received a single CBT and 124 (60%) had a double CBT. Thirty percent of CBU had 0-1 HLA mismatch (A, B, DRB1), 67% received CyFluTBI2Gy and median infused TNC was $3.7x10^7$/Kg. Pre-transplant serum was tested for HLA-Ab with a panel of fluorescent beads coated with single HLA-antigen using Luminex™ platform. Results were interpreted as fluorescence intensity (MFI) against donor-specific mismatch (>1000 MFI was the threshold for positivity). Overall 48 pts (23%) had anti-HLA-Ab before CBT and those were donor specific anti-HLA-Ab (DSA) in 16 pts. Among the 16 pts with DSA (11 females, 5 males), 9 had single and 7 double-CBT (none had DSA directed to both CB units). Seven pts had DSA vs to HLA-Class-I, 5 vs to HLA-Class-II and 4 to both HLA-Class-I and-II. DSA threshold ranged from 1620-17629 MFI. **Results.** Cumulative incidence (CI) of day-60 neutrophil engraftment was 76%. It was 44% for recipients with DSA and 81% in pts without DSA (p=0.006). There was no difference for pts with anti-HLA-Ab non donor-specific (77% vs 69%). Multivariate model showed DSA (RR 2.7, p=0.01) and CBT before 2008 (RR 1.49, p=0.03) independently associated with GF.Seven pts with DSA engrafted, 4 after double CBT and chimerism analysis showed the engraftment of the CBU with DSA in 1 case.Among 50 pts who failed engraftment,· 9 (20%) pts had DSA specific for donor HLA-Class-I(n=4) or Class-II(n=2) or both Class-I and Class-II (n=3).CI of platelet recovery at day-180 was 62%, 12 of 16 patients with DSA did not achieve platelet recovery. CI of 1-year TRM was 35%. DSA was associated with higher TRM (p=0.002). Overall survival at 3-years was 44%, it was 41% and 45% for pts with non-malignant and malignant disease respectively. OS was 47% for recipients without DSA and 25% for those with DSA, p=0.006. In multivariate analysis, the absence of DSA was the only factor associated with better survival (RR 2.41, p=0.005). **Conclusions**. Donor-specific anti-HLA-Ab in recipients of CBT is associated with failed engraftment and lower survival. Screening for DSA may be included in the algorithm of donor choice for cord blood transplantation.

0447

ENDOTHELIAL STRESS IN STEROID-REFRACTORY GVHD: A BURDEN STATINS CANNOT LIFT

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Graft-versus-host disease (GvHD) is the major complication of allogeneic stem cell transplantation (alloSCT) and its therapy-resistant form causes significant morbidity and mortality. The pathomechanism of steroid resistance is currently not completely understood; however, we have recently suggested that endothelial dysfunction seems to play an important role (Luft et al., Blood 2011). The aim of this study was to validate in a large cohort of patients that steroid refractory acute GvHD is associated with endothelial stress. Secondly we assessed if this endothelial stress can be overcome by statins, which are known to have endothelial protective effects. **Patients and Methods.** For this retrospective study, 393 patients were eligible who had undergone alloSCT between 09/2001 and 08/2010 at our institution. Serum levels of endothelial stress markers (Angiopoietin-2: Ang-2, soluble Thrombomodulin: sTM, Interleukin-8: IL-8 and Vascular endothelial growth factor: VEGF) were compared between patients with no GvHD (n=221), grade1-2 (n=103), steroid sensitive grade 3-4 (n=27) and steroid refractory grade 3-4 (n=41) GvHD, and correlated with outcome. Serum levels of these markers were also compared between patients with and without concomitant statin treatment. **Results.** Landmark analyses at days +50 and +100 after alloSCT showed that NRM was dramatically high in the steroid refractory group but was equivalently low in the no GvHD-, sensitive grade 1-2 - and grade 3-4 - groups (p<0.0001, Figure 1). Steroid-refractory patients showed higher serum levels of Ang-2 (p=0.03) prior alloSCT and significantly stronger rises in IL-8 (day 50: p=0.006; day 100: p=0.003) and sTM (day 50: p=0.04; day 100: P=0.006) levels post alloSCT than sensitive grade 3-4 GvHD patients. High levels of sTM, IL-8, Ang-2 were significantly associated with increased NRM rates (day +50: IL8 p=0.06, sTM p=0.0008, Ang-2 p=0.0001; day +100: IL8 p=0.0007, day +100 sTM p<0.0001, Ang-2: p=0.05); even after multivariate adjustment for donor, conditioning intensity, disease status at alloSCT, and sex mismatch. In contrast, VEGF was elevated in steroid sensitive grade 3-4 GvHD patients at day 100 but did impact on NRM rates.When comparing these markers in patients who had (n=84) or had not (n=309) received concomitant therapy with statins, no difference was seen for any endothelial cell stress markers neither in the whole cohort nor in patients with grade 3-4 GvHD. Patients with or without statins had similar NRM, relapse rates and overall survival. **Conclusions.** This study supports the hypothesis that steroid-refractory GvHD is associated with progressive microangiopathy. Statins, although reported to have protective effects on endothelial cells, were inefficient to alleviate endothelial stress in this context and accordingly, did not change the outcome of acute GvHD patients.

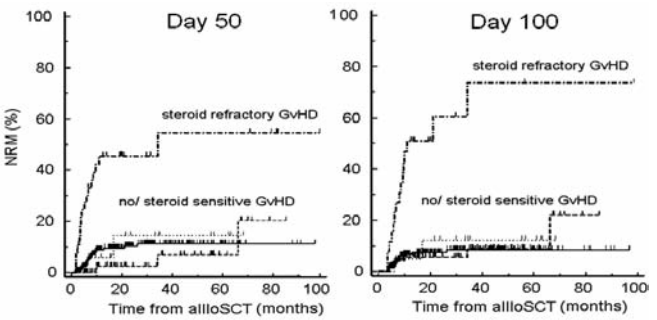

Figure 1. NRM rate by GvHD.

0448

COMPARISON OF IMMUNE RECONSTITUTION AFTER HEMATOPOIETIC STEM CELL TRANSPLANTATION WITH FLU-TBI VERSUS TLI-ATG CONDITIONING

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Background. The impact of the type of reduced intensity conditioning regimen used on immune recovery after allogeneic hematopoietic cell transplantation (allo-HCT) is poorly determined. **Aims.** We analyzed immune reconstitution in patients enrolled in a BHS-HCT sponsorised randomized study comparing two non-myeloablative conditioning regimens for allo-HCT for which cell samples were prospectively collected. **Patients and Methods.** The conditioning regimen consisted of either 2 Gy TBI with 90 mg/m² fludarabine (=TBI arm, n=21), or 8 Gy TLI plus thymoglobulin (ATG) 7.5 mg/kg (=TLI arm, n=19). Median ages at HCT were 59 yrs and 61 yrs in the TBI and TLI arms, respectively. Immune reconstitution was assessed by flow-cytometry phenotyping, signal joint T-cell Receptor Excision Circle (sjTREC) quantification, and T-cell spectratyping. Written informed consent has been obtained for each patient included. **Results.** Absolute T cell counts were lower in the TLI arm than in the TBI arm on day 28 after HSCT (P=0.04) but not thereafter. Further, B cells, as well as CD4+, CD4+CD45RA+ and CD4+CD45RO+ T cell reconstitution lagged behind in the TLI arm compared to the TBI arm the first year after HCT (B cells: p=0.0295 and others: p>0.0001). In contrast, reconstitution of CD8+ T cells, NK cells, Tregs and iNKT cells were similar in the 2 groups. For the thymic function, while sjTREC levels were higher in the TBI arm than in the TLI arm on day 100 (P=0.002) and on day 365 (not significant) after HCT, the increase in sjTREC levels from day 100 to day 365 was similar in the 2 groups of patients. The diversity of the TCR repertoire was similar in the 2 groups of patients on day 100 after HCT. Finally, we found that ATG persists in patients up to 17 days after allo-HCT in TLI patients (median of [ATG] at day 17=0.62 mg/l and for one patient at day 20=0.53). **Conclusions.** These preliminary results suggest that ATG may be responsible for the delay of immune reconstitution of CD4+ T cells in the TLI arm. Furthermore, ATG probably destroyed grafted sjTREC+ T cells, explaining the difference of sjTRECs level at days 100 and 365 between the two groups while sjTREC increment from day 100 to day 365 was similar in the 2 groups. Finally, TLI conditioning has no impact on immune regulatory populations (Treg and iNKT) after the transplantation.