Impact of red blood cell alloimmunization on sickle cell disease mortality: a case series

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BACKGROUND: Although red blood cell (RBC) transfusion represents an integral component of sickle cell disease (SCD) care, transfusion support for some patients can result in alloimmunization to RBC antigens. Alloimmunized patients with SCD appear to experience worse survival compared to nonalloimmunized patients. While this difference in mortality may in part be due to underlying immunologic differences related to disease severity, it may also reflect direct clinical consequences of RBC alloimmunization. Alloimmunized patients have an increased risk of serious hemolytic transfusion reactions (HTRs) and may not receive adequate RBC transfusion support due to lack of compatible RBC units.
CASE REPORT: This study reports on five RBC alloimmunized patients with SCD who died, to illustrate the concept that RBC alloimmunization itself contributes to premature death.
RESULTS: The clinical course for each of the reported patients provides insight into the direct and indirect consequences of RBC alloimmunization, where patients experienced delayed HTRs or did not receive needed RBC transfusions.
CONCLUSION: Future work examining the clinical impact of RBC alloimmunization should not only consider HTRs but should also address the potential consequences associated with difficulties in obtaining compatible blood.

Red blood cell (RBC) transfusion often represents a lifesaving intervention for individuals with sickle cell disease (SCD). Transfusion is critical in the management of acute SCD complications such as aplastic crisis, splenic sequestration, and acute chest syndrome (ACS). Chronic transfusion to maintain a hemoglobin (Hb)S percentage less than 30% effectively prevents stroke and improves quality of life for certain patients.[1-3] A well-recognized complication of transfusion for patients with SCD is alloimmunization to RBC antigens. The incidence of RBC alloimmunization in SCD can be decreased by prophylactic matching for certain minor RBC antigens (specifically C/c, E/e, and K),[4] but this practice does not completely prevent alloimmunization. Chou

ABBREVIATIONS: ACS = acute chest syndrome; AVN = avascular necrosis; DHTR(s) = delayed hemolytic transfusion reaction(s); HSCT = hematopoietic stem cell transplant; HTR(s) = hemolytic transfusion reaction(s); ICU = intensive care unit; MMA = monocyte monolayer assay; SCD = sickle cell disease.

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and colleagues[5] demonstrated that despite antigen matching for D, C, E, and K via serologic phenotyping, 58% of chronically transfused and 15% of episodically transfused patients with SCD became alloimmunized. Alloimmunization can occur even with serologically determined antigen matching because of variant *RH* alleles.[5] Furthermore, it is currently not feasible to phenotypically match for all potentially significant RBC antigens for all patients with SCD.

RBC alloimmunization can cause serious hemolytic transfusion reactions (HTRs), which can be fatal and lead to a hyperhemolysis syndrome. While many have described this specific complication in patients with SCD,[6-17] few have comprehensively studied the clinical impact of RBC alloimmunization on patients with SCD. Aygun and coworkers[11] attempted to evaluate the clinical significance of RBC alloantibodies by retrospectively reviewing the records of 140 transfused patients with SCD who received a total of 3239 RBC units over a 10-year period. RBC alloimmunization was common (29% of pediatric patients, 47% of adult patients) in this cohort; however, less than 4% of all patients had reported clinical evidence of a HTR, and only one patient died secondary to this complication. These authors thereby concluded that "clinically, RBC alloimmunization and its sequela may be acceptable." This study, however, may have failed to fully evaluate the clinical implications of RBC alloimmunization. Some clinically significant, delayed HTRs (DHTRs) may not have been recognized as a DHTR, given that DHTRs can present similarly to vasoocclusive pain crises. Vidler and colleagues,[12] in a retrospective study of 2158 RBC transfusion episodes in 220 SCD adults, illustrate this concept with the finding that 48% of DHTR episodes were not diagnosed as a DHTR at the time of the event. This study also demonstrated that clinical DHTRs were not rare (occurring in 8% of transfused patients) and were associated with prolonged hospitalizations and the need for critical care support.

HTRs are not the only clinical complication of RBC alloimmunization. Obtaining compatible RBCs for patients with certain RBC alloantibodies may be difficult or impossible. The resulting delay associated with obtaining safe blood for transfusion or inability to safely transfuse may have harmful clinical consequences for patients and may contribute to mortality. This idea is consistent with a recent study involving alloimmunized SCD patients who were found to have significantly decreased survival compared to nonalloimmunized SCD patients (median life expectancy of 54 years vs. 65 years).[18] In this study of 319 adult SCD patients, those who were alloimmunized also had a significantly higher prevalence of avascular necrosis (AVN) and daily home opioid use. The authors proposed that immune system activation might contribute to both alloimmunization and chronic pain development. This chronic pain may then lead to decreased survival. It is important to note that in this study, alloimmunized patients had a similar frequency of acute vasoocclusive pain episodes to nonalloimmunized patients, suggesting that alloimmunized patients do not inherently have more severe disease. Thus, RBC alloimmunization itself may directly contribute to decreased survival through HTRs and problems obtaining compatible blood. To illustrate this proposed concept, we present case reports of five alloimmunized patients with SCD who died, four before 20 years of age.

CASE REPORTS

Patient 1

Patient 1 was a 19-year-old male with HbSS disease on hydroxyurea with severe daily pain from hip AVN. He had a history of multiple RBC alloantibodies (anti-C, -E, -K, -Fya, -Fyb, -Jkb, and -M) and RBC autoantibodies (cold and warm). He experienced three prior DHTRs with bystander hemolysis at ages 10, 12, and 17. The reaction at age 17 occurred during an episode of ACS when he was transfused with antiglobulin test crossmatch-compatible RBCs lacking the antigens he was known to have alloantibodies against (pretransfusion Hb 5.5 g/dL, posttransfusion Hb 7.0 g/dL). Twelve days later he was admitted to the intensive care unit (ICU) with severe pain, hemoglobinuria, hyperbilirubinemia, and a direct antibody test (DAT) positive for complement; his Hb nadir was 2.6 g/dL. He was treated with intravenous immune globulin (IVIG) and steroids and recovered from that episode. Given his history of severe DHTRs, further transfusion was avoided. The patient suffered daily pain and ambulatory difficulties from his hip AVN, and therefore, at age 19, desired hip replacement surgery, which required preoperative transfusion. In an effort to decrease the risk of a DHTR, rituximab was administered preoperatively at 375 mg/m^2 weekly for 4 weeks. He then received 2 units of antiglobulin test crossmatch-compatible RBCs lacking the C, E, K, Fya, Fyb, Jkb, and M antigens (pretransfusion Hb 7.1 g/dL, posttransfusion Hb 9.5 g/dL). Three days after this transfusion, on the day of his scheduled surgery admission, he developed severe whole body pain and had a Hb of 7.3 g/dL, hemoglobinuria, and hyperbilirubinemia. Indirect antibody testing showed new reactivity in two of seven screening cells that were negative for the antigens against which he had previously identified antibodies. His surgery was canceled and he was admitted for supportive care. He subsequently developed respiratory failure and required transfer to the ICU. The day after admission he also developed fever and a peripheral blood culture grew *Salmonella*. Despite treatment with antibiotics, methylprednisolone (30 mg/kg × 1), IVIG (1 g/kg × 1), and epoetin alfa (50 units/kg ordered three times weekly), he continued to worsen clinically. On the second day of the

hospitalization his Hb decreased to 5 g/dL. On the third day of the hospitalization an additional antiglobulin test crossmatch-compatible RBC transfusion was started, but it was stopped after only transfusing 40 mL due to concerns of a transfusion reaction because of increased patient agitation and peaked T-waves. The patient died later that day. The last tested Hb was 2 g/dL.

Patient 2

Patient 2 was an 18-year-old male with HbSS disease who at age 10 was started on chronic transfusions at an outside institution after an acute stroke. The outside institution did not prospectively C/c, E/e, and K antigen match RBCs and he developed anti-C, -e, -K, and -Fya alloantibodies (patient's serologic phenotype C−, c+, E+, e−, Fya−, Fyb−, Jka+, Jkb−, K−) as well as suffered a DHTR a few months after initiation of chronic transfusions. Because of these problems, he was taken off chronic transfusion therapy and treated with hydroxyurea. At age 18, the patient was admitted to a different outside hospital for a foot ulcer where he received an antiglobulin test crossmatch-compatible, non–antigen-matched RBC transfusion. This other institution's blood bank did not seek out information about previously identified alloantibodies that were no longer serologically detectable (even though the family had informed clinicians that the patient had a history of many RBC alloantibodies). His posttransfusion Hb was 9.0 g/dL and he was discharged in stable condition. Five days after this transfusion, he presented to his primary institution with leg pain. His admission Hb was 7.2 g/dL. Over the next 3 days, his Hb decreased to 4.7 g/dL. He had a negative DAT, but had hyperbilirubinemia and hemoglobinuria. After contacting the outside facility where he was recently transfused, it was determined that he had received units that were e+ despite his historic anti-e alloantibody. He was treated with methylprednisolone (initially 1 mg/kg every 6 hr × 3 days and then weaned) and transfused phenotypically matched (e–), antiglobulin test crossmatch-compatible RBCs. After a 7-day hospitalization, he was discharged on an 18-day prednisone taper (discharge Hb 6.9 g/dL). At clinic follow-up 3 weeks later, his Hb was 7.1 g/dL with a reticulocyte count of 10.0%, quantification of HbS was 89.2% and HbA was 3.4%. Five days after this visit, he was found at home unresponsive and in asystole. His Hb was 1.9 g/dL with a reticulocyte of 9.4%. No sample was obtained for DAT, but his plasma was negative for hemolysis. With intensive cardiopulmonary resuscitation that included epinephrine × 4 and emergent transfusions, he briefly regained cardiac function but was neurologically devastated from the event. He suffered cardiac arrest again later that same day and died. Significant findings on autopsy included hepatic sequestration, marrow necrosis, extensive marrow thrombotic emboli, and pulmonary hypertension with cor pulmonale.

Patient 3

Patient 3 was a 15-year-old female with HbSS disease on hydroxyurea therapy due to a history of frequent episodes of vasoocclusive pain and ACS as well as multiple RBC transfusions. She also had chronic abdominal pain, loose stools, occult blood on stool guaiac testing, and weight less than the 5th percentile, concerning for gastrointestinal disease. During a hospitalization for pain (14 months before death), RBC transfusion was ordered because of a decrease in Hb from 8.5 g/dL (baseline) to 6.0 g/dL; antibody screen was positive but 1 crossmatch-compatible unit was transfused. Three months later she was scheduled for endoscopy and colonoscopy with preoperative transfusion at a different hospital, but these procedures were canceled because of a positive RBC antibody screen with e-like specificity, subsequently determined to be anti-hr(B). *RHCE* and *RHD* molecular typing with the BioArray human erythrocyte antigen (HEA) BeadChip (Immucor, Norcross, GA) showed Dce(733G)/Dce(48C, 733G) with the predicted phenotype: D+, C–, partial c+, E–, partial e+, VS+, V+, hr^{B}–/+^{w}. The patient never pursued further evaluation of her gastrointestinal disorder despite ongoing symptoms (pain and melena) as well as attempts to reschedule her evaluation by the treating hematologist. Two months later, during hospitalization for vasoocclusive pain, she was transfused 1 unit of RBCs (e–) due to a decline in Hb to 5.9 g/dL. Posttransfusion Hb was 8.2 g/dL at 1 day posttransfusion and 8.8 g/dL at 6 days posttransfusion; DAT was positive for immunoglobulin (Ig)G at 6 days posttransfusion. Seven months after this transfusion, antibody screen identified a new anti-E. Given the presence of anti-E and anti-hr(b) alloantibodies, transfusion was avoided despite chronic worsening anemia. Over several months, the patient's Hb declined from approximately 8 to approximately 6 g/dL with microcytosis (decrease in MCV from 80-90 fL to approx. 55 fL), raising concern for iron deficiency due to ongoing gastrointestinal blood loss. Oral iron was prescribed, hydroxyurea was stopped temporarily, and subcutaneous erythropoietin was administered during hospitalization. During her final hospitalization for extremity pain, intravenous (IV) ferric gluconate was given as her Hb continued to decrease from 6.5 to 4.6 g/dL with inadequate erythropoiesis (absolute reticulocyte counts, 110×10^9−233×10^9/L). Three days after discharge from this hospitalization and after experiencing gross melena and new abdominal pain, she was found unresponsive at home. She was transported to the hospital by emergency medical services in asytole, and intensive resuscitation was unsuccessful. Autopsy showed acute appendicitis with peritonitis and transmural necrosis of the cecum and ascending colon consistent with inflammatory bowel disease.

Patient 4

Patient 4 was a 41-year-old male with HbSβ^0 thalassemia and end-stage renal disease secondary to SCD nephropathy. He had a history of multiple RBC alloantibodies, including anti-Jkb, -Fya, -Fyb, -Jsa, -K, -E, -C, -S, -V, -N, and -Leb, and a DHTR that required hospitalization. The patient was being evaluated for renal transplant, but the transplant team was concerned that his RBC alloimmunization would seriously complicate transplantation. He also had a history of cholethiasis and recurrent biliary colic. After a several-month delay due to concerns about finding compatible blood, 7 units of antiglobulin test crossmatch-compatible RBC units negative for the antigens to which he had historic RBC alloantibodies were obtained, and he was scheduled to undergo laparoscopic cholecystectomy and arteriovenous graft repair. He was admitted preoperatively for IV hydration and transfusion of 5 RBC units, increasing his Hb from 4.7 to 9.8 g/dL. The patient tolerated surgery well with minimal blood loss; however, a few hours after surgery, he developed hypotensive shock and was found to have an Hb of 3.3 g/dL. He received the remaining 2 compatible RBC units and underwent emergent exploratory laparotomy, which demonstrated intraperitoneal hemorrhage from a bleeding umbilical arteriole. After RBC transfusion and surgery, his Hb was 5.7 g/dL. Additional RBC units negative for antigens against all historic antibodies were not available and his Hb decreased to 2.0 g/dL in less than 24 hours. Because of his life-threatening status, he was transfused with least incompatible units by serology, one S+ and another V+ unit. He initially tolerated these transfusions (posttransfusion Hb of 5.4 g/dL). With intensive support, including 100 μg of darbopoietin alfa weekly and 50 mg of hydrocortisone every 12 hours for 4 days, and transfusion of 2 fully compatible units, he clinically stabilized and 7 days after his surgery had a Hb of 7.1 g/dL. Over the next 5 days, however, he had a decline in his Hb to 2.9 g/dL with hyperbilirubinemia and a positive DAT (IgG+ and complement+). No additional compatible units could be immediately obtained and the patient subsequently developed ischemic colitis. After national and international searches, 2 additional compatible units were eventually obtained and transfused. By this time, he had already developed multisystem organ failure and 2 days after the transfusion of these units, on Hospital Day 17, he died.

Patient 5

Patient 5 was a 14-year-old female with HbSS disease and systemic lupus erythematosus, on hydroxyurea and mycophenolate mofetil, and with multiple complications including chronic pain, chronic lung disease, and feeding intolerance. At age 8, 2 weeks after receiving C/E/K phenotypically matched RBCs for ACS (posttransfusion Hb 11.5 g/dL), she suffered a DHTR with bystander hemolysis (Hb nadir 3.7 g/dL, DAT positive; new anti-Jkb, anti-Fya, and anti-S alloantibodies were identified). She was hospitalized for 4 days and received steroid treatment and an antiglobulin test crossmatch-compatible RBC transfusion. At age 10, during another episode of ACS, she received antiglobulin test crossmatch-compatible C/E/K phenotypically matched RBCs that were also lacking the Jkb, Fya, and S antigens (posttransfusion Hb 9.0 g/dL). Two weeks later, she suffered yet another DHTR with bystander hemolysis (Hb nadir 4.8 g/dL) requiring hospitalization for 8 days. During this episode she was noted to have a new alloantibody against a high-incidence antigen. A monocyte monolayer assay (MMA), completed using two sets of Jkb–, Fya–, and S– RBCs, was significant at 9 and 33% reactive monocytes. Extensive genetic analysis revealed that the patient lacked a high-prevalence antigen in the Cromer system (Tc(a–b+)), and she was determined to have an anti-Tca alloantibody. Additionally, she was noted to genotype as Sl(a–) and as KCAM–. Only a single compatible unit was identified in an international search. Given the patient's discordant MMA test results concerning for an additional unidentified antibody as well as her propensity to develop DHTR with subsequent worsening anemia, transfusion even with the single identified unit was avoided. Over the next few years, she had multiple prolonged hospitalizations for pain and required nightly home biphasic positive airway pressure support. She also was treated with 400 units/kg/dose epoetin alfa weekly for 9 months in an attempt to improve her baseline Hb, but it remained between 7 and 8 g/dL. Given the severity of her disease, the family was interested in hematopoietic stem cell transplant (HSCT). However, the inability to obtain compatible RBCs that would have been necessary to support her through HSCT prevented the patient and her family from pursuing transplant further. At age 14, she was admitted for pain and fever, and had a Hb of 7.0 g/dL. During the course of her hospitalization she had a worsening of her respiratory status and a progressive decline in her Hb (nadir 3.6 g/dL) despite resuming epoetin alfa and iron therapy as well as 24 mg/m^2 dexamethasone $\times$ 2 doses. During this time the patient's complement levels were normal (C3 125 mg/dL, C4 32 mg/dL) and rheumatology felt that her lupus was well controlled. Her antibody screen was positive to a high-incidence antigen, consistent with the previously characterized anti-Tca. Thirty-four days after her admission, she died in the ICU due to high output cardiac and respiratory failure.

DISCUSSION

RBC alloimmunization likely contributed to the deaths of each of the reported patients (Table 1). Despite efforts to prevent a DHTR with prophylactic rituximab treatment and the transfusion of RBCs lacking antigens against

TABLE 1. Summary of patient cases illustrating the clinical consequences of RBC alloimmunization

Patient	RBC alloantibodies prevented patient from receiving	Cause of death	Association of RBC alloimmunization with death
1	Hip replacement surgery	DHTR with bystander hemolysis, *Salmonella* sepsis	Fatal DHTR occurred despite pretransfusion treatment with rituximab
2	Chronic transfusion therapy	Hepatic sequestration/embolic event, chronic organ damage due to SCD	Death occurred after severe DHTR episode
3	Sedation to work up underlying gastrointestinal disease	Acute appendicitis	Underlying severe anemia from no transfusion likely contributed to death
4	Renal transplantation	Complications from cholecystectomy bleed	Inability to safely transfuse emergently post-op likely contributed to death
5	Chronic transfusion therapy, HSCT	High-output cardiac and respiratory failure	Inability to safely transfuse in setting of progressive, severe anemia likely contributed to death

known alloantibodies, Patient 1 still suffered a severe DHTR with bystander hemolysis that, along with *Salmonella* sepsis, led to his death. Patient 2 did not have evidence of an active DHTR at the time of his death, but his severe DHTR just a few weeks earlier may have triggered a cascade of events that ultimately caused his death. In particular, the pathogenesis of hepatic sequestration is not well understood.[19,20] While it is possible that his fatal hepatic sequestration occurred completely independently of his recent DHTR, it is also possible that hyperbilirubinema and tissue hypoxia from the prior DHTR initiated intrahepatic vasoocclusion that then led to hepatic sequestration. Patient 3 was not transfused because of her alloantibodies and, due to her resultant severe anemia, she likely decompensated rapidly from acute appendicitis, a treatable problem. Patient 4 died as a result of his iatrogenic bleed after cholecystectomy: a complication he likely would have recovered from with aggressive RBC transfusion support if compatible RBCs had been available. Finally, Patient 5 had multiple medical problems that contributed to her death, but the inability to safely transfuse her was paramount.

In addition to implicating RBC alloimmunization as a significant contributor to their deaths, these patient cases also illustrate notable concepts related to the clinical consequences of RBC alloimmunization. First, some institutions may not appreciate the importance of seeking a patient's RBC antibody history to match for alloantibodies that are no longer detectable (Patient 2). If alloimmunized patients are given RBC units positive for antigens against historic alloantibodies, then these patients may suffer a DHTR even if the units appeared to be antiglobulin test crossmatch compatible. Patients who have RBC alloantibodies detected at one point in time often subsequently have negative antibody screens due to antibody evanescence,[21-23] so transfusion histories noting previously detected alloantibodies are critical. While some regions have a centralized transfusion database,[23] in many areas no registry exists that could alert providers about historic alloantibodies identified at other institutions. In addition, a centralized donor registry of rare phenotyped RBC units would also be helpful. Currently multiple blood procurement services often need to be contacted to find rare units that can delay care.

These cases also demonstrate that difficulties in obtaining blood or fear of further alloimmunization may delay or even prevent patients from receiving needed medical procedures (Patients 1-3). Such difficulties may also prevent alloimmunized patients from receiving chronic transfusions (Patients 2 and 5). RBC alloimmunization concerns may also prevent patients from being considered as candidates for solid organ transplant or curative HSCT (Patients 4 and 5). Alternatively, significant RBC alloimmunization can be an appropriate indication for HSCT if a sufficient number of compatible RBC units can be acquired to support a patient through transplant.[24]

These cases also illustrate the concept that the decision to transfuse or not to transfuse an alloimmunized patient with SCD can be very difficult. This decision is further complicated by the fact that not all RBC alloantibodies will cause hemolysis if antigen-positive RBC units are transfused.[25] Better methods are needed to predict the clinical significance of specific alloantibodies to appropriately guide transfusion restrictions for individual patients.[26,27] One method that appears to be helpful in this setting is the MMA, a cellular-functional assay that seeks to determine if an antibody is clinically significant.[28] Noumsi and coworkers[29] recently reported that 103 RBC units that were incompatible by serology but compatible by MMA (monocyte index $< 5\%$) were effectively transfused to 31 patients with no report of an adverse clinical reaction. For alloimmunized patients in which antigen-negative RBC units are difficult to obtain, the use of the MMA to select RBC units for transfusion is thus an alternative strategy to exhaustive searches for antigen-negative blood. This assay, however, is not widely available or simple to perform. In addition, some alloimmunized patients will still be deemed "untransfusable," with incompatible MMA results for all tested RBC units (Patient 5).

The use of RBC genotyping in the initial transfusion management of patients with SCD may help decrease the incidence of RBC alloimmunization in the future. Casas

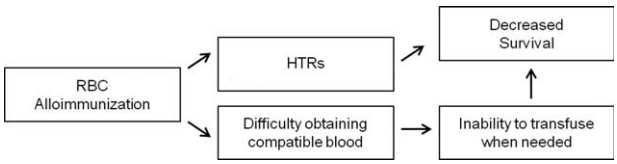

Fig. 1. Proposed pathway involving RBC alloimmunization and decreased survival in patients with SCD.

and coworkers[30] recently demonstrated in 494 patients with SCD that DNA-based RBC typing was more accurate than serologic phenotyping and, additionally, identified patients who lacked or had altered high-prevalence antigens. However, the use of RBC genotyping in the transfusion management of SCD patients is unlikely to eliminate the problem of RBC alloimmunization, given the limited number of blood donors (mostly from minority groups) with certain needed rare RBC genotypes.[31,32] Prospective RBC antigen matching for all patients with SCD may also not be cost-effective.[33,34]

Strong evidence does not exist to guide the management of HTRs in SCD. For example, while a few reports have suggested that rituximab is effective in preventing or treating HTRs in patients with SCD,[35-38] Patient 1 demonstrates that this therapy is not always effective. Corticosteroids and IVIG have been traditionally recommended in severe cases, but they have never been formally studied in this context.[17] A recent case report also documented the successful treatment of a SCD patient with a life-threatening DHTR by using the C5 inhibitor eculizumab.[39] While such cases are informative, reporting bias is possible. Clinical trials should be designed to study immunosuppressive agents in the management of HTRs.

Additional work is also needed to understand the underlying immunology that causes some, but not all, patients to become alloimmunized. Studies have reported that alloimmunized patients with SCD have received a greater number of RBC units than nonalloimmunized patients, but alloimmunization does not strongly correlate with the number of RBC units received.[5,18] Some patients become alloimmunized after only a few transfusions (responders), while other patients never develop a RBC alloantibody after even hundreds of transfusions (nonresponders). Variables other than transfusion burden thus clearly mediate alloimmunization. Genetic factors related to B lymphocyte signal modulation may play a role.[40] Studies have also found differences in T cells between responder and nonresponder patients with SCD.[41-43] Recipient inflammatory state at time of transfusion also appears to affect alloimmunization.[44] SCD patients transfused during a proinflammatory complication (particularly ACS) appear to be at increased risk for subsequent RBC alloantibody development.[45]

A potential critique of this case series is that these patients represent rare scenarios associated with RBC alloimmunization and that most patients with RBC alloantibodies do not have problems. In support of this thinking, the number of fatalities reported to the US Food and Drug Administration (FDA) that were determined to be transfusion-related due to non-ABO HTRs has consistently been very low over the past few years (<8 deaths/year).[46] It is critical to realize, though, that some cases in which RBC alloimmunization may have contributed to the death of a patient do not actually involve a transfusion reaction (Patients 3 and 5) or only indirectly involve a possible transfusion reaction (Patients 2 and 4) so would not be reported to the FDA. In addition, the patients presented in this case series were not compiled after an exhaustive search, but rather from the clinical experiences of the authors caring for patients at two institutions over just a 2-year time period (January 2013 to March 2015). Finally, while we believe that RBC alloimmunization contributed significantly to the five described deaths, at least in one case (Patient 2) this contribution may be overstated. The proposed link between RBC alloimmunization and decreased survival (Fig. 1) suggested by these cases merits further study. In particular, future studies should investigate premature deaths in a large cohort of patients with SCD to explicitly determine the actual incidence of the proposed problem that alloimmunization contributes to death.

Death in childhood from SCD today is rare in high-income nations compared to several decades ago.[47,48] As people with SCD are living longer, the problem of RBC alloimmunization for this population is likely to only increase. The use of RBC transfusion for SCD has increased over the past few years[49,50] and recent evidence suggests that a greater number of patients could benefit from chronic transfusion therapy.[2] If RBC alloimmunization directly contributes to premature mortality in SCD, then further efforts to prevent and manage RBC alloimmunization are clearly justified. Even if such a direct link is not established, it is likely that RBC alloimmunization delays appropriate medical care, causing increased morbidity and medical costs. Additional research on the biology underlying RBC alloimmunization[51-53] is needed to help develop new therapies to mitigate the dangers of this complication.

CONFLICT OF INTEREST

CDJ is a consultant for Immucor and Octapharma. The other authors have disclosed no conflicts of interest.

REFERENCES

1. Adams RJ, McKie VC, Hsu L, et al. Prevention of a first stroke by transfusions in children with sickle cell anemia and abnormal results on transcranial Doppler ultrasonography. N Engl J Med 1998;339:5-11.

2. DeBaun MR, Gordon M, McKinstry RC, et al. Controlled trial of transfusions for silent cerebral infarcts in sickle cell anemia. N Engl J Med 2014;371:699-710.
3. Beverung LM, Strouse JJ, Hulbert ML, et al. Health-related quality of life in children with sickle cell anemia: impact of blood transfusion therapy. Am J Hematol 2015;90:139-43.
4. Vichinsky EP, Luban NL, Wright E, et al. Prospective RBC phenotype matching in a stroke-prevention trial in sickle cell anemia: a multicenter transfusion trial. Transfusion 2001;41:1086-92.
5. Chou ST, Jackson T, Vege S, et al. High prevalence of red blood cell alloimmunization in sickle cell disease despite transfusion from Rh-matched minority donors. Blood 2013; 122:1062-71.
6. King KE, Shirey RS, Lankiewicz MW, et al. Delayed hemolytic transfusion reactions in sickle cell disease: simultaneous destruction of recipients' red cells. Transfusion 1997;37:376-81.
7. Win N, New H, Lee E, et al. Hyperhemolysis syndrome in sickle cell disease: case report (recurrent episode) and literature review. Transfusion 2008;48:1231-8.
8. Win N. Hyperhemolysis syndrome in sickle cell disease. Expert Rev Hematol 2009;2:111-5.
9. Talano JA, Hillery CA, Gottschall JL, et al. Delayed hemolytic transfusion reaction/hyperhemolysis syndrome in children with sickle cell disease. Pediatrics 2003;111(6 Pt 1):e661-5.
10. Chadebech P, Habibi A, Nzouakou R, et al. Delayed hemolytic transfusion reaction in sickle cell disease patients: evidence of an emerging syndrome with suicidal red blood cell death. Transfusion 2009;49:1785-92.
11. Aygun B, Padmanabhan S, Paley C, et al. Clinical significance of RBC alloantibodies and autoantibodies in sickle cell patients who received transfusions. Transfusion 2002;42:37-43.
12. Vidler JB, Gardner K, Amenyah K, et al. Delayed haemolytic transfusion reaction in adults with sickle cell disease: a 5-year experience. Br J Haematol 2015;169:746-53.
13. Santos B, Portugal R, Nogueira C, et al. Hyperhemolysis syndrome in patients with sickle cell anemia: report of three cases. Transfusion 2015;55:1394-8.
14. de Montalembert M, Dumont MD, Heilbronner C, et al. Delayed hemolytic transfusion reaction in children with sickle cell disease. Haematologica 2011;96:801-7.
15. Petz LD, Calhoun L, Shulman IA, et al. The sickle cell hemolytic transfusion reaction syndrome. Transfusion 1997;37: 382-92.
16. Ipe TS, Wilkes JJ, Hartung HD, et al. Severe hemolytic transfusion reaction due to anti-D in a D+ patient with sickle cell disease. J Pediatr Hematol Oncol 2015;37:e135-7.
17. Gardner K, Hoppe C, Mijovic A, et al. How we treat delayed haemolytic transfusion reactions in patients with sickle cell disease. Br J Haematol 2015;170:745-56.
18. Telen MJ, Afenyi-Annan A, Garrett ME, et al. Alloimmunization in sickle cell disease: changing antibody specificities and association with chronic pain and decreased survival. Transfusion 2015;55:1378-87.
19. Ahmed N, Chizhevsky V. Acute hepatic sequestration associated with pneumococcal infection in a 5-year-old boy with sickle beta degrees -thalassemia: a case report and review of the literature. J Pediatr Hematol Oncol 2007;29:720-4.
20. Gardner K, Suddle A, Kane P, et al. How we treat sickle hepatopathy and liver transplantation in adults. Blood 2014;123: 2302-7.
21. Tormey CA, Stack G. The persistence and evanescence of blood group alloantibodies in men. Transfusion 2009;49:505-12.
22. Rosse WF, Gallagher D, Kinney TR, et al. Transfusion and alloimmunization in sickle cell disease. The Cooperative Study of Sickle Cell Disease. Blood 1990;76:1431-7.
23. Harm SK, Yazer MH, Monis GF, et al. A centralized recipient database enhances the serologic safety of RBC transfusions for patients with sickle cell disease. Am J Clin Pathol 2014; 141:256-61.
24. Fasano RM, Monaco A, Meier ER, et al. RH genotyping in a sickle cell disease patient contributing to hematopoietic stem cell transplantation donor selection and management. Blood 2010;116:2836-8.
25. Stowell SR, Liepkalns JS, Hendrickson JE, et al. Antigen modulation confers protection to red blood cells from antibody through Fcγ receptor ligation. J Immunol 2013;191:5013-25.
26. Stowell SR, Girard-Pierce KR, Smith NH, et al. Transfusion of murine red blood cells expressing the human KEL glycoprotein induces clinically significant alloantibodies. Transfusion 2014;54:179-89.
27. Stowell SR, Winkler AM, Maier CL, et al. Initiation and regulation of complement during hemolytic transfusion reactions. Clin Dev Immunol 2012;2012:307093.
28. Arndt PA, Garratty G. A retrospective analysis of the value of monocyte monolayer assay results for predicting the clinical significance of blood group alloantibodies. Transfusion 2004; 44:1273-81.
29. Noumsi GT, Billingsley KL, Moulds JM. Successful transfusion of antigen positive blood to alloimmunised patients using a monocyte monolayer assay. Transfus Med 2015;25: 92-100.
30. Casas J, Friedman DF, Jackson T, et al. Changing practice: red blood cell typing by molecular methods for patients with sickle cell disease. Transfusion 2015;55:1388-39.
31. Shaz BH, Hillyer CD. Minority donation in the United States: challenges and needs. Curr Opin Hematol 2010;17:544-9.
32. Karafin MS, Field JJ, Gottschall JL, et al. Barriers to using molecularly typed minority red blood cell donors in support of chronically transfused adult patients with sickle cell disease. Transfusion 2015;55:1399-406.
33. Kacker S, Ness PM, Savage WJ, et al. Cost-effectiveness of prospective red blood cell antigen matching to prevent alloimmunization among sickle cell patients. Transfusion 2014;54:86-97.
34. Kacker S, Ness PM, Shirey RS, et al. The future of red blood cell alloimmunization risk reduction. Transfusion 2015;55: 222-4.

35. Bachmeyer C, Maury J, Parrot A, et al. Rituximab as an effective treatment of hyperhemolysis syndrome in sickle cell anemia. Am J Hematol 2010;85:91-2.
36. Noizat-Pirenne F, Bachir D, Chadebech P, et al. Rituximab for prevention of delayed hemolytic transfusion reaction in sickle cell disease. Haematologica 2007;92:e132-5.
37. Cattoni A, Cazzaniga G, Perseghin P, et al. An attempt to induce transient immunosuppression pre-erythrocytapheresis in a girl with sickle cell disease, a history of severe delayed hemolytic transfusion reactions and need for hip prosthesis. Hematol Rep 2013;5:36-8.
38. Delmonte L, Cantini M, Olivieri O, et al. Immunoglobulin-resistant delayed hemolytic transfusion reaction treated with rituximab in an adult sickle cell patient. Transfusion 2013;53: 688-9.
39. Boonyasampant M, Weitz IC, Kay B, et al. Life-threatening delayed hyperhemolytic transfusion reaction in a patient with sickle cell disease: effective treatment with eculizumab followed by rituximab. Transfusion 2015;55:2398-403.
40. Tatari-Calderone Z, Tamouza R, Le Bouder GP, et al. The association of CD81 polymorphisms with alloimmunization in sickle cell disease. Clin Dev Immunol 2013;2013:937846.
41. Bao W, Zhong H, Li X, et al. Immune regulation in chronically transfused allo-antibody responder and nonresponder patients with sickle cell disease and beta-thalassemia major. Am J Hematol 2011;86:1001-6.
42. Vingert B, Tamagne M, Habibi A, et al. Phenotypic differences of CD4(+) T cells in response to red blood cell immunization in transfused sickle cell disease patients. Eur J Immunol 2015;45:1868-79.
43. Zhong H, Bao W, Friedman D, et al. Hemin controls T cell polarization in sickle cell alloimmunization. J Immunol 2014;193:102-10.
44. Hendrickson JE, Desmarets M, Deshpande SS, et al. Recipient inflammation affects the frequency and magnitude of immunization to transfused red blood cells. Transfusion 2006;46:1526-36.
45. Fasano RM, Booth GS, Miles M, et al. Red blood cell alloimmunization is influenced by recipient inflammatory state at time of transfusion in patients with sickle cell disease. Br J Haematol 2015;168:291-300.
46. Fatalities reported to FDA following blood collection and transfusion: annual summary for fiscal year 2013. Silver Spring (MD): U.S. Food and Drug Administration; 2014.
47. Telfer P, Coen P, Chakravorty S, et al. Clinical outcomes in children with sickle cell disease living in England: a neonatal cohort in East London. Haematologica 2007;92:905-12.
48. Quinn CT, Rogers ZR, McCavit TL, et al. Improved survival of children and adolescents with sickle cell disease. Blood 2010;115:3447-52.
49. Raphael JL, Oyeku SO, Kowalkowski MA, et al. Trends in blood transfusion among hospitalized children with sickle cell disease. Pediatr Blood Cancer 2013;60:1753-8.
50. Drasar E, Igbineweka N, Vasavda N, et al. Blood transfusion usage among adults with sickle cell disease: a single institution experience over ten years. Br J Haematol 2011;152:766-70.
51. Stowell SR, Henry KL, Smith NH, et al. Alloantibodies to a paternally derived RBC KEL antigen lead to hemolytic disease of the fetus/newborn in a murine model. Blood 2013; 122:1494-504.
52. Zimring JC, Spitalnik SL. Pathobiology of transfusion reactions. Annu Rev Pathol 2015;10:83-110.
53. Girard-Pierce KR, Stowell SR, Smith NH, et al. A novel role for C3 in antibody-induced red blood cell clearance and antigen modulation. Blood 2013;122:1793-801.