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CASE REPORT

Acute Hemolytic Transfusion Reaction Due to the "Anti-E" Rhesus Antibody in a Patient with Crohn's Disease

Ari Ahn [1] and Sang-Hyun Hwang [2]

[1] *Department of Laboratory Medicine, College of Medicine, The Catholic University of Korea, Seoul, Korea*
[2] *Department of Laboratory Medicine, University of Ulsan College of Medicine and Asan Medical Center, Seoul, Korea*

SUMMARY

Background: **The rhesus (Rh) system is the second most important blood group system after ABO, with highly immunogenic antigens. Although the anti-E Rh antibody has been reported to cause hemolytic disease of the newborn and delayed hemolytic transfusion reactions, acute hemolytic transfusion reactions (AHTR) have been rarely reported.**
Methods: **Peripheral blood (PB) samples were screened for irregular antibodies using a commercial ID-Diacell I - II antibody screening Panel (Bio-Rad Laboratories, Glattbrugg, Switzerland) and ID-cards "LISS/Coombs" (Bio-Rad, Switzerland). The antibody was confirmed using ID DiaPanel, an antibody identification panel (Bio-Rad, Switzerland). Rh phenotyping was performed for RhC/c and RhE/e antigens using an immediate-spin tube test with monoclonal anti-C, -c, -E, and -e (OrthoClinical Diagnostics, High Wycombe, UK) in saline-filled test-tubes.**
Results: **The patient was negative for antibody screening test before transfusion. After receiving a total of 6 units of cross-matching negative RBC transfusion, the antibody screening test result increased to 2+ after showing traces and the antibody was confirmed as anti-E Rh antibody. The Rh phenotype of the patient was C (+), c (+), E (-), and e (+). In addition, we verified that all the six units of RBCs transfused were E (+) except for the two units transfused before surgery.**
Conclusions: **Here is an unusual case of an AHTR due to the anti-E Rh antibody after E-positive RBC transfusion in a patient with Crohn's disease. Because anemia is common in patients with Crohn's disease, it is important to determine the cause of the anemia and necessary to examine the Rh phenotype before transfusions because of the high need for transfusion due to any cause. Awareness of this possibility will ensure safe blood transfusion with special care to screen for antibodies and perform Rh phenotyping, thereby minimizing morbidity and preventing potential mortality.**
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Correspondence:
Sang-Hyun Hwang, MD, PhD
Department of Laboratory Medicine
University of Ulsan College of
Medicine and Asan Medical Center
88 Olympic-ro 43-gil
Songpa-gu, Seoul, 05505
Korea
Phone: +82 2-3010-4502
Fax: +82 2-478-0884
Email: mindcatch@amc.seoul.kr

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KEY WORDS

acute hemolytic transfusion reaction, anti-E, Rh antibody, Crohn's disease, adalimumab

CASE PRESENTATION

On July 27, 2017, a 40-year-old male with a 17-year history of Crohn's disease was admitted to the hospital because of a one-month history of intermittent, diffuse, and crampy abdominal pain accompanied by progressive early satiety, anorexia, and weight loss of approxi-

mately 2.0 kg. He had a surgical history of segmentectomy of the ileus in 2000 and 2012 due to Crohn's disease with small bowel stricture. He reported that he had been receiving biweekly adalimumab (D2E7, Humira; Abbott Laboratories, Maidenhead, UK) injections for the previous five years as treatment for his Crohn's disease. In addition, the last prescription he received, in April 2016, was a daily dose of 50 mg azathioprine. Physical examination demonstrated fullness in the epigastric region. Complete blood count (CBC) revealed a hemoglobin level of 10.2 g/dL (mean corpuscular volume, 71.3 fL; mean corpuscular hemoglobin concentration, 27.5 g/dL), a hematocrit of 37.1%, white blood cell count of 5,000/μL, and platelet count of 333,000/μL. Differential counts for white blood cells identified 54.5% neutrophils, 35.9% lymphocytes, 7.2% monocytes, 2.2% eosinophils, and 0.2% basophils. Basic metabolic panels showed 142 mmol/L sodium, 3.9 mmol/L potassium, 102 mmol/L chloride, 7 mg/dL BUN, 0.66 mg/dL creatinine, and 0.8 mg/dL total bilirubin. Aspartate aminotransferase level was 16 IU/L and alanine aminotransferase level was 9 IU/L. The erythrocyte sedimentation rate and C-reactive protein level were in the normal ranges.

Six days after admission, a blood sample was screened for irregular antibodies per our standard protocol using a commercial ID-Diacell I - II antibody screening Panel (Bio-Rad Laboratories, Switzerland) and ID-cards "LISS/Coombs" (Bio-Rad, Switzerland). The test result was negative. The direct antiglobulin test (DAT) was performed using "LISS/Coombs" (Bio-Rad, Switzerland) and this result was negative as well.

Computed tomography (CT) revealed stricture in the ileal anastomosis and another short-segmental stricture proximally with dilatation of the intervening segments. The patient was started on prophylactic antibiotics including 200 mg ciprofloxacin twice a day, 500 mg metronidazole once daily, and 2 g cefoxitin once a day for surgery. Ciprofloxacin and metronidazole were maintained for nine and four days, respectively. No additional doses of adalimumab were given after the last administration on July 25, 2017 and no other medication history was reported.

Seven days after admission, the patient underwent an ileocolic resection of 175 cm to 190 cm below the ligament of Treitz for stenoplasty of the anastomotic site strictures.

On postoperative day (POD) 1, the patient complained of thirst and developed hypotension and tachycardia with a blood pressure (BP) of 93/68 mmHg and a pulse rate of 128/min. CBC and arterial blood gas analysis (ABGA) were performed to identify the cause of hypotension and tachycardia. Low hemoglobin (8.6 g/dL) was observed and ABGA was normal. The patient was administered 500 mL of normal saline, but the patient continued to have hypotension and tachycardia and received an additional 1,000 mL of normal saline. Subsequently, the patient suddenly lost consciousness after changing position; his BP was not measured. Approximately one minute later, the patient returned to consciousness and complained of dizziness. His BP was measured and found to be 60/40 mmHg. Follow-up CBC revealed very severe anemia (6.4 g/dL) and postoperative CT demonstrated a large acute hematoma from arterial bleeding near the ileocolic anastomosis. A blood bank test was performed for blood transfusion and the antibody screening test was negative. Depending on the patient's blood type, two units of cross-matching negative O+ RBC were delivered. The patient's dizziness improved, and his vital signs became stable.

Laparotomy was then performed, the hematoma was removed, and hemostasis and arterial bleeder ligation of the anastomosis and colonic mesentery were achieved. After an additional four units of RBC transfusion (six units in total) on POD 0, laboratory investigation findings were as follows; hemoglobin 9.7 g/dL, hematocrit 29.7%, platelet count 99,000/μL, total bilirubin 6.7 mg/dL, direct bilirubin 2.1 mg/dL, and lactate dehydrogenase (LD) 220 IU/L. PT was 17.4 second and fibrinogen, D-dimer, and fibrinogen degradation production level were 218 mg/dL, 1.47 μg/mL FEU, and 27.9 μg/mL, respectively. A peripheral blood smear showed mild anisocytosis with normocytic normochromic red blood cells but no significant spherocytes or shistocytes. The clinical and hematological features favored a diagnosis of hemolytic anemia and disseminated intravascular coagulation, although the reticulocyte production index was 0.55% and an antibody screening test and DAT were negative. However, the patient had no acute hemolytic transfusion reaction (AHTR)-related symptoms such as fever, chills, hypotension, decreased urine output, or pain at the injection site.

On POD 3, the antibody screening test result increased to 2+ after showing traces and the antibody was confirmed as anti-E rhesus (Rh) antibody using ID DiaPanel, an antibody identification panel (Bio-Rad, Switzerland). The Rh phenotype of the patient was C (+), c (+), E (-), and e (+). In addition, we verified that all the six units of RBCs transfused were E (+) except for the two units transfused before surgery. There was a decrease in haptoglobin (< 7.8 mg/dL) and an increase in plasma hemoglobin (15.1 mg/ dL), with a total bilirubin of 7.2 mg/dL and LD of 341 IU/L. However, DAT showed only one trace immediately after transfusion and continued to be negative afterwards.

Hematuria and albuminuria were observed in urinalysis at POD 8; occult blood showed a result of 4+, bilirubin showed a result of 2+, and albumin showed a result of 1+. Following a further decrease in the hemoglobin level to 6.7 g/dL and increase in the total bilirubin level to 6.1 mg/dL with the potential possibility of non-immune hemolytic anemia caused by ciprofloxacin, the Ciprofloxacin was discontinued and an additional two units of E-negative RBC were transfused due to the anti-E antibodies. Subsequently, the anemia improved, the bilirubin decreased, and the patient was discharged without any complications.

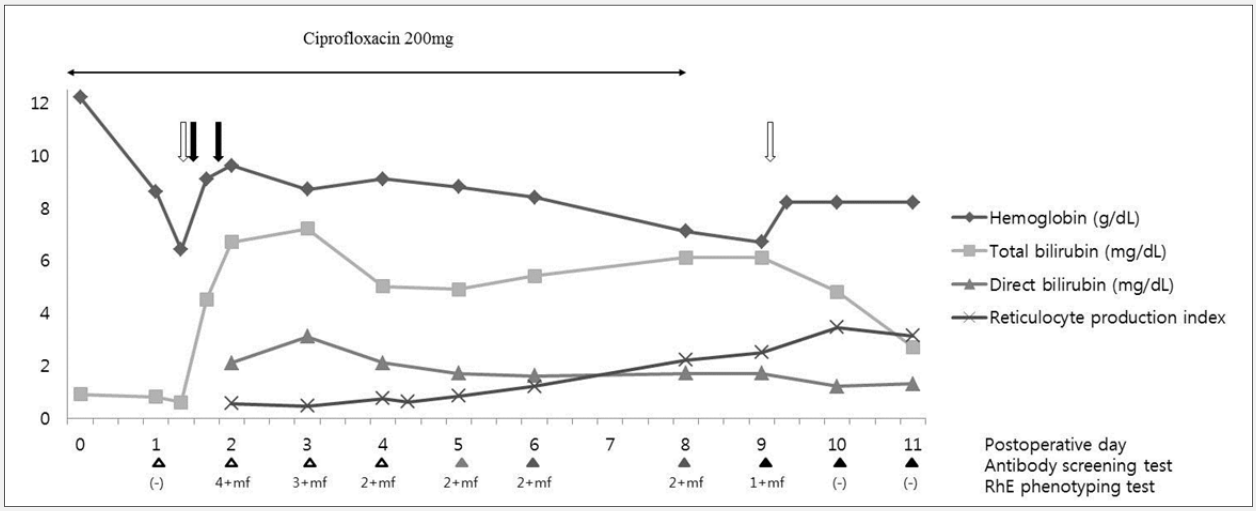

Figure 1. Antibiotic treatments, laboratory and transfusion-related medical findings in the case.

Solid triangles, positive for antibody screening test; open triangles, negative for antibody screening test.
White arrows, E-negative RBC transfusions, two units; black arrow, E-positive RBC transfusions, two units.

DISCUSSION

The Rh system is the second most important blood group system after ABO, with highly immunogenic antigens. Rh system-induced hemolytic transfusion reactions are immunoglobulin G (IgG)-mediated and usually result in extravascular hemolysis and delayed hemolytic transfusion reactions (DHTR). The most commonly reported non-ABO AHTR include those due to Kidd, Diego, and P antigens [1]. Although the anti-E Rh antibody has been reported to cause hemolytic disease of the newborn, and DHTR and AHTRs have been rarely reported [2]. Further, the possibility of Rh antibodies being the cause of intravascular hemolysis is still under debate [3].

The present report describes an unusual case of AHTR due to the anti-E antibodies causing intravascular hemolysis. Although the patient did not show AHTR symptoms such as fever, hypotension, and hematuria, bilirubin level was markedly increased within 24 hours after transfusion and DAT, especially IgG, was trace positive, and hemoglobin did not rise as expected.

Other Rh antigens other than RhD are known to be of lesser clinical significance and blood without an antigen should be transfused only when the antibodies have already been sensitized to the antigen [4]. Most of the antibodies against Rh antigen are known to cause delayed hemolytic transfusion reactions and extravascular hemolysis [4]. Very few cases of Rh antibodies causing intravascular hemolysis and acute presentation have been reported; two cases involving anti-c [1,3], one involving anti-C [5], and one involving anti-E [2].

Drug-induced hemolytic anemia (DIHA) could also be considered in the present case. Adalimumab is a recombinant, fully human IgG1 monoclonal antibody specific to TNF-α and is efficacious in severe and refractory CD [6], although there remain significant therapy- and disease-related risks of serious complications [7]. There have been several reports that autoimmune hemolytic anemia could be caused due to long-term administration of adalimumab [8]. However, pre-operative laboratory findings such as DAT-negative and antibody screening-negative results did not support adalimumab-induced hemolytic anemia.

In addition, ciprofloxacin was administered in this patient. Hemolytic anemia is a rare adverse drug reaction to ciprofloxacin and several case reports have reported an association between ciprofloxacin and immune hemolytic anemia [9].

The common presentation of DIHA is associated with hyperbilirubinemia and raised ferritin levels, occurring 3 - 12 days after administration of the drugs [9]. Some of the patients with ciprofloxacin-induced hemolytic anemia have glucose-6-phosphate dehydrogenase (G6PD) deficiency [10]. The mechanism of DIHA appears to involve oxidant damage, and the side effects are thus more common in patients with G6PD deficiency [10]. In particular, the presence of bile cells in the peripheral blood smear should raise the possibility of a drug reaction. The DAT in our case was positive only once for

IgG after transfusion of four units of E-positive RBC and subsequently remained negative, suggesting that the etiology was unlikely to be ciprofloxacin-induced-anemia. G6PD deficiency could not be confirmed because he was discharged without G6PD testing.

CONCLUSION

We described an unusual case of AHTR due to the anti-E Rh antibody. The patient was successfully treated with discontinuation of ciprofloxacin and transfusion of E-negative RBCs, and he experienced good outcomes. Because anemia is common in patients with Crohn's disease, it is important to determine the cause of the anemia and necessary to examine the Rh phenotype before transfusions because of the high need for transfusion due to any cause. The case also emphasizes the critical role of blood banks in early diagnosis and treatment of AHTR, especially due to antibodies in individuals with multiple transfusions. Awareness of this possibility will ensure safe blood transfusion with special care to screen for antibodies and perform Rh phenotyping, thereby minimizing morbidity and preventing potential mortality.

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Ari Ahn and Sang-Hyun Hwang drafted and revised the manuscript.

Declaration of Interest:
The authors have no competing interests.

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