Multipotent stem cells (MSCs) can be isolated from various sources including bone marrow. They hold huge potential for cell-based therapy for a wide array of diseases [4-7].

They are heterogeneous cell populations and isolated in homogeneity for clinical applications and basic research has been quite a challenge. There is even more differentiation into the three mesenchymal lineages, adipogenic, chondrogenic, and osteogenic cells, which has been described [4-7].

Despite the intensity of focus to theirs in regenerative medicine, their biology has not been fully described. The background in the areas of genomics, proteomics, and pharmacogenomics have offered opportunities to explore MSC’s in a systematic and integrated ways.

Studies dealing with MSCs so far have been based only on proteins that are highly abundant and are insufficient to reveal important cellular events ([4-7]).

The present work aimed to:

1. Significantly improve the large scale proteomic profiling of MSCs derived from bone marrow (hBM-MSCs)
2. Explore the results of the extensive quantitative mass spectrometric analysis of cell lines from six donors. The work highlights the proteomic basis of donor-to-donor variability of hBM-MSCs.
3. Perform an extensive bioinformatic analysis to gain insights into the highly enriched signaling events and protein-protein interaction networks that can potentially influence the biology of culture-expanded and undifferentiated hBM-MSCs.
4. Study the change due to de-differentiation processes when hBM-MSCs are subjected to two expansion passages.

The analytical workflow involves proteomic analysis: protein extraction, an overall 2D proteomic fractionation steps, and multiple MS, namely LC-ESI-MS/MS, nanoLC-ESI-MS, and LC/MALDI-MS/MS for peptide sequencing and protein identification.

**Summary of the hBM-MSCs proteome**

- Proteins that have been identified in hBM-MSCs (characterized by Table 1) are classified using cellular or subcellular localization and sexual and developmental pattern. The proteome of hBM-MSCs is highly enriched with various signaling events and protein-protein interaction networks that can potentially influence the biology of cell differentiated cells.

**Comparison between cell lines**

- The proteomic basis of hBM-MCS.

**Results**

- Quantitative profiling of the expression of proteins across three passages (P3, P7, and P14).

**Conclusions**

- The present work employed a significantly modified analytical method to sample preparation that involves pressure cycling technology and further protein fractionation using HPLC IPFREE protein fractionation system prior to proteomic analysis.

- An overall 2D proteomic fractionation with subsequent multiple complementary MS identification techniques, namely ESI-MS/MS, ESI-MS/MS, and MALDI-MS/MS, led to comprehensive indexing of proteins expressed by hBM-MSCs.

- The workflow improved the proteome coverage and 761 proteins were confidently identified (ID=1). The majority of proteins are of nuclear origin and some are predicted to appear at more than one subcellular location.

- The proteome are of various molecular classes including 50% transcription and translation regulators, 32% kinases, 11% receptors, and 3% cytoskeletal.

- The molecular profiles of hBM-MSCs across passages was investigated and the results highlighted the extensive changes in the molecular programming upon passage.

- The identified proteins could be further investigated as potential markers for hBM-MSCs.

- Overall, we present for the first time a comprehensive proteomic dataset that also contains detailed analyses to help the molecular characterization of undifferentiated and culture-expanded hBM-MSCs. This dataset could help to generate proteomic reference database for these cell types. It also serves as a basis for further exploration of hBM-MSCs with respect to self-renewal, differentiation, and characterization.

**References**