Hematopoietic Transplant-Associated Thrombotic Microangiopathy: Case Report and Review of Diagnosis and Treatments

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Abstract: Transplant-associated thrombotic microangiopathy (TA-TMA) refers to inflammatory and thrombotic diseases of the microvasculature characterized by hemolytic anemia, thrombocytopenia, and evidence of organ damage, particularly acute renal failure. This syndrome occurs in 10% to 20% of patients with allogeneic hematopoietic stem cell transplants (HSCTs). It is much less frequent in the autologous setting. TA-TMAs present diagnostic challenges because they may not clearly fall into one of the categories of the 2 major TMAs: atypical hemolytic uremic syndrome (aHUS) and thrombotic thrombocytopenic purpura (TTP). In addition, complications of the transplant itself, including infection, graft-versus-host disease, and disseminated intravascular coagulation, as well as the side effects of immunosuppressive drugs, can mimic a TMA. Because the pathophysiology of TA-TMA is poorly understood, current treatment options are suboptimal, and the condition carries a very high mortality rate. In 3 recent case summaries, the median acute response rate to plasma exchange was as high as 55%, but this therapy failed to alter underlying disease pathology and had little impact on overall mortality, which was approximately 80%. Indeed, the vast majority of TA-TMA patients lack suppression of ADAMTS13 (a disintegrin and metalloproteinase with a thrombospondin type 1 motif, member 13) activity to less than 5% to 10% of normal and do not have a complete response to plasma exchange, characteristics indicating that a TTP-like disorder is not involved. Recent advances in the treatment of aHUS may offer a therapeutic option in the aHUS-like TMAs associated with HSCTs. These issues are discussed in the context of a patient recently evaluated and treated at our institution; the case serves to illustrate the difficulties associated with the diagnosis and treatment of TA-TMA.

Case Report

The patient was a 33-year-old white man who had a history of a JAK2 (Janus kinase 2)–negative myeloproliferative disorder in childhood that had progressed to myelofibrosis with splenomegaly. The patient underwent a matched unrelated allogeneic hematopoietic stem cell transplant (HSCT) with fludarabine and busulfan (Myleran,

Aspen Global) conditioning at our institution. He had known anti–HLA-DP antibodies, which were treated with bortezomib (Velcade, Millennium Pharmaceuticals), intravenous immunoglobulin, and plasma exchange before transplant. He was started on tacrolimus after the transplant to prevent graft-versus-host disease (GVHD). Engraftment occurred by day 15 after transplantation, but the patient remained thrombocytopenic and required periodic platelet transfusions. His splenomegaly decreased slightly after the transplant, but 2 months later, he became anemic and transfusion-dependent without a clear cause, requiring weekly transfusions of packed red blood cells. On day 168 after transplantation, he developed fevers, jaundice, and worsening hemolysis, with an increased transfusion requirement. He had a persistent sinus tachycardia, but his blood pressure was not elevated. The results of an ultrasound examination of the liver were negative for vaso-occlusive disease, and the results of infectious studies were also negative. On day 172 after transplantation, he developed a pruritic rash on his lower extremities, and his platelet count was 20,000/mm^3. His hemoglobin level remained low, at 7.6 g/dL. The results of additional laboratory studies were consistent with hemolysis: indirect bilirubin, 8.7 mg/dL; lactate dehydrogenase (LDH), 439 IU/L (normal, 98-192 IU/L); and undetectable haptoglobin. Rare schistocytes were found on his peripheral blood smear. A direct Coombs test was negative. His creatinine level and estimated glomerular filtration rate remained normal. Urinalysis showed trace protein, with no red cells or casts.

The patient received red cell transfusions without an increase in his hemoglobin level, and his LDH peaked at 585 IU/L. He developed a rash consistent with GVHD on biopsy, and high-dose prednisone therapy was initiated. Because of a progressive Coombs-negative hemolysis, increasing transfusion requirements, and thrombocytopenia, transplant-associated thrombotic microangiopathy (TA-TMA) was diagnosed.

Tacrolimus was discontinued, ADAMTS13 (a disintegrin and metalloproteinase with a thrombospondin type 1 motif, member 13) studies were done, and daily plasma exchange was started. Some improvement in the hemoglobin level and reduction in hemolysis were noted, as judged by a decrease in the bilirubin and LDH levels, but the patient remained thrombocytopenic with increasing splenomegaly. His ADAMTS13 activity was normal (88%), and ADAMTS13 inhibitors, characteristic of a thrombotic thrombocytopenic purpura (TTP)–like TMA, were not detected. All of the 3 punch skin biopsy specimens that were obtained, 1 in an area of GVHD and 2 in regions of normal-appearing skin, revealed focal deposition of C3, C3d, C4d, and C5b–9 (membrane attack complex, or MAC) in small vessels at the dermal–epidermal junction (Figure).

The diagnosis of TTP was excluded based on the normal ADAMTS13 level and the failure to mount an adequate response to plasma exchange. A splenectomy was planned to palliate the patient's symptoms and possibly to reduce hemolysis and improve platelet recovery. The evidence of terminal complement component (MAC) deposition in his skin biopsy specimen was consistent with an atypical hemolytic uremic syndrome (aHUS)–like TMA. Given that surgery is a potent activator of the alternative complement pathway involved in aHUS pathology, anti-C5 therapy with eculizumab (Soliris, Alexion) was initiated just before surgery on day 187 after transplantation. The patient underwent splenectomy, distal pancreatectomy, and liver biopsy without complications. Pathology revealed significant extramedullary sites of hematopoiesis in the liver and spleen. In the spleen, 74% of the cells were of donor origin. Eculizumab was continued; weekly intravenous infusions of 900 mg were given for a total of 4 weeks, followed by 1200 mg every other week. Rapid and sustained improvement in the patient's platelet count was noted, as well as mitigation of his anemia and indirect hyperbilirubinemia. Currently, more than 1 year after transplant, the patient is doing well on minimal immunosuppression. He has had no further symptoms of TA-TMA or GVHD.

Overview of TA-TMA

Approximately 11,000 autologous and 7000 allogeneic HSCTs are performed annually in the United States.[1] TMA, defined by thrombocytopenia, microangiopathic hemolytic anemia, and evidence of organ (renal, central nervous system, gastrointestinal, pulmonary, cardiac, endocrine, and ophthalmic) dysfunction, is a well-recognized complication of HSCT, with significant mortality. TMAs appear to complicate 10% to 20% of stem cell transplants.[2-6] The clinical manifestations of TA-TMA are similar to those of other TMAs, such as TTP and aHUS. Common features include thrombocytopenia, a Coombs-negative hemolysis, the presence of schistocytes on peripheral blood smear, and acute renal failure or mental status changes, or both. However, in virtually all instances, TTP is not a part of the TA-TMA syndrome because ADAMTS13 activity is above 5% to 10% (the exact cutoff value is assay dependent) and autoantibody inhibitors of this von Willebrand factor–cleaving protease are not found.[7-9] However, true estimates of the rate at which TMAs occur in the HSCT setting are difficult to make because of inconsistencies in the diagnostic criteria applied and because of the overlap of clinical signs and laboratory abnormalities with post-transplant complications that mimic TA-TMAs.

Various causes of TA-TMA have been postulated, including endothelial cell damage resulting from the chemo-

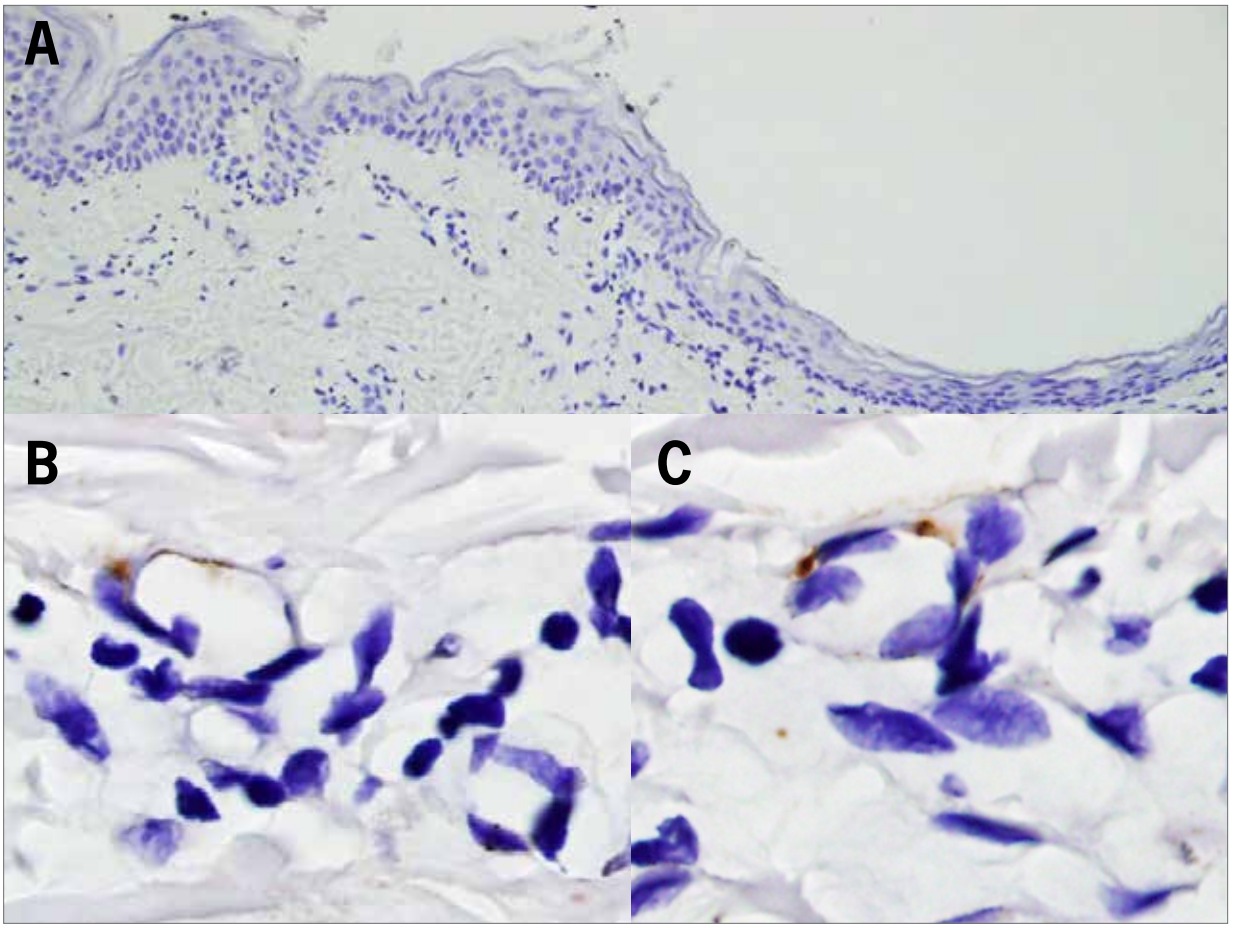

Figure. Immunohistochemical staining of the epidermis did not reveal the terminal complement components C5b-9 (MAC) (A), but strong C5b-9 deposition was detectable along the small vessels of the superficial dermis (B, C; magnification 100 ×).

MAC, membrane attack complex.

therapy and radiation used in conditioning regimens, side effects of immunosuppressive medications, viral infections, and GVHD.[2] In general, these factors are of little help in deciding upon a therapy. Distinct pathologic features, including the nature of the microvascular thrombus (primarily platelets, or a "white clot," vs fibrin, or a "red clot") and the nature of the endothelial cell damage (relatively bland apoptotic injury vs apoptosis and necrosis with an inflammatory component), have been documented for decades in different TMAs in general, and in TA-TMA specifically.[10-13] However, until recently, the absence of a specific therapy for aHUS rendered such pathologic distinctions largely academic, as everyone was given a trial of plasma exchange.

It is critical to remember that classic TTP and aHUS are characterized by thrombi in the microvasculature and in renal arterioles. However, fibrin-rich (red) clots with an inflammatory component and C5b–9 deposition, a result of the inability to control the alternative complement pathway when it is super-activated, are most characteristic of an aHUS lesion, whereas von Willebrand factor deposition in platelet-rich (white) clots in the absence of inflammation is typical of TTP.[11-13] (For unclear reasons, microthrombi are virtually never found in the lungs in TTP, whereas the lungs are involved in some 30% of cases in aHUS.[11]) In terms of TA-TMA pathology, although early autopsy series failed to recognize microthrombi,[2] more recently intrarenal and extrarenal microthrombi have been described at autopsy in patients with TA-TMA.[14,15] Based on the ADAMTS13 activity above 5% to 10% found in most patients with TA-TMA and, as in our patient reported here, complement staining in the microvasculature, the TMA lesions in most transplant cases appear to represent a form of aHUS, not TTP.

These observations also call into question the involvement of transplant-associated medications in TMAs occurring in the allogeneic HSCT setting. Some of these agents may directly damage endothelium, activate complement, and alter ADAMTS13 activity and/or secretion. For example, calcineurin inhibitors, such as cyclosporine and sirolimus, have been implicated in TA-TMAs.[16] Cyclosporine

may suppress ADAMTS13 through direct endothelial toxicity, with the release of von Willebrand factor multimers that can form complexes with ADAMTS13, decreasing plasma levels of the enzyme. Cyclosporine can also inhibit ADAMTS13 secretion.[17] However, these effects do not appear to reduce ADAMTS13 activity to TTP levels (ie, <5%-10%) in vivo, nor do they induce ADAMTS13 inhibitors. The fact that most TA-TMAs do not show a complete (ie, hematologic and organ) response to plasma exchange is another indication that any such drug-induced alterations in ADAMTS13 are unlikely to be of clinical relevance,[17] even though they may set up a scenario for thrombus formation on an injured endothelium, unrelated to control of ADAMTS13 or complement, and are often responsive to drug discontinuation. The serum levels of cyclosporine are not predictive of TA-TMA development or of its severity.[3,16,18]

TA-TMA is not a variant of acute GVHD, an immunologic perturbation that occurs in allogeneic HSCT as a result of host–donor mismatch. The predominant pathology in GVHD results from a T-cell–mediated attack on donor somatic cells. However, GVHD is an independent risk factor for TA-TMA, and both conditions have been associated with involvement of the gastrointestinal tract, causing symptoms that include diarrhea, which can be bloody.[19,20] In contrast to TA-TMAs, which occur a median of 150 days (mean 90 days) after transplant, acute GVHD tends to occur much earlier, before day 100.[2] Finally, although much less commonly than in the allogeneic setting, TA-TMA has been reported in autologous HSCT, in which GVHD does not occur.[14,21,22]

Risk Factors

The most significant predictors of the development of TMA in the HSCT setting are advanced age, GVHD, the use of preconditioning radiation, and female sex.[2,3,18] Calcineurin inhibitors and sirolimus are also risk factors, with mechanisms that may be multifactorial.[4,23-25] Their effect may be a consequence of their ability to damage endothelium directly, as previously described, as well as to activate the alternative complement pathway in an individual with a genetically based inability to control that system. The association of TA-TMAs with autologous transplants, although infrequent, makes it harder to implicate conditioning regimens, calcineurin inhibitors, and GVHD as general risks for the development of TA-TMA because these are neither components nor consequences of an autologous transplant.

Other transplant-related parameters have also been evaluated in terms of increasing risk for TA-TMA. The extent of HLA mismatch is one factor.[26] In some cases, specific conditioning regimens and intensities of conditioning appear to be predictive of TA-TMA.[5] Corticosteroid exposure is not predictive of TA-TMA. Higher grades of acute GVHD are associated with TA-TMA, although chronic GVHD is not.[18] Finally, in one series, no increase in TA-TMA was noted at the time of second transplant, suggesting that TA-TMA may occur with equal frequency after a first or after additional transplants, and that the risk does not diminish with further immunologic challenge. Stem cell source is not a factor; the incidence rates of TA-TMA are similar with peripherally mobilized and with bone marrow–derived cells.[27]

Infectious agents have been proposed as risk factors, including *Aspergillus*, cytomegalovirus, and adenovirus.[2,6,18,23,28] Infections occur with high frequency in the allogeneic HSCT setting. No direct pathologic mechanism has been proposed for infection-mediated TA-TMAs. However, infections are a major activator of the alternative complement pathway, and the inability to control this pathway is the underlying pathophysiology of aHUS, as is discussed later.[11,29-32]

Diagnosis

TA-TMA is difficult to diagnose because the principal manifestations of a TMA are common sequelae of the underlying diseases that are treated with transplants, transplant conditioning regimens, and opportunistic infections, particularly those caused by cytomegalovirus, Epstein-Barr virus, human herpesvirus 6, parvovirus, *Aspergillus*, and adenovirus. There are numerous reasons for schistocytes to be found in a patient with a transplant, including infection and underlying hematologic malignancies accompanied by low-grade disseminated intravascular coagulation (DIC). Bilirubin levels are also elevated in GVHD and vaso-occlusive disease of the liver. One consistent finding in TA-TMA is LDH elevation, which is also a key characteristic of classic TTP and aHUS.[33,34] LDH levels have consistently been elevated over baseline, and Fuge and colleagues reported that most patients in their series had an LDH level that was increased 3-fold or more over normal.[18] Isoenzyme analysis reveals that the source of LDH elevation includes not only red cell lysis but also tissue damage secondary to ischemia related to thrombosis and endothelial injury.[33]

One study attempted to categorize TMAs occurring in the HSCT setting based on exposures and pathology. Four TA-TMA categories were proposed: multifactorial fulminant TMA, conditioning-associated aHUS, cyclosporine-associated neurotoxicity with microangiopathic hemolytic anemia, and cyclosporine-associated nephrotoxicity with microangiopathic hemolytic anemia.[35] However, this classification system falls short of standardizing TMA as a diagnosis because it was developed before

Table. Definition of Thrombotic Microangiopathy Occurring in Hematopoietic Stem Cell Transplants: Comparison of 3 Different Sets of Criteria

Parameter	LeukemiaNet International Working Group[36]	Blood and Marrow Transplant Clinical Trials Network[28]	Overall Thrombotic Microangiopathy (O-TMA) Grouping[34]
Schistocytes	>4%	>2 per high-power field	>2 per high-power field
Platelet count	<50,000/mm^3 or <50% of normal baseline	NS	<50,000/mm^3 or <50% of normal baseline
Lactate dehydrogenase	Increased	Increased	Increased
Haptoglobin	Decreased	NS	Decreased
Transfusions	Increased	NS	NS
Creatinine	NS	2 × baseline	NS
Direct Coombs test	NS	Negative	Negative
Coagulation studies	Normal	Normal	Normal

NS, not specified.

the widespread use of ADAMTS13 testing and the availability of anti-C5 monoclonal antibody therapy.

Several professional societies have also attempted to create criteria for diagnosing TA-TMAs (Table). The European Group for Blood and Bone Marrow Transplantation and the European LeukemiaNet International Working Group proposed a definition based on expert consensus. Five criteria must be met for the diagnosis of TA-TMA: more than 4% schistocytes in peripheral blood; de novo prolonged or progressive thrombocytopenia (platelet count <50,000/mm^3 or 50% reduction from baseline level); sudden and persistent increase in LDH; decrease in serum haptoglobin; and decreased hemoglobin concentration or increased transfusion requirement.[36] (It should be recalled, however, that because haptoglobin is an acute phase reactant, the serum haptoglobin level may be an unreliable marker of the presence or extent of hemolysis.)

A second set of criteria was created by the Blood and Marrow Transplant Clinical Trials Network (see Table)[28]: more than 2 schistocytes per high-power field, LDH level above baseline, renal dysfunction (assessed by a doubling of the creatinine level from baseline before hydration or the institution of a conditioning regimen) or neurologic symptoms, and a negative direct Coombs test. This group did not include the platelet count, recognizing that thrombocytopenia is common in all HSCTs. Neither of these scoring systems has been prospectively analyzed in validation cohorts. A third system, proposed by Cho and colleagues[34] (see Table), includes the category of "probable TMA" to address the issue of a lack of some defining characteristics at the time of initial diagnosis.

In terms of organ system manifestations, renal dysfunction is not consistently part of the clinical presentation of a TA-TMA, and reports vary on the actual prevalence of neurologic involvement in patients with TA-TMA.[6,34] The absence of nephropathy seems to predict a good outcome and improved overall survival.[19,27,34] However, the assessment of renal failure by the creatinine level alone is of limited utility in TA-TMA. Even in classic acute aHUS, 20% of individuals have a normal serum creatinine level,[11] and the creatinine level may not correspond to the histologic findings on the kidney biopsy specimen.[3] Other tests to detect potential renal dysfunction, such as urinalysis for protein, red cells, and casts, and blood pressure measurement for hypertension, should also be done.[18]

Schistocytes may not be identifiable in all TA-TMAs, and in any event, they are a nonspecific finding in this population unless followed serially. The recommendations of some transplant societies, requiring at least 4% schistocytes, may specify too high a clinical cutoff.[34,37]

Additional questions remain regarding these scoring systems. At least one system recommended excluding DIC before a diagnosis of a TMA could be entertained. We agree that it is difficult to diagnose a TMA in the setting of active DIC. A direct Coombs test should yield a negative result before TA-TMA is diagnosed, but in this heavily transfused population, Coombs positivity may be a false reassurance. In our opinion, the greatest omission is that no professional group or society has recommended that ADAMTS13 testing be required in order to exclude, in the vast majority of instances, TTP as defined by an ADAMTS13 activity level of less than 5% to 10%. This is the case even though most series suggest that TTP rarely occurs in the transplant setting. This problem has been addressed by a recent study of 39 children and young adults undergoing HSCT who had a TMA and ADAMTS13 activity greater than 5%.[38] Elevated LDH levels, hypertension, and proteinuria on routine urinalysis were the earliest markers of a TMA, and elevated serum C5b-9 levels with proteinuria were associated with very poor survival (<20% at 1 year). The authors concluded

that terminal complement activation represents a likely therapeutic target in TA-TMA patients with such high-risk features, and we concur.

Complement and complement regulatory protein mutation analyses were not considered in these schemes. Such findings could be useful in documenting aHUS-like TMA occurring in the transplant setting because most adult cases of aHUS have been linked to congenital mutations of one or more complement-related proteins.[11] However, we do not recommend their routine use. Such tests are expensive, results may take weeks, and available commercial platforms fail to identify known mutations in classic aHUS at least 30% of the time.[11] We have reported that tissue biopsy to assess complement deposition in the microvasculature, along with mutational analysis, can be useful in difficult diagnostic situations in which TTP has been defined by ADAMTS13 testing but the patient still does not respond by both hematologic and clinical criteria to plasma exchange and immune-suppressive regimens.[39-41]

Classic Therapies

Until recently, a diagnosis of TA-TMA—which is usually made within the first 3 months after an allogeneic HSCT (range, 32-733 days)[2,19]—was associated with increased mortality in more than two-thirds of patients. The 3-year survival rate was a dismal 11%.[16] In addition to being difficult to diagnose, TA-TMA is difficult to treat, and future therapy-directed research should be a priority. Plasma exchange has been a standard, effective treatment for TTP since 1991,[42] but it has also been widely used for any form of TMA, including TA-TMA, for decades. Although the efficacy of plasma exchange and immune suppression has been clearly established in TTP, plasma exchange has no long-term role in aHUS[43] or in aHUS-like TMAs occurring in situations that can unmask a patient's inability to control complement activation, such as the transplant setting.[44]

Eculizumab, the humanized monoclonal antibody against complement protein C5, is a therapy for aHUS approved by the US Food and Drug Administration. It has been available only since September 2011 for this indication,[43] and it was therefore logical that common clinical practice would dictate a trial of plasma exchange for TA-TMA before that date. But, as previously noted, there is little evidence that it is effective beyond perhaps a temporary, acute response, and a large number of reports and meta-analyses suggest that it is not effective in the long term.[2,22,44-46] In addition, the risks associated with plasma exchange are not trivial; they include vascular catheter infections, alloimmunization, and life-threatening infusion reactions.[47] Plasma exchange had no clinical benefit for the patient in our case report, nor has our experience with other such patients persuaded us to think differently.

Defining the "response" to plasma exchange in aHUS, or in TA-TMA resembling aHUS clinically and pathologically, is important. Up to 80% of patients who have classic, complement mutation–documented aHUS may improve by hematologic criteria—showing complete responses in terms of platelet count and hemoglobin and haptoglobin levels, and significant partial responses in LDH levels—with plasma exchange alone. There are sufficient levels of soluble complement regulatory proteins in fresh frozen plasma to effect such short-term remissions. However, ongoing tissue damage and mortality are not altered.[18,19,34] This was highlighted by a recent review summarizing 260 TA-TMA patients who had a median "response rate" to plasma exchange of 37% to 55%, but an 80% overall mortality.[45] Nine of 10 consecutive pediatric TA-TMA patients followed by one group showed normalization of hematologic parameters following plasma exchange, but only 5 recovered and survived, for an overall mortality of 50%.[46]

Elevations of inflammatory cytokines and thrombotic markers were reported in patients with TA-TMA at presentation, and reductions were noted after a few sessions of plasma exchange.[48] However, all of these patients died within a year after transplant, 2 deaths occurring within 130 days.[48] TA-TMA therefore should not be expected to respond significantly to plasma exchange unless an ADAMTS13-deficient state with activity of less than 5% to 10% is present, instances of which appear to be quite rare.

As for potentially drug-induced cases of TA-TMA, when calcineurin inhibitors are involved, they should be discontinued or the dose reduced. This will lead to improvement in some patients. Response rates of 63% have been observed when these drugs are withheld, which is better than the rates of response to plasma exchange.[49] However, this strategy may also increase the risk of transplant rejection and inadequately control GVHD.

Anti-CD20 treatment with rituximab (Rituxan, Genentech/Biogen Idec) can impact TTP refractory to plasma exchange,[50-53] and it has been used in those few patients with TA-TMA who had TTP as defined by ADAMTS13 deficiency.[51] However, like plasma exchange, rituximab is of limited or no clinical value in the vast majority of patients with TA-TMA, who have ADAMTS13 activity levels above 5% to 10%. Other immunosuppressive strategies have also been tried, including anti–interleukin 2 monoclonal antibody.[54] It is conceivable that immunosuppressive strategies could have an effect in aHUS-like TA-TMA associated with complement factor H autoantibodies, although this intervention has been attempted in only a small number of individuals in the pediatric setting, with limited follow-up.[55] It is discussed in greater detail below. The anticoagulant defibrotide, which has been approved for use in Europe, has been used with some response in patients having mild manifestations of disease.[56]

Complement Dysregulation and the Use of Anti-C5 Therapy

TTP is an acquired disorder. It is characterized by a deficiency of the von Willebrand factor–cleaving protease ADAMTS13 that is caused by an immunoglobulin G autoantibody. This deficiency results in an accumulation of ultralarge von Willebrand factor multimers tethering platelets, with uncontrolled clotting. More than 90% of patients with TTP have a complete response to plasma exchange.[42] By contrast, a congenital inability to control the alternative complement system drives the pathophysiology of aHUS, and the humanized anti-C5 monoclonal antibody eculizumab has drastically altered the morbidity and mortality associated with this disorder.[43,57] Plasma exchange has no role in the long-term management of aHUS. As previously noted, in the vast majority of patients, an ADAMTS13 activity level of less than 5% to 10% defines TTP, with only rare patients who have TTP defined by their ADAMTS13 activity level failing to respond to plasma exchange and having complement mutations characteristic of aHUS.[39,40]

Complement activation has also been reported in TTP, although its pathophysiologic significance here is uncertain. Specifically, elevated circulating levels of terminal complement components C5a and MAC occur in acute aHUS and TTP.[58-60] Although one recent report suggests that MAC levels are significantly greater in aHUS than in TTP,[60] the utility of this assay to distinguish between the 2 conditions, arising spontaneously or following TA-TMA, requires further study.

Given that the vast majority of patients with TA-TMA who are tested have ADAMTS13 activity levels above 5% to 10%, it is reasonable to hypothesize that complement mutations characteristic of classic aHUS would also be found in patients with TA-TMA. Complement activation, including C4d and C5b–9 deposition in tissues, has been demonstrated in the renal arterioles and other organs of patients with TA-TMA, as in those with classic aHUS, in reports dating from 1986.[11,61,62] Complement mutations have also been reported in TA-TMA. Jodele and colleagues described 6 pediatric patients in whom TA-TMA with acute renal failure developed in the allogeneic HSCT setting.[63] The majority were found to have deletions of complement factor H–related proteins (CFHRs) 1 and 3, and 3 of the 6 also had autoantibodies to CFHR. This pattern is similar to that seen in an autoimmune TMA known as DEAP-HUS (deficiency of CFHR plasma protein and factor H-HUS).[64] The patients of Jodele and colleagues had poor responses to plasma exchange and elevated circulating MAC levels, and most had thrombosis on biopsy. Of the 6 patients, 4 achieved therapeutic plasma levels of eculizumab and clinical responses to the drug.[63]

These patients had also previously responded to an antibody depletion strategy with rituximab.[14] This treatment was initiated because the authors had identified autoantibodies to complement factor H, the soluble complement regulatory factor most often involved in diagnoses of uncomplicated aHUS,[11] along with the complement mutations. Complement factor H autoantibodies were not detected in 18 children undergoing HSCT in whom TMA did not develop.

Jodele and colleagues proposed that the CH50, a hemolytic complement assay, might be used as a surrogate for adequate plasma levels of eculizumab, and that the dose might need to be increased if the CH50 level remained at or above 4%. Indeed, 2 of their patients with aHUS-like TA-TMA did not respond to eculizumab, failed to attain therapeutic plasma levels despite further dosing, and died. The patients required at least 4 to 6 weeks of treatment with eculizumab, but the duration of treatment remains uncertain.[63]

Discussion and Plans for Future Research

TA-TMA is a frequent and serious problem in patients with allogeneic HSCTs. At the time of this writing, there appears to be no clear strategy to prevent or mitigate the risk of TA-TMA. Our case represents the first published use of anti-complement therapy with eculizumab in an adult patient with TA-TMA. It illustrates the difficulties encountered in making the diagnosis, so that an appropriate treatment can be chosen. Better diagnostic standards are clearly needed.

We propose the following strategy for the evaluation of a patient with TA-TMA. Because the incidence is quite high, up to 10% to 20% in the setting of allogeneic HSCT, the transplant physician should keep the diagnosis in mind for any patient with the new onset of thrombocytopenia, hemolytic anemia, and renal failure. However, it is important to note that these TMAs can occur, if much less commonly, following an autologous HSCT. The risk for TA-TMA is increased in older patients, female patients, and those with acute grade 2 through 4 GVHD. The clinical features associated with the 2 classic TMAs, TTP and aHUS, including altered mental status, renal dysfunction, fever, and diarrhea, are nonspecific in a transplant population and should not be relied upon for diagnosis. A baseline peripheral blood smear should be obtained before transplant, and the blood should be periodically monitored throughout the transplant setting to follow peripheral schistocytosis. However, some red cell fragmentation is also a nonspecific finding in the transplant setting.

Part of the diagnostic evaluation of a patient with TA-TMA should include ADAMTS13 activity testing before the initiation of plasma therapy. Although all patients with

a TA-TMA commonly undergo plasma exchange initially, there is now a large body of evidence to suggest that this treatment is not effective, should not be considered a standard of care, and should be discontinued once an ADAMTS13 value above 5% to 10% is obtained.[28,43]

A work-up for infections should be instituted. It is also reasonable to hold or reduce the dose of calcineurin inhibitors. The pathophysiologic role of terminal complement components in TA-TMA is becoming clearer and is a field in need of further study. A growing body of evidence, including this report, suggests that the inability to control the alternative complement pathway is related to at least some cases of TA-TMA, and anti-complement therapy with eculizumab may result in complete remission. The dosing and duration of eculizumab therapy, and the monitoring of patients, are still to be defined and should be analyzed in clinical trials. The evaluation of complement activation by staining skin, bone marrow, or kidney biopsy specimens for C5b–9, C5a, and C4d may provide useful information to increase our understanding of the pathology of this TMA. The bone marrow niche has not been systematically studied in patients with TA-TMA,[65] and our group is actively pursuing this.

Genetic testing for complement mutations is not widely available, and even when testing is done by clinical centers, the platform of mutations analyzed, typically 12 genes encompassing 200 mutations, will fail to capture at least 30% of known aHUS-associated changes. Indeed, in clinical trials used to support the US Food and Drug Administration approval of eculizumab for uncomplicated aHUS, an identifiable mutation did not correlate with response to eculizumab.[66] Because these complement abnormalities involve germline mutations, if complement mutation testing is performed in the allogeneic transplant setting, it must be done with DNA isolated from nonhematologic sources (eg, a buccal swab).

There is also a need for a complement activity metric predictive of response to anti-complement therapy, not only in the TA-TMA setting but also in a broad range of diseases in which complement damage has been implicated. As previously noted, measurement of circulating C5b–9, C5a, and other complement components may help. For the past several years, our institution has been using immunofluorescence and immunohistochemistry to assess MAC deposition in the microvasculature of biopsy specimens of skin, ileum, and kidney from patients with putative aHUS.[11,39,43] However, the clinical utility of this test, in terms of its sensitivity and specificity for complement-mediated TMA, has not been established in routine practice. Clearly, further in vitro and in vivo studies are needed to identify new treatments and further elucidate the role of complement dysregulation in TA-TMA.

Disclosures

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