P874 DECIPHERING PLASMA CELL HETEROGENEITY AND TUMOR MICROENVIRONMENT IN LIGHT-CHAIN AMYLOIDOSIS USING SINGLE CELL RNA-SEQUENCING

Topic: 13. Myeloma and other monoclonal gammopathies - Biology & Translational Research

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Background:

Immunoglobulin light-chain amyloidosis (AL) is a rare disease caused by plasma cell secretion of misfolded light chains that assemble as amyloid fibrils, which deposit on vital organs causing organ dysfunction. Similar to AL, multiple myeloma (MM) is also caused by atypical plasma cell (PC) expansion in the bone marrow. Although, much research has been done on MM to understand disease mechanisms and to develop effective treatments, knowledge related to AL is still limited. Notably, there is little information available regarding the associated tumor microenvironment (TME) of AL. As the TME plays an important role in disease development and treatment response, comprehensive investigation of the AL TME could improve disease diagnosis and treatment. Furthermore, information distinguishing PCs from AL and MM patients is relatively sparse, but could help determine if treatments developed for MM would also be applicable for the treatment of AL.

Aims:
The main objective of our study was to decipher the underlying disease mechanisms of the AL at cellular and molecular level by analyzing AL patient samples using single cell RNA sequencing (scRNAseq). To achieve this, we aimed to understand heterogeneity within AL vs MM plasma cells and the TME at the cellular and gene expression level.

Methods:
The Finnish Hematology Research Biobank provided 23 viably frozen bone marrow mononuclear cell (BM-MNC) samples from 22 AL patients and 24 samples from 23 MM patients. The BM-MNCs were thawed and sorted based on their CD138 expression and cell viability (7AAD dead cell marker). The viable CD138+ and CD138- cells were mixed in a ratio where CD138+ were enriched, but not exceeding 50%. The Chromium Single Cell 3’RNAseq run and library preparation were done using Chromium™ Single Cell 3’ Reagent version 3 chemistry. 10x Genomics Cell Ranger v3.0.1 pipelines were used for the initial data processing. Further, the quality control and data preprocessing was performed using Seurat 4.1.0. For cell type annotations to identify the clusters, specific cell type markers for the immune cells were obtained from scType database, GSEA-MSigDB and manual curation.

Results:

From the integrated analysis of scRNAseq data from 22 AL patient samples, we identified a total of 27 distinct clusters of cells belonging to 10 unique cell types. Out of the 27 clusters, 10 clusters were identified as PC sub-populations. The proportions of PCs in the samples negatively correlated to the other major cell types such as NK cells, T cells, gamma delta T cells and monocytes. High transcriptional variability was observed between the PC clusters of the AL patients indicating a high degree of heterogeneity among the AL PCs. Analysis of the MM scRNAseq data resulted in 14 PC clusters, however variability between these clusters was not much distinct as compared to AL. A comparison between AL and MM PC clusters resulted in overlapping transcriptional profiles of 2 AL PC clusters with one MM PC cluster. 5 out of 10 clusters of PCs in AL patients were found to be associated with protein processing in the endoplasmic reticulum and apoptosis pathway.

Summary/Conclusion:

Within the AL TME, we observed negative correlation of major immune cell types with PCs, suggesting that immune cells may contribute to controlling tumor cell burden in AL. In addition, PC sub-populations in amyloidosis patients...
are very heterogeneous at the inter-individual level compared to MM patients further indicating that AL patients may benefit from more tailored treatment approaches.