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Markers of coagulation activation and acute kidney injury in patients after hematopoietic cell transplantation

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Abstract

Acute kidney injury (AKI) is common after hematopoietic cell transplant (HCT). The etiology of AKI is unknown because biopsies are rarely performed. The pathophysiology of injury is inferred from clinical data. Thrombotic microangiopathy (TMA) is often invoked as the cause of renal injury.

Patients > 2 years undergoing their first HCT at Fred Hutchinson Cancer Research Center (FHCRC) participated in this study. We prospectively measured plasma markers of coagulation activation, (PAI-1 and tPA) and fibrinolyis (D-dimer) weekly in 149 patients during the first 100 days post-transplant. Cox proportional hazards modeling was used to determine associations between these markers and AKI (doubling of baseline serum creatinine). Kruskal-Wallis test was used to determine associations between day 100 urinary albumin to creatinine ratios (ACR) and these markers.

Thirty one percent of patients developed AKI. Though elevations in these markers occurred frequently, neither PAI-1 nor tPA were associated with development of AKI. D-dimer was associated with a slightly increased risk of AKI (RR=1.76; p-value 0.04). None of these markers were associated with micro- or macroalbuminuria at day 100.

The lack of an association with AKI suggests that endothelial injury in the form of TMA is not a common cause of AKI early after transplant.

Keywords

acute kidney injury; endothelial injury; acute graft vs host disease

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Introduction

Hematopoietic cell transplantation (HCT) has evolved as a primary therapy for refractory hematological malignancies, conditions of bone marrow failure, certain metabolic and autoimmune diseases, and some congenital immunodeficiencies. Over time, survival of patients undergoing HCT has improved with serial refinements of procedures, donor selection and toxicity management. However, kidney injury continues to be an ongoing problem, with an overall incidence of acute kidney injury (AKI) of 33% and a 5% incidence of renal injury that requires dialysis [1].

The exact etiology of AKI after HCT is unknown because biopsies are rarely performed and thus, the pathophysiology of injury must be inferred from clinical data. Potential causes of AKI include sepsis, renal toxicity from medications, portal hypertension resulting from liver injury, and thrombotic microangiopathy (TMA). TMA as an explanation for AKI suffers from several problems: clinical definitions do not correlate with serum creatinine measurements or renal pathology [2] and thrombocytopenia, anemia and hemolysis are unreliable markers of TMA in the early post-HCT period when most AKI occurs.

In the study reported here, we prospectively studied the predictive association between biomarkers of coagulation activation and fibrinolysis (PAI-1, tPA and D-dimer) and development of AKI. Plasminogen activator inhibitor (PAI-1), a glycoprotein produced by endothelial cells, hepatocytes and platelets, is normally present at low concentrations in plasma. A member of the serine protease inhibitor family, PAI-1 has been implicated in development of thrombotic microangiopathy, proliferative glomerulonephritis, and progressive renal disease. PAI-1 may also behave as an acute phase protein [3]. Tissue plasminogen activator is a serine protease found in the blood, brain parenchyma and at the interface of the two. The vascular activity of tPA is primarily controlled by PAI-1 and the balance between the two regulates fibrinolysis in vessels [4]. D-dimer, a fibrin degradation product present in the plasma when there is activation of the coagulation system in the setting of thrombosis and/or disseminated intravascular coagulation, is a measure of endogenous fibrinolysis. In hematopoietic cell transplant patients, PAI-1 elevations have been associated with multiple post-transplant complications including hepatic sinusoidal obstruction syndrome, sepsis, and thrombotic thrombocytopenic purpura [5,6,7].

Our hypotheses in this study were first, that endothelial injury (triggering a cascade of events leading to activation of the coagulation system with thrombin generation, fibrin formation, and platelet aggregation) is the proximate cause of AKI after HCT and second, that activation of the coagulation system, reflected by increases in plasma levels of PAI-1, tPA, and D-dimer, will be associated with subsequent development of AKI.

Methods

Patient Selection

Patients over the age of 2 years undergoing their first HCT participated in this study if they met the following eligibility criteria: a) a baseline creatinine within the limits of normal for age in children, <1.3 mg/dL in women, and < 1.5 mg/dL in men; b) not currently taking

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angiotensin receptor blockers or angiotensin converting enzyme inhibitors; c) no history of diabetes at time of enrollment into the study; and d) consenting to a protocol approved by the Institutional Review Board of Seattle Children's Hospital (SCH). All consents were obtained prior to enrollment.

Technique of HCT

Patients undergoing HCT received a preparative conditioning regimen followed by infusion of hematopoietic cells from an allogeneic donor or from their own stored cells (autologous HCT). Myeloablative regimens were typically cyclophosphamide-based (with either total body irradiation (TBI) or targeted busulfan) for allogeneic transplants; autologous graft recipients received a number of different regimens. Reduced intensity conditioning regimens consisted of fludarabine and TBI 2-4 Gy[8]. The kidneys were not shielded during TBI. Allogeneic graft recipients received prophylaxis against acute GVHD with immunosuppressive drugs, usually cyclosporine or tacrolimus plus methotrexate or mycophenolate mofetil [9]. Prophylaxis for infections included acyclovir to prevent herpes simplex virus and varicella zoster infection, trimethoprim/ sulfamethoxazole to prevent *Pneumocystic jivecii* infection, oral fluconazole or itraconazole for prophylaxis of fungal infection, and pre-emptive ganciclovir for cytomegalovirus disease among viremic patients [10-14]. Prophylactic oral ursodiol was given routinely to prevent cholestatic liver injury [15].

Specimen Collection and Analytical Methods

Blood was collected from a Hickman central venous access catheter and placed in a citrated tube between the hours of 8-10 a.m. at baseline (prior to the conditioning regimen), and then weekly through day 100 post-HCT. Blood was centrifuged at 2500 rotations per minute at 4 degrees Celsius for 15 minutes and plasma was aspirated and frozen (-70°C) in 2 mL aliquots until analysis. At the time of analysis, plasma was rapidly thawed and the concentrations of PAI-1 activity (Chromolize, Biopool, Ventura, CA*), t-PA antigen (Asserachrom,* Diagnostica Stago, Parsippany, NJ , and D-dimer (Asserachrom, Diagnositca Stago, Parsippany, NJ) were determined by immunoassay. The intra-assay and inter-assay coefficient of variation is 6-8% for these analytes. Normal values were PAI-1 <20.4 IU/mL; tPA 1.8-12.5 ng/mL. and D-dimer <590 μg/mL[16]

Urine was also collected between the hours of 8-10 a.m., immediately placed on ice, separated into 2 mL aliquots and frozen at -80 degrees until time of analysis.

Urinary albumin was determined using an immunoturbidimetric assay using a Cobas c 11 analyzer in aliquots of untreated urine samples. The inter-assay coefficient of variation (CV) of the assay is 0.7-2.2% and intra-assay CV is 1.0-1.6%. A quantitative determination of urine creatinine was measured on Roche/Hitachi modular automated clinical chemistry analyzers.

Clinical and outcome variables

Clinical data included baseline patient characteristics: age, gender, race/ethnicity, indication for HCT, preparative regimen, total body irradiation (TBI), use of busulfan or

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cyclophosphamide as part of the conditioning regimen, and development of sinusoidal obstruction syndrome (SOS). Primary indications for transplant were categorized as acute leukemia, chronic leukemia, myelodysplastic syndrome and all other groups. AKI was defined as the doubling of baseline serum creatinine measurement in the first 100 days post-HCT; the onset of AKI was the day of first doubling of baseline serum creatinine. SOS was scored individually on each patient by GBM based on published criteria [17]. Additional week-specific clinical information included exposure to calcineurin inhibitors, exposure to amphotericin in any form, presence of hypertension (blood pressure >140/90 in adults and >95%tile for age, gender and height for children or use of antihypertensives), presence of acute graft-versus-host disease (aGVHD), and presence of culture-positive bacteremia. Bacteremia was defined as a positive blood culture and aGVHD was scored based on consensus criteria [18]. The degree of albuminuria was expressed as a urinary albumin-to-creatinine ratio (ACR). Normal ACR was <30 mg/g creatinine; microalbuminuria was 30-299 mg/g creatinine; and macroalbuminuria was defined as ≥ 300 mg/g creatinine. ACR values closest to day 100 that fell within in the window of day 70-100 post-HCT were used for the analysis.

Statistical methods

Cox-regression modeling was used to determine the association between elevations in the markers of coagulation activation and fibrinolysis and development of AKI. All Cox regression models include the covariate information in a time-dependent fashion. For PAI-1, the time-dependent covariate at time *t* contains the last observation of PAI-1 obtained in the interval [0,*t*]. If the weekly PAI-1 observation on a subject was missing, the previous PAI-1observation was carried forward for a maximum of two weeks. Similar methods were used for t-PA and D- dimer models. The proportion of at-risk time in the Cox models for which a PAI-1, t-PA or D-dimer value was carried forward beyond one week in order to fill in for missing measurements was between 10% and 13%, depending on the marker and outcome event being analyzed. To evaluate another potentially important aspect of the longitudinal data profile, a secondary set of models were also run where the time-dependent covariate at time *t* contains the running peak value of PAI-1, i.e. the maximum PAI-1 observation obtained during the interval [0,*t*]. Similar parameters were used for t-PA and D-dimer models. The relative risk estimates for PAI-1 are expressed per 10 unit change in PAI-1. The relative risk estimates for t-PA are expressed per 1 unit change in t-PA. The relative risk estimates for D-dimer are expressed per 1000 unit change in D-dimer value.

The multivariable model for the outcome AKI includes aGVHD status (grade 2-4 vs grade 0-1), SOS, and patient age at transplant (age ≥40 yrs vs age <40 yrs). Receipt of amphotericin was also considered for inclusion in the multivariable model. However the model did not converge properly for this covariate due to the rarity of amphotericin usage and it was subsequently removed from the multivariable models. Sensitivity analyses were run for each of the Cox models to assess the influence of the two-week restriction on the length of time in which the last observation for each of the three markers of coagulation activation could be carried forward in cases with missing covariate data. Analyses allowing covariate observations to be carried forward for cases indefinitely for cases with missing covariate data produced similar results (data not shown). Peak PAI-1, tPA and D-dimer were

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summarized by day 100 ACR group (normal, micro- and macroalbuminuria) with box plots and compared among the groups using Kruskal-Wallis tests. Data analysis was completed with STATA 10 (Statacorp LP, College Station, Texas).

Results

Demographic information

One hundred and forty-nine patients were included in this study on the basis of baseline eligibility and complete blood sample collection. Pre-transplant demographic information by development of AKI is presented in Table 1. Forty-six patients developed AKI (31%). Use of high-dose TBI (≥12 Gy) was more common in the AKI group, but the difference was consistent with the increased proportion of patients with acute leukemia in the AKI group. No subject undergoing autologous HCT developed AKI. The proportion of patients with hypertension at baseline was similar between the groups. One patient in the AKI group had diabetes at the time of enrollment and was included in the analyses.

Markers of coagulation activation and fibrinolysis and association with AKI

There was no difference in baseline values of markers of coagulation activation between the patients who developed AKI compared to those who did not develop AKI (Table 1). Box plots of the weekly values for PAI-1, tPA and D-dimer are shown in Figures 1-3, respectively. Neither PAI-1, nor tPA nor D-dimer plasma levels in the 2 weeks prior to the development AKI (or a comparable time post-transplant) differed between patients who did and did not develop AKI (Table 2). Median values for each of these analytes were not higher in patients who subsequently developed AKI; in fact median values were higher in those who never developed AKI (Table 2). Neither PAI-1 nor tPA were associated with development of AKI post-HCT in the Cox regression analyses. However, D-dimer was associated with a slightly increased risk of development of AKI after adjusting for aGVHD, age at transplant and presence of SOS (RR=1.76; p-value 0.04) (Table 3). The secondary set of Cox models utilizing the running peak value as the time-dependent covariate for each TMA marker did not identify any statistically significant associations between the markers and AKI (data not shown).

Clinical associations with AKI

We examined known risk factors for the development of AKI during the first 100 days post HCT in our 149- patient cohort (Table 4). The AKI group had a higher proportion of subjects with SOS, acute GVHD, blood stream infections, vancomycin use, and calcineurin inhibitor use. Amphotericin in any form was rarely used. In our previous studies [19], we had defined TMA based on laboratory criteria of a hematocrit < 30, platelet count <100,000 and an LDH value above the upper limits of normal in the 2 weeks prior to the development of AKI. Of the 46 patients in the current cohort who developed AKI, 9 met these criteria for laboratory evidence of TMA prior to development of AKI and 35 did not. Two patients who developed AKI on day 0 and 1 post-transplant were excluded from this analysis.

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Markers of coagulation activation and fibrinolysis and association with ACR at Day 100

Among the 149 subjects, 111 had both day 100 ACR data and post-transplant PAI-1, tPA and D-dimer data. Thirty-seven patients had a normal ACR, 55 patients had microalbuminuria and 19 patients had macroalbuminuria at day 100. We found no significant association between the peak values of PAI-1 (p=0.56), tPA (p=0.10) and D-dimer (p=0.27) in the first 100 days post-HCT and ACR category at day 100 (Figure 4).

Discussion

The primary focus of this prospective study of HCT patients was to determine if endothelial injury (as reflected by plasma markers of coagulation activation (PAI-1 and tPA) and fibrinolysis (D-dimer)) is associated with the development of AKI after transplant. Much of the earlier literature regarding kidney injury after HCT attributed it to thrombotic microangiopathies (TMA), specifically hemolytic uremic syndrome (HUS). Though HUS is often described as occurring 6-12 months after transplant, there is a wide range of onset, anywhere from 3 days to 3 years after HCT [20]. In our prior autopsy study of kidney pathology of HCT patients, approximately 20% of the patients had evidence of TMA in their kidneys at time of death (median survival time 43 days post HCT) [2]. In studies of diarrhea-associated HUS in a non-HCT population, markers of coagulation activation and fibrinolysis identical to the ones measured in this study were elevated prior to the clinical diagnosis of HUS [16]. This prospective study measuring markers of coagulation activation and fibrinolysis found little evidence to support the hypothesis that activation of the coagulation system with thrombin generation, fibrin formation, and platelet aggregation, is the proximate cause of AKI after HCT. Although increases in plasma levels of PAI-1, tPA, and D-dimer are commonly found in other situations where renal endothelial injury occurs, we found no such evidence of activation of the coagulation system in patients developing AKI after HCT.

Although levels of these markers fluctuate post-transplant and suggest a state of coagulation activation throughout the transplant process, these levels did not correlate with clinical events [21]. Similarly, although we found elevated levels of PAI-1, tPA and D-dimer in patients after transplant, they were not associated with development of AKI. There is evidence that activation of coagulation and fibrinolytic pathways may be part of the normal process of hematopoietic reconstitution that is going on in all HCT patients. For example, in animal models of hematopoietic regeneration, fibrinolytic pathways are necessary for hematopoietic cell proliferation and differentiation after myelosuppression and/or stress [22]. Thus, during the time of engraftment, proliferation and differentiation of hematopoietic cells, elevated levels of these markers may be expected as a marker of developing hematopoiesis.

There are a few limitations to our study. We did not have kidney biopsy specimens with which to correlate plasma levels of PAI-1, tPA and D-dimer with histologic evidence of TMA. However, in an autopsy study of renal histopathology, we found no correlation between standard laboratory parameters of TMA, serum creatinine and histologic evidence of TMA in the kidney [2]. We had only 9 patients with laboratory evidence of TMA in this study as LDH is not routinely drawn. Therefore, we were not able to correlate a laboratory diagnosis of TMA with these markers.

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Although endothelial injury may still be playing a role in post-transplant renal complications, such injury may not be reflected in plasma levels of PAI-1, tPA and D-dimer. Other mechanisms may be involved in the AKI seen after transplant. We have shown that albuminuria occurs early after transplant and is associated with the development of renal injury[23]. It is postulated that albuminuria reflects a systemic, generalized endothelial injury that affects multiple organs [24]. We have not found a correlation between albuminuria and serum levels of PAI-1 or other markers of coagulation activation in this patient population. Either PAI-1 is not a good marker for endothelial injury in these patients, or albuminuria reflects the inflammatory state of GVHD and not endothelial injury. The inflammatory and cytokine cascades activated by GVHD can affect the kidney without invoking the direct T-cell mediated injury that is implicated in GVHD involving skin, gut and liver [25, 26]. In support of this indirect hypothesis, increased plasma cytokine levels correlate with post-transplant complications and organ dysfunction in the HCT population [27, 28]. In animal models of GVHD, tissue destruction by acute GVHD does not require alloantigen expression on target epithelium for cellular cytotoxicity; injury can be mediated by inflammatory cytokines [29]. Perhaps the inflammatory milieu of GVHD after transplant contributes to the early renal injury. Another possible mechanism is that the ischemia reperfusion injury commonly associated with AKI in other hospitalized patient populations also leads to acute tubular necrosis in patients early after HCT. If this is the case, than the lack of PAI-1 expression demonstrated in normal human kidneys and in the case of acute tubular necrosis, is consistent with the findings of our study [30].

In summary, markers of coagulation activation and fibrinolysis are not associated with the development of AKI after HCT. These findings suggest that alternate pathophysiologic processes cause AKI during the first 100 days after transplant.

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REFERENCES

1. Gooley TA, Chien JW, Pergam SA, Hingorani S, Sorror ML, Boeckh M, et al. Reduced mortality after allogeneic hematopoietic-cell transplantation. N Engl J Med. 2010; 363(22):2091–101. [PubMed: 21105791]
2. Changsirikulchai S, Myerson D, Guthrie KA, McDonald GB, Alpers CE, Hingorani SR. Renal thrombotic microangiopathy after hematopoietic cell transplant: role of GVHD in pathogenesis. Clin J Am Soc Nephrol. 2009; 4(2):345–53. [PubMed: 19144762]
3. Eddy AA. Plasminogen activator inhibitor-1 and the kidney. Am J Physiol Renal Physiol. 2002; 283(2):F209–20. [PubMed: 12110504]
4. Lemarchant S, Docagne F, Emery E, Vivien D, Ali C, Rubio M. tPA in the injured central nervous system: different scenarios starring the same actor? Neuropharmacology. 2012; 62(2):749–56. [PubMed: 22079561]
5. Nurnberger W, Michelmann I, Burdach S, Gobel U. Endothelial dysfunction after bone marrow transplantation: increase of soluble thrombomodulin and PAI-1 in patients with multiple transplant-related complications. Ann Hematol. 1998; 76(2):61–5. [PubMed: 9540759]

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6. Anthony MT, Zeigler ZR, Lister J, Raymond JM, Shadduck RK, Kramer RE, et al. Plasminogen activator inhibitor (PAI-1) antigen levels in primary TTP and secondary TTP post-bone marrow transplantation. Am J Hematol. 1998; 59(1):9–14. [PubMed: 9723570]
7. Salat C, Holler E, Reinhardt B, Kolb HJ, Seeber B, Ledderose G, et al. Parameters of the fibrinolytic system in patients undergoing BMT: elevation of PAI-1 in veno-occlusive disease. Bone Marrow Transplant. 1994; 14(5):747–50. [PubMed: 7889007]
8. Carella AM, Champlin R, Slavin S, McSweeney P, Storb R. Mini-allografts: ongoing trials in humans. Bone Marrow Transplant. 2000; 25(4):345–50. [PubMed: 10723575]
9. Doroshow, JH.; Synold, TW. Thomas' Hematopoietic Cell Transplantation. Wiley-Blackwell; 2009. Pharmacologic Basis for High-dose Chemotherapy.; p. 287-315.
10. Leather, HL.; Wingard, JR. Thomas' Hematopoietic Cell Transplantation. Wiley-Blackwell; 2009. Bacterial Infections.; p. 1325-1345.
11. Zaia, JA. Thomas' Hematopoietic Cell Transplantation. Wiley-Blackwell; 2009. Cytomegalovirus Infection.; p. 1367-1381.
12. Brown, JMY. Thomas' Hematopoietic Cell Transplantation. Wiley-Blackwell; 2009. Fungal Infections after Hematopoietic Cell Transplantation.; p. 1346-1366.
13. Ito, JI. Thomas' Hematopoietic Cell Transplantation. Wiley-Blackwell; 2009. Herpes Simplex Virus Infections.; p. 1382-1387.
14. Ho, DY.; Arvin, AM. Thomas' Hematopoietic Cell Transplantation. Wiley-Blackwell; 2009. Varicella-zoster Virus Infections.; p. 1388-1409.
15. Ruutu T, Eriksson B, Remes K, Juvonen E, Volin L, Remberger M, et al. Ursodeoxycholic acid for the prevention of hepatic complications in allogeneic stem cell transplantation. Blood. 2002; 100(6):1977–83. [PubMed: 12200355]
16. Chandler WL, Jelacic S, Boster DR, Ciol MA, Williams GD, Watkins SL, et al. Prothrombotic coagulation abnormalities preceding the hemolytic-uremic syndrome. N Engl J Med. 2002; 346(1): 23–32. [PubMed: 11777999]
17. McDonald GB, Hinds MS, Fisher LD, Schoch HG, Wolford JL, Banaji M, et al. Veno-occlusive disease of the liver and multiorgan failure after bone marrow transplantation: a cohort study of 355 patients. Ann Intern Med. 1993; 118(4):255–67. [PubMed: 8420443]
18. Przepiorka D, Weisdorf D, Martin P, Klingemann HG, Beatty P, Hows J, et al. 1994 Consensus Conference on Acute GVHD Grading. Bone Marrow Transplant. 1995; 15(6):825–8. [PubMed: 7581076]
19. Hingorani SR, Guthrie K, Batchelder A, Schoch G, Aboulhosn N, Manchion J, et al. Acute renal failure after myeloablative hematopoietic cell transplant: incidence and risk factors. Kidney Int. 2005; 67(1):272–7. [PubMed: 15610251]
20. Pettitt AR, Clark RE. Thrombotic microangiopathy following bone marrow transplantation. Bone Marrow Transplant. 1994; 14(4):495–504. [PubMed: 7858524]
21. Brandao LR, Kletzel M, Boulad F, Kurtzberg J, Maloney K, Fligman I, et al. A prospective longitudinal multicenter study of coagulation in pediatric patients undergoing allogeneic stem cell transplantation. Pediatr Blood Cancer. 2008; 50(6):1240–6. [PubMed: 18273869]
22. Heissig B, Lund LR, Akiyama H, Ohki M, Morita Y, Romer J, et al. The plasminogen fibrinolytic pathway is required for hematopoietic regeneration. Cell Stem Cell. 2007; 1(6):658–70. [PubMed: 18371407]
23. Hingorani SR, Seidel K, Lindner A, Aneja T, Schoch G, McDonald G. Albuminuria in hematopoietic cell transplantation patients: prevalence, clinical associations, and impact on survival. Biol Blood Marrow Transplant. 2008; 14(12):1365–72. [PubMed: 19041058]
24. Toto RD. Microalbuminuria: definition, detection, and clinical significance. J Clin Hypertens (Greenwich). 2004; 6(11 Suppl 3):2–7. [PubMed: 15538104]
25. Couriel D, Caldera H, Champlin R, Komanduri K. Acute graft-versus-host disease: pathophysiology, clinical manifestations, and management. Cancer. 2004; 101(9):1936–46. [PubMed: 15372473]
26. Krenger W, Hill GR, Ferrara JL. Cytokine cascades in acute graft-versus-host disease. Transplantation. 1997; 64(4):553–8. [PubMed: 9293864]

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27. Takatsuka H, Wakae T, Mori A, Okada M, Suehiro A, Okamoto T, et al. Thrombotic thrombocytopenic purpura and hemolytic uremic syndrome following allogeneic bone marrow transplantation. Bone Marrow Transplant. 2002; 29(11):907–11. [PubMed: 12080356]

28. Seconi J, Watt V, Ritchie DS. Nephrotic syndrome following allogeneic stem cell transplantation associated with increased production of TNF-alpha and interferon-gamma by donor T cells. Bone Marrow Transplant. 2003; 32(4):447–50. [PubMed: 12900785]

29. Teshima T, Ordemann R, Reddy P, Gagin S, Liu C, Cooke KR, et al. Acute graft-versus-host disease does not require alloantigen expression on host epithelium. Nat Med. 2002; 8(6):575–81. [PubMed: 12042807]

30. Xu Y, Hagege J, Mougenot B, Sraer JD, Ronne E, Rondeau E. Different expression of the plasminogen activation system in renal thrombotic microangiopathy and the normal human kidney. Kidney Int. 1996; 50(6):2011–9. [PubMed: 8943484]

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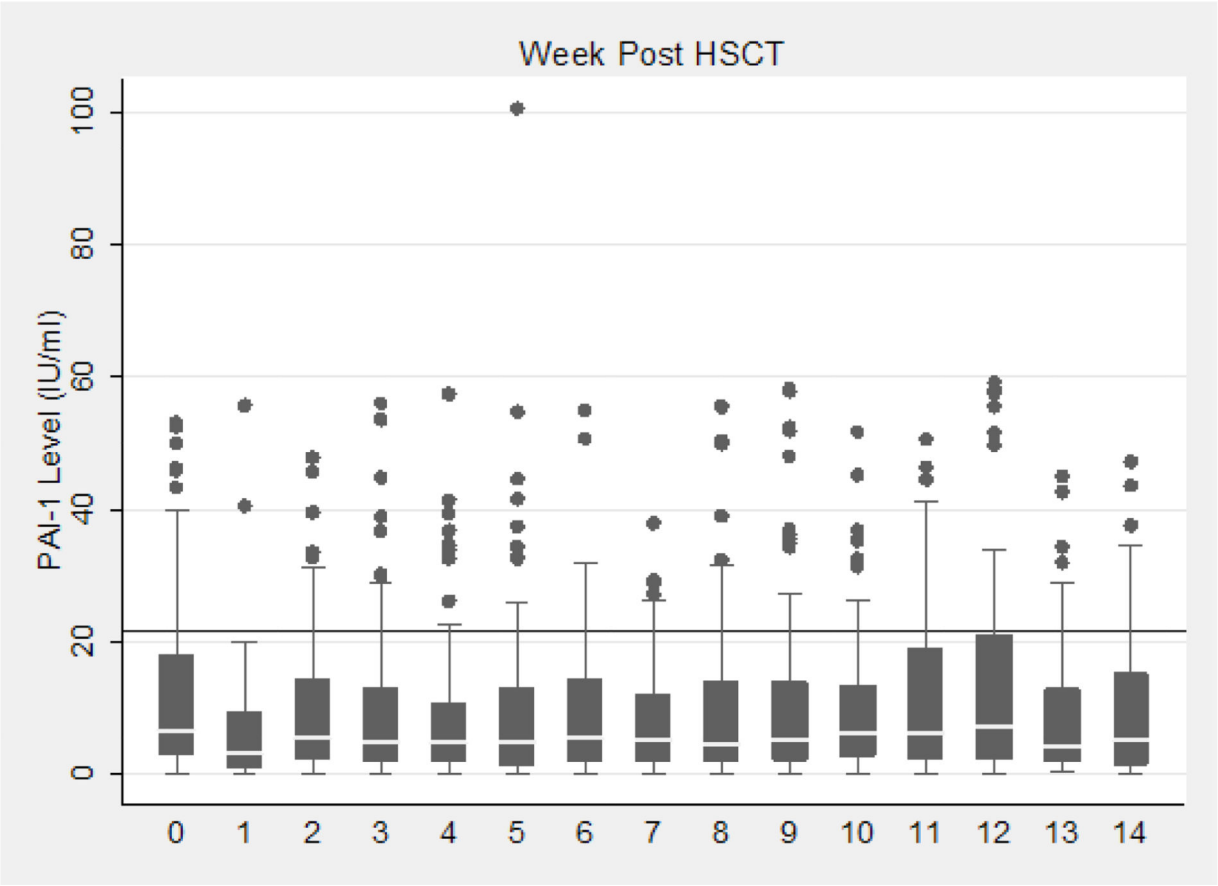

Figure 1. Serum PAI-1 levels by Week

Bar at 21.5 IU/ml indicates the upper limit of normal for PAI-1 levels. White bar is median value. Lower and upper limits of black box are 25th and 75th interquartile ranges.

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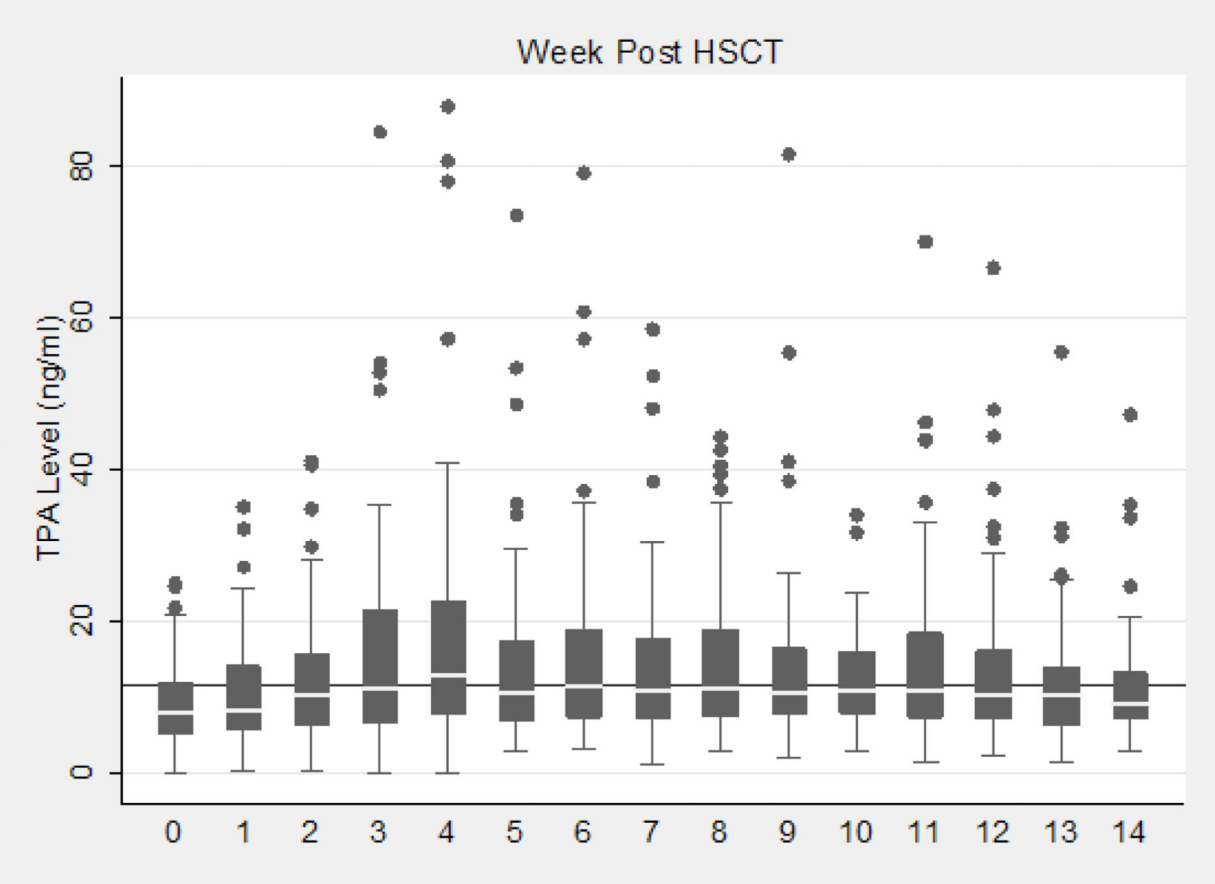

Figure 2. Serum tPA levels by Week

Bar at 11.5 ng/ml indicates the upper limit of normal for TPA levels. White bar is median value. Lower and upper limits of black box are 25th and 75th interquartile ranges.

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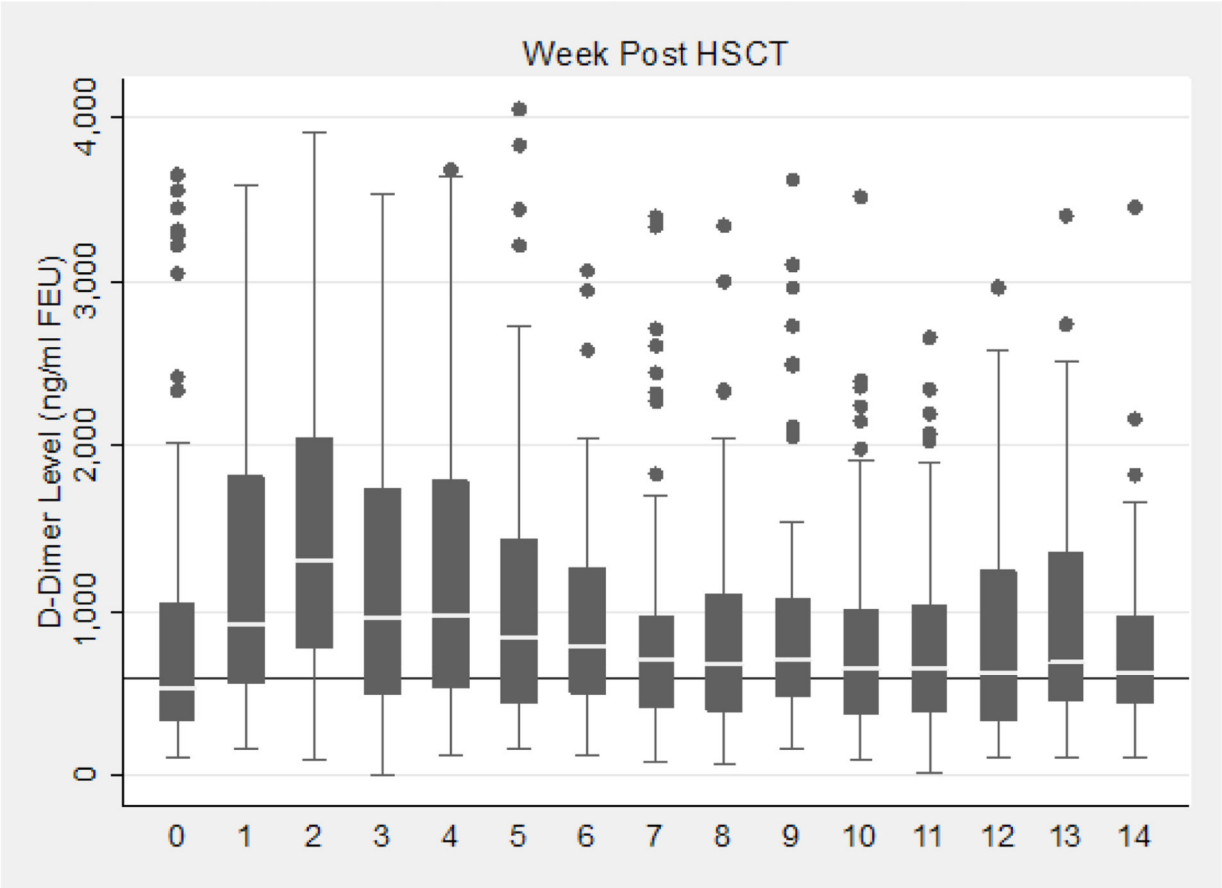

Figure 3. Serum D-dimer Levels by Week

Bar at 590 ug/ml FEU indicates upper limit of normal for D-dimer levels. White bar is median value. Lower and upper limits of black box are 25th and 75th interquartile range.

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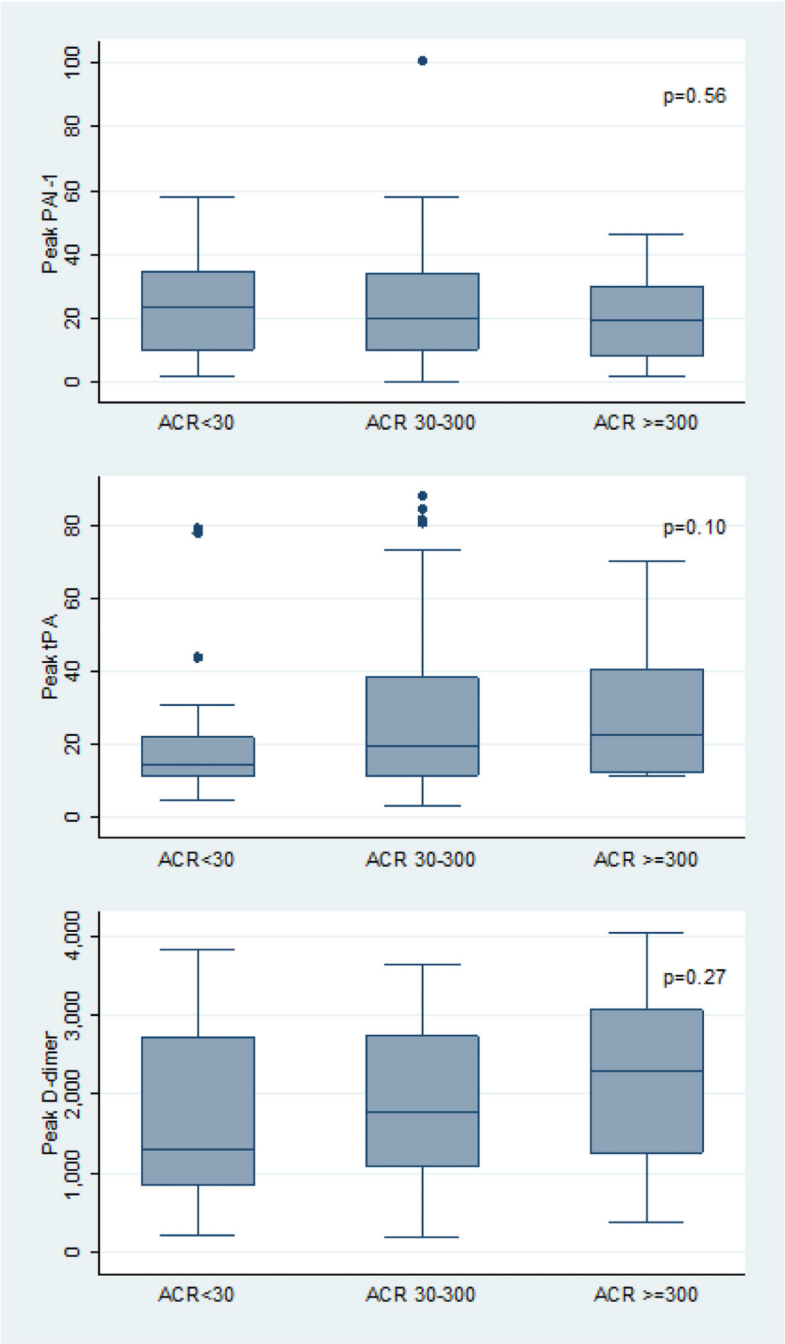

Figure 4. Box-plots of peak PAI-1, tPA and D-dimer values within the first 100 days by ACR category

p-values generated from Kruskal-Wallis test.

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Table 1

Demographic Information, by AKI Status

	Acute Kidney Injury (≥2x Baseline Creatinine)	No Acute Kidney Injury (Creatinine < 2x Baseline)	p-value
Number (%)	46 (30.9%)	103 (69.1%)	
Sex			
Male	27 (58.7%)	68 (66.0%)	0.39
Female	19 (41.3%)	35 (34.0 %)	
Age (years)			
<20	6 (13.0%)	8 (7.8%)	0.44
20-39	7 (15.2%)	21 (20.4%)	
40-59	23 (50.0%)	59 (57.3%)	
60+	10 (21.7%)	15 (14.6%)	
Race/Ethnicity			
Caucasian	35 (76.1%)	83 (81.4%)	0.44
Black	4 (8.7%)	3 (2.9%)	
Hispanic	3 (6.5%)	4 (3.9%)	
Other	3 (6.5%)	11 (10.8%)	
Missing	1 (2.2%)	1 (1.0%)	
Indication for HSCT			
Aplastic Anemia	2 (4.3%)	3 (2.9%)	0.20
Acute Lymphocytic Leukemia	5 (10.9%)	3 (2.9%)	
Acute Nonlymphocytic Leukemia	19 (41.3%)	36 (35.0%)	
Chronic Lymphocytic Leukemia	2 (4.3%)	2 (1.9%)	
Chronic Myelogenous Leukemia	3 (6.5%)	16 (15.5%)	
Myelodysplastic Syndrome	9 (19.6%)	20 (19.4%)	
Multiple Myeloma	3 (6.5%)	4 (3.9%)	
Non-Hodgkin's Lymphoma	2 (4.3%)	13 (12.6%)	
Other	1 (2.2%)	6 (5.8%)	
Preparative Regimen			
Busulfan (BU) + Cyclophosphamide (CY)	17 (37.0%)	46 (44.7%)	0.02
CY/TBI > 12 Gy	11 (23.9%)	16 (15.5%)	
Reduced-intensity conditioning	17 (37.0%)	23 (22.3%)	
Other non-TBI regimens	1 (2.2%)	18 (17.5%)	
Total Body Irradiation dose			
≥12 Gy	11 (24.4%)	16 (15.3%)	0.03
2 Gy		23 (22.3%)	
None	17 (36.9%)	64 (62.1%)	
	18 (39.1%)		
Type of Donor Marrow			
Autologous	0 (0%)	19 (18.4%)	0.01
Allogenic related	20 (43.5%)	40 (38.8%)	

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	Acute Kidney Injury (≥2x Baseline Creatinine)	No Acute Kidney Injury (Creatinine < 2x Baseline)	p-value
Allogeneic unrelated	26 (56.5%)	44 (42.7%)	
Hypertension at enrollment			
Elevated BP &/or BP medications	8 (29.6%)	16 (22.5%)	0.47
BP not elevated & not on medications	19 (70.4%)	55 (77.5%)	
Missing data	19	32	
Baseline TMA Marker, mean (SD)			
D-Dimer (μg/mL)	934 ± 802	806± 847	
PAI-1 (lU/ml)	11.6 ± 14.5	12.0 ± 12.5	
TPA (ng/ml)	8.50 ± 4.89	9.27 ± 5.01	

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Table 2

Median values and range for markers of coagulation activation in the 2 weeks prior to the development of each clinical event.

Clinical event	PAI-1 (lU/ml)	tPA (ng/mL)	D-dimer (μg/mL)
AKI			
-yes	2.5 (0.2-55.7)	9.8 (2.8-37.1)	787.9 (333.2-3323.5)
-no†	5.7 (0-57.5)	12 (0-87.9)	1075.7 (1.7-3643.4)

† For the cohort of patients who did not experience the event in question, the median time for those who did develop AKI.

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Table 3

Cox regression analysis of association of markers of coagulation activation and development of AKI after HCT

All models are Cox regression models which include the TMA marker information in a time-dependent fashion. E.g, for PAI-1, the time-dependent covariate at time t contains the last observation on PAI-1 obtained in the interval [0,t]. The relative risk estimates for D-dimer are expressed per 1000 unit change in D-dimer value. The relative risk estimates for PAI-1 are expressed per 10 unit change in PAI-1. The relative risk estimates for tPA are expressed per 1 unit change in tPA.

	Model			
	Univariable Model		Multivariable Model *	
	Relative risk (95% CI)	p-value	Relative risk (95% CI)	p-value
PAI-1	0.75 (0.49, 1.15)	0.19	0.91 (0.59, 1.41)	0.69
D-dimer	1.43 (0.93, 2.20)	0.11	1.76 (1.02, 3.05)	0.04
tPA	0.98 (0.94, 1.03)	0.48	1.00 (0.95, 1.05)	0.99

* The multivariable model for the AKI outcome includes AGVHD status (grade 2-4 vs grade 0-1), VOD/SOS and patient age at transplant (age>40 yrs vs age<40 yrs).

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Table 4

Clinical Events by AKI status in the first 100 Days Post HCT

	Acute Kidney Injury 2x Baseline Creatinine (N=46)	No Acute Kidney Injury (N=103)
SOS *		
Yes	14 (30.4%)	6 (5.8%)
No	32 (69.6%)	97 (94.2%)
Acute Graft Vs. Host Disease		
aGVHD grade ≤ 1	20 (43.5%)	42 (40.8%)
aGVHD grade ≥2	26 (56.5%)	61 (59.2%)
Blood Stream Infections		
Number at <100 days	16 (35.6%)	25 (24.3%)
Nephrotoxins		
Use at anytime in first 100 days		
Amphotericin	1 (2.2%)	2 (1.9%)
Calcineurin Inhibitors	34 (75.6%)	64 (62.1%)
Gentamicin	2 (4.4%)	3 (2.9%)
Vancomycin	20 (44.4%)	31 (30.1%)
TMA diagnosis based on standard laboratory criteria (see Methods)		
-yes	9 (20.0%)	6 (5.8%)
-no*	35 (77.8%)	97 (94.2%)

* 2 patients developed AKI early after transplant, day 0 and 1