SINGLE-CELL ANALYSIS REVEALS IMMUNE PATHWAYS LINKED TO CLINICAL OUTCOMES AFTER ALLOGENEIC STEM CELL TRANSPLANTATION

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Allogeneic stem cell transplantation (allo-SCT) offers curative potential for haematological malignancies but remains limited by complications like graft-versus-host disease (GvHD) and disease relapse. Early immune reconstitution plays a pivotal role in determining post-transplant outcomes, however cellular and molecular programs governing divergent clinical trajectories are not well understood.

To capture this process at single-cell resolution, we performed RNAseq on 82,552 cells from 48 longitudinal samples from 11 allo-SCT recipients. These included -infused donor stem cell product (SCP) and patient samples from pre-conditioning (day –7), week 2, and months 1–4 post-transplant. Patients were selected based on clinical outcomes: four developed acute GvHD (grade>2), four had moderate to severe chronic GvHD (one with both), three relapsed, and two remained disease-free for over three years. Single-cell transcriptomic data were analysed to assess immune cell composition, transcriptional divergence, pathway enrichment, and lineage-specific chimerism.

Key immune shifts included early NK cell expansion and reduced naïve $CCR7^+$ $CD4^+$ T cells across all patients. Acute GvHD was marked by delayed donor engraftment in T, B, and myeloid cells, reduced NK/B cell proportions at 0–1 month, and $CD8^+CCL5^+$ T cell expansion after one month, alongside enrichment of interferon, TNF-α, and allograft rejection pathways. Chronic GvHD showed reduced activated NK cells and increased $CD4^+CCR6^+$ T cells by month 2, with transcriptional changes from month 1–3 in B cells, monocytes, and T cells, including modest IL-2/STAT5 and TGF-β activation. Relapse was associated with high pre-transplant T cells, poor donor chimerism at one-month, early divergence in T/B cells from the SCP, and late enrichment of MYC, G2M, hypoxia, and p53 in T cells. Disease-free patients showed rapid multilineage donor chimerism and minimal inflammatory signalling.

These findings highlight critical early windows and immune signatures that could inform biomarker-led risk stratification and enable timely, outcome-specific interventions to improve allo-SCT success.