Human Mesenchymal Stromal Subcellular Composition Depends on Bone Mass Density: A Single Cell Level Study by Mass Cytometry

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Purpose: Osteoporosis is a major health concern in societies with aging populations. In general, osteoporosis is a disease characterized by low bone mineral density (BMD) and increased skeletal fragility leading to increased fracture risk, primarily in the vertebra, wrist, and the hip. Osteoporosis-related fractures will cause costs of more than $28.5 billion USD by 2025, representing a serious economical burden. Osteoporosis is caused by an activity imbalance between osteoblast-mediated bone formation and osteoclastic bone resorption processes. Various studies reported protein expression patterns that are either associated with high or low BMD. However, there is a lack of insight into the molecular and cellular biology of the bone remodeling process at single cell resolution. In this study we aimed at analyzing at single cell level by mass cytometry differences in bone marrow stromal cell subpopulations between healthy patients and patients with low BMD.

Methods: Mesenchymal stromal cells (MSCs), which are the origin of osteoblasts, were the subject of this study. MSCs were isolated from patients either with a healthy BMD (dual energy x-ray absorptiometry [DEXA] T-score >–1) or with a reduced BMD (DEXA T-score <–1). We characterized MSCs on a phenotypic level based on their in vitro differentiation potential towards osteoblasts, chondroblasts, and adipocytes as well as on a molecular level using single cell mass cytometry.

Results: We report that phenotypical characterization revealed that MSCs from healthy BMD individuals showed a full trilineage potential toward osteoblasts, chondroblasts, and adipocytes. MSCs from low BMD individuals showed strong osteoblast differentiation potential, moderate chondrogenic potential, and poor adipogenic potential, reconfirming that MSCs are highly heterogeneous. Further, a large variation in the MSC subset composition between donors with healthy BMD and reduced BMD was detected. Single cell mass cytometry revealed two distinct MSC subsets. One of the subsets is primarily present in healthy BMD individuals and the other subset is mainly present in patients with low BMD.

Conclusion: In summary, our study shows, at single cell level, a high intra- and interpatient heterogeneity in protein expression and cell composition. The molecular expression indicates that specific MSC subpopulations are involved in the changed molecular mechanisms that are associated with low BMD diseases like osteopenia or osteoporosis. On a phenotypic level, our data showed that low-BMD individuals are still capable of forming bone extracellular matrix in vitro. These observations lead to the conclusion that the treatment of osteoporosis requires an individual approach with a focus on bone resorption processes.