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Next Generation Sequencing Study of Mesenchymal Stem Cells in *de novo* Myelodysplastic Syndromes

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Myelodysplastic Syndromes (MDS) are a complex spectrum of bone marrow stem cell disorders. Mesenchymal stem cells (MSCs) are considered as precursors of many cell types in marrow stroma that regulate haematopoiesis and thought to be altered in MDS. The molecular basis for altered properties of MDS derived MSCs is poorly examined. In this study, we determined the mutation profiles of *de novo* MDS- MSCs by comprehensive analysis of 54 genes by next generation sequencing on MiSeq platform (Illumina).

Our study showed that the MDS-MSCs have mutations in *DNMT3A, TET2, EZH2, ASXL1, BCOR/BCORL1* and *KDM6A* genes. These genes were found to be mutated in more than 30% of patients. *NRAS, KIT, RAD21, ATRX, KMT2A* and *CUX1* genes were also frequently mutated. Refractory anaemia with excess blasts (RAEB) showed the highest number of mutations and *KDM6A* and *KIT* were common to all RAEB patients. *ETV6, CUX1, SMC3* mutations were found in MSCs of patients with low hemoglobin levels. *KIT* and *PDGFRA* genes were observed to be co-mutated in two patients. Marrow fibrosis was observed in one RAEB patient who showed a *CALR* mutation. T665P in PDGRFA, M541L & H485P of KIT, H1367P and H1542P of BCOR, L1742W of TET2, H588P of DNMT3A and T181P and T833P of KDM6A were observed recurrently.

The study showed mutations particularly affecting the epigenetic mechanisms and signaling pathways of MSCs. The mechanisms of these mutations are yet to be elucidated. Presence of mutations in gene regions that are known to be mutated in MDS-blasts, suggest that the causative mutagen(s) may induce parallel mutations in the MSC compartment. The knowledge of the mutation patterns of both hematopoietic and mesenchymal cell compartments are equally important in understanding the disease pathogenesis and before directing patients to marrow transplantation or targeted therapies.

Keywords: Myelodysplastic syndromes (MDS), mesenchymal stem cells (MSCs), mutations, next generation sequencing.