

## REVIEW ARTICLE

Dan L. Longo, M.D., *Editor*

## Hemolytic Transfusion Reactions

Sandhya R. Panch, M.D., M.P.H., Celina Montemayor-Garcia, M.D., Ph.D.,  
and Harvey G. Klein, M.D.

From the Department of Transfusion Medicine, Warren G. Magnuson Clinical Center, National Institutes of Health Clinical Center, Bethesda, MD. Address reprint requests to Dr. Panch at the Center for Cellular Engineering, Bldg. 10, 3C-720D, Department of Transfusion Medicine, Warren G. Magnuson Clinical Center, National Institutes of Health, Bethesda, MD 20892, or at sandhya.panch@nih.gov.

N Engl J Med 2019;381:150-62.

DOI: 10.1056/NEJMra1802338

Copyright © 2019 Massachusetts Medical Society.

**B**LOOD TRANSFUSION IS THE MOST COMMON THERAPEUTIC PROCEDURE performed in hospitalized patients; some 15% of inpatients receive blood components during their stay. Approximately 1% of transfused products result in serious adverse reactions,<sup>1</sup> including hemolytic transfusion reactions, which account for up to 5% of these serious adverse reactions.<sup>2</sup> Although technical and administrative controls to prevent transfusion of ABO-mismatched blood have reduced transfusion-related deaths, immune-mediated hemolysis remains an important, if underappreciated, risk. Deaths attributed to emergency transfusion in patients with an unknown antibody history and hemolysis related to non-red-cell components such as platelets, plasma, and intravenous immune globulin constitute a small but serious problem. Delayed hemolytic transfusion reactions and life-threatening “bystander hemolysis” (i.e., hemolysis of autologous red cells), particularly in patients with hemoglobinopathies who have received multiple transfusions, present unique diagnostic challenges with regard to the timing of presentation, predictability, symptom overlap with other complications, and antibody identification and management.<sup>3</sup> Owing to increases in solid-organ and hematopoietic stem-cell transplantation, donor lymphocyte-mediated immune hemolysis is no longer a rare event.<sup>4-6</sup>

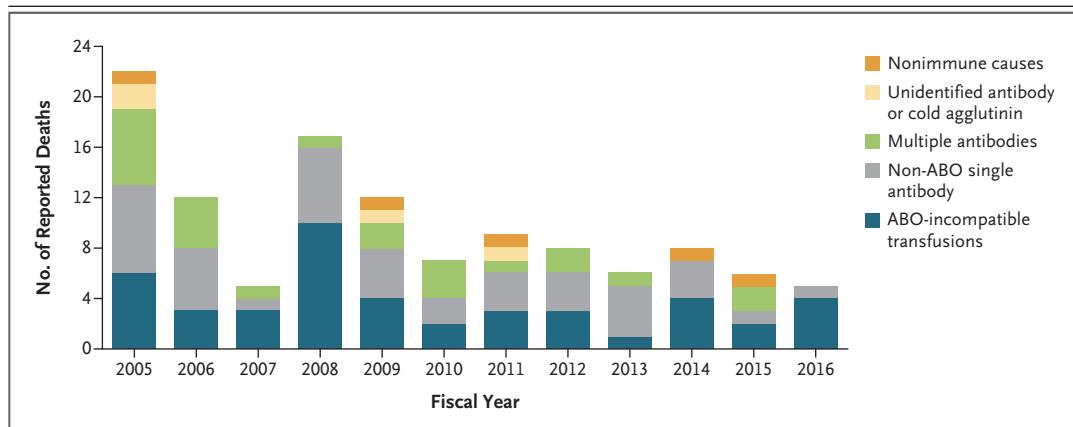
## HISTORY

The earliest description of an incompatible hemolytic transfusion reaction dates to the experimental start of transfusion therapy in the mid-17th century. While treating a nobleman who had episodes of violent mental derangement with infusions of “soothing” calf blood, Jean-Baptiste Denis described what has become the classic reaction:

The patient was transfused with 5-6 ounces of calves’ blood. During the procedure, the patient complained that the vein in his right arm became quite painful. The procedure was repeated 2 days later; a larger transfusion was given. Following the transfusion, however, the patient complained of pain in the arm vein; his pulse rose, he vomited, and he had a severe nosebleed, pain over the kidney, and an “oppressive sensation in the chest.” The next day, he “made a great glass of urine with a color as black as if it had been mixed with the soot of a chimney.”<sup>7</sup>

The severity of the reaction prompted the Parlement of Paris (the appellate court), along with most of Europe, to ban all human transfusions. These signs and symptoms have come to define acute immune-mediated hemolysis.

With Landsteiner’s discovery of ABO blood groups in 1900, red-cell agglutination patterns became the recognized laboratory method for typing blood. Ottenberg



**Figure 1. Annual Reported Deaths in the United States from Hemolytic Transfusion Reactions.**

The data, reported by the Food and Drug Administration for fiscal years 2005 through 2016,<sup>9</sup> show an overall decline in deaths related to hemolytic transfusion reactions, with persistently low numbers of reported deaths in more recent years.

applied this technique to routine pretransfusion testing as a way to prevent hemolytic transfusion reactions.<sup>8</sup> The development of anticoagulant-preservative solutions allowed not only storage of typed blood but also sufficient time to perform extended laboratory testing before transfusion. A combination of serologic techniques and molecular identification of the corresponding red-cell genes is currently used to provide extended compatibility testing, to select rare compatible units, and to help establish the diagnosis when hemolytic reactions are suspected.

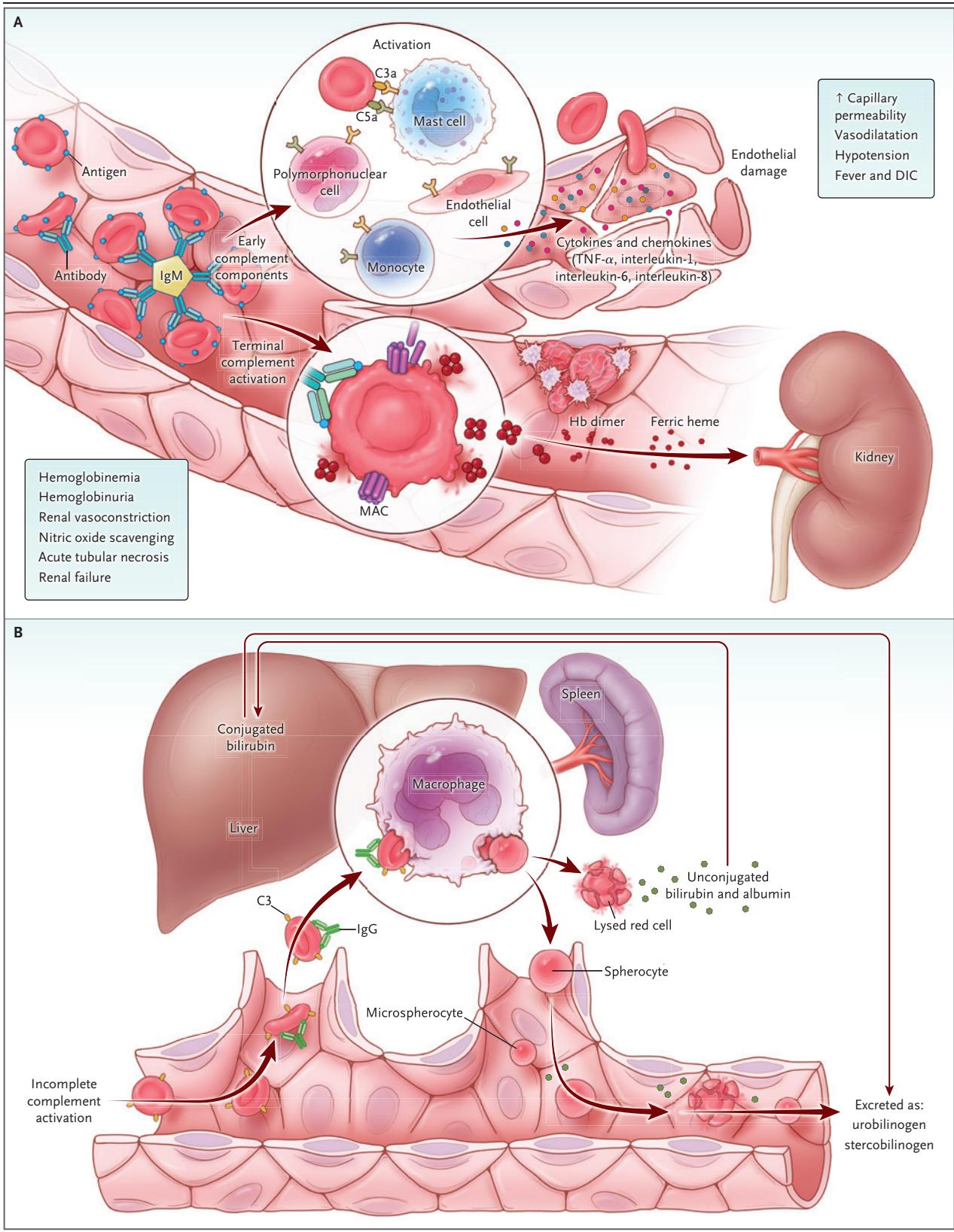
#### EPIDEMIOLOGIC FEATURES

The Food and Drug Administration (FDA) requires reporting of transfusion-related deaths in the United States under the Code of Federal Regulations, Title 21, Section 606.170(b), and publishes annual compilations of hemolysis-associated deaths (Fig. 1).<sup>9</sup> Any effort to discern national trends should take into account that the classification was modified in fiscal year 2015 to ensure consistency with other national and international agencies and that deaths attributed to transfusion are probably underreported. The United States does not require reporting of severe, nonlethal hemolytic reactions.

Hemolysis was the most commonly cited cause of transfusion-associated death during the 1976–1985 period<sup>10</sup> but has become one of the least common fatal complications of transfusion, with an estimated risk of one death per 1,972,000

red-cell units transfused in 2016.<sup>11</sup> Much of this decline can be attributed to a reduction in transfusion of ABO-incompatible blood. Fatal hemolytic transfusion reactions continued to decrease from fiscal year 2005 through fiscal year 2010, but one to four deaths continue to be reported annually (Fig. 1).<sup>9</sup> The risk of fatal ABO-incompatible transfusion is still estimated to exceed the combined risks of infection with human immunodeficiency virus and hepatitis B and C viruses.<sup>12,13</sup>

The most frequent preventable cause of lethal hemolysis remains misidentification of the patient or mislabeling of the blood sample from the identified recipient, commonly referred to as “wrong blood in the tube.”<sup>14</sup> In the United Kingdom, ABO-incompatible events that were nearly fatal were reported for 25 units per 100,000 issued in 2017.<sup>15</sup> Deaths from hemolysis have also been documented after emergency transfusions in patients with an unknown antibody history.<sup>9</sup> The rate of acute hemolytic transfusion reactions among patients receiving emergency transfusions of blood that has not been cross-matched, a common practice in the management of trauma, is estimated at 1 reaction per 2000 transfusions.<sup>2</sup> Robust data regarding nonfatal hemolytic transfusion reactions are not available, largely because of subclinical presentations and lack of rigorous reporting. Consequently, the incidence of delayed hemolytic transfusion reactions is estimated to range from 1 in 500 transfusions to 1 in 10,000 transfusions. Certain patients, such as those with sickle cell disease, appear to be at increased risk,



**Figure 2 (facing page). Pathophysiological Features of Acute and Delayed Hemolytic Transfusion Reactions.**

Panel A shows the pathophysiological features of acute hemolytic transfusion reactions. Immunologic incompatibility between donor and recipient results in foreign blood-group antigen recognition and binding by circulating IgM, activating terminal complement and leading to formation of the membrane attack complex (MAC). The MAC destroys red-cell membranes, releasing free hemoglobin (Hb) into the intravascular space, which results in end-organ damage, including acute tubular necrosis and renal failure. Early complement components cause endothelial damage and increased capillary permeability through activation of mast cells, polymorphonuclear cells, monocytes, and endothelial cells, which release cytokines and interleukins. DIC denotes disseminated intravascular coagulation, and TNF- $\alpha$  tumor necrosis factor  $\alpha$ . Panel B shows the pathophysiological features of delayed hemolytic transfusion reactions. Incomplete complement activation through IgG and C3b opsonization mediates splenic and hepatic erythrophagocytosis, resulting in spherocytes and microspherocytes. Lysis of red cells releases unconjugated bilirubin, which is transported to the liver. Hepatic conjugated bilirubin is excreted as urobilinogen and stercobilinogen. Anemia from red-cell destruction and jaundice from excess unconjugated and conjugated bilirubin are the primary clinical manifestations of delayed hemolytic transfusion reactions.

with the incidence of delayed hemolytic transfusion reactions and hyperhemolysis ranging from 1 to 20% of transfusions.<sup>16</sup> On the basis of an international hemovigilance database, delayed hemolytic transfusion reactions account for 4.3% of all transfusion reactions and 16% of all serious reactions.<sup>17,18</sup>

#### PATHOPHYSIOLOGICAL AND CLINICAL MANIFESTATIONS

Immunologic incompatibility between donor and recipient cell types is the most common cause of clinically significant hemolytic transfusion reactions. Acute reactions (i.e., those occurring within 24 hours after transfusion) develop in response to red cells transfused in patients with preexisting antibodies. Naturally occurring antibody reactions against ABO-incompatible transfusions have been implicated in a majority of the fatal cases.<sup>19</sup>

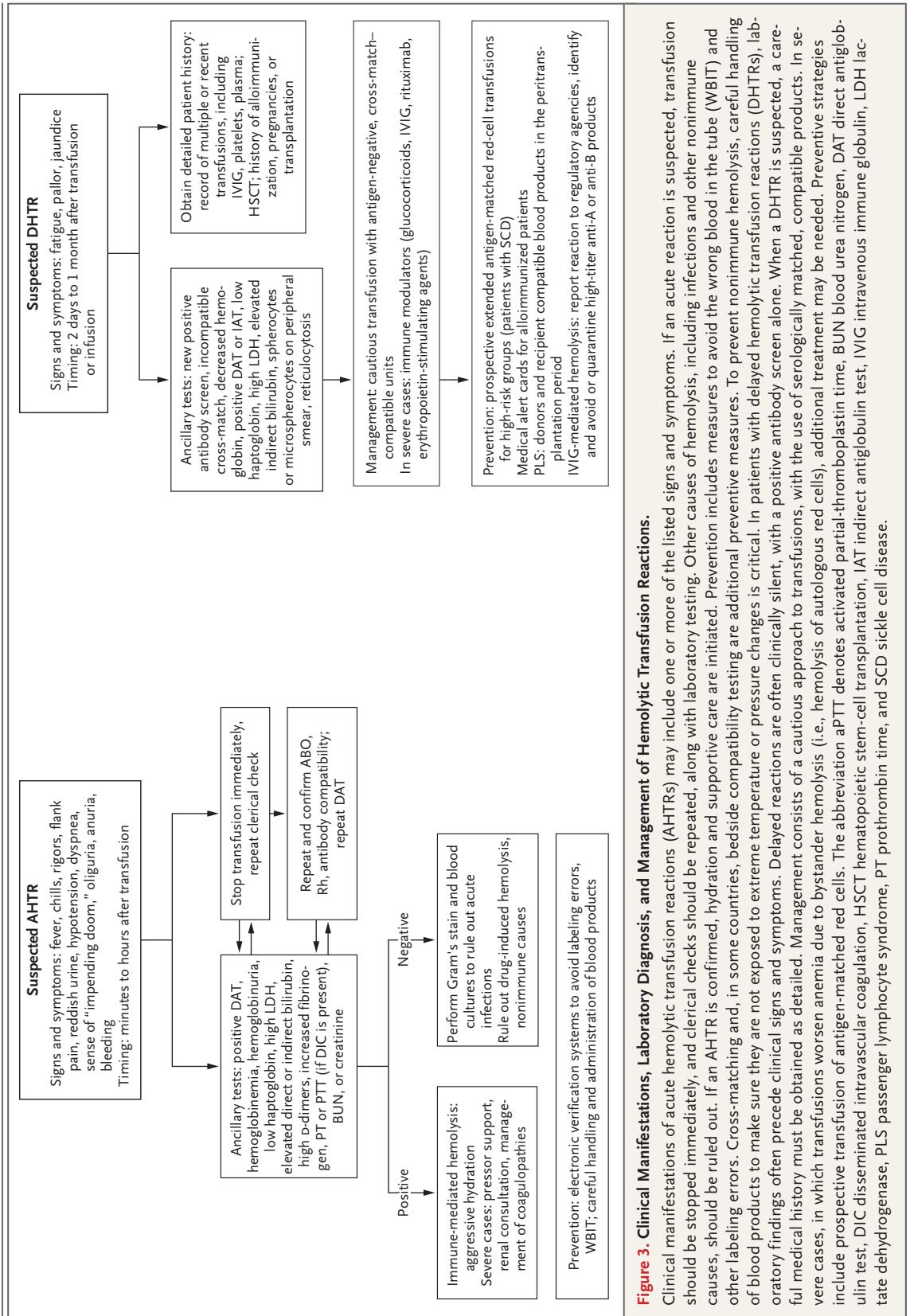
Incompatible A and B blood-group antigens interact with preexisting IgM antibodies and less commonly with hemolytic IgG antibodies, both of

which fix and activate complement. Formation of excessive terminal membrane attack complexes consisting of components C5 through C9 creates multiple pores in the transfused red-cell membranes, initiating intravascular osmolysis. The resulting excess cell-free hemoglobin overwhelms the binding capacity of plasma albumin, haptoglobin, and hemopexin and can be measured with assays of hemoglobinemia and hemoglobinuria. Free heme induces renal vasoconstriction through nitric oxide scavenging. Acute tubular necrosis and renal failure may ensue.<sup>20</sup>

Incomplete complement activation generates the anaphylatoxins C3a and C5a, which activate mast cells, releasing histamine and serotonin. These cells, along with the by-products of hemolysis, including residual red-cell stromal components, activated monocytes and leukocytes, enzymes, and anaphylatoxins, mediate the release of proinflammatory cytokines and chemokines (tumor necrosis factor  $\alpha$  and interleukin-8). Furthermore, activation of the bradykinin and kallikrein systems and coagulation pathways results in a systemic inflammatory response syndrome of increased capillary permeability, vasodilatation, hypotension, and fever, as well as disseminated intravascular coagulation. In extreme cases, the syndrome progresses to shock, with multiorgan failure and death (Fig. 2A).<sup>20</sup>

Incomplete complement activation also destroys incompatible red cells through C3b opsonization and monocyte-and-macrophage-induced erythrophagocytosis in the liver and spleen. Complement-coated red cells are phagocytosed in stages, with gradual removal of red-cell membrane and surface area resulting in spherocytes and microspherocytes. This extravascular destruction process, with minimal release of free hemoglobin in the plasma, may also be mediated by immunoglobulins that are recruited by B-cell growth and differentiation factors (interleukin-1 $\beta$  and interleukin-6). Reactions to Rh antibodies and to other non-ABO antigens may be manifested in this manner. Such hemolytic reactions typically occur 3 to 30 days after transfusion but may be immediate (Fig. 2B).<sup>21</sup>

Unlike acute hemolytic transfusion reactions, delayed hemolytic transfusion reactions are almost invariably caused by secondary (anamnestic) immune responses in patients immunized by previous transfusions, allogeneic stem-cell transplants,



**Figure 3. Clinical Manifestations, Laboratory Diagnosis, and Management of Hemolytic Transfusion Reactions.**

Clinical manifestations of acute hemolytic transfusion reactions (AHTRs) may include one or more of the listed signs and symptoms. If an acute reaction is suspected, transfusion should be stopped immediately, and clerical checks should be repeated, along with laboratory testing. Other causes of hemolysis, including infections and other nonimmune causes, should be ruled out. If an AHTR is confirmed, hydration and supportive care are initiated. Prevention includes measures to avoid the wrong blood in the tube (WBIT) and other labeling errors. Cross-matching and, in some countries, bedside compatibility testing are additional preventive measures. To prevent nonimmune hemolysis, careful handling of blood products to make sure they are not exposed to extreme temperature or pressure changes is critical. In patients with delayed hemolytic transfusion reactions (DHTRs), laboratory findings often precede clinical signs and symptoms. Delayed reactions are often clinically silent, with a positive antibody screen alone. When a DHTR is suspected, a careful medical history must be obtained as detailed. Management consists of a cautious approach to transfusions, with the use of serologically matched, compatible products. In severe cases, in which transfusions worsen anemia due to bystander hemolysis (i.e., hemolysis of autologous red cells), additional treatment may be needed. Preventive strategies include prospective transfusion of antigen-matched red cells. The abbreviation aPTT denotes activated partial-thromboplastin time, BUN blood urea nitrogen, DAT direct antiglobulin test, DIC disseminated intravascular coagulation, HSCT hematopoietic stem-cell transplantation, IAT indirect antiglobulin test, IVIG intravenous immune globulin, LDH lactate dehydrogenase, PLS passenger lymphocyte syndrome, PT prothrombin time, and SCD sickle cell disease.

or pregnancy. These reactions rarely constitute a medical emergency. In many instances, alloantibodies appear on routine testing in the blood bank (reported as “delayed serologic transfusion reactions”) and are not associated with clinical events. Clinical manifestations, if they occur, include anemia and jaundice due to extravascular red-cell destruction, followed by hemoglobin degradation and liberation of bilirubin into the plasma. Fever, hemoglobinuria, and hemoglobinemia are even less frequent.<sup>21,22</sup> Differences in the clinical presentations of acute hemolytic transfusion reactions and delayed hemolytic transfusion reactions are detailed in Figure 3.

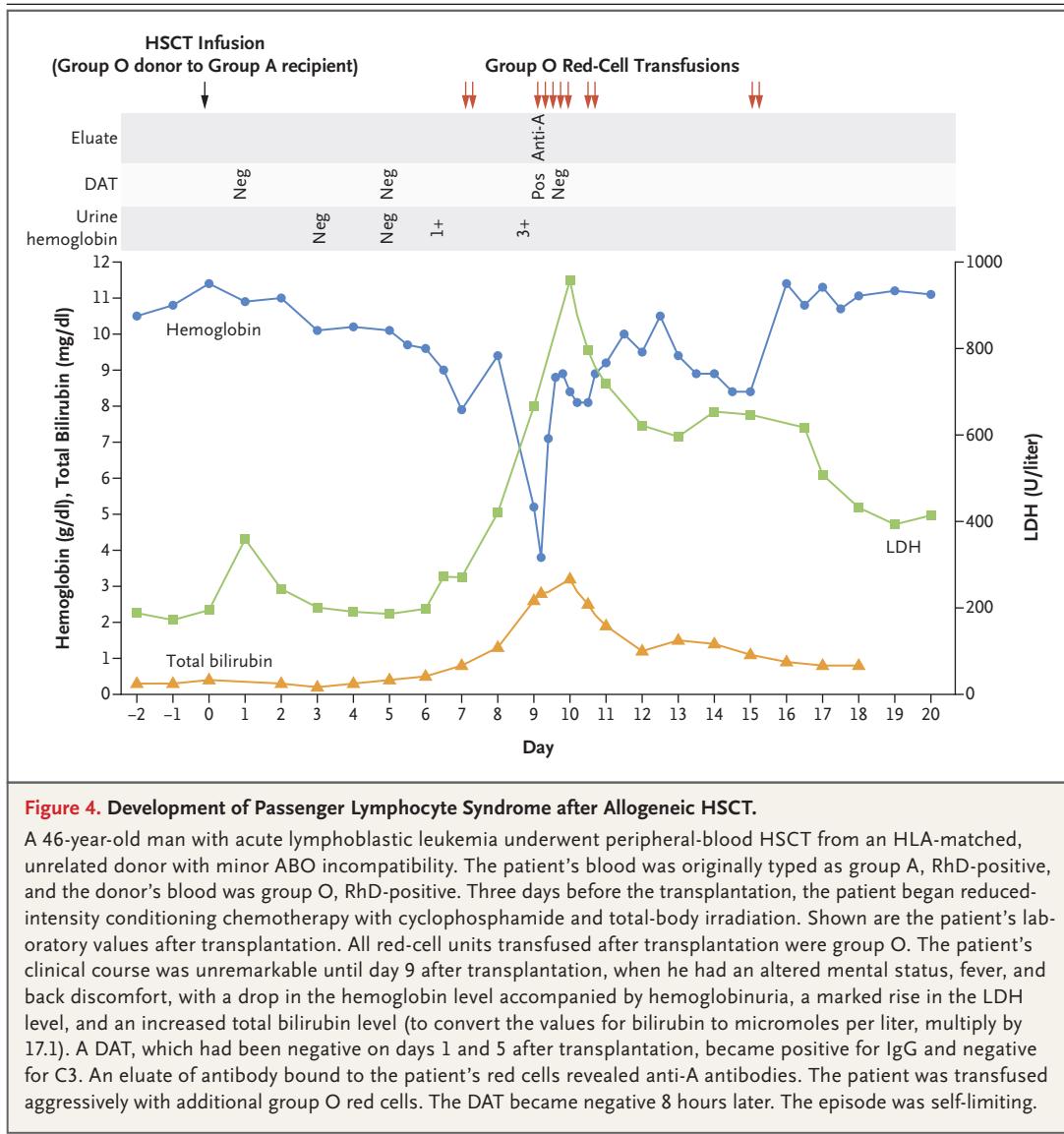
Occasionally, severe hemolytic reactions in patients receiving long-term transfusions for hematologic conditions such as sickle cell disease, thalassemia, or malaria can precipitate bystander hemolysis, in addition to clearing transfused red cells. The mechanisms are not well understood. This hyperhemolytic transfusion reaction may be mediated in part by the release of cell-free hemoglobin, which further activates leukocyte-driven inflammasome pathways and causes endothelial dysfunction through nitric oxide scavenging.<sup>23</sup> The decrease in reticulocytes in this context probably results from contact lysis of red-cell precursors by macrophages. This process may be immediate or delayed, with post-transfusion hemoglobin levels falling below the pretransfusion values, often to life-threatening levels. Further red-cell transfusion typically exacerbates ongoing hemolysis, with the exogenous (transfused) antigen probably triggering the development of a temporary pseudo-autoimmune state.<sup>24,25</sup>

Immune-mediated hemolysis may also occur after infusion of hematopoietic cells for transplantation or after solid-organ transplantation. Incompatibility between the donor’s plasma and the recipient’s red cells, termed minor ABO incompatibility, with subsequent red-cell destruction in the recipient, is the most common cause of clinically significant hemolysis in such cases.<sup>5</sup> However, viable donor B lymphocytes, termed “passenger lymphocytes,” are also transferred passively with the graft and produce isohemagglutinins that target recipient red cells. Life-threatening hemolysis due to passenger lymphocyte syndrome has been reported to develop 5 to 14 days after heart-lung,<sup>15</sup> liver,<sup>26</sup> kidney,<sup>4</sup> and intestinal<sup>27</sup> transplantations, as well as after hematopoietic stem-cell infusions.<sup>28</sup> Reduced-intensity conditioning regi-

mens and cyclosporine as prophylaxis against graft-versus-host disease (GVHD) or rejection have been associated with an increased risk of passenger lymphocyte syndrome.<sup>29</sup> Umbilical cord blood hematopoietic stem-cell grafts with predominantly naive T cells have not been associated with minor ABO-incompatible hemolysis. An illustrative case of delayed hemolysis after allogeneic stem-cell transplantation is shown in Figure 4. The evolution of preparative regimens (e.g., fludarabine), newer immunosuppressive agents, and modified combinations for GVHD prophylaxis (methotrexate-containing regimens) has significantly reduced the incidence of passenger lymphocyte syndrome.<sup>28</sup> Hematopoietic transplantation may also result in acute hemolysis due to incompatible red-cell destruction in the graft by recipient antibodies (major ABO incompatibility). Prolonged destruction of graft red-cell precursors in the recipient’s bone marrow may result in pure red-cell aplasia for up to 1 year after transplantation.<sup>30</sup>

Most platelets transfused in the United States are collected by means of apheresis and suspended in donor plasma, which contains antibodies complementary to blood type. Use of apheresis platelets is often prioritized according to the date of expiration, without regard for donor–recipient (plasmatic) ABO compatibility. Consequently, antibodies in group O platelets have been implicated in several hemolytic transfusion reactions.<sup>31,32</sup> Some blood collectors screen group O platelets for “high titer” antibodies and restrict transfusion of implicated units from “dangerous donors” in group O recipients. However, there is no universal definition of high-titer antibodies.<sup>33</sup> Similarly, plasma<sup>34</sup> and blood that has not been cross-matched<sup>2</sup> may result in clinically significant minor ABO-incompatible hemolysis.

Acute hemolysis may develop in patients treated with high-dose intravenous immune globulin, particularly patients with blood group A or AB.<sup>35</sup> One explanation involves the higher density of group A antigens than of group B antigens on the red-cell surface and the generally higher anti-A antibody titers in intravenous immune globulin products. Methods of manufacturing intravenous immune globulin differ in the extent to which they can remove these isoagglutinins.<sup>36</sup> Children with Kawasaki’s disease appear to be at particularly high risk.<sup>37-40</sup> Intravascular hemolysis has also been observed with intravenous infusion of anti-D antibody concentrates used for the treatment of



**Figure 4. Development of Passenger Lymphocyte Syndrome after Allogeneic HSCT.**

A 46-year-old man with acute lymphoblastic leukemia underwent peripheral-blood HSCT from an HLA-matched, unrelated donor with minor ABO incompatibility. The patient's blood was originally typed as group A, RhD-positive, and the donor's blood was group O, RhD-positive. Three days before the transplantation, the patient began reduced-intensity conditioning chemotherapy with cyclophosphamide and total-body irradiation. Shown are the patient's laboratory values after transplantation. All red-cell units transfused after transplantation were group O. The patient's clinical course was unremarkable until day 9 after transplantation, when he had an altered mental status, fever, and back discomfort, with a drop in the hemoglobin level accompanied by hemoglobinuria, a marked rise in the LDH level, and an increased total bilirubin level (to convert the values for bilirubin to micromoles per liter, multiply by 17.1). A DAT, which had been negative on days 1 and 5 after transplantation, became positive for IgG and negative for C3. An eluate of antibody bound to the patient's red cells revealed anti-A antibodies. The patient was transfused aggressively with additional group O red cells. The DAT became negative 8 hours later. The episode was self-limiting.

immune thrombocytopenia in RhD-positive patients.<sup>39</sup> Although hemolysis with the administration of intravenous immune globulin is generally modest and is detected most often in retrospect by laboratory methods, the occasional reports of severe cases should alert clinicians to the importance of this association.

Hemolysis in association with transfusion is attributed almost reflexively to immune mechanisms. However, a variety of nonimmune causes of hemolysis have been reported, and these cases differ from immune-mediated hemolysis with respect to diagnosis, management, and outcome. Nonimmune mechanisms of hemolysis include

transfusion of blood concurrently with hypo-osmolar solutions,<sup>41</sup> transfusion of overheated blood,<sup>42</sup> and transfusion of accidentally frozen blood.<sup>43</sup> Blood transfusion under pressure through small-bore needles<sup>44</sup> or with the use of leukoreduction filters during processing<sup>45</sup> may result in mechanical lysis of red cells. Autoimmune hemolytic anemias<sup>46</sup> and drug-induced hemolytic anemias<sup>47,48</sup> may be exacerbated by transfusion and can therefore mimic hemolytic transfusion reactions. Transfusion of blood contaminated with hemolytic bacteria and transfusion in patients with sepsis may mimic immune-mediated hemolysis,<sup>49</sup> as can transfusion of donor red cells with intrinsic

sic defects (e.g., glucose-6-phosphate dehydrogenase deficiency<sup>50</sup>) or transfusion in recipients with these red-cell defects.<sup>51</sup> Categories of hemolytic transfusion reactions are listed in Table 1.

#### DIAGNOSTIC CONSIDERATIONS

An acute hemolytic transfusion reaction is considered to be a medical emergency. Although fever, flank pain, and reddish urine represent the classic triad of an acute hemolytic transfusion reaction, this type of reaction may also be suspected if one or more of the following signs or symptoms appears within minutes to 24 hours after a transfusion: a temperature increase of 1°C or more, chills, rigors, respiratory distress, anxiety, pain at the infusion site, flank or back pain, hypotension, or oliguria. One fascinating early symptom, a “sense of impending doom,” has been reported by numerous patients and is possibly the equivalent of the “oppressive sensation in the chest” reported in the 17th century by Denis’s patient; it should not be ignored.

The severity of acute hemolytic transfusion reactions may be related to the titer strength of anti-A antibodies, anti-B antibodies, or both in the recipient’s plasma, as well as the volume of incompatible blood transfused and the rate of transfusion. Most deaths have been associated with infusions of 200 ml or more of incompatible blood, although volumes as small as 25 ml have been fatal, particularly in children. Laboratory testing does not predict the severity of the reaction.

When an acute hemolytic transfusion reaction is suspected, the transfusion should be stopped immediately, and the blood being transfused should be saved for analysis. Laboratory testing should include repeat ABO and Rh compatibility testing, along with additional antibody testing for non-ABO incompatibility. Visual inspection of urine and plasma, as well as testing for urine and plasma free hemoglobin, is standard. Timing is critical, since free hemoglobin is cleared rapidly from the circulation. Simultaneously, alternative causes, including infectious agents, must be ruled out by means of Gram’s staining and cultures of the remaining transfused component.

A newly identified positive direct antiglobulin test (direct Coombs’ test), which detects IgG or complement bound to the red-cell membrane, is pathognomonic of immune-mediated hemolysis (Fig. 5). Conversely, the indirect antiglobulin test

**Table 1. Categories of Hemolytic Transfusion Reactions.\***

#### Immune-mediated reactions

- Acute hemolytic transfusion reaction due to clerical error and consequent ABO or Rh incompatibility
- Acute hemolytic transfusion reaction due to emergency transfusion of blood that was not cross-matched
- Delayed hemolytic transfusion reaction due to prior (evanescent) antibodies
- Hyperhemolysis (with bystander hemolysis [i.e., hemolysis of autologous red cells]) in patients receiving long-term transfusions (for SCD or thalassemia)
- Hemolysis due to ABO-incompatible platelet or plasma infusions
- Hemolysis due to intravenous immune globulin or Rh immune globulin
- Passenger lymphocyte syndrome after hematologic or solid-organ transplantation
- Pure red-cell aplasia after transplantation (destruction of erythroid precursors in bone marrow)

#### Non-immune-mediated reactions

- Thermal injury (excess heat or cold)
- Osmotic lysis (from dextrose or inadequate deglycerolization)
- Mechanical injury (from pressurized or rapid infusions or infusion through a narrow leukodepletion filter)

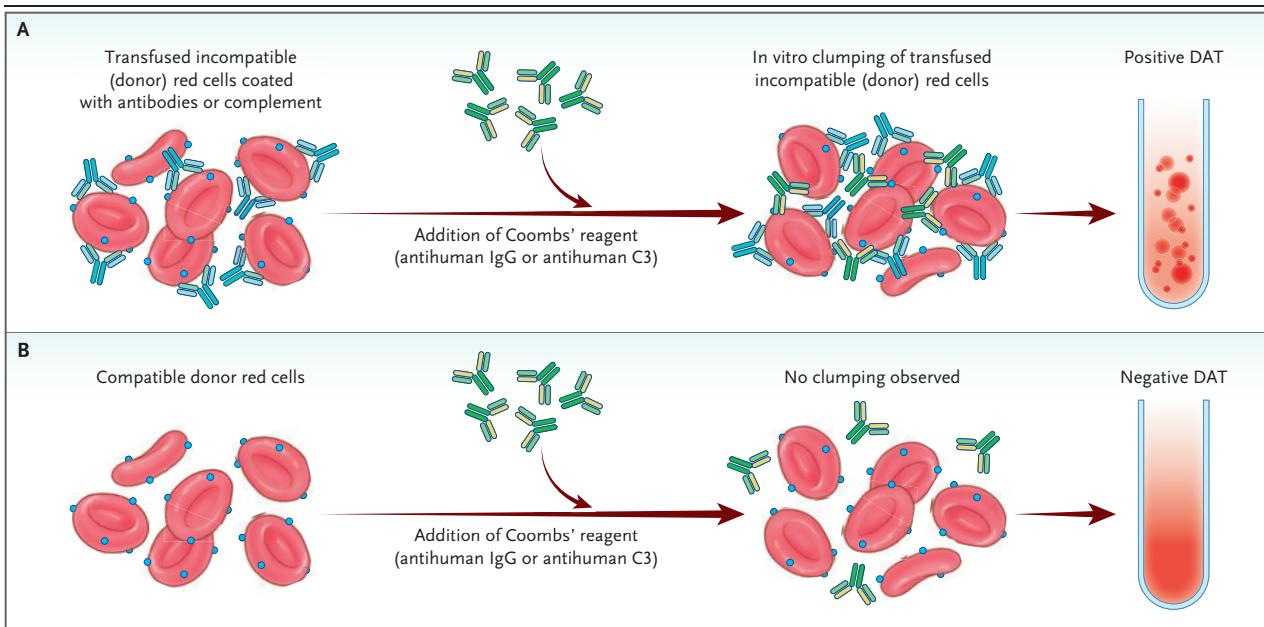
#### Conditions with exacerbated hemolysis after transfusion

- Autoimmune hemolytic anemia (warm or cold agglutinin disease)
- Drug-induced immune-mediated hemolytic anemia
- Sepsis in recipient or infusion of infected donor blood
- Red-cell membrane defects in donor or recipient

\* The listed mechanistic categories may overlap in clinical scenarios. SCD denotes sickle cell disease.

(indirect Coombs’ test) detects the presence of antibodies in the patient’s serum. Although severe hemolytic episodes produce strong reactions to the direct antiglobulin test, the strength of the reactions does not correlate with the degree of hemolysis. The test result may occasionally be negative in a patient with acute severe immune-mediated hemolysis if the antigen–antibody complexes are cleared from the circulation before the test sample is obtained.

Delayed hemolysis, occurring days to a month after transfusion, is less evident than an acute hemolytic transfusion reaction, since the temporal relationship to transfusion is often overlooked. New-onset anemia, jaundice, elevated lactate dehydrogenase and bilirubin levels, a decreased haptoglobin level in a patient who has had prior transfusions or is a transplant recipient, or the likelihood of preformed but often evanescent antibodies due to pregnancy should prompt ad-



**Figure 5. DAT for the Diagnosis of Immune-Mediated Hemolytic Transfusion Reactions.**

A DAT (direct Coombs' test) is performed by mixing the patient's (transfused) red cells with Coombs' reagent (antihuman globulin) in vitro. If the transfused red cells are incompatible and coated in vivo with IgG or complement, the resulting agglutination after the addition of Coombs' reagent is defined as a positive reaction (Panel A). In the absence of IgG- or complement-coated red cells, no agglutination is seen after the addition of antihuman globulin (negative reaction) (Panel B). A false negative reaction may occur in cases in which hemolysis is brisk and short-lived and the resulting IgG- or complement-coated red cells are cleared from the circulation before testing. In the IAT (indirect Coombs' test), plasma (containing antibodies) from the patient with suspected immune-mediated hemolysis is mixed with donor red cells, followed by the addition of antihuman globulin. In vitro agglutination represents a positive IAT.

ditional evaluation for delayed hemolytic transfusion reactions. A direct or indirect antiglobulin test may be positive in the event of ongoing immune-mediated hemolysis. A peripheral-blood smear may reveal spherocytes and microspherocytes. Symptoms in patients with sickle cell disease merit a high level of suspicion lest a delayed hemolytic transfusion reaction go unrecognized, since signs and symptoms overlap with those of vaso-occlusive crises<sup>52</sup> and the results of serologic tests for alloimmunization are often delayed. The distinction between delayed hemolytic transfusion reactions and vaso-occlusive crises is important, since further transfusion can result in life-threatening hemolysis in cases of delayed hemolytic transfusion reactions. In patients with a delayed hemolytic transfusion reaction, serial electrophoretic analyses of hemoglobin may indicate the degree of destruction of transfused red cells, as measured by the asymmetric decline in levels of hemoglobin A as compared with hemoglobin S.<sup>3</sup> Classic laboratory findings are shown in Figure 3.

## MANAGEMENT

Clinically significant acute hemolytic transfusion reactions often occur in situations in which clinicians are unfamiliar with these high-risk incidents. Once an immune-mediated acute hemolytic transfusion reaction has been recognized, management is mainly supportive. Prompt interruption of the transfusion, saving of the remaining blood in the unit for testing, early blood and urine sampling to establish baseline values, and a thorough clerical check to interdict a possible second misidentified transfusion are crucial initial steps. Management must occur in an intensive care unit, along with a renal consultation, since dialysis may be required. Vigorous hydration with isotonic saline to maintain urine output at a rate above 0.5 to 1 ml per kilogram of body weight per hour is recommended to minimize the effects of free heme-mediated renal and vascular injury. The common practice of mannitol administration is not evidence based and should be used cautiously, if at all, in patients with ane-

mia and limited cardiac reserve. Supplemental diuretics (a 40-mg intravenous bolus of furosemide, followed by a continuous infusion at a dose of 10 to 40 mg per hour in the absence of hypotension) are helpful in such cases. Forced alkaline diuresis may be helpful. Sodium bicarbonate (130 mmol per liter in 5% dextrose or water) is administered through a separate intravenous line at a starting rate of 200 ml per hour to achieve a urinary pH of more than 6.5. The infusion is discontinued if either the arterial pH exceeds 7.5 or the urinary pH fails to increase after 2 to 3 hours. Electrolyte abnormalities such as hyperkalemia are common and warrant swift correction. In the event of hypotension, pressor support with a dopamine infusion (2 to 10  $\mu$ g per kilogram per minute) is commonly used. In patients with disseminated intravascular coagulation and severe bleeding, platelets, fresh-frozen plasma, and cryoprecipitate infusions may be required to maintain a platelet count of more than 20,000 per cubic millimeter, an international normalized ratio of less than 2.0, and a fibrinogen level of more than 100 mg per deciliter, respectively.

No evidence supports the routine use of therapeutic high-dose glucocorticoids, intravenous immune globulin, or plasma exchange. However, when transfusion of incompatible units is necessary, prophylaxis with glucocorticoids (hydrocortisone at a dose of 100 mg, administered just before transfusion and repeated 24 hours later) and intravenous immune globulin (1.2 to 2.0 g per kilogram, administered over a period of 2 to 3 days, with the first dose given just before the incompatible transfusion) has been used.<sup>53</sup> Acute hemolytic reactions to transfusion of incompatible units, although frightening and potentially lethal, are self-limited in most instances.

The most important aspect of management is prevention. Rates of acute hemolytic transfusion reactions due to erroneous patient identification and specimen collection or labeling have decreased significantly as hospitals have instituted relatively inexpensive, safe, and efficient preventive strategies.<sup>54,55</sup> Errors due to mislabeling of samples have been reduced by “zero tolerance” policies for accepting blood samples without core identifiers (i.e., full name of recipient, date of birth, and a unique identification number) on electronically generated labels and identification bands.<sup>56</sup> Repeating ABO checks is an additional method to prevent acute hemolytic transfusion reactions due

to sample-collection errors and is required by the College of American Pathologists and by the AABB (the American Association of Blood Banks).<sup>55</sup> Centralized transfusion databases help track blood types and transfusion requirements and can identify errors involving the wrong blood in the tube.<sup>56</sup>

Delayed hemolytic transfusion reactions are often clinically silent and are revealed by a positive antibody screen alone on routine laboratory testing. These episodes do not require intervention but must always be reported to the transfusion facility in order to reduce the risk of reactions to future transfusions. For patients receiving multiple transfusions who are at risk for more serious delayed hemolytic transfusion reactions (especially patients with hemoglobinopathies), phenotypically matched red-cell transfusions or units that are negative for antigens known to be immunogenic and clinically significant, such as those in the Rh system, are desirable.<sup>52</sup>

Guidelines for the extent of matching for minor red-cell antigens to prevent delayed hemolytic transfusion reactions have been published.<sup>57</sup> Identification of and tailored transfusions for recipients at particular risk, such as those of African ancestry with a high prevalence of partial Rh system antigens, are prudent measures to prevent alloimmunization. To optimize alloantibody detection, antibody testing should be repeated after transfusion, preferably 1 to 3 months later.<sup>53</sup> The integration of mass-scale red-cell genotyping into the blood supply chain has enabled timely provision of antigen-negative red-cell units beyond ABO and Rh types.<sup>58-60</sup> Red-cell genotyping may be of particular value in certain instances (e.g., for patients with multiple myeloma and anemia who receive daratumumab, an anti-CD38 monoclonal antibody known to interfere with and delay serologic testing and transfusion support).<sup>61</sup> Among patients who are already heavily alloimmunized and require long-term transfusion support, prophylactic rituximab (one or two 1000-mg doses administered intravenously, 2 weeks apart in the case of two doses, along with 10 mg of intravenous methylprednisolone, with the last dose given 10 to 30 days before transplantation) has been used with some success.<sup>62</sup>

For patients with sickle cell disease who have hyperhemolysis,<sup>3</sup> directed treatment strategies have included immune modulators such as glucocorticoids, intravenous immune globulin, and rituximab,<sup>62</sup> as well as erythropoiesis-stim-

ulating agents, since the endogenous erythropoietic response may be inadequate or delayed.<sup>63</sup> Blood transfusions are generally avoided, except in patients with profound anemia and symptoms of hypoperfusion.<sup>64</sup> Plasma exchange,<sup>32,65</sup> hemoglobin-based red-cell substitutes,<sup>66</sup> eculizumab,<sup>67</sup> and tocilizumab, an anti-interleukin-6 monoclonal antibody,<sup>68</sup> have been used in instances of life-threatening hemolysis but are of uncertain benefit. Newer agents such as haptoglobin and hemopexin concentrates are being explored as free heme scavengers in preclinical models.<sup>69,70</sup>

Strategies for preventing passenger lymphocyte syndrome include transfusion of red cells and plasma products compatible with donor and recipient blood types in the pretransplantation period. A recipient with blood group A receiving a transplant from a group O donor should receive group O red cells and group AB plasma.<sup>6</sup> Severe cases have been managed with prophylactic plasma reduction in the donor graft, partial red-cell exchange in the recipient before transplantation, or both. However, the results of these interventions are equivocal.<sup>71,72</sup> Red-cell aplasia due to immune-mediated lysis of donor red-cell precursors by recipient isohemagglutinins has been managed with transfusions, plasma exchange, rapid discontinuation of cyclosporine, donor lymphocyte infusions, erythropoietin, azathioprine, and rituximab with some success.<sup>73</sup> Recently, daratumumab, a human IgG1 $\kappa$  monoclonal antibody targeting CD38 (expressed at high levels on antibody-secreting plasma cells), was successfully used in a case of treatment-refractory pure red-cell aplasia after ABO-mismatched allogeneic stem-cell transplantation.<sup>74</sup>

ABO- or Rh-incompatible platelet transfusions due to passive antibody transfer may be mitigated by a plasma-matching donor inventory or by

screening donors for high-titer antirecipient antibodies and avoiding transfusion of units from such donors. In addition, plasma reduction or resuspension of platelets in platelet additive solutions may mitigate hemolytic transfusion reactions.<sup>31</sup> Hemolytic reactions to intravenous immune globulin are also managed supportively. Reactions should be reported to regulatory agencies expeditiously, and quarantine of batches with high-titer anti-A or anti-B antibodies should be considered. Key points in the diagnosis and management of acute hemolytic transfusion reactions and delayed hemolytic transfusion reactions are summarized in Figure 3.

## SUMMARY

Hemolytic transfusion reactions are recognized as an important cause of transfusion-associated reactions and may be subclinical, mild, or lethal. Acute, immune-incompatible reactions to ABO-mismatched transfusions have declined dramatically with the introduction of electronic verification systems. Other reactions, including delayed hemolytic transfusion reactions, hyperhemolysis, and passenger lymphocyte syndrome in transplant recipients, pose diagnostic and therapeutic challenges. Preventive strategies have been effective in reducing hemolysis-associated morbidity and mortality in all categories of hemolytic transfusion reactions. Established, systematic protocols for quickly identifying and responding to suspected reactions, as well as reporting them, remain the cornerstone of timely management of hemolytic transfusion reactions.

No potential conflict of interest relevant to this article was reported.

Disclosure forms provided by the authors are available with the full text of this article at NEJM.org.

## REFERENCES

- Hendrickson JE, Roubinian NH, Chowdhury D, et al. Incidence of transfusion reactions: a multicenter study utilizing systematic active surveillance and expert adjudication. *Transfusion* 2016;56:2587-96.
- Fiorellino J, Elahie AL, Warkentin TE. Acute haemolysis, DIC and renal failure after transfusion of uncross-matched blood during trauma resuscitation: illustrative case and literature review. *Transfus Med* 2018 February 19 (Epub ahead of print).
- Pirenne F, Yazdanbakhsh K. How I safely transfuse patients with sickle-cell disease and manage delayed hemolytic transfusion reactions. *Blood* 2018;131:2773-81.
- ElAnsary M, Hanna MO, Saadi G, et al. Passenger lymphocyte syndrome in ABO and Rhesus D minor mismatched liver and kidney transplantation: a prospective analysis. *Hum Immunol* 2015;76:447-52.
- Petz LD. Immune hemolysis associated with transplantation. *Semin Hematol* 2005;42:145-55.
- Yazer MH, Triulzi DJ. Immune hemolysis following ABO-mismatched stem cell or solid organ transplantation. *Curr Opin Hematol* 2007;14:664-70.
- Myhre BA. The first recorded blood transfusions: 1656 to 1668. *Transfusion* 1990;30:358-62.
- Ottenberg R. Studies in isoagglutination: I. Transfusion and the question of intravascular agglutination. *J Exp Med* 1911;13:425-38.
- Transfusion/donation fatalities. Silver Spring, MD: Food and Drug Administration (<https://www.fda.gov/vaccines-blood-biologics/report-problem-center>)

- biologics-evaluation-research/transfusiondonation-fatalities).
10. Sazama K. Reports of 355 transfusion-associated deaths: 1976 through 1985. *Transfusion* 1990;30:583-90.
  11. Carson JL, Guyatt G, Heddle NM, et al. Clinical practice guidelines from the AABB: red blood cell transfusion thresholds and storage. *JAMA* 2016;316:2025-35.
  12. Bolton-Maggs PHB. Serious hazards of transfusion — conference report: celebration of 20 years of UK haemovigilance. *Transfus Med* 2017;27:393-400.
  13. AuBuchon JP, Kruskall MS. Transfusion safety: realigning efforts with risks. *Transfusion* 1997;37:1211-6.
  14. Dzik WH. Emily Cooley Lecture 2002: transfusion safety in the hospital. *Transfusion* 2003;43:1190-9.
  15. Prosser AC, Kallies A, Lucas M. Tissue-resident lymphocytes in solid organ transplantation: innocent passengers or the key to organ transplant survival? *Transplantation* 2018;102:378-86.
  16. Garratty G. What do we mean by “hyperhaemolysis” and what is the cause? *Transfus Med* 2012;22:77-9.
  17. Politis C, Wiersum JC, Richardson C, et al. The International Haemovigilance Network database for the surveillance of adverse reactions and events in donors and recipients of blood components: technical issues and results. *Vox Sang* 2016;111:409-17.
  18. Delaney M, Wendel S, Bercovitz RS, et al. Transfusion reactions: prevention, diagnosis, and treatment. *Lancet* 2016;388:2825-36.
  19. Simmons DP, Savage WJ. Hemolysis from ABO incompatibility. *Hematol Oncol Clin North Am* 2015;29:429-43.
  20. Davenport RD. Pathophysiology of hemolytic transfusion reactions. *Semin Hematol* 2005;42:165-8.
  21. Garratty G. The James Blundell Award Lecture 2007: do we really understand immune red cell destruction? *Transfus Med* 2008;18:321-34.
  22. Zimring JC, Spitalnik SL. Pathobiology of transfusion reactions. *Annu Rev Pathol* 2015;10:83-110.
  23. Guarda CCD, Santiago RP, Fiuza LM, et al. Heme-mediated cell activation: the inflammatory puzzle of sickle cell anemia. *Expert Rev Hematol* 2017;10:533-41.
  24. Win N. Hyperhemolysis syndrome in sickle cell disease. *Expert Rev Hematol* 2009;2:111-5.
  25. Petz LD. Bystander immune cytotoxicity. *Transfus Med Rev* 2006;20:110-40.
  26. Gniadek TJ, McGonigle AM, Shirey RS, et al. A rare, potentially life-threatening presentation of passenger lymphocyte syndrome. *Transfusion* 2017;57:1262-6.
  27. Foell D, Glasmeyer S, Senninger N, Wolters H, Palmes D, Bahde R. Successful management of passenger lymphocyte syndrome in an ABO-compatible, non-identical isolated bowel transplant: a case report and review of the literature. *Transfusion* 2017;57:1396-400.
  28. Worel N. ABO-mismatched allogeneic hematopoietic stem cell transplantation. *Transfus Med Hemother* 2016;43:3-12.
  29. Bolan CD, Childs RW, Procter JL, Barrett AJ, Leitman SF. Massive immune haemolysis after allogeneic peripheral blood stem cell transplantation with minor ABO incompatibility. *Br J Haematol* 2001;112:787-95.
  30. Vaezi M, Oulad Dameshghi D, Souiri M, Setarehdan SA, Alimoghaddam K, Ghavamzadeh A. ABO incompatibility and hematopoietic stem cell transplantation outcomes. *Int J Hematol Oncol Stem Cell Res* 2017;11:139-47.
  31. Fontaine MJ, Mills AM, Weiss S, Hong WJ, Viele M, Goodnough LT. How we treat: risk mitigation for ABO-incompatible plasma in plateletpheresis products. *Transfusion* 2012;52:2081-5.
  32. Josephson CD, Castillejo MI, Grima K, Hillyer CD. ABO-mismatched platelet transfusions: strategies to mitigate patient exposure to naturally occurring hemolytic antibodies. *Transfus Apher Sci* 2010;42:83-8.
  33. Fung MK, Eder AF, Spitalnik SL, Westhoff CM, eds. Technical manual. 19th ed. Bethesda, MD: AABB Press, 2017.
  34. Berséus O, Boman K, Nessen SC, Westerberg LA. Risks of hemolysis due to anti-A and anti-B caused by the transfusion of blood or blood components containing ABO-incompatible plasma. *Transfusion* 2013;53:Suppl 1:114S-123S.
  35. Branch DR, Hellberg Å, Bruggeman CW, et al. ABO zygosity, but not secretor or Fc receptor status, is a significant risk factor for IVIG-associated hemolysis. *Blood* 2018;131:830-5.
  36. Scott DE, Epstein JS. Hemolytic adverse events with immune globulin products: product factors and patient risks. *Transfusion* 2015;55:Suppl 2:S2-S5.
  37. Flegel WA. Pathogenesis and mechanisms of antibody-mediated hemolysis. *Transfusion* 2015;55:Suppl 2:S47-S58.
  38. Padmore R. Possible mechanisms for intravenous immunoglobulin-associated hemolysis: clues obtained from review of clinical case reports. *Transfusion* 2015;55:Suppl 2:S59-S64.
  39. Gaines AR. Disseminated intravascular coagulation associated with acute hemoglobinemia or hemoglobinuria following Rh(0)(D) immune globulin intravenous administration for immune thrombocytopenic purpura. *Blood* 2005;106:1532-7.
  40. Berg R, Shebl A, Kimber MC, Abraham M, Schreiber GB. Hemolytic events associated with intravenous immune globulin therapy: a qualitative analysis of 263 cases reported to four manufacturers between 2003 and 2012. *Transfusion* 2015;55:Suppl 2:S36-S46.
  41. Decesare WR, Bove JR, Ebaugh FG Jr. The mechanism of the effect of iso- and hyperosmolar dextrose-saline solutions on *in vivo* survival of human erythrocytes. *Transfusion* 1964;4:237-50.
  42. McCullough J, Polesky HF, Nelson C, Hoff T. Iatrogenic hemolysis: a complication of blood warmed by a microwave device. *Anesth Analg* 1972;51:102-6.
  43. Lanore JJ, Quarré MC, Audibert G, et al. Acute renal failure following transfusion of accidentally frozen autologous red blood cells. *Vox Sang* 1989;56:293.
  44. MacDonald WB, Berg RB. Hemolysis of transfused cells during use of the injection (push) technique for blood transfusion. *Pediatrics* 1959;23:8-11.
  45. Ma SK, Wong KF, Siu L. Hemoglobinemia and hemoglobinuria complicating concomitant use of a white cell filter and a pressure infusion device. *Transfusion* 1995;35:180.
  46. Liebman HA, Weitz IC. Autoimmune hemolytic anemia. *Med Clin North Am* 2017;101:351-9.
  47. Garratty G. Immune hemolytic anemia caused by drugs. *Expert Opin Drug Saf* 2012;11:635-42.
  48. Mayer B, Bartolmäs T, Yürek S, Salama A. Variability of findings in drug-induced immune haemolytic anaemia: experience over 20 years in a single centre. *Transfus Med Hemother* 2015;42:333-9.
  49. Felix CA, Davey RJ. Massive acute hemolysis secondary to Clostridium perfringens sepsis in a recently transfused oncology patient with multiple alloantibodies. *Med Pediatr Oncol* 1987;15:42-4.
  50. Renzaho AM, Husser E, Polonsky M. Should blood donors be routinely screened for glucose-6-phosphate dehydrogenase deficiency? A systematic review of clinical studies focusing on patients transfused with glucose-6-phosphate dehydrogenase-deficient red cells. *Transfus Med Rev* 2014;28:7-17.
  51. Sazama K, Klein HG, Davey RJ, Corash L. Intraoperative hemolysis: the initial manifestation of glucose-6-phosphate dehydrogenase deficiency. *Arch Intern Med* 1980;140:845-6.
  52. Diamond WJ, Brown FL Jr, Bitterman P, Klein HG, Davey RJ, Winslow RM. Delayed hemolytic transfusion reaction presenting as sickle-cell crisis. *Ann Intern Med* 1980;93:231-4.
  53. Win N, Needs M, Thornton N, Webster R, Chang C. Transfusions of least-incompatible blood with intravenous immunoglobulin plus steroids cover in two patients with rare antibody. *Transfusion* 2018;58:1626-30.
  54. Bolton-Maggs PH, Wood EM, Wiersum-Osselton JC. Wrong blood in tube — potential for serious outcomes: can it be prevented? *Br J Haematol* 2015;168:3-13.
  55. Standards for blood banks and transfusion services. 31st ed. Bethesda, MD: AABB Press, 2018.
  56. Delaney M, Dinwiddie S, Nester TN, Aubuchon JA. The immunohematologic

- and patient safety benefits of a centralized transfusion database. *Transfusion* 2013;53:771-6.
57. Narbey D, Habibi A, Chadebec P, et al. Incidence and predictive score for delayed hemolytic transfusion reaction in adult patients with sickle cell disease. *Am J Hematol* 2017;92:1340-8.
58. Flegel WA, Gottschall JL, Denomme GA. Integration of red cell genotyping into the blood supply chain: a population-based study. *Lancet Haematol* 2015;2(7):e282-e289.
59. Gehrie EA, Ness PM, Bloch EM, Kacker S, Tobian AAR. Medical and economic implications of strategies to prevent alloimmunization in sickle cell disease. *Transfusion* 2017;57:2267-76.
60. Fasano RM, Chou ST. Red blood cell antigen genotyping for sickle cell disease, thalassemia, and other transfusion complications. *Transfus Med Rev* 2016;30:197-201.
61. Lancman G, Arinsburg S, Jhang J, et al. Blood transfusion management for patients treated with anti-CD38 monoclonal antibodies. *Front Immunol* 2018;9:2616.
62. Noizat-Pirenne F, Habibi A, Mekontso-Dessap A, et al. The use of rituximab to prevent severe delayed haemolytic transfusion reaction in immunized patients with sickle cell disease. *Vox Sang* 2015;108:262-7.
63. Habibi A, Mekontso-Dessap A, Guillaud C, et al. Delayed hemolytic transfusion reaction in adult sickle-cell disease: presentations, outcomes, and treatments of 99 referral center episodes. *Am J Hematol* 2016;91:989-94.
64. Gardner K, Hoppe C, Mijovic A, Thein SL. How we treat delayed haemolytic transfusion reactions in patients with sickle cell disease. *Br J Haematol* 2015;170:745-56.
65. Uhlmann EJ, Shenoy S, Goodnough LT. Successful treatment of recurrent hyperhemolysis syndrome with immunosuppression and plasma-to-red blood cell exchange transfusion. *Transfusion* 2014;54:384-8.
66. Plastini T, Locantore-Ford P, Bergmann H. Sanguinate: a novel blood substitute product. *Blood* 2017;130:Suppl 1:1120. abstract.
67. Dumas G, Habibi A, Onimus T, et al. Eculizumab salvage therapy for delayed hemolysis transfusion reaction in sickle cell disease patients. *Blood* 2016;127:1062-4.
68. Shetty A, Hanson R, Korsten P, et al. Tocilizumab in the treatment of rheumatoid arthritis and beyond. *Drug Des Devel Ther* 2014;8:349-64.
69. Immenschuh S, Vijayan V, Janciauskiene S, Gueler F. Heme as a target for therapeutic interventions. *Front Pharmacol* 2017;8:146.
70. Schaer DJ, Buehler PW, Alayash AI, Belcher JD, Vercellotti GM. Hemolysis and free hemoglobin revisited: exploring hemoglobin and heme scavengers as a novel class of therapeutic proteins. *Blood* 2013;121:1276-84.
71. Cunard R, Marquez II, Ball ED, et al. Prophylactic red blood cell exchange for ABO-mismatched hematopoietic progenitor cell transplants. *Transfusion* 2014;54:1857-63.
72. Worel N, Greinix HT, Supper V, et al. Prophylactic red blood cell exchange for prevention of severe immune hemolysis in minor ABO-mismatched allogeneic peripheral blood progenitor cell transplantation after reduced-intensity conditioning. *Transfusion* 2007;47:1494-502.
73. Helbig G, Stella-Holowiecka B, Wojnar J, et al. Pure red-cell aplasia following major and bi-directional ABO-incompatible allogeneic stem-cell transplantation: recovery of donor-derived erythropoiesis after long-term treatment using different therapeutic strategies. *Ann Hematol* 2007;86:677-83.
74. Chapuy CI, Kaufman RM, Alyea EP, Connors JM. Daratumumab for delayed red-cell engraftment after allogeneic transplantation. *N Engl J Med* 2018;379:1846-50.

Copyright © 2019 Massachusetts Medical Society.

**TRACK THIS ARTICLE'S IMPACT AND REACH**

Visit the article page at [NEJM.org](https://www.nejm.org) and click on Metrics for a dashboard that logs views, citations, media references, and commentary.  
[www.nejm.org/about-nejm/article-metrics](https://www.nejm.org/about-nejm/article-metrics).