

Mathematical Modeling to Predict Bone Forming Potency of Human Mesenchymal Stem Cells

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INTRODUCTION: Increasing evidence supports the clinical use of mesenchymal stem cells (MSCs) for bone repair. A major problem is the structural and clinical outcome variability, which is at least partly due to donor-related factors and inconsistency of MSC preparations. In addition, MSCs isolated from different tissues have distinct differentiation properties. All this limits standardization and impedes comparison of clinical study outcomes. There is therefore a pressing clinical need for assays that allow quantitative estimation of the "potency" of stem cell preparations. Such potency assays would allow development of quality controls for efficacy in stem cell therapies.

In this proof of concept study we set out a strategy to allow prediction of bone-forming potency of human MSC preparations, independent of donor-related variables and tissue source of MSCs.

METHODS: MSCs were isolated from synovium and periosteum of adult human individuals of various ages and culture expanded. Phenotypic analysis was carried out by FACS and real-time RT-PCR. Telomere lengths were determined by Southern blot. In vitro osteogenesis was assessed quantitatively by measurements of ALP activity and calcium deposits. To investigate bone formation in vivo, MSCs were seeded onto osteoinductive scaffolds and implanted subcutaneously in immunodeficient mice. Bone was assessed by histology and the human origin investigated by in situ hybridization for human Alu genomic repeats. Quantitation was achieved by histomorphometry and real-time RT-PCR for human bone markers. Analysis at the single-cell level was performed with clonal populations obtained by limiting dilution. Multiple regressions were used to explore the incremental predictive value of the markers.

RESULTS: We first quantified bone-forming potency of matched human MSCs from

synovium and periosteum using in vitro and in vivo osteogenesis assays. Notably, the outcomes of these two independent assays were tightly correlated. Second, we analyzed the sources of variability in osteogenic outcome. We identified the tissue of origin of MSCs as the main source of variability, since MSCs from periosteum had significantly greater osteogenic potency than MSCs from synovium inherent to the single multipotent MSC. A second component of variability was donor-related. Third, to identify predictors of osteogenic potency of MSC preparations, we measured the expression levels of osteoblast-lineage genes in synovial and periosteal clonal MSCs prior to osteogenic treatment. We identified biomarkers that correlated with osteogenic outcome and developed a mathematical model that allows accurate prediction of bone-forming potency of human MSC preparations, independent of donor-related variables and tissue source.

DISCUSSION & CONCLUSIONS: This quality-control mathematical model could be used to estimate bone-forming potency of human MSC preparations for bone repair. We anticipate that such a strategy based on potency assays and related quality controls will enhance the consistency of results, will allow standardized comparison of outcomes from different clinical studies, and will increase the chances of success of individual clinical trials. In addition, it will provide solid ground to establishing batches of certified MSC products with specific clinical indications, which will then be investigated for allogeneic transplantation. This will increase consistency and decrease costs of stem cell therapies.

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