Figure. Genetic variations in complement associated genes (HSCT recipient DNA). Figure shows a "heat map" of gene variants in recipients with and without TMA: each column represents a gene, with heterozygous variants shaded light blue and homozygous variants shaded red. Each row is a single patient, illustrating the wide distribution of variants in multiple genes in the TMA cases, while few variants are seen in the cases without TMA.

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The Genetic Fingerprint of Susceptibility to Transplant Associated Thrombotic Microangiopathy

Sonata Jodele [1], *Benjamin Laskin* [2], *Kejian Zhang* [3],
Fanggeng Zou [3], *Christopher E. Dandoy* [1], *Kasiani C. Myers* [1],
Javier El-Bietar [1], *Gregory Wallace* [1], *Michael S. Grimley* [1],
Jack Bleesing [1], *Jens Goebel* [4], *Diane Kissell* [5],
Shannon Nortman [5], *Adam Lane* [1], *Stella M. Davies* [1]. [1] *Bone Marrow Transplantation and Immune Deficiency, Cincinnati Children's Hospital Medical Center, Cincinnati, OH;*
[2] *Nephrology and Hypertension, Cincinnati Children's Hospital Medical Center, Cincinnati, OH;* [3] *Human Genetics, Cincinnati Children's Hospital Medical Center, Cincinnati, OH;*
[4] *Nephrology and Hypertension, Cincinnati Children's Hospital Medical Center, Cincinnati, OH;* [5] *Molecular Genetics, Cincinnati Children's Hospital Medical Center, Cincinnati, OH*

Background: Transplant associated thrombotic micro-angiopathy (TMA) is frequent after HSCT, and in severe cases causes significant morbidity and mortality. Currently there are no data addressing individual susceptibility to transplant associated TMA. Frequent genetic variants in ADAMTS13 and complement genes have been described in other micro-angiopathies such as aHUS and TTP. We hypothesized that polymorphic variants in complement genes increase susceptibility to transplant associated TMA.

Methods: 90 consecutive allogeneic HSCT patients were enrolled on a prospective TMA biomarker study and categorized as having TMA or no TMA using rigorous diagnostic criteria (Jodele et al, Blood 2014). Genomic DNA was isolated from pre-HSCT recipient blood. We used a candidate gene approach to identify 12 genes within the complement pathway likely to play a role in terminal complement activation, the likely effector mechanism for vascular damage in TMA. All exons, flanking intronic and untranslated regions of ADAMTS13, CFH, CFI, CFB, MCB, THBD, C3, C5, CFD, CFHR1, CFHR3 and CFHR5 were sequenced using next generation sequencing technology. The resulting sequence reads were aligned against the reference DNA sequence and the variants were detected using NextGENe

software. Observed variants were compared against dbSNP (NCBI). Novel variants were further evaluated using laboratory developed bioinformatic tools. Eight variants previously described as likely pathogenic and 34 variants of unknown pathogenic significance were identified. Results were compared in patients with and without TMA.

Results: 77 patients had DNA available for analysis (34 with TMA and 43 without TMA); 23 of 34 (68%) subjects with TMA had complement gene variants as compared to 4 of 43 (9%) controls without TMA (p<0.0001). Figure shows a "heat map" of gene variants in recipients with and without TMA illustrating the wide distribution of variants in multiple genes in the TMA cases, while few variants are seen in those without TMA. Likely pathogenic variants previously described in other microangiopathies were seen in ADAMTS13 (n=2), CFH (n=2), CFB (n=1), CFHR5 (n=1) and CFI (n=3), all in recipients with TMA. No known pathogenic variants were seen in subjects without TMA. Two recipients with TMA had homozygous deletion of CFHR3/R1, which was not seen in the recipients without TMA. The median number of gene variants (of known or unknown significance) seen in recipients with TMA was 1 (0-7), and 0 (0-2) in those without TMA (p<0.0001). Two subjects had 5 variants and one 7 variants and all three had fatal TMA.

Conclusion: The incidence of complement gene variants was higher in patients with TMA as compared to patients without TMA, indicating that genetic susceptibility importantly alters risk. Additional larger studies are needed to define the mechanistic importance of variants of unknown clinical significance.

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High Hematopoietic Transplantation Comorbidity Index Is Not Associated with Increased Transplant Related Mortality: Review of a Large Cohort of Pediatric Patients Undergoing Allogeneic Stem Cell Transplantation Following Busulfan-Based Regimens for Malignant and Non-Malignant Diseases

Justine Kahn [1], *Naima Al Mulla* [2], *Mahvish Qureshi* [1],
Grace Kim [1], *Zhezhen Jin* [3], *Monica Bhatia* [4], *Esra Karamehmet* [5],

James Garvin [4], Diane George [4], Andrew Kung [6], Prakash Satwani [7]. [1] *Pediatric Hematology/Oncology/Stem Cell Transplantation, New York Presbyterian Hospital-Columbia University Medical Center, New York, NY;* [2] *Pediatric Hematology/Oncology/Stem Cell Transplantation, New York Presbyterian Hospital-Columbia University Medical Center, New York, NY;* [3] *Biostatistics, Columbia University, New York, NY;* [4] *Division of Pediatric Hematology, Oncology, and Stem Cell Transplantation, Department of Pediatrics, Columbia University, New York, NY;* [5] *Columbia University Medical Center, New York, NY;* [6] *Pediatrics, Columbia University, New York, NY;* [7] *Division of Pediatric Hematology, Oncology, and Stem Cell Transplantation, Department of Pediatrics, Columbia University Medical Center, New York, NY*

Introduction: Identification of patients at high risk for transplant related mortality (TRM) prior to allogeneic hematopoietic cell transplantation (alloHCT) is crucial. However, the heterogeneity of alloHCT indications, conditioning regimens, patient characteristics and center-specific practices makes defining generalizable risk factors for TRM a perpetual challenge. In a retrospective study, Smith et al. (Blood, 2011) concluded that Sorror's 17-item pre-alloHCT adult comorbidity index was also an effective tool for risk-stratifying pediatric alloHCT patients. In this study, we attempt to re-validate the HCT-CI published by Smith et al. in a cohort of patients receiving well-defined Busulfan (Bu)-based conditioning regimens.

Methods: Retrospective cohort study of 165 patients (<22y) who underwent alloHCT. Patients conditioned with 1 of 3 Bu-based regimens were included: reduced intensity (RIC): Bu (6.4-8mg/kg)+ Fludarabine (Flu) (150mg/m2), reduced toxicity (RTC): Bu (12.8-16mg/kg)+ Flu (180mg/m2) and myeloablative (MAC): Bu (12.8-16mg/kg)+ cyclophosphamide (120-200mg/kg) or melphalan (135mg/m2).

Results: Median age at alloHCT was 9.5y (0.1-22y), malignant group n=102, non-malignant group n=63. Pre-alloHCT comorbidities were scored using HCT-CI and risk group numbers were as follows: 32 (19.4%) group 0, 69 (41.8%) group 1-2, 64 (38.8%) group ≥3. The difference in incidence of TRM at 1y in each HCT-CI group was not statistically significant for malignant vs non-malignant patients. 1yr TRM of patients receiving RIC, RTC and MAC regimens was also not significantly impacted by pre-transplant HCT-CI scores in each of these cohorts. Data outlined in supplemental tables.

Conclusion: Preliminary results suggest that the HCT-CI as it stands may not be a generalizable risk stratification tool for all pediatric alloHCT patients. Contrary to aforementioned studies, we were unable to demonstrate poorer outcomes in patients with HCT-CI score ≥3. One potential reason could be a smaller sample size in our study (n=166 vs. 252). We excluded total body irradiation as it is only used in MAC regimens at our center which might have skewed the comparison between RIC/RTC vs. MAC thus making findings generalizable. Further

Table 1

HCT-CI Score	1y TRM All Patients			p-value
	Number	Deceased	Living	
0	32	9 (28.1%)	23 (71.9%)	0.299
1-2	69	19 (27.5%)	50 (72.5%)	
≥3	64	11 (17.2%)	53 (82.8%)	
All	165	39 (23.6%)	126 (76.4%)	

Table 2

HCT-CI Score	1y TRM: Conditioning Regimens			p-value
	RIC	RTC	MAC	
0	16.7% (1/6)	0% (0/11)	20% (3/15)	0.376
1-2	5% (1/20)	13% (3/23)	15.4% (4/26)	0.576
≥3	0% (0/18)	10.7% (3/28)	11.1% (2/18)	0.576

Table 3

1yr TRM: HCT-CI Score Comparisons	p-value
0 vs 1-2	0.180
1-2 vs ≥3	0.238
≥3	0.706

analysis is currently being conducted to identify risk factors distinct from the HCT-CI that may have a higher impact on TRM in pediatric patients undergoing alloHCT.

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Peripheral Blood Expansion of CD38 Bright CD8+ Effector Memory T-Cells Predicts Acute Graft Versus Host Disease with a Diagnostic Accuracy of 87%

Pooja Khandelwal [1], Adam Lane [1], Erika Owsley [1], Vijaya Chaturvedi [1], Michael B. Jordan [1], Stella M. Davies [1], Daniel J. Marmer [2], Alexandra Filipovich [3], Rebecca A. Marsh [1]. [1] *Bone Marrow Transplantation and Immune Deficiency, Cincinnati Children's Hospital Medical Center, Cincinnati, OH;* [2] *Cancer and Blood Diseases Institute Laboratories, Cincinnati Children's Hospital Medical Center, Cincinnati, OH;* [3] *Division of Bone Marrow Transplantation & Immune Deficiency, Cincinnati Children's Hospital Medical Center, Cincinnati, OH*

Acute graft versus host disease (aGVHD) is an important complication of allogeneic hematopoietic stem cell transplantation (HSCT). A need exists for validated biomarkers

Table 1

Grade and timing of aGVHD relative to absolute CD38 bright CD8+TEM cells/ μL prior to aGVHD onset

Patient	Maximum Absolute CD38 Bright CD8+ TEM (cells/μL)	Maximum Absolute CD38 Bright CD8+ TEM (Day Post HSCT)	Onset of acute GVHD (Day Post HSCT)	Onset of CD38 Bright CD8+ TEM >35cells/ uL (Day Post HSCT)	Maximum grade of aGVHD	Organ(s) involved
1	64	26	33	26	1	Skin
2	69	48	58	37	3	GI Liver
3	32	34	41	34	1	Skin
4	304	34	47	20	1	Skin
5	231	7	34	7	1	Skin
6	41	24	34	24	2	Skin
7	99	25	32	25	4	Skin Liver
8	98	16	23	16	3	Skin GI
9	90	52	69	17	3	Skin Liver
10	183	37	52	20	3	Skin GI Liver
11	42	33	42	33	3	Skin GI
12	36	11	15	11	4	Skin GI
13	82	15	29	15	4	GI
14	37	43	44	12	2	Skin
15	151	45	79	15	3	Skin GI
16	63	23	33	23	4	Skin GI
17	47	25	31	25	1	Skin
18	87	17	18	17	3	GI
19	69	56	59	56	1	Skin
20	2	25	28	NA	1	Skin
21	12	36	37	NA	1	Skin
22	70	28	38	21	1	Skin
23	128	42	45	28	3	Skin GI