Poster 12: Mesenchymal stem cells (MSCs) from non-fibrotic and fibrotic MPN patients have distinct differentiation capacities that are confirmed by a mnltiomics study unraveling dysregulated pathways

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Introduction: BCR-ABL negative myeloproliferative neoplasms (MPN) are hematological diseases characterized by an overproduction of myeloid cells with constitutively activated JAK-STAT signaling. It is now established that mutations in the JAK2, CALR and MPL (TPOR) genes drive the onset of MPN. Polycythemia vera (PV) and essential thrombocythemia (ET) are non-fibrotic diseases but can evolve to a fibrotic stage called secondary myelofibrosis in which the bone marrow undergoes a loss ofhematopoietic function. Myelofibrosis can also occur as a primary disease (PMF). Both primary and secondary myelofibrosis are aggressive diseases, can evolve to acute myeloid leukemia and cause reduced survival.

Although the mutations of the hematopoietic compartment drive the onset of the disease in MPN, changes in the microenvironment especially in MSCs have also been reported. Hence, we launched a study on bone marrow MSCs from non-fibrotic (NF) and fibrotic (F) patients for a better understanding of the evolution ofMPN towards a fibrotic destiny.

Methods: MSCs isolated from NF and F MPN patients were studied by immunophenotyping, osteoblastic and adipocytic differentiation, RNA-Seq and ATAC-Seq. Moreover, we performed co-cultures of MPN CD34+ cells with the HS-5 stromal cell line. Finally, using a customized 16 cytokine Luminex assay and a TGF-β assay, we investigated the secretome of both MSC populations.

Results: Firstly, we show that F-MSCs differ in surface marker expression from NF-MSCs. Secondly, they show increased osteogenic and decreased adipogenic potential. By RNA-Seq we confirm upregulation of osteoblastic genes as well as TGF-β isoforms. Using DAVID analysis, we reveal several pathways differentially expressed between both MSC populations. An ATAC-Seq approach also highlights an increased accessibility to fibrosis related transcription factors. Moreover, our co-culture assay shows an upregulation of the TAGLN gene, also linked to osteoblastic differentiation, in the HS-5 cell line following coculture with both NF and F CD34+ cells. Finally, the Luminex assay shows an upregulation of PDGF-CC, VEGF and lipocalin in F-MSC supematants.

Conclusion: The study pro vides a better understanding of the mechanisms that might contribute to the evolution of a NF-MSC to a F-MSC and paves the way for exploring new therapeutic strategies.