Case Report

Anti-Jk3 in a Filipino man

S. McCaskill, S. Wise, and S. Tinsley

A 62-year-old Filipino man with a history of chronic obstructive pulmonary disease, hypertension, and hyperlipidemia was admitted to the emergency department at Hospital A with recurrent fevers, weakness, and jaundice. The patient was evaluated and eventually discharged with a diagnosis of possible drug-induced hepatitis. One month later, the patient was admitted to Hospital B for recurrent fevers and weakness. The patient's hemoglobin was 3.8 g/dL. Six units of packed red blood cells (RBCs) were ordered for transfusion. The patient's sample typed as group B, D+, and the antibody screen was negative. All six units of packed RBCs appeared compatible (at immediate spin) and were transfused to the patient. His hemoglobin level 4 days post-transfusion was 9.3 g/dL, and the patient was discharged. The patient returned after a week for follow-up and his hemoglobin was found to have dropped to 8.5 g/dL, which continued to fall until it reached 7.0 g/dL. Additional packed RBCs were ordered for transfusion. During subsequent pre-transfusion compatibility testing, the antibody screen was found to be positive (all screening cells reactive at the antihuman globulin phase). An antibody identification panel was performed. The patient's serum was found to react with all panel cells tested, including the autocontrol tube. A direct antiglobulin test revealed the presence of both anti-IgG and anti-C3 coating the patient's RBCs. The specimen was then sent to a reference laboratory for further testing. Results from the reference lab testing revealed the presence of anti-Jk3 in the patient's serum. The patient was placed on steroids, and his reticulocyte count increased with no further signs of extravascular hemolysis. No additional transfusions were necessary. He was eventually discharged with a hemoglobin of 13.6 g/dL. The purpose of this case study is to report the findings of an extremely rare but clinically significant antibody, anti-Jk3.
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Key Words: magnetic resonance cholangiopancreatography, MRCP, antihuman globulin, AHG, direct antiglobulin test, DAT, intravenous immunoglobulin, IVIG, delayed hemolytic transfusion reaction, DHTR, breakpoint cluster region protein, BCR, Janus kinas 2 gene, JAK2, Abelson murine leukemia viral oncogene homolog 1, ABL1

The Kidd blood group system is defined by two antigens, Jk^a and Jk^b, that yield four different phenotypes: Jk(a+b+), Jk(a+b–), Jk(a–b+), and Jk(a–b–). Jk(a–b–) is also known as Jk:–3 or the Jk_{null} phenotype. The antigens Jk^a and Jk^b were discovered by Allen, Diamond, and Niedziela in 1951[1] and the Jk(a–b–) phenotype was discovered soon after by Pinkerton and Mermod in 1959.[2] Individuals with the Jk(a–b–) phenotype lack both Jk^a and Jk^b. The Jk(a–b–) phenotype can result from the homozygous inheritance of recessive alleles that produce neither Jk^a nor Jk^b or from the inheritance of a dominant inhibitor gene, *In(Jk),* reported to have no association with the *Jk* locus but causes a suppression of the expression of Kidd antigens.[3] The Jk(a–b–) phenotype is seen more frequently in people of Polynesian, Filipino, and Chinese descent.[4]

The first reported occurrence of the Jk(a–b–) phenotype was detected in a Filipino woman with Chinese and Spanish ancestry.[5] The Jk(a–b–) phenotype is rare among white and black populations and is more common among Polynesians, Filipinos, and Chinese.[3,6] The Jk(a–b–) phenotype frequency among the white population is less than 0.1 percent.[6] In Taiwan, the Jk(a–b–) phenotype was found to be 1.0 percent among the Rukai tribe and 1.2 percent among the Paiwan tribe.[7] Among Polynesians, the Jk(a–b–) phenotype frequency is 1.4 percent.[6] The Jk(a–b–) phenotype has also been reported to occur more frequently in Indian, Brazilian, Japanese, and Thai populations.[8]

Alloantibodies to Kidd antigens are clinically significant in transfusion recipients and women who are pregnant. The antibodies are produced in reaction to antigen exposure during a previous transfusion or pregnancy and remain in low titer in plasma until secondary exposure occurs. Kidd antibodies are capable of binding complement and are known to cause both immediate and delayed hemolytic transfusion reactions (DHTRs) as well as hemolytic disease of the fetus and newborn (HDFN). The production of anti-Jk3 occurs only in individuals who have inherited recessive alleles causing the Jk(a–b–) phenotype; those who have inherited the dominant inhibitor gene *In(Jk)* cannot make anti-Jk3.[8] Anti-Jk3 is best detected by the indirect antiglobulin test (IAT) and can be enhanced with enzyme pretreatment of RBCs used in antibody identification procedures.

Case Report

A 62-year-old Filipino man with a history of chronic obstructive pulmonary disease, hypertension, and hyperlipidemia was admitted to the emergency department at Hospital A with recurrent fevers, weakness, and jaundice. His liver function tests were elevated: aspartate aminotransferase (AST) = 64 IU/L (normal 1–40 IU/L),

alanine aminotransferase (ALT) = 85 IU/L (normal range 7–567 IU/L), alkaline phosphatase (ALP) = 561 IU/L (normal range 44–147 IU/L), total bilirubin = 1.8 mg/dL (normal range 0.3–1.9 mg/dL), and an ultrasound showed some evidence of liver disease of uncertain cause. He underwent magnetic resonance cholangiopancreatography (MRCP), which showed no evidence of biliary obstruction/dilation or stones. Other serologic testing for infectious disease markers and autoimmune disorders was negative.

His hemoglobin was low, between 7 and 9 g/dL (normal range elderly male: 12.4–14.9 g/dL), and serum ferritin was greater than 2,000 (normal range male: 12–300 ng/mL). The patient's peripheral blood smear was unremarkable with the exception of leukocytosis and the presence of target cells. Target cells were suggestive of biliary obstruction/dilation or stones, but this was ruled out by MRCP. The patient also had a high serum ferritin level, which is a common finding in patients with liver disease. In a patient experiencing a DHTR, schistocytes and reticulocytosis may be present on the peripheral blood smear; they were absent in this patient. The bone marrow biopsy did show hypocellularity with atypical megakaryocytic hyperplasia and erythroblastopenia. An IgG plasma cell dyscrasia was also noted with 13 percent to 15 percent plasma cells (flow and immunofixation positive, fluorescence in situ hybridization [FISH] negative) with serum protein electrophoresis of 1.5 (reference range: 0.7–1.5 g/dL). Results of genetic testing revealed the patient to be JAK2-negative and FISH for BCR/ABL–negative, but positive for three copies of ABL1 w/trisomy 9/9q, which can be associated with BCR/ABL1-negative chronic myeloproliferative disease, myelodysplastic syndrome, acute myelogenous leukemia, and, rarely, B-cell acute lymphoblastic leukemia. Tests for parvovirus B19 (serum IgM and immunohistochemistry) on the bone marrow were negative, but the presence of intranuclear eosinophilic inclusions in the bone marrow suggested possible parvovirus infection.

The patient was subsequently discharged. The patient was admitted to Hospital B 1 month later with recurrent fevers and weakness. His hemoglobin was 3.8 g/dL. A peripheral blood smear showed signs of leukocytosis (29K) (normal range male: 4.5–11.0K) and thrombocytosis (normal range: 150–450K). A bone marrow biopsy showed hypercellularity with atypical megakaryocytes, hyperplasia, and erythroblastopenia. IgG plasma cell dyscrasia was noted with 13 percent to 15 percent plasma cells and the presence of eosinophilic inclusions suggestive of parvovirus infection. Serologic tests for parvovirus were negative. A type and crossmatch for six units of blood was ordered. The patient was found to be group B, D+–; the antibody screen was negative; and all six units ordered appeared compatible (immediate spin crossmatch). The patient received all six units of type-specific, packed red blood cells (RBCs) with no signs of adverse reaction. The patient was also given intravenous immunoglobulin (IVIG) for the suspected parvovirus infection. His hemoglobin level 4 days post-transfusion was 9.3 g/dL, and he was discharged 10 days from date of admittance.

The patient returned to Hospital B 1 week later for follow-up with an oncologist, and his hemoglobin was found to have dropped to 8.5 g/dL, which continued to fall until it reached 7.0 g/dL (approximately 19 days after transfusion). Additional packed RBCs were ordered for transfusion. During subsequent compatibility testing procedures, the antibody screen was found to be positive with all screening cells at the antihuman globulin (AHG) phase of testing. An antibody identification panel was performed. The patient's serum was found to react with all panel cells tested including the autocontrol tube at the AHG phase of testing. A direct antiglobulin test (DAT) panel was performed using polyspecific (anti-IgG + anti-C3) and monospecific AHG reagents (anti-IgG, anti-C3). The results of the DATs showed both anti-IgG and anti-C3 coating the patient's RBCs. The specimen was then sent to a reference laboratory for further testing. Results from the reference lab testing using a polyethylene glycol (PEG)-IAT method revealed the presence of anti-Jk3 in the patient's serum. An elution was not performed because the patient had not been transfused within the previous 2 weeks. Aliquots of the patient's serum were adsorbed using allogeneic cells. Clinically significant antibodies to other major blood group antigens were excluded by the PEG-IAT using the adsorbed plasma. Phenotyping was performed on autologous neocytes recovered by microhematocrit centrifugation. Results of the phenotyping showed the patient to be Jk(a–b–). Genomic sequencing was also performed on the patient's RBCs to determine the genotype that would allow for the prediction of the patient's phenotype. Results of the genetic testing revealed the presence of two variant alleles that were detected by JK-cDNA analysis and genomic sequencing. *Jk*02N.01* carried the intron 5c.342-1g>a variant that is associated with exon 6 skipping and a null Jk(b–) phenotype. The *Jk*01* allele carried the two known changes of c.130G>A and c.893G>A, not normally reported on the same allele. The JKc.130A single-nucleotide polymorphism (SNP) is associated with weakened antigen expression, and the c.893A SNP is associated with a null Jk(a–) phenotype. It was recommended, if transfusion was necessary, to select ABO/Rh compatible units negative for both Jk^a and Jk^b.

Discussion

This case report focuses on a clinically significant but infrequent antibody known as anti-Jk3. The clinical significance of anti-Jk3 is that it displays weak reactivity in vitro but has the capacity to induce severe RBC destruction in vivo in the presence of Jk^a and/or Jk^b.[3] Anti-Jk3 will present as a "panagglutinin" because the antibody will react with all panel cells, since all panel cells are Jk(a+b–), Jk(a–b+), or Jk(a+b+).[9,10] The autocontrol tube may be either positive or negative depending on whether or not the patient has been recently transfused. The DAT will usually be positive for both anti-IgG and anti-C3. If the patient has been recently transfused (within the last 3 months), the use of allogeneic cells for adsorption procedures is necessary to rule out antibodies to other major blood group antigens. The decision to perform an elution rests with a facility's protocol(s). Some facilities may elect to do an elution on any patient specimen with positive DAT results, whereas other facilities may be more selective in terms of number of days following a recent transfusion. The patient was questioned concerning transfusion of blood at other facilities and reported that he had never received a transfusion prior to the units that were given at Hospital B. The timing of this DHTR is consistent with the formation of a new antibody rather than an evanescent form.

Another laboratory test that can be helpful in the identification of the Jk(a–b–) phenotype is the urea lysis test. In 1982, Heaton and McLoughlin showed that Jk(a–b–) RBCs were resistant to lysis in the presence of 2M urea. RBCs with normal Kidd phenotypes rapidly swell and will lyse in the presence of 2M urea.[11] In 1995, another group showed that the Kidd blood group antigens and urea transport function were carried on the same protein structure. As a result, individuals with the Jk(a–b–) phenotype may not be able to concentrate urine as well as individuals with normal Kidd antigens.[12]

Serologic phenotyping is most commonly used to identify specific antigens on RBCs. A patient with the Jk(a–b–) phenotype should be nonreactive when tested with anti-Jk^a and anti-Jk^b typing sera. It is important to check the patient's transfusion history, since recent transfusion could invalidate phenotyping results. If the patient has been recently transfused, autologous neocytes prepared through microhematocrit centrifugation can be used for phenotyping. Genotyping for Kidd variants may also be used to support serologic typing and to predict Kidd phenotypes when antisera are unavailable.[13] This particular patient was shown to have inherited two variant alleles producing the Jk(a–b–) phenotype. Jk(a–b–) RBCs are recommended for transfusion in patients with anti-Jk3. Because of the rareness of the Jk(a–b–) phenotype, it is difficult to find blood for these patients. Finding compatible units for such patients will require the use of rare donor registries. Siblings of the patient can also be tested to see if they might be of the same phenotype as the patient.

In this case, IVIG was given to the patient because of the suspected parvovirus infection. This is one of the off-label uses of IVIG as described by Hillyer et al.[14] Also, because IVIG is prepared from pools of donor plasma, it may contain antibodies to blood group antigens (e.g., anti-A, -B, -D, and -K; panagglutinins). In this particular case, the IVIG did not interfere in the serologic testing used to identify the antibody present in the patient's serum.

Conclusion

This case report presents the findings of an extremely rare but clinically significant antibody. Anti-Jk3 is capable of causing severe DHTRs and HDFN. This antibody is difficult to identify in that it will present as a "panagglutinin," meaning that it will react with all panel cells tested, since they are typically either Jk(a+) or Jk(b+) or both. The autocontrol may be positive if the patient has been recently transfused. If the autocontrol is positive, a DAT should be performed to characterize the protein coating the patient's RBCs. An elution should also be performed to identify the antibody coating the patient's RBCs, since it is not safe to assume that the antibody present in the patient's serum is the same antibody that is coating the patient's RBCs. Because most blood banks and transfusion services have limited resources, confirmation of the antibody specificity and phenotyping of the patient's RBCs may need to be performed by a reference laboratory. Only blood that is Jk(a–b–) should be transfused to patients with anti-Jk3. Because of the difficulty in finding compatible units for such patients, rare donor registries will need to be consulted in the event that transfusion is required. The use of IVIG for patient treatment must be taken into consideration when performing serologic workup for suspected transfusion reactions because of contaminating antibodies that may be present in the formulation that could interfere with serologic testing.

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