Hemolysis Due to the Simultaneous Occurrence of Passenger Lymphocyte Syndrome and a Delayed Hemolytic Transfusion Reaction in a Liver Transplant Patient

Lisa B. Jacobs, MD; R. Sue Shirey, MS, MT(ASCP)SBB; Paul M. Ness, MD

• **Hemolysis due to donor-derived antibodies produced by "passenger" B lymphocytes, called passenger lymphocyte syndrome, has been described in ABO-unmatched solid organ and bone marrow transplant recipients. Delayed hemolytic transfusion reactions occur within a similar time frame and have similar clinical and serologic findings. To our knowledge, we report the first case of hemolysis due to the simultaneous occurrence of passenger lymphocyte syndrome (donor-derived anti-A) and a delayed hemolytic transfusion reaction (recipient-produced anti-E) in a liver transplant patient.**

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Alloantibodies of anti-A or anti-B specificity directed against recipient red blood cells have been detected in the posttransplantation eluates and sera of patients receiving ABO-unmatched (defined as a group O organ transplanted to a non–group O recipient or a group A or B organ transplanted to a group AB recipient) solid organ and bone marrow transplants.[1–13] In some cases, these alloantibodies have been shown by Gm allotyping to be derived from "passenger" lymphocytes present in the transplanted organ.[12,14–16] Hemolysis due to donor-derived antibodies gives rise to a syndrome characterized clinically by hemoglobinemia, hyperbilirubinemia, reticulocytosis, and an excessive red cell transfusion requirement occurring 1 to 3 weeks after the transplant. These clinical findings are accompanied by the laboratory findings of a positive direct antiglobulin test and the detection of unexpected antibodies in the patient's red cell eluate and serum. Both the hemolytic anemia and serology resolve over the course of weeks to months.

Delayed hemolytic transfusion reactions (DHTR) may also be diagnosed based on clinical evidence of hemolysis, a positive direct antiglobulin test, and the detection of alloantibodies.[17,18] Delayed hemolytic transfusion reactions are usually due to an anamnestic or secondary response to an antigenic stimulus, and alloantibodies, as well as clinical signs of hemolysis, are generally detected 3 to 14 days following transfusion.[17,18] Anecdotal cases of DHTR following liver transplantation have been reported.[17,19]

To our knowledge we report the first case of the simultaneous occurrence of passenger lymphocyte syndrome and a delayed hemolytic transfusion reaction in a liver transplant recipient.

MATERIALS AND METHODS

All serologic tests were performed in accordance with the procedures described in the *American Association of Blood Banks Technical Manual.*[20] Acid eluates (Gamma Biologicals, Inc, Houston, Tex) were prepared from the patient's red cells and tested with selected cells by standard methods or with the addition of polyethylene glycol. Serum tests were performed using low ionic strength solution and/or polyethylene glycol methods.

REPORT OF A CASE

The patient was a 54-year-old, group A, Rh-positive, E-negative, white man with alcoholic cirrhosis and a history of multiple red blood cell transfusions. The patient received a liver transplant from a group O male donor who had never been transfused and had a negative antibody screening test.

Prior to transplantation, the patient had a negative antibody screening test, a hematocrit of 33%, total bilirubin of 3.2 mg/dL (55 μmol/L), and reticulocyte count of 2.0%. Perioperatively, the patient was transfused 23 units of group A red cells, 34 units of group A fresh frozen plasma, and 2 plateletpheresis units (one group A and one group AB).

Sixteen days after receiving the liver transplant, the patient developed an increased red cell transfusion requirement (6 units over 7 days), hemoglobinemia (7 g/dL or 700 g/L), and hyperbilirubinemia (6.4 mg/dL or 109 μmol/L). There was no evidence of bleeding.

Direct antiglobulin tests were positive with both anti-IgG (2+) and anti-C3 (1+) on day 16 posttransplant. Anti-A and anti-E were identified in the patient's eluate and serum. The patient was subsequently transfused with group O, E-negative red cells. Three months later, the patient's hemolytic anemia was completely resolved; direct antiglobulin tests and antibody screening tests were negative.

COMMENT

Hemolysis of autologous red cells due to blood group antibodies derived from donor passenger lymphocytes (ie, passenger lymphocyte syndrome) occurs frequently in recipients of ABO-unmatched liver transplants. In one study, 56% of patients at risk had detectable donor-de-

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From the Department of Pathology (Dr Jacobs) and the Transfusion Medicine Division (Ms Shirey and Dr Ness), The Johns Hopkins Hospital, Baltimore, Md.

Reprint requests to The Johns Hopkins Hospital, 600 N Wolfe St, Baltimore, MD 21287-6667 (Ms Shirey).

rived antibodies and clinical evidence of hemolysis occurring approximately 7 to 21 days posttransplantation.[13] The severity of hemolysis varies; the majority of cases present with unexplained decreases in hemoglobin and hematocrit and increased blood transfusion requirements.[13] However, substantial morbidity associated with immune hemolysis, including acute renal failure, disseminated intravascular hemolysis, hypotension, multiorgan failure, and death, have been reported in this setting.[1,7,13,21,22]

The clinical and serologic features, as well as the time course of passenger lymphocyte syndrome, are quite similar to the manifestations of DHTRs. The etiologies of immune hemolysis differ in that passenger lymphocyte syndrome is caused by antibodies produced by the organ donor lymphocytes, whereas DHTR results from antibody production by the recipient's lymphocytes in an anamnestic response to an antigenic stimulus via red cell transfusions. Delayed hemolytic transfusion reactions with significant clinical sequelae following liver transplantation have been documented.[17,19] Hyma et al[19] described DHTR due to anti-JK^a, -c, and -S, in a liver transplant recipient, and Ness et al[17] included a patient with DHTR due to anti-JK^a and anti-E following liver transplantation in a series they studied. These cases indicate that liver transplant recipients are capable of mounting an anamnestic immune response to blood group antigens despite immunosuppressive therapy.

The differential diagnosis of passenger lymphocyte syndrome and delayed hemolytic transfusion reactions is usually based on the antibody specificities identified and the medical and transfusion histories of the organ recipient and donor. In some instances, immunoglobulin allotyping (Gm allotyping) has been useful in confirming the origin of the antibodies.[14-16]

In the present case, the patient presented with hemolytic anemia and the serologic findings of a positive direct antiglobulin test 16 days following transplantation of an ABO-unmatched liver. The serologic findings were consistent with hemolysis of autologous red cells due to the presence of donor-derived anti-A (ie, passenger lymphocyte syndrome). However, in addition to donor derived anti-A, anti-E was identified in the patient's eluate and serum studies. A diagnosis of DHTR due to anti-E derived from the recipient's lymphocytes was made based on the following evidence: (1) The liver donor was a man with negative antibody screening test results who had no transfusion history prior to his terminal admission. Therefore, the donor was not immunized to red cell antigens either through transfusion or pregnancy. (2) The transplant recipient was E-negative and had a history of multiple red cell transfusions prior to organ transplantation. Consequently, anti-E, one of the most commonly implicated antibody specificities in DHTR,[17] was most likely produced by the patient's own lymphocytes in an anamnestic response to red cell products transfused during his transplant surgery. The remote possibility that either anti-A or anti-E was passively acquired through transfusion of blood products was excluded because all the transfused donor products were ABO-compatible and negative for unexpected antibodies.

Unfortunately, further confirmation of the diagnosis in this case was not possible; the donor's blood sample was not available for Rh phenotyping or Gm allotyping. It could be argued that the anti-E was produced by donor passenger lymphocytes as "naturally occurring" antibodies or as a primary response to E-positive red cell transfusions of the recipient. Although anti-E can be a naturally occurring or non–red-cell-induced antibody,[17,18] this explanation seems unlikely because the donor's antibody screening tests were negative, indicating that there was no serologically detectable anti-E prior to harvesting the organ. A primary antibody response by passenger lymphocytes to E-positive red cells cannot be entirely excluded; however, primary responses usually require several weeks or months before antibody titers rise to detectable levels.

In summary, the development of posttransplant hemolytic anemia in this case, coupled with the serologic findings of anti-A and anti-E and the concordant recipient and donor medical histories, represents, to our knowledge, the first report of the coincident occurrence of passenger lymphocyte syndrome and a delayed hemolytic transfusion reaction. In our previous studies, passenger lymphocyte syndrome was identified in 56% of recipients who received ABO-unmatched liver transplants.[13] Delayed hemolytic transfusion reactions occur in approximately 1 (0.66%) of 151 patients who receive red cell transfusions at our hospital.[17] Therefore, the incidence of simultaneous passenger lymphocyte syndrome and DHTR can be calculated (0.56×0.0066) to occur in approximately 4 of every 1000 patients who receive ABO-unmatched liver transplants and multiple red cell transfusions.

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