example of the high proportion of EBV-induced secondary HLH in Asian population.[19] Aside from EBV, various other organisms are also the cause of HLH as reported by previous studies.[13,16] In all the patients of this study, infection was the main cause of HLH in children. However, neoplasm (mainly lymphoma) was the main cause of HLH in adults which is consistent with other

international data.[13,16] In terms of clinical manifestations, hepatomegaly with increased aminotransferase was predominant in pediatric patients with HLH; conversely, splenomegaly was more apparent in adult patients with HLH in this study. This clinical aspect is also considered to be related to the causative disease between children and adults.

Although an accurate comparison is difficult considering the very small number of primary HLHs in this study, the most common cause of primary HLH was FHL type 3 with UNC13D variants as with the previous Korean data.[13] Interestingly, one patient with FHL type 2 (PRF1 variant), in this study, was a 28.3-year-old man. Initially, this patient was diagnosed with T-cell lymphoma that progressed aggressively, and he met the criteria of HLH with multiorgan failure. Because of this extraordinary disease course, the attending physician performed a genetic test and he was eventually diagnosed with adult FHL. Although primary HLH is commonly known to be diagnosed in children, several examples of genetically confirmed HLH in adults have recently been reported.[20-22] Allogenic stem cell transplantation is a recommended therapy in adult primary HLH.[22] Furthermore, a 9.2-year-old pediatric patient with XLP1 (SH2D1A variant) was initially diagnosed with EBV-associated lymphoproliferative disease, which also demonstrated very aggressive features of HLH with multiorgan failure. As there was no pathogenic variant based on the conventional genetic tests for PRF1 and UNC13D, the pediatrician ordered next-generation sequencing, and finally he was diagnosed with XLP1. These two patients with FHL would have been classified as neoplasm-associated secondary HLH unless additional genetic testing was performed. Therefore, the physician must not only perform all tests in the diagnostic criteria, but also consider genetic testing for the accurate diagnosis of HLH. In this study, some patients with FHL may have

been classified as the patients with neoplasm-associated HLH or uncertain cause-HLH because genetic testing has not been thoroughly conducted. Additionally, patients with uncertain cause-HLH may include those who have not been thoroughly screened for underlying infective organisms.

In all the patients with HLH, the 5-year OS rate was 59.9% in this study. Univariate analysis showed that the OS rate of HLH patients was affected by the age of diagnosis, treatment with HLH protocol, anemia, thrombocytopenia, or abnormality on brain MRI. However, the underlying cause, primary or neoplasm-induced HLH, was the only independent risk factor for death by the multivariate analysis. Therefore, it is important to accurately identify the underlying cause for improving the survival rate in patients with HLH. Treatment algorithms for HLH, such as HLH-94 and HLH-2004, are mainly based on pediatric protocols, which may result unnecessary toxicity or overtreatment in adults.[23] Thus, an age-dependent modified therapeutic approach such as individualized tailoring or reduction of treatment duration must be considered.[23]

This study has some limitations. First, this study was performed as retrospectively, thus a possibility of selection bias among the study populations exists. During the review of the chart, several children, as well as adults, had not undergone tests such as soluble CD25 or NK cell activity. Although several patients had the characteristics of HLH, they were not enrolled in this study because they did not undergo the tests and had not met the diagnostic criteria. Therefore, HLH may have been underdiagnosed during the study period. Second, adult patients with HLH were enrolled according to the HLH-2004 criteria, which focused on pediatric patients to clarify the scope of the study subjects. The HLH-2004 criteria has not yet been formally validated for adults,

and thus it continues to be based on expert opinion.[23] Various case series have used modified HLH-2004 criteria.[24]

5. Summary

This is the first Korean study that analyzed the underlying causes and patients' survival in both children and adults with HLH. Additionally, we compared the clinical characteristics according to the age group and analyzed the survival rate according to the HLH subtypes. Primary or neoplasm-induced HLH was found to be an independent risk factor for poor prognosis in patients with HLH. Therefore, it is essential to accurately identify the genetic cause in patients with HLH, and the optimal treatment regimen for neoplasm-induced HLH requires further discussion.

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Analysis of causes and survival of patients with hemophagocytic lymphohistiocytosis at tertiary medical centers in Korea

Kwak, Abraham

Department of Pediatrics
Graduate School

Keimyung University

(Supervised by Professor Shim, Ye Jee)

(Abstract)

Hemophagocytic lymphohistiocytosis (HLH) is a rare but severe life-threatening inflammatory condition if untreated. We aimed to review the causes and survival of both pediatric and adult patients with HLH.

The data of patients who met the HLH criteria from January 2001 to December 2019 were investigated. Their medical records were retrospectively reviewed. In this study, 60 patients with HLH (male:female sex, 35:25) were included. When divided by patient age, the number of adult patients with HLH showed an increase with each year from 2001 to 2019. The median age at diagnosis and median follow-up duration were 7.0 years (range, 0.1 - 83 years) and 8.5 months (range, 0 - 204 months), respectively. Four patients had primary HLH, 48 patients had secondary HLH (20 infection-associated, 18 neoplasm-associated, and 10 autoimmune-associated HLH), and there were eight patients with unknown causes. Further, 28 patients were treated with HLH-2004/94 immunochemotherapy, whereas others were primarily treated with corticosteroids. The 5-year overall survival (OS) rate for all HLH patients was 59.9% (95% confidence intervals (CI): 46.6 - 73.2). The 5-year OS rates for patients with primary,

infection-associated, neoplasm-associated, autoimmune-associated, and unknown cause HLHs were 25% (95% CI: 0 - 67.5), 85% (95% CI: 69.3 - 100), 26.7% (95% CI: 3.6 - 49.8), 87.5% (95% CI: 64.6 - 100), and 62.5% (95% CI: 29 - 96), respectively. According to the multivariate analysis, primary or neoplasm-induced HLHs were a dependent risk factor for death (P=0.036). Although HLH seems to occur primarily in children, its occurrence in adult patients has increased recently. The underlying genetic cause or neoplasm in HLH was a risk factor for death; optimal management for these patients requires to be discussed.

국내 혈구탐식성림프조직구증식증 환자의 원인과 생존률에 대한 분석

곽 아 브 라 함

계명대학교 대학원

의학과 소아청소년과학 전공

(지도교수 심 예 지)

(초록)

혈구탐식성림프조직구증식증(HLH;Hemophagocytic lymphohistiocytosis)은 희귀하지만 매우 위험한 질환으로 치료하지 않을 시 사망에 이를 수 있는 염증성 질환이다. 이에 저자들은 HLH 소아와 성인 환자에서의 원인과 생존률에 대해 살펴보았다. 2001 년 1 월부터 2019 년 12 월까지 HLH 진단 기준에 부합하는 환자군을 대상으로 하였다. 이들의 의무기록을 후향적으로 조사하였다. 대상 기간 동안 HLH 환자들은 전체 60 명(남:녀비, 35;25)이었다. 진단 당시 나이 중앙값은 7 세(0.1-8.3 세 범위)를 보였고 추적 기간의 중앙값은 8.5 개월(0-204 개월 범위)이었다. 전체 환자들 중 일차성 HLH 환자는 4 명, 이차성 HLH(감염 관련 20 명, 신생물 관련 18 명, 자가면역 관련 10 명) 환자는 48 명이었고 8 명의 HLH 환자들의 원인은 불명확하였다. HLH 환자들을 나이에 따라 분류하였을 때에 성인에서 진단된 HLH 환자들의 수가 2001 년도부터 2019 년까지 매년 증가하는 경향을 보였다. 전체 HLH 환자들 중에서 28 명의 환자들이 HLH-2004/94 항암 치료를 하였던 데에 반해 다른 환자들은 스테로이드를 주류로 하는 치료를 시행하였다. 5 년 생존률은 전체 환자들에서 59.9% (95% 신뢰 구간:46.6-73.2)을 보였다. 원인에 따른 5 년 생존률은 일차성, 감염 관련, 신생물 관련, 자가면역과 원인 불명의 원인에 따라 각각 25% (95% 신뢰 구간: 0 - 67.5), 85% (95% 신뢰 구간: 69.3 - 100), 26.7% (95% 신뢰 구간: 3.6 - 49.8), 87.5% (95% 신뢰 구간: 64.6 - 100), 62.5% (95% 신뢰 구간: 29 - 96)를 보였다. 사망률에 영향을 끼치는 요인들로 다변량 분석을 실시

하였을 때에 일차성 혹은 신생물 관련 HLH 여부가 유의미한 독립 요인이었다(P 값= 0.037). 비록 HLH는 주로 소아에서 발생하는 질환이지만 최근 성인에서도 HLH의 발생률이 증가됨을 보이고 있다. 기저 유전 결함이 있거나 신생물 관련 이상으로 인한 HLH 발생 여부가 사망률에 유의미한 독립 요인이었다. 이러한 원인을 가진 HLH 환자들에 있어서 최적의 치료에 대한 고찰이 더욱이 요구되는 바이다.