

Etoposide as an Effective Drug for Adult Macrophage Activation Syndrome: A Retrospective Study

Lingbo He

Capital Medical University Affiliated Beijing Friendship Hospital

Zhili Jin

Capital Medical University Affiliated Beijing Friendship Hospital

Menghan Liu

Capital Medical University Affiliated Beijing Friendship Hospital

Tingting Cui

Capital Medical University Affiliated Beijing Friendship Hospital

Lin Wu

Capital Medical University Affiliated Beijing Friendship Hospital

Zhao Wang

Capital Medical University Affiliated Beijing Friendship Hospital

Yini Wang (✉ wangyini@ccmu.edu.cn)

Capital Medical University Affiliated Beijing Friendship Hospital <https://orcid.org/0000-0001-8545-2660>

Research

Keywords: Hemophagocytic lymphohistiocytosis, Autoimmune disease, Macrophage activation syndrome, Etoposide

Posted Date: January 14th, 2021

DOI: <https://doi.org/10.21203/rs.3.rs-144071/v1>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License.

[Read Full License](#)

Abstract

Background

Autoimmune disease related hemophagocytic syndrome, in other words, macrophage activation syndrome(MAS), is a rare, but lethal complication of autoimmune disease. At present, specific treatment guidelines for adult MAS have not been formulated, most experience are derived from children, researches about etoposide are scarce. As the importance of etoposide in the initial treatment had been proved in other subtypes of hemophagocytic syndrome, the objective of this study is to investigate the effectiveness of etoposide in the treatment of the adult macrophage activation syndrome.

Result

74 patients with autoimmune disease related hemophagocytic syndrome were involved in this study, they were divided into two groups based on initial treatment, group 1(n=53): initial therapy did not contain etoposide, group 2(n=21): initial therapy contained etoposide. The overall response rate and complete response rate of group 2 were significantly higher than group 1(ORR 90.5% vs 24.5%, CRR 33.3% vs 3.8%, $P<0.05$). Patients with different HLH remission states have significantly different prognosis($P<0.001$).

Conclusion

Adopting VP-16 in initial treatment can significantly increase the OR rate and CR rate of adult MAS patients, and the HLH states influenced the prognosis significantly.

Background

Hemophagocytic syndrome(HPS) is a hyperinflammatory condition with high mortality, characterized by the cytokine storm originate from excessive activated macrophages and T cells[1], also known as Hemophagocytic lymphohistiocytosis(HLH). HLH is defined into two forms as primary and secondary, secondary hemophagocytic lymphohistiocytosis(sHLH) can be triggered by infection, malignancy and autoimmune diseases(AID)[2], autoimmune disease associated hemophagocytic syndrome(AID-HLH), in other words, macrophage activation syndrome is a rare, but lethal complication of AID. Approximately the mortality of sHLH is 41%[3], although prognosis of MAS is better than other subtypes, for AID patients, accompanying HLH reduced the survival rate dramatically[4]. Currently, specific treatment guidelines for adult MAS have not been formulated, glucocorticoid pulse therapy is the major treatment for MAS patients according to experience derived from children. As a key drug in HLH-94/2004 regimens, the importance of etoposide in the initial treatment had been proved in other subtypes of HLH, studies on etoposide(VP-16) application in adult MAS are still limited. Our research aims to explore the importance of VP-16 in the treatment of adult MAS by analyzing the clinical characteristics, laboratory indicators, regimens and prognosis of adult MAS.

Methods

Patients

Clinical data of patients over 18 years old with autoimmune disease related hemophagocytic syndrome diagnosed in Beijing Friendship Hospital affiliated to Capital Medical University from December 2014 to November 2019 were collected in this study. All patients met HLH-2004 diagnostic criteria[21]. The survival time was calculated from the time patients were diagnosed as HLH until death or February 2020.

Assessment of therapy

Efficacy was evaluated every 2 weeks after initiating therapy, Complete response(CR) was defined as the normalization of all quantifiable symptoms and laboratory markers of HLH, including levels of soluble CD25, ferritin, triglyceride, hemoglobin levels, neutrophil and platelet counts, and alanine aminotransferase(ALT). Partial response(PR) was defined as improvement in two or more of the following quantifiable symptoms and laboratory markers by 2 weeks: 1.5-fold decrease in soluble CD25 response; ferritin and triglyceride decreases of at least 25%; an increase of at least 100% to $>0.5 \times 10^9/L$ in patients with an initial neutrophil count of $<0.5 \times 10^9/L$; an increase of at least 100% to $>2.0 \times 10^9/L$ in patients with an initial neutrophil count of $0.5-2.0 \times 10^9/L$; and a decrease of at least 50% in patients with initial ALT levels $>400u/l$. In addition, the body temperature must have reverted to normal ranges for either CR or PR to be diagnosed. No response(NR):Failure to achieve PR was defined as no response[23, 24]. The overall response rate was defined as the ratio of patients with CR and PR to all patients.

Research design

Patients were divided into two groups according to initial therapy. Group 1: VP-16 was excluded in initial regimen, Group 2: VP-16 was included in initial regimen. The gender, age, clinical characteristics, laboratory indicators, underlying disease, regimens and prognosis of two groups were analyzed retrospectively.

Statistical analysis

SPSS 24.0 statistical software was adopted, all normally distributed data were represented by means \pm standard deviations, all data that were not distributed normally were represented by median and range. T test is used for normally distributed data and rank sum test is used for non-normal distribution data. $P < 0.05$ was considered to denote a significant difference. Some patients lack of sCD25 and NK cell activity results, missing data was deleted.

Results

General information

A total of 74 patients with autoimmune disease related hemophagocytic syndrome were involved in this study, female patients representing 78.4% of the total number (58/74), male representing 21.6% (16/74). The male to female ratio was 1:3.6. The median age of the patients was 31 years (16~72 years). Among

these cases, underlying diseases included AOSD, SLE, undifferentiated connective tissue disease(UCTD), rheumatoid arthritis(RA) and other variety of autoimmune diseases(Fig 1).

Clinic Features

All patients(74/74) had fever at diagnosis, 60.8%(45/74) of patients had skin rashes, 55.4%(41/74) of patients had joint pain. Splenomegaly was found in 85.1%(63/74) patients, and hepatomegaly had a lower rate at 9.4%(7/74). Leukocytopenia, anemia and thrombocytopenia were found in 41.9%(31/74), 51.4%(38/74), 59.5%(44/74) of patients, respectively. Hepatic transaminases elevation was shown in 63.5%(47/74) patients, the incidence of hyperlipidemia, hyperbilirubinemia and hypofibrinogenemia was 43.2%(32/74), 50.0%(37/74) and 40.5%(30/74). The incidence of elevated serum ferritin was 97.3% (72/74), which is extremely high. Only part of patients were able to accomplish the tests of soluble CD25(sCD25) or NK cell activity by the time of HLH diagnosis, 67.3%(33/49) of them showed sCD25 elevation, and 75.0%(30/40) showed low NK cell activity(Fig 2).

Treatment

Patients were defined into two groups based on the initial regimens. Group 1(n=53): the initial treatment plan didn't contain VP-16, the specific treatment includes corticosteroids, corticosteroids plus cyclosporine A, corticosteroids plus methotrexate and corticosteroids plus hydroxychloroquine. Group 2(n=21): the initial treatment included VP-16, the specific treatment options includes HLH-94, HLH-2004 and DEP regimen. Group 2 patients had lower ALT and T-BIL than group 1 patients, other baseline data showed no significant difference(Table 1).

Table 1. Initial clinical characteristics of the patients according to group			
	Group 1	Group 2	P value
Gender			
Male	12	4	0.980
Female	41	17	
Age	33(18-62)	27(18-72)	0.118
Fever(>38.5°C,n)	53	21	
Splenomegaly (n)	44	19	0.652
Hepatomegaly (n)	6	1	0.668
Skin rashes (n)	33	12	0.886
Joint pain (n)	30	11	0.944
Haemophagocytosis (n)	32	13	1.000
WBC (x10 ⁹ /L)	5.94±5.28	6.78(0.50~29.30)	0.300
NEU (x10 ⁹ /L)	3.10(0.02~25.68)	5.24(0.25~27.10)	0.260
HGB (g/L)	91(48~145)	96.76±26.30	0.820
PLT (x10 ⁹ /L)	95.81±65.59	135.00(25.00~471.00)	0.170
ALT (U/L)	67.00(2.10~2252.00)	23.00(5.00~926.00)	0.022
AST (U/L)	75.50(7.65~2057.00)	23.6(12.00~1579.00)	0.164
T-BIL (µmol/L)	16.87(5.82~554.80)	11.89(4.05~28.41)	0.003
TG (mmol/L)	2.80±1.07	2.72±0.83	0.679
Fbg (g/L)	1.76(0.56~4.49)	2.20±1.50	0.838
Fer (ng/mL)	2000.00(466.00~67830.00)	3343.00(109.60~100000.00)	0.527
sCD25 (pg/mL)	11934.00(639.00~40435.00)	9790.00 (2337.00~26732.00)	0.401
NK cell activity (%)	12.05±5.18	12.55±3.92	0.727

WBC, white blood cell count; NEU, neutrophil count; HGB, haemoglobin concentration; PLT, platelet count; ALT, alanine aminotransferase; AST, aspartate aminotransferase; T-BIL, serum Total Bilirubin; TG, triglycerides; Fbg, fibrinogen; Fer, serum ferritin; sCD25, soluble CD25.

After initial therapy, of 53 patients in group 1, 2 achieved CR(2/53,3.8%), 11 achieved PR(11/53,20.8%), the overall response rate was 24.5%(13/53), 40 patients with no response were confirmed as refractory

MAS. Among patients in group 2, the overall response rate was 90.5%(19/21), with a CR rate of 33.3% (7/21) and a PR rate of 57.1%(12/21). There were significant differences in ORR($P<10^{-6}$) and CRR($P=0.002$) between two groups. 37 refractory MAS patients received salvage treatment contains VP-16, 24.3%(9/37) of them achieved CR and 67.6%(25/37) achieved PR, the ORR was 91.9%(34/37).

A total of 65 patients reached at least PR eventually, according to whether VP-16 was used or not, we divided this part of patients into two groups, group A: VP-16 was not included during the whole treatment, and group B: VP-16 was administrated in the treatment. We compared laboratory indicators of two groups at the remission stage of HLH, the data suggested patients in group B had lower sCD25 and higher NK cell activity than group A, while blood cell counts had no significant differences between two groups(Table 2).

Table 2. Clinical characteristics of the patients at remission stage according to group.			
	Group A	Group B	P value
WBC ($\times 10^9/L$)	6.79(0.60~16.94)	7.71 \pm 4.74	0.768
NEU ($\times 10^9/L$)	4.29(0.13~14.91)	6.01 \pm 4.32	0.491
HGB (g/L)	96(58~132)	101.94 \pm 13.40	0.987
PLT ($\times 10^9/L$)	197(42~395)	194.50(60.00~426.00)	0.993
ALT (U/L)	43.00(6.00~283.00)	24.00(6.00~189.00)	0.053
AST (U/L)	41.11 \pm 28.71	24.77 \pm 16.23	0.008
T-BIL ($\mu\text{mol/L}$)	12.31(6.78~171.80)	13.16 \pm 8.82	0.286
TG (mmol/L)	2.05 \pm 1.88	2.23 \pm 1.36	0.754
Fbg (g/L)	2.03(0.77~3.68)	2.43 \pm 1.14	0.394
Fer (ng/mL)	1198.00(62.40~8234.90)	374.60(6.40~17300.00)	0.071
sCD25 (pg/mL)	4442.00(742.00~22826.00)	1441.00 (139.00~9177.00)	0.014
NK cell activity (%)	13.54(9.89~16.44)	16.25(6.38~18.7)	0.010

Survival

The survival time was calculated from the time patients were diagnosed HLH until death or January 2020. A total of 9 deaths occurred in 74 patients with a mortality of 12.16%. The mortality rate of group 1 was 13.2%(7/53), and that of group 2 was 9.5%(2/21). Of patients who achieved PR, 1 patient died(1/48,2.1%), nonresponsive patients were all died within 6 weeks after diagnosis with a median survival of 29 days, the HLH states influenced the prognosis significantly(Fig 3, $P<0.01$).

Discussion

HLH is a rare, but lethal complication of adult autoimmune diseases patients, most common in AOSD, accounting for 58.1% in our study, consistent with literature reports[5]. HLH may occur at each stage of autoimmune diseases, even becomes the initial manifestation, but the specific mechanism is still not clear, polymorphism of *UNC13D* is possibly a predisposing factor[6, 7].

The treatment strategy of HLH contains two parts, short term and long term. The short-term strategy focuses on the control of cytokine storm to suppress the excessive inflammatory status, and the long-term strategy targets on the underlying diseases[8]. Although the prognosis of adult acquired HLH was mainly determined by the underlying diseases, fail to stabilize the inflammatory condition may lead to a rapid death[9, 10].

In order to achieve short-term goal, diagnosis should be established promptly and effective regimens should be taken. Fever and abnormal hemogram are common in patients with autoimmune disease[11, 12], makes it difficult to distinguish HLH at the early stage[13]. Studies proves that NK cell activity and sCD25 are sensitive indicators for early diagnosis of HLH, a ferritin level above 10,000ug/L appears to be specific and sensitive for HLH diagnosis[14]. In addition to assessing the activity, examination of NK cell activity, sCD25 and serum ferritin are valuable for patients with rapid condition deterioration. Meanwhile, the significance of sCD25 in autoimmune diseases has gradually attracted attention[15, 16], whether routine monitoring of sCD25 is needed or not in patients with autoimmune diseases still needs further study.

At present, glucocorticoid pulse therapy is the major treatment for MAS patients[17, 18], but most researches were based on children, although adult HLH presented similar clinical features as in children, adult HLH still has different characteristics with pediatric HLH, so far there is no unified therapy for adult MAS. Song et al[19] found that in EBV-HLH patients, the use of VP-16 in initial treatment has no significant relationship with the prognosis for patients under the 18 years old, but for adult patients, the prognosis was improved, our data demonstrated that VP-16 contained initial therapy reveals same results in adult MAS patients. In our study, the mortality of MAS patients was 12.16%, and the prognosis of patients with different HLH remission status was significantly different, only 24.5% patients were responsive to glucocorticoid pulse therapy, adopting VP-16 in the initial regimen reached a significantly higher ORR and CRR. Even for patients nonresponsive to glucocorticoid pulse therapy, VP-16 contained therapies lead to a high response rate as 91.9%.

A significant portion of patients was transferred to our center as a result of poor treatment outcome or difficulties in diagnosis, consequently, referral bias existed in this study, which may influence the overall response rate of glucocorticoid pulse therapy. We believe it reflected effectiveness of the etoposide.

VP-16 is one of cell cycle specific drugs which acts on topoisomerase II. By blocking the reconnection of DNA strand by forming drug enzyme DNA complex, anti-tumor effect was achieved. VP-16 can selectively bypass the over activated T cells to inhibit the activation of monocyte macrophages, thereby controlling

HLH by reducing the generation of cytokine storm without compromising quiescent phase and memory T cells[20]. In accordance with whether VP-16 was used or not, we analyzed patients' laboratory indicators, and came to a same conclusion with Henter et al[21], Arca et al[22], efficacy of VP-16 on HLH is far outstrip than the risk of hemogram deterioration.

Conclusion

Determined by the underlying disease, MAS patients had a better prognosis than other type of HLH patients, but occurrence of HLH significantly reduced the survival rate of patients with autoimmune diseases[4], and we demonstrated that remission of HLH, especially CR, is the key to improve the prognosis of MAS patients. Currently, there is no unified treatment guideline for adult MAS patients, our study has confirmed that admitting VP-16 in the initial regimens significantly increase the CR and OR rate of adult MAS patients, and did not cause severe adverse reactions such as bone marrow suppression. VP-16 is safe and effective for MAS patients.

Declarations

Ethics approval and consent to participate

The study was approved by the Ethics Committee at Beijing Friendship Hospital

Consent for publication

Not applicable.

Availability of data and materials

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Competing interests

The authors declare that they have no competing interests.

Funding

This work was supported by Beijing Municipal Administration of Hospital Clinical Medicine Development of Special Funding Support (XMLX201823); National Natural Science Foundation of China (81871633); Beijing Natural Science Foundation (7181003); Beijing Municipal Administration of Hospitals' Ascent Plan (DFL20180101).

Author's contributions

Yini Wang and Zhao Wang designed the study manuscript and Lingbo He wrote the initial draft. All authors were involved in data acquisition and analysis. All authors read and approved the final manuscript.

Acknowledgements

We sincerely thank all patients and their families for participating in the study. We further thank the Beijing Friend hospital for the support.

References

1. Janka GE, Lehmborg K. Hemophagocytic syndromes—an update. *Blood Rev.* 2014;28(4):135-42; doi: 10.1016/j.blre.2014.03.002.
2. Al-Samkari H, Berliner N. Hemophagocytic Lymphohistiocytosis. *Annu Rev Pathol.* 2018;13:27-49; doi: 10.1146/annurev-pathol-020117-043625.
3. Ramos-Casals M, Brito-Zerón P, López-Guillermo A, Khamashta MA, Bosch X. Adult haemophagocytic syndrome. *Lancet.* 2014;383(9927):1503-16; doi: 10.1016/s0140-6736(13)61048-x.
4. Ahn SS, Yoo BW, Jung SM, Lee SW, Park YB, Song JJ. Application of the 2016 EULAR/ACR/PRINTO Classification Criteria for Macrophage Activation Syndrome in Patients with Adult-onset Still Disease. *J Rheumatol.* 2017;44(7):996-1003; doi: 10.3899/jrheum.161286.
5. Atteritano M, David A, Bagnato G, Beninati C, Frisina A, Iaria C, et al. Haemophagocytic syndrome in rheumatic patients. A systematic review. *Eur Rev Med Pharmacol Sci.* 2012;16(10):1414-24.
6. Zhang M, Behrens EM, Atkinson TP, Shakoory B, Grom AA, Cron RQ. Genetic defects in cytolysis in macrophage activation syndrome. *Curr Rheumatol Rep.* 2014;16(9):439; doi: 10.1007/s11926-014-0439-2.
7. Zhang K, Biroschak J, Glass DN, Thompson SD, Finkel T, Passo MH, et al. Macrophage activation syndrome in patients with systemic juvenile idiopathic arthritis is associated with MUNC13-4 polymorphisms. *Arthritis Rheum.* 2008;58(9):2892-6; doi: 10.1002/art.23734.
8. Janka GE, Lehmborg K. Hemophagocytic lymphohistiocytosis: pathogenesis and treatment. *Hematology Am Soc Hematol Educ Program.* 2013;2013:605-11; doi: 10.1182/asheducation-2013.1.605.
9. Huang W, Wang Y, Wang J, Zhang J, Wu L, Li S, et al. [Clinical characteristics of 192 adult hemophagocytic lymphohistiocytosis]. *Zhonghua Xue Ye Xue Za Zhi.* 2014;35(9):796-801; doi: 10.3760/cma.j.issn.0253-2727.2014.09.003.
10. Rosado FG, Kim AS. Hemophagocytic lymphohistiocytosis: an update on diagnosis and pathogenesis. *Am J Clin Pathol.* 2013;139(6):713-27; doi: 10.1309/ajcp4zdkj4icouat.
11. Justiz Vaillant AA, Goyal A, Bansal P, Varacallo M. Systemic Lupus Erythematosus (SLE). In: *StatPearls.* Treasure Island (FL): StatPearls Publishing Copyright © 2020, StatPearls Publishing LLC.; 2020.

12. Kadavath S, Efthimiou P. Adult-onset Still's disease-pathogenesis, clinical manifestations, and new treatment options. *Ann Med*. 2015;47(1):6-14; doi: 10.3109/07853890.2014.971052.
13. Boom V, Anton J, Lahdenne P, Quartier P, Ravelli A, Wulffraat NM, et al. Evidence-based diagnosis and treatment of macrophage activation syndrome in systemic juvenile idiopathic arthritis. *Pediatr Rheumatol Online J*. 2015;13:55; doi: 10.1186/s12969-015-0055-3.
14. Allen CE, Yu X, Kozinetz CA, McClain KL. Highly elevated ferritin levels and the diagnosis of hemophagocytic lymphohistiocytosis. *Pediatr Blood Cancer*. 2008;50(6):1227-35; doi: 10.1002/pbc.21423.
15. Zhang RJ, Zhang X, Chen J, Shao M, Yang Y, Balaubramaniam B, et al. Serum soluble CD25 as a risk factor of renal impairment in systemic lupus erythematosus - a prospective cohort study. *Lupus*. 2018;27(7):1100-6; doi: 10.1177/0961203318760993.
16. Russell SE, Moore AC, Fallon PG, Walsh PT. Soluble IL-2R α (sCD25) exacerbates autoimmunity and enhances the development of Th17 responses in mice. *PLoS One*. 2012;7(10):e47748; doi: 10.1371/journal.pone.0047748.
17. Gavand PE, Serio I, Arnaud L, Costedoat-Chalumeau N, Carvelli J, Dossier A, et al. Clinical spectrum and therapeutic management of systemic lupus erythematosus-associated macrophage activation syndrome: A study of 103 episodes in 89 adult patients. *Autoimmun Rev*. 2017;16(7):743-9; doi: 10.1016/j.autrev.2017.05.010.
18. Carter SJ, Tattersall RS, Ramanan AV. Macrophage activation syndrome in adults: recent advances in pathophysiology, diagnosis and treatment. *Rheumatology (Oxford)*. 2019;58(1):5-17; doi: 10.1093/rheumatology/key006.
19. Song Y, Wang Y, Wang Z. Requirement for etoposide in the initial treatment of Epstein-Barr virus-associated haemophagocytic lymphohistiocytosis. *Br J Haematol*. 2019;186(5):717-23; doi: 10.1111/bjh.15988.
20. Johnson TS, Terrell CE, Millen SH, Katz JD, Hildeman DA, Jordan MB. Etoposide selectively ablates activated T cells to control the immunoregulatory disorder hemophagocytic lymphohistiocytosis. *J Immunol*. 2014;192(1):84-91; doi: 10.4049/jimmunol.1302282.
21. Henter JI, Horne A, Aricó M, Egeler RM, Filipovich AH, Imashuku S, et al. HLH-2004: Diagnostic and therapeutic guidelines for hemophagocytic lymphohistiocytosis. *Pediatr Blood Cancer*. 2007;48(2):124-31; doi: 10.1002/pbc.21039.
22. Arca M, Fardet L, Galicier L, Rivièrè S, Marzac C, Aumont C, et al. Prognostic factors of early death in a cohort of 162 adult haemophagocytic syndrome: impact of triggering disease and early treatment with etoposide. *Br J Haematol*. 2015;168(1):63-8; doi: 10.1111/bjh.13102.
23. Marsh RA, Allen CE, McClain KL, Weinstein JL, Kanter J, Skiles J, et al. Salvage therapy of refractory hemophagocytic lymphohistiocytosis with alemtuzumab. *Pediatr Blood Cancer*. 2013;60(1):101-9; doi: 10.1002/pbc.24188.
24. Wang Y, Huang W, Hu L, Cen X, Li L, Wang J, et al. Multicenter study of combination DEP regimen as a salvage therapy for adult refractory hemophagocytic lymphohistiocytosis. *Blood*.

Figures

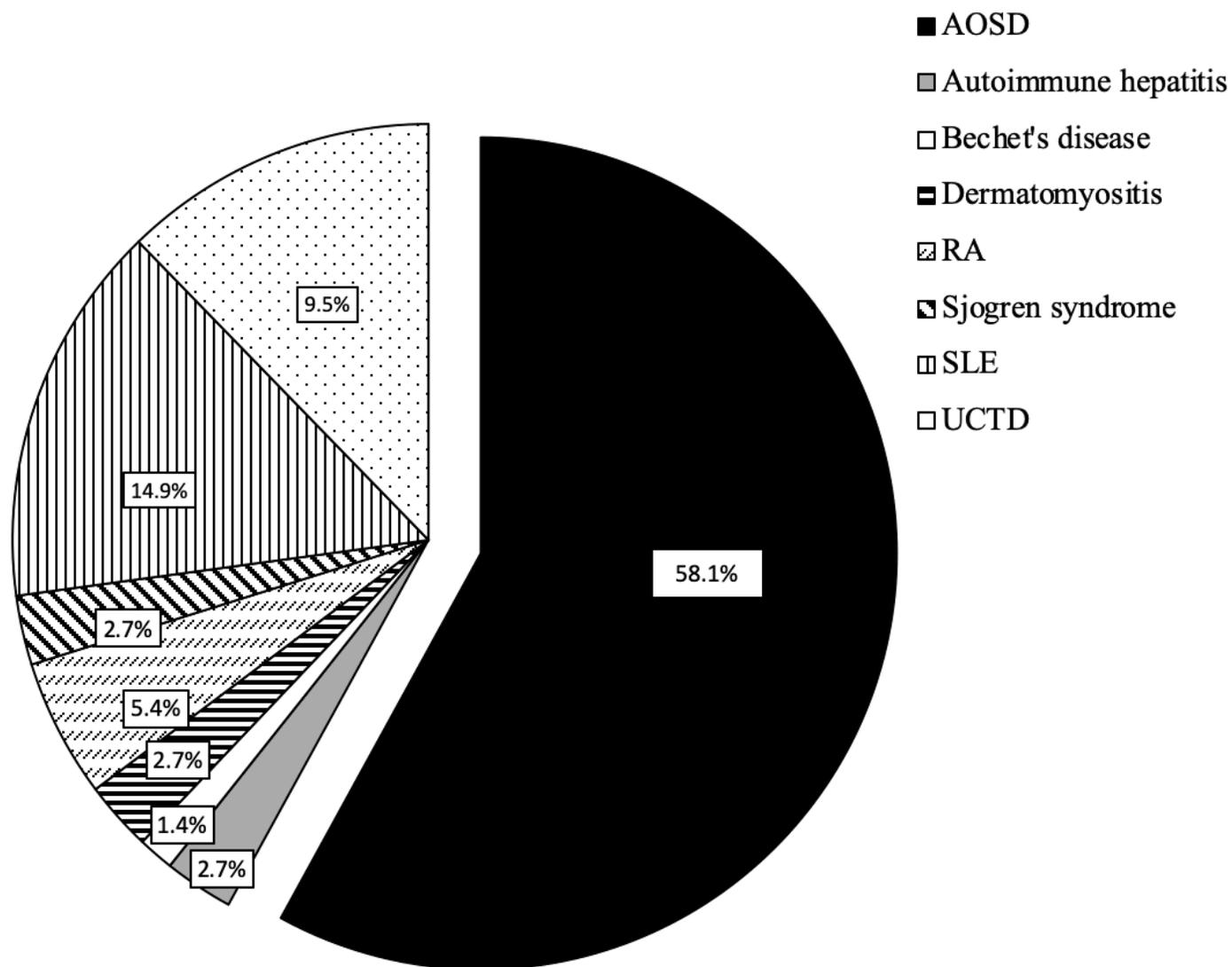


Figure 1

Underlying autoimmune diseases of 74 AAHS patients. 43 with AOSD, 11 with SLE, 9 with UCTD, 4 with RA, 2 patients each with autoimmune hepatitis, dermatomyositis and sjogren syndrome, 1 with Bechet's disease.

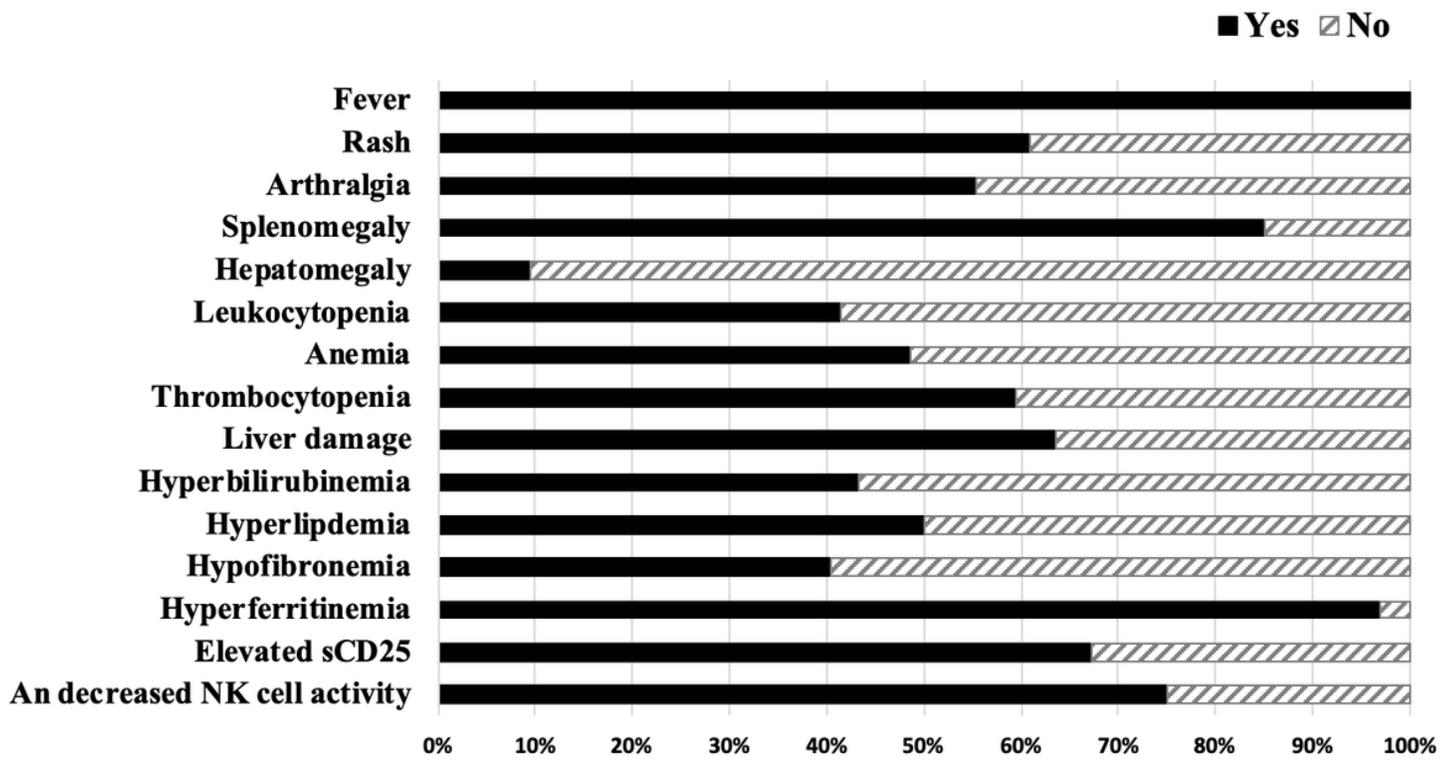


Figure 2

Clinical characteristics of 74 patients.

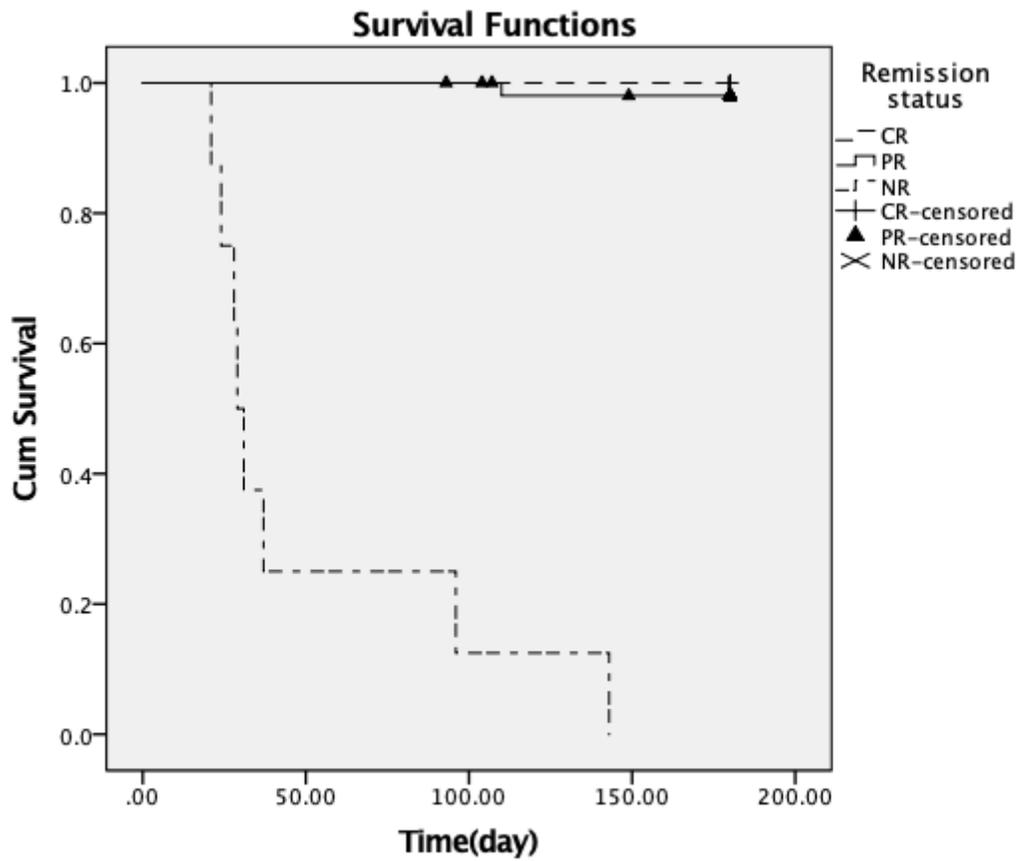


Figure 3

The survival curves of patients based on their remission status of HLH.