Efficacy of hemoadsorption using HA330-II perfusion column in secondary HLH patients: two cases report and literature review.

Qian Gao[1], Chun Zhao[1], Yujuan Wang[1], Wei Wang[1], Yi Yin[1], Xiaowei Xin[1], Xiaoru Wang[1], and Youpeng Jin[1]

[1]Shandong Provincial Hospital

March 26, 2021

Abstract

Abstract: Hemophagocytic lymphohistiocytosis (HLH) is a severe and potentially lethal disorder characterized by overwhelming immune activation and inflammation. Several blood purification methods have been reported to be successfully practiced as adjuvant therapy for hypercytokinamia complicated with HLH. We firstly reported two cases of secondary HLH children received hemoadsorption therapy with HA330-II perfusion cartridge combined with CVVHDF. The marked decrease in IL-6 plasma levels and stabilization of organ function were observed after treatment, which proved the efficiency of HA330-II perfusion column.
Keywords: hemoadsorption, blood purification, Hemophagocytic lymphohistiocytosis, pediatric

Efficacy of hemoadsorption using HA330-II perfusion column in secondary HLH patients: two cases report and literature review.

Full author list: Youpeng Jin[1], Qian Gao[2]; Chun Zhao[1]; Yujuan Wang[1]; Wei Wang[1]; YI Yin[1]; Xiaowei Xin[1] and Xiaoru Wang[1];

1 Pediatric Intensive Care Unit, Shandong Provincial Hospital Affiliated to Shandong First Medical University; Shandong Provincial Hospital, Cheeloo College of Medicine, Shandong University, Jinan, Shandong, 250021, China.

2 Pediatric Intensive Care Unit, Shandong Provincial Hospital, Cheeloo College of Medicine, Shandong University, Jinan, Shandong, 250021, China.

Correspondence: Youpeng Jin, Pediatric Intensive Care Unit, Shandong Provincial Hospital Affiliated to Shandong First Medical University; Shandong Provincial Hospital, Cheeloo College of Medicine, Shandong University, Jinan, Shandong, 250021, China.

Phone: 15168863809

Email:*jinyp79@126.com*

Address: 324 Jingwu Road, Jinan, Shandong, China.

Funding

Jinan Clinical Medical Science and Technology Innovation Program (201704071)

Jinan Clinical Medical Science and Technology Innovation Program (202019177)

Abstract

Hemophagocytic lymphohistiocytosis (HLH) is a severe and potentially lethal disorder characterized by overwhelming immune activation and inflammation. Several blood purification methods have been reported

to be successfully practiced as adjuvant therapy for hypercytokinamia complicated with HLH. We firstly reported two cases of secondary HLH children received hemoadsorption therapy with HA330-II perfusion cartridge combined with continuous veno-venous hemodiafiltration (CVVHDF). The marked decrease in interleukin (IL)-6 plasma levels and stabilization of organ function were observed after treatment, which proved the efficiency of HA330-II perfusion column.

Keywords: hemoadsorption, CBP, Hemophagocytic lymphohistiocytosis, pediatric

1 INTRODUCTION

Hemophagocytic lymphohistiocytosis (HLH) is a severe and potentially lethal disorder characterized by overwhelming immune activation and inflammation [1]. Primary HLH mostly occurs in childhood and is caused by genetic defects leading to impaired function of natural killer cells and cytotoxic T cells which lead to bouts of increased cytokine release and severe inflammation. Secondary HLH is encountered in association with some viral infections, auto-inflammatory/autoimmune diseases, or lymphoma [2]. For both primary and secondary HLH, there exist excessive activation of T cells and macrophages and therefore lead to "cytokine storm" as well as multiple organ disorder. So the most important thing is to control the inflammatory cytokine storm in the treatment of HLH. The conventional therapy included steroid, intravenous immunoglobulin (IVIG) and immunosuppressant. Recently, several blood purification methods have been reported to be successfully practiced as adjuvant therapy to resolve hypercytokinamia complicated with HLH [3, 4].

In the current study, we firstly reported two cases of secondary HLH children received hemoadsorption therapy with HA330-II perfusion column.

2.1 Case 1

A 4-year-old male was admitted to hospital due to diarrhea and seizure caused by unknown pathogen. He received antibiotic (meropenem) and anticonvulsive treatment (carbamazepine) for two days. However, the patient presented hyperpyrexia, erythra and multiple organ dysfunction syndrome with coagulopathy (INR 5.46, APTT 108s), respiratory insufficiency, and hypotension. He was subsequently transferred to pediatric intensive care unit (PICU). With regard to the erythra and fever secondary to antiepileptic drug (carbamazepine), antiepileptic Drug-Induced Hypersensitivity Syndrome was diagnosed. The patient suffered from hemoglobinopenia, thrombopenia, hepatosplenomegaly, hyperferritinemia (ferritin 726.9 ng/ml, normal range 11.0-306.8 ng/mL), hypofibrinogenemia (fibrinogen 0.60 g/L, normal range 1.50-4.35 g/l), hypertriglyceridemia (triglycerides 4.2 mmol/L, normal range 0.4-1.8 mmol/L) and low NK cell activity (2.06 %, normal range 5-26 %) (Table 1). Five of eight diagnostic criteria were found to be fulfilled, and the patient was diagnosed with HLH based on HLH-2004 guideline. Bone marrow biopsy showed a high number of activated macrophages that incorporated erythrocytes and granulocytes, and the diagnosis of HLH was confirmed. Gene examination was performed to identify the primary HLH. Carbamazepine was out of use, dexamethasone and norepinephrine was administrated.

White blood cell (WBC) (40.8 $\times 10^9$/L, normal range 3.5-9.5 $\times 10^9$/L) and procalcitonin (PCT) (¿100 ng/ml, normal range 0-0.05 ng/ml) were significantly elevated. In addition, damage of liver function occurred with a highly increased in transaminases (Table 1). Sonography of the abdomen showed formerly detected hepatosplenomegaly but regular portal vein and hepatic vein flow in Doppler. Multiple blood and urine cultures as well as virus polymerase chain reaction (PCR) for Epstein-Barr virus (EBV) and cytomegalovirus (CMV) were negative. X-ray of the thorax was remarkable and showed no signs of infection. Continuous therapy with meropenem did not improve circulatory dysfunction or laboratory parameters of inflammation.

In view of largely increased interleukin (IL)-6 (1467.6 pg/ml, normal range 0-7 pg/ml) and ferritin (726.9 ng/ml) as well as the diagnosis of acute kidney injury, plasma exchange (PE) and continuous veno-venous hemodiafiltration (CVVHDF) were considered to start. However, during that period, plasma separator was scared and unavailable for us because of coronavirus disease 2019 (COVID-19) epidemic. Therefore we tried to use the hemoadsorption combined with CVVHDF. A hemoadsorption cartridge (HA330-II perfusion column,

Zhuhai Health Sails Biotechnology Co.,Ltd., Zhuhai, China) was integrated into the extracorporeal circuit to promote cytokine adsorption. The hemoadsorption treatment lasted for 3 hours every time and then the HA330-II perfusion column was moved off from the hemodiafiltration circuit. Hemoadsorption was performed once a day and totally for three times. CVVHDF (substitute flow 20 mL/kg.h, dialysate flow 20 mL/kg.h and blood flow 3-5mL/kg.min) lasted for about 72 hours and the filter was changed when it was considered clotted. Anticoagulation was performed with heparin sodium. After 72 hours of treatment, the concentration of IL-6 and ferritin separately fall to 119.25 pg/ml and 295.3 ng/ml (Figure 1). The norepinephrine (NE) (0.1 μg/kg/min) could be wean off as well as erythra alleviated significantly. However, fever and erythra recurrenced 48 hours after the cease of CVVHDF and hemoadsorption. So single hemoadsorption (HA330-II perfusion column, duration of 3 hours) by perfusion machine was performed three times (once a day) again. Subsequently, no further deterioration of the patient's clinical condition was seen. Gene examination exclude the primary HLH. The patient was discharged from PICU on day 10 and at that time his PCT level declined from more than 100 ng/ml to 0.12 ng/ml.

2.2 Case 2

A 10-year-old male was hospitalized due to respiratory system infection of unknown pathogen. The patient's clinical state deteriorated quickly and display respiratory failure as well as capillary leak syndrome and hypotension. So he was admitted to PICU.

Initial laboratory studies revealed excessive hyperferritinemia (1434 ng/mL), fever up to 40.0, low NK cell activity (0.44 %), hypofibrinogenemia(1.12 g/l), hemoglobinopenia (87 g/L) and thrombopenia (68 $\times 10^9$/L) (Table 2). According to these manifestations, the patient was suspected of HLH and subsequent bone marrow biopsy supported the diagnosis. In consideration of high PCT level of 139.3 ng/ml and high IL-6 level of 1549.5 pg/ml, HLH was most likely triggered by acute bacterial infection. Multiple treatments with meropene, norepinephrine, IVIG and dexamethasone were initiated. However, hypotension could not be improved and the patient developed acute renal failure, CVVHDF (substitute flow 20 mL/kg.h, dialysate flow 20 mL/kg.h and blood flow 3-5mL/kg.min) was started. PE was expected to be initiated simultaneously. However, we could not get plasma separator because of COVID-19 epidemic too. So we tried hemoadsorption (HA330-II perfusion column, Zhuhai Health Sails Biotechnology Co.,Ltd., Zhuhai, China) combination with CVVHDF again. The anticoagulation was performed with heaprin sodium. At the meantime, platelets, fibrinogen prothrombin complex concentrate were infused into the patient to improve coagulation function. The hemoadsorption was performed once a day and was continuously done three times. CVVHDF lasted for about 7 days. From the initiation of hemoadsorption combination with CVVHDF, the dosage of NE (0.5 μg/kg/min) reduced to 0.3 μg/kg/min after 24 h of treatment and 0.2 μg/kg/min after 72 h later and was weaned off 6 days later (Figure 2). The patient's IL-6 level decreased to 15.87 pg/mL and PCT decreased to 0.12 ng/ml after 72 h of therapy (Table 2, Figure 2). On day 11, the patient was discharged from PICU.

3 DISCUSSION

The main pathophysiologic feature of HLH is excessive activation and expansion of T lymphocytes (mainly cytotoxic CD8+ T cells) and macrophages. These activated immune cells produce large amounts of pro-inflammatory cytokines, creating hypercytokinemia/hyper- inflammation and subsequent multiple organ failure [5]. Patients with severe HLH present high levels of cytokines such as IL-1, IL-2, IL-6, IL-18, tumor necrosis factor (TNF)-α, interferon (IFN)-γ. Among these cytokines, IL-6 plays a major role in HLH and appear a good target molecule for a cytokine storm. Excessive expression of IL-6 leads to the excessive activation and expansion of CD8+T cells and macrophages. It results in acute severe systemic inflammatory response known as 'cytokine storm' and can activate the coagulation pathway and vascular endothelial[6].

Since HLH can be rapidly fatal without specific intervention, it is recommended that treatment should be started when there is a high clinical suspicion, even when results of diagnostic studies are still pending. Except for EBV-driven HLH, there are no specific treatment guidelines for HLH secondary to infections. Conventionally, in addition to the management of infection, immunoglobulins and methylprednisolone were administered to attenuate inflammation, which could only reduce the production of cytokines rather than

remove any cytokines that had been caused [7].

Several studies have demonstrated the use of blood purification for patients with HLH. DiCarlo et al [8] reported that hemofiltration when properly applied could relieve severe metabolic acidosis and reduce the level of cytokine activity with multiple organ dysfunction syndrome (MODS). High volume hemofiltration (HVHF) was used by Cui Y et al [9] to reduce cytokines levels and restore organ function. Demirkol et al [10] reported that patients with secondary hemophagocytic syndrome can be successfully treated with PE, IVIG, and methylprednisolone. Additionally, hemoadsorption by CytoSorb^{TM} column [11] or endotoxin-binding polymyxin-B- immobilized fiber column[12] was tried for the treatment of HLH in a few researches.

For HLH patients, we used to carry out PE as the adjuvant treatment to control the hypercytokinemia. When the patients complicated with multi-organ dysfunction, CVVHD(F) was combined with PE. PE can efficiently remove big molecule such as pathogenic cytokines and toxic substances [13, 14]. However, PE is often limited due to an inadequate plasma supply [15] and can carry transfusion-related risks. The two cases met the lack of plasma separator. So we tried the new combination to create a unique treatment, combined hemoadsorption therapy by HA330-II perfusion column (Zhuhai Health Sails Biotechnology Co.,Ltd., Zhuhai, China) with CVVHDF. HA330-II perfusion column is a macroporous resin hemoperfusion device which remove cytokines via small polymer beads. Originally, HA330-II perfusion column was reported as one part of DPMAS and successfully used in liver failure patients [16, 17]. It can effectively reduce inflammatory cytokine. Combined CVVHDF with HA330-II perfusion column could not only reduce the total amount of blood needed for PE, but obtained some very good curative effect.

As far as we know, this is the first case series report involving the clinical application of HA 330-II perfusion column in children suffering from HLH. In two cases we reported, high level of IL-6 was decreased to normal after treatment of the hemoadsorption combined with CVVHDF. As inflammatory cytokine level had returned to normal, patients' signs and symptom fade away. In a word, the marked decrease in IL-6 plasma levels, decrease of inflammatory cytokines and stabilization of liver function were observed after treatment, which proved the efficiency of HA330-II perfusion column. Importantly, treatment was safe and well-tolerated, without any adverse events.

In conclusion, hemoadsorption by HA330-II column is safe and ffective in HLH patients. A limitation of this study was that the number of cases was too small. Further studied need to do in the future.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

1. Fraga-Rodriguez GM, Brio-Sanagustin S, Turon-Vinas E, Dixon BP, Carreras-Gonzalez E. Eculizumab in a child with atypical haemolytic uraemic syndrome and haemophagocytic lymphohistiocytosis triggered by cytomegalovirus infection. *BMJ Case Rep* . Apr 26 2017;2017doi:10.1136/bcr-2016-219065

2. George MR. Hemophagocytic lymphohistiocytosis: review of etiologies and management. *J Blood Med* . 2014;5:69-86. doi:10.2147/JBM.S46255

3. Ishikawa Y, Nishizawa H, Kasuya T, et al. Successful treatment of fatal macrophage activation syndrome and haemophagocytic lymphohistiocytosis by combination therapy including continuous haemodiafiltration with a cytokine-adsorbing haemofilter (AN69ST) in a patient with systemic lupus erythematosus %J Modern Rheumatology Case Reports. 2018;2(1)

4. Rademacher JG, Wulf G, Koziolek MJ, Zeisberg M, Wallbach M. Cytokine adsorption therapy in lymphoma-associated hemophagocytic lymphohistiocytosis and allogeneic stem cell transplantation. *J Artif Organs* . Jan 18 2021;doi:10.1007/s10047-020-01244-2

5. Schulert GS, Grom AA. Pathogenesis of macrophage activation syndrome and potential for cytokine-directed therapies. *Annu Rev Med* . 2015;66:145-59. doi:10.1146/annurev-med-061813-012806

6. Tanaka T, Narazaki M, Kishimoto T. Immunotherapeutic implications of IL-6 blockade for cytokine storm %J Immunotherapy. 2016;8(8)

7. Henter JI, Horne A, Arico M, et al. HLH-2004: Diagnostic and therapeutic guidelines for hemophagocytic lymphohistiocytosis.*Pediatr Blood Cancer* . Feb 2007;48(2):124-31. doi:10.1002/pbc.21039

8. DiCarlo J, Lui WYS, Frankel L, Howell W, Schiffman J, Alexander S. THE HEMOPHAGOCYTIC SYNDROME: Titrating Continuous Hemofiltration to the Degree of Lactic Acidosis %J Pediatric Hematology-Oncology. 2006;23(7)

9. Cui Y, Zhang YC, Kang YL, Ren YQ, Miao HJ, Wang F. High-Volume Hemofiltration in Critically Ill Patients With Secondary Hemophagocytic Lymphohistiocytosis/Macrophage Activation Syndrome: A Prospective Study in the PICU. *Pediatr Crit Care Med* . Oct 2016;17(10):e437-e443. doi:10.1097/PCC.0000000000000896

10. Demirkol D, Yildizdas D, Bayrakci B, et al. Hyperferritinemia in the critically ill child with secondary hemophagocytic lymphohistiocytosis/sepsis/multiple organ dysfunction syndrome/macrophage activation syndrome: what is the treatment? %J BioMed Central. 2012;16(2)

11. Greil C, Roether F, La Rosee P, Grimbacher B, Duerschmied D, Warnatz K. Rescue of Cytokine Storm Due to HLH by Hemoadsorption in a CTLA4-Deficient Patient. *J Clin Immunol* . Apr 2017;37(3):273-276. doi:10.1007/s10875-017-0377-7

12. Tatara R, Sato M, Fujiwara S, et al. Hemoperfusion for Hodgkin lymphoma-associated hemophagocytic lymphohistiocytosis. *Intern Med* . 2014;53(20):2365-8. doi:10.2169/internalmedicine.53.2457

13. Gerard C, Violeta O, Thomas G, et al. Therapeutic plasma exchange in children: One center's experience. %J Journal of clinical apheresis. 2017;32(6)

14. Patale D, Bajpai M, Maiwall R, Kumar G. Hemodynamic stability in liver failure patients undergoing therapeutic plasma exchange %J Journal of Clinical Apheresis. 2020;35(2)

15. DNA Viruses - Hepatitis B Virus; Data on Hepatitis B Virus Described by Researchers at Shanxi Dayi Hospital (Therapeutic Effect of Double Plasma Molecular Adsorption System and Sequential Half-dose Plasma Exchange In Patients With Hbv-related Acute-on-chronic Liver Failure) %J Biotech Week. 2020;

16. S Z, N W, J Z, et al. [Plasma exchange combined with double plasma absorption therapy improve the prognosis of acute-on-chronic liver failure]. %J Zhonghua gan zang bing za zhi = Zhonghua ganzangbing zazhi = Chinese journal of hepatology. 2018;26(10)

17. Yang CF, Zhang Z, Zhang XY, Li YM. Artificial liver support system in pediatric acute liver failure due to mushroom poisoning: Case series.*Ann Hepatol* . Nov 19 2020;23:100290. doi:10.1016/j.aohep.2020.100290

Abbreviations Tanle

word	abbreviation
hemophagocytic lymphohistiocytosis	HLH
continuous veno-venous hemodiafiltration	CVVHDF
Continuous Blood Purification	CBP
international normalized ratio	INR
activated partial thromboplastin time	APTT
pediatric intensive care unit	PICU
white blood cell	WBC
procalcitonin	PCT
polymerase chain reaction	PCR
Epstein-Barr virus	EBV
cytomegalovirus	CMV

word	abbreviation
interleukin-6	IL-6
plasma exchange	PE
coronavirus disease 2019	COVID-19
norepinephrine	NE
intravenous immunoglobulin	IVIG
tumor necrosis factor -α	TNF -α
interferon -γ	IFN -γ
multiple organ dysfunction syndrome	MODS
high volume hemofiltration	HVHF

Hosted file

TABLE 1.pdf available at https://authorea.com/users/404064/articles/515371-efficacy-of-hemoadsorption-using-ha330-ii-perfusion-column-in-secondary-hlh-patients-two-cases-report-and-literature-review

Hosted file

TABLE 2.pdf available at https://authorea.com/users/404064/articles/515371-efficacy-of-hemoadsorption-using-ha330-ii-perfusion-column-in-secondary-hlh-patients-two-cases-report-and-literature-review

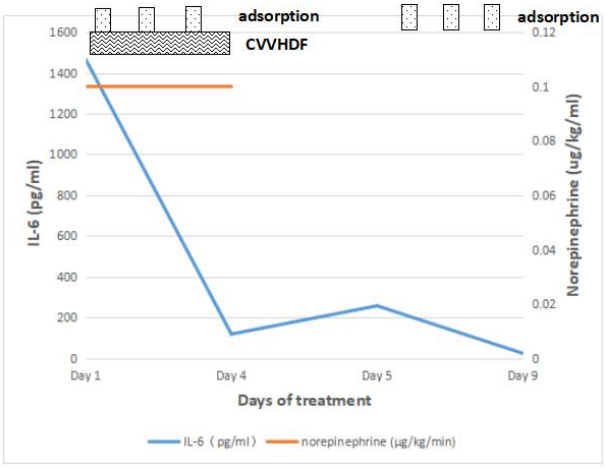

Figure 1. Case 1: interleukin-6 levels and dosage of norepinephrine under treatment. Day 1: Before first hemoadsorption combined with CVVHDF. Day 4: after 72 h of first hemoadsorption combined with CVVHDF. Day 5: Before second hemoadsorption. Day 9: After 72 h of second hemoadsorption.

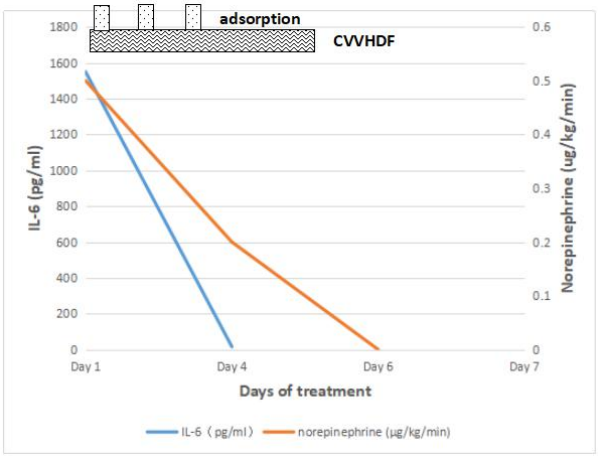

Figure 2. Case 2: interleukin-6 levels and dosage of norepinephrine under treatment. Day 1: Before first hemoadsorption combined with CVVHDF. Day 4: after 72 h of first hemoadsorption combined with CVVHDF.