

# A case of acute hemolytic transfusion reaction due to anti-Di<sup>a</sup> antibody

## -A case report-

Sung Ha Mun, Seung Hwan Lee, and Min Young No

Department of Anesthesiology and Pain Medicine, Kangbuk Samsung Hospital, Sungkyunkwan University School of Medicine, Seoul, Korea

Many medical institutions in Korea have recently been performing an antibody screening test as one of the essential elements of a pre-transfusion test. The Di<sup>a</sup> antigen is well known as one of the antigens with low incidence among Caucasians; however, it has been discovered with a relatively higher incidence among Mongoloid populations. The frequency of the Di<sup>a</sup> antigen among the Korean population is estimated to be 6.4–14.5%. But in Korea, a screening panel of cells from abroad without Di<sup>a</sup> positive cells has been commonly used when a patient has an unexpected antibody screening test. Here we report a case of acute hemolytic transfusion reaction due to Anti-Di<sup>a</sup> antibody. To prevent other transfusion reaction by anti-Di<sup>a</sup> antibody, addition of Di<sup>a</sup> positive cells as unexpected antibody screening test is recommended. (Korean J Anesthesiol 2012; 63: 353-356)

**Key Words:** Anti-Di<sup>a</sup> antibody, Antibody screening test, Hemolytic transfusion reaction.

Pre-transfusion test consists of ABO blood grouping, RhD genotyping test, antibody screening test and cross-matching. Antibody screening test, among these, is to detect unexpected antibody present in the patient's blood to prevent hemolytic transfusion reaction by alloantibody. Unexpected antibody, differently from expected antibodies such as anti-A or anti-B from ABO blood groups, is a case where the existence of antibody to a specific antigen in the serum is not detectable until testing, so it is also called irregular antibody. Most blood-group antibodies including ABO other than some of P blood

group belong to unexpected antibody. Although unexpected antibody possibly occurs naturally, most of important unexpected antibodies are immune antibody produced when exposed to different antigens through pregnancy, transfusion, etc. In Korea, the frequency of unexpected antibody has been reported to range from 0.26% to 1.11%, depending on subjects and test methods [1].

Antibody screening panels for an unexpected antibody screening test currently used in Korea are mostly imported abroad, in which it is impossible to detect anti-Di<sup>a</sup> antibody

Received: October 5, 2011. Revised: November 21, 2011. Accepted: December 2, 2011.

Corresponding author: Sung Ha Mun, M.D., Department of Anesthesiology and Pain Medicine, Kangbuk Samsung Hospital, Sungkyunkwan University School of Medicine, 108, Pyoung-dong, Jongno-gu, Seoul 110-746, Korea. Tel: 82-2-2001-2001, Fax: 82-2-2001-2326, E-mail: [anesmun@hanmail.net](mailto:anesmun@hanmail.net)

© This is an open-access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (<http://creativecommons.org/licenses/by-nc/3.0/>), which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

because these panels for screening test usually does not contain  $D_i^a$  antigens that are seldom or in low frequency found in Caucasians [2-6]. However, Koreans show relatively high frequency of  $D_i^a$  antigen and anti- $D_i^a$  antibody, so there is high risk of transfusion reaction.

We report a current episode of intraoperative acute hemolytic transfusion reaction due to anti- $D_i^a$  antibody not detected by preoperative antibody screening test.

## Case Report

A 75-year-old woman (height 150 cm, weight 63.5 kg) was admitted to the hospital for phased total knee replacement on the both sides under the diagnosis of bilateral degenerative arthritis. She had no other specific diseases than osteoporosis. She had a history of a right shoulder joint replacement and a lumbar discectomy. Blood test, EKG, chest x-ray, and pulmonary function test performed prior to surgery were within normal ranges, and echocardiography showed ejection fraction of 60%, and manifested diastolic dysfunction of the left atrium. The patient's blood type was O Rh-positive and the preoperative cross matching and antibody screening test turned out to be negative. Under general anesthesia, right total knee replacement arthroplasty was first performed and after surgery she received 2 units of packed red blood cells (RBCs). After transfusion, there was no episode of transfusion reaction.

Two weeks after surgery, preoperational test for left total knee replacement indicated that hemoglobin level was 10.2 g/dl and hematocrit was 29.2%, and the antibody screening test was negative. On surgery day, she received 0.2 mg of glycopyrrolate intramuscularly one hour before induction of anesthesia. In the operating room, EKG, noninvasive blood pressure monitoring and a pulse oximetry monitor were placed. Anesthesia was induced with propofol 120 mg and rocuronium 50 mg, and maintained using  $O_2$  2 L/min and  $N_2O$  2 L/min, and sevoflurane 2 vol% after tracheal intubation. For continuous monitoring of blood pressure and arterial blood gas analysis, 20 G catheter was placed to the left radial artery and central venous catheter was inserted to the internal jugular vein for central venous pressure (CVP).

One hour and 20 minutes after the onset of surgery when the total amount of blood loss was estimated more than 800 ml, transfusion of packed RBCs was started. A cross-matching test turned out to be negative again, suitable for transfusion. During transfusion, the patient showed stable vital signs. Five minutes after 1 unit of packed RBCs was transfused, red-colored urine, which was suspected as hemoglobinuria, was observed. For immediate treatment, fluid was administered while 10 mg furosemide was injected. Then we asked the Department of Laboratory Medicine to conduct ABO-Rh blood typing test of

the patient's and the donor's blood, cross-matching, antibody screening test, direct anti-globulin test (DAT) and indirect anti-globulin test (IAT), and urinalysis. Hereafter, no big blood loss developed and the surgery was completed without further transfusion. Except for the incidence of red-colored urine sustained during surgery, patient's vital signs were stably maintained. At the completion of surgery when her spontaneous respiration and consciousness were confirmed, she was extubated and transported to the recovery room.

Red-colored urine sustained even in the recovery room, but the color was waning and the patient's vital signs were normal, so she was referred to a ward. Post-operative test results showed that the transfused blood and patient's blood were compatible and that antibody screening test result was negative. IAT was negative while DAT was positive. Cross-matching turned out to be negative again. Urinalysis findings indicated 3+ blood, 0 RBC/HPF, and 2+ protein. Examination of patient's blood tested by an independent laboratory to determine more exact cause revealed that anti- $D_i^a$  antibody was identified in an unexpected antibody screening test.

Since the next day of surgery, red-colored urine was not observed, which made additional urinalysis unnecessary. Hemoglobin level measured right after surgery was 8.7 g/dl, showing a decrease, but thereafter no significant change further took place and BUN and creatinine level also sustained in the normal range, not followed by any other transfusion reaction. Then, she was discharged without any complications.

## Discussion

Potential adverse effects from blood transfusion involve contamination, acute immunologic hemolytic reaction, delayed extravascular hemolysis, febrile allergic transfusion reaction, erythrocytolysis, etc. Among these reactions, acute immunologic hemolytic reaction is mainly developed by incompatible blood transfusion of ABO blood group or by unexpected antibody present in the blood.

For the patient in this case, cross-matching tests conducted before transfusion and immediately after red-colored urine developed were both negative. The main purposes of cross matching are: 1) to prevent acute hemolytic transfusion reaction due to ABO blood group incompatibility by detecting patient's anti-A and anti-B antibodies against RBCs to be transfused; 2) to prevent acute hemolytic transfusion reaction developed by the response of recipient's unexpected antibody to the counterpart antigen of donor's blood. What is definitely necessary for the 2nd purpose is the indirect anti-globulin method including an antiglobulin phase. When detecting IgM that reacts even at room temperature such as anti-A or anti-B antibody in 1), it is possible to identify agglutination reaction by centrifugal

sedimentation following after reaction of RBCs and serum at room temperature, which is called "immediate spin phase" or "saline phase". The main purpose of the immediate spin phase is to prevent ABO blood group incompatibility. When the absence of unexpected antibody in recipient's specimen is confirmed (by negative antibody screening result) or when an emergency situation makes it impossible to perform a test for incompatibility by unexpected antibody (in cross-matching test of anti-globulin method, it takes at least 15 to 30 minutes for the reaction time of serum), it is a routine for RBCs to be taken out only after immediate spin phase [7]. In the present case, unexpected antibody screening test performed at our hospital was negative while the patient had no abnormal medical history. As bleeding during surgery is regarded as emergency transfusion, cross-matching test was performed only with immediate spin phase and the result turned out to be negative (compatible) when transfusion was performed.

For the causes of acute hemolytic reaction developed during surgery in the present case, while the results of the tests performed by an independent blood center were unknown, the following three scenarios were to be considered:

1) Drug-induced hemolysis: this assumption is supported by the fact that IAT was positive while antibody screening test was negative. Even though there is no abnormality in previous transfusions, such drugs as acetaminophen, antihistamine, cephalosporin, etc. can induce hemolytic reactions or positive results of DAT by forming immune complex in combining with the red cell membrane of blood product [8].

2) Hemolysis induced by unexpected antibody of low potency: In presence of low-potency antibody, it is possible that although antibody screening test is negative, hemolytic reaction can develop when antibody is activated by transfusion. Even if a test performed is still negative after exhibition of hemolytic reaction, such possibility cannot be completely excluded. It is because there are some cases in which the manifestation of unexpected antibodies such as 'kidd' can be identified late or clinical manifestation can be presented at even low potency when the antibody screening test is negative [2,9].

3) Hemolysis induced by anti-Di<sup>a</sup> antibody: among the antigens of Diego blood group, Di<sup>a</sup> and Di<sup>b</sup> are the most significant ones. In the frequency of Diego phenotypes, Di(a-b+) is found 90% of Korean population, followed by Di(a+b+), 9.75%, and Di(a+b-), 0.25% [10]. Di<sup>b</sup> is classified as a high-frequency antigen regardless of ethnicity while the prevalence of Di<sup>a</sup> differs among races. The Di<sup>a</sup> antigen is rare in Caucasians, but occurs with high incidence in Asians and American Indians. For Koreans, it is reported that 6.4–14.5% of the population tests positive [2]. Anti-Di<sup>a</sup> antibody has relative high positive reaction rate in Koreans [3-6]. It has been reported that anti-Di<sup>a</sup> antibody was detected in a patient who was given transfusion

repeatedly in Korea [11,12], besides incidences of delayed hemolytic transfusion reaction due to anti-Di<sup>a</sup> antibody [13]. Because antibody screening panels for an unexpected antibody screening test currently used in Korea are mostly imported abroad, there is a high probability that the Di<sup>a</sup> antigen is not contained. Even though anti-Di<sup>a</sup> antibody is present in patient's serum, it is not detected in unexpected antibody screening test, which keeps transfusion reaction from being prevented.

On the basis of the lab results from the independent blood center that the patient was negative for Di<sup>a</sup> antigen and positive for anti-Di<sup>a</sup> antibody, we made a diagnosis of acute hemolytic reaction due to anti-Di<sup>a</sup> antibody. Considering that she had a history of a surgery previously performed at another hospital when she may have received transfusions and that she was given a transfusion two weeks ago, it is highly likely that anti-Di<sup>a</sup> antibody was produced by transfusion. Since she had no specific adverse effects of transfusion performed two weeks ago, we assume that she would be first exposed to Di<sup>a</sup> antigen at the time of transfusion or that there was no side effect because there would be no anti-Di<sup>a</sup> antigen in the donor's blood even if she had already have anti-Di<sup>a</sup> antibody. If our hospital had performed an unexpected antibody screening test with antibody screening panels containing Di<sup>a</sup> antigen, it would have been possible to detect anti-Di<sup>a</sup> antibody and to prevent transfusion reaction by preparing blood free of Di<sup>a</sup> antigen at transfusion.

On the contrary to the reports that anti-Di<sup>a</sup> antibody is an immune antibody, generally causing delayed hemolytic transfusion reaction, the present case experienced acute hemolytic transfusion reaction. Fortunately, the patient was recovered without much adverse effect because the amounts of blood loss and transfusion were not large. If acute hemolytic transfusion reactions due to anti-Di<sup>a</sup> antibody develop when emergency transfusion or massive transfusion is required, however, it will take a long time to diagnose and prepare for blood. This will greatly damage the patient's treatment procedure.

Therefore, considering that Koreans have a high incidence rate of positive reactions for Di<sup>a</sup> antigen and anti-Di<sup>a</sup> antibody, it is necessary that unexpected antibody screening test before transfusion should include a test to identify anti-Di<sup>a</sup> antibody.

## References

1. Oh DJ, Kim MJ, Seo DH, Song EY, Han KS, Kim HO. The frequency of unexpected antibodies in blood donors and transfusion candidates in Korea. *Korean J Blood Transfus* 2003; 14: 160-72.
2. Han KS, Park MH, Cho HI. *Transfusion medicine*. 3rd ed. Seoul, Korea Medical Publishing Co. 2006, pp 228-31.
3. Chae SL, Cho HI, Kim SI. A study on the frequencies of U, Diegoa, and kell blood group antigens and anti-Dia and anti-K in Koreans. *Korean J Hematol* 1988; 23: 183-8.

4. Park TS, Oh SH, Choi JC, Lee DD, Kim HH, Chang CL, et al. The clinical significance of antibody screening test including Di(a+) panel cell in Asian-Mongoloid populations. *J Korean Med Sci* 2003; 18: 669-72.
5. Sung NH, Jeon TY, Lee EY, Chung JS, Cho GJ, Kim HH. Prevalence and specificity of unexpected antibody using antibody screening test including Dia and Mia cells. *Korean J Lab Med* 2005; 25: 340-6.
6. Lee SM, Lim YA, Oh JS. An experience of including ID DiaCell Dia cell in unexpected antibody screening test. *Korean J Blood Transfus* 2005; 16: 32-7.
7. Cooling L, Downs T. Immunohematology. In: Henry's clinical diagnosis and management by laboratory methods. 22nd ed. Edited by McPherson RA, Pincus MR: Philadelphia, Elsevier Saunders. 2011, pp 721-2.
8. Cooling L, Downs T. Immunohematology. In: Henry's clinical diagnosis and management by laboratory methods. 22nd ed. Edited by McPherson RA, Pincus MR: Philadelphia, Elsevier Saunders. 2011, pp 729-30.
9. Cha YJ. Hematology. Seoul, Epublic. 2006, pp 574-7.
10. Choi SY, Kim SI, Cho HI. Study on genefrequencies of blood groups in Koreans. *Korean J Hematol* 1984; 19: 63-75.
11. Kim HO, Song KS, Lee SY. A case of anti-Diegoa. *Korean J Clin Pathol* 1987; 7: 357-61.
12. Kim HK, Cho YJ, Kim YK, Oh YC, Choi BR, Cho MJ. A case of anti-Dia antibody. *Korean J Hematol* 1989; 24: 155-9.
13. Chung HR, Lee JW, Kim HO, Lee SY, Kim HJ. A case of delayed hemolytic transfusion reaction in a patient with anti-Diegoa and anti-Lewisa. *Korean J Clin Pathol* 1989; 9: 557-62.