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Chondrogenic Potential Of Haemarthrosis-Derived Mesenchymal Stem Cells For Cartilage Repair After Abrasion Arthroplasty

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Summary:
Recently we have shown that bone marrow stimulation technique reveal different chondrogenic growth factors and MSCs into the postoperative haemarthrosis. These MSCs are able to differentiate into chondrocyte-like cells and produce a collagen proteoglycan rich matrix in pellet-culture. Our study provides the evidence for a therapeutic benefit of opening bone marrow for cartilage repair.

Abstract:
Objectives: Mesenchymal stem cells (MSC) from bone marrow are a very attractive tool in the context of repair and regeneration of cartilage lesion. Arthroscopic treatment of OA includes bone marrow stimulation technique such as abrasion arthroplasty (AAP) and microfracturing (MF). These procedures are proved to stimulate cartilage regeneration. Recently we have shown the release of growth factors such as insulin-like growth factor-1 (IGF-1) and transforming growth factor beta-1 (TGF-ß1) in the postoperative haemarthrosis depending on the choice of arthroscopic procedure. They play a pivotal role in the regeneration process of chondral defects and chondrogenic differentiation of MSC. The aim of the current study was to characterize the mononuclear cells after bone marrow stimulation techniques and to determine their regeneratory potential.
Methods: Haemarthrosis was collected from the drainage bottle 22 hours (h) (n=164) and one week (n=10) after different arthroscopic knee procedures. Mononuclear cells were isolated by ficoll density gradient centrifugation. Adherent cells were were characterized using fluorescence-activated cell-sorting (FACS) analysis, immunohistochemistry (IHC) and immunofluorescence for characteristic stem cell markers. Thereafter, MSC were seeded in a high-density culture. To determine the chondrogenic potential of different serum media, MSC were either cultured with basal stimulation medium, chondrogenic differentiation medium or with haemarthrosis-serum. The 3D cell pellet was characterized (e.g. collagen type II, chondroitin-4-sulfate, SOX-9) by IHC.
Results: After 22 h AAP release more cells comparing to chondral procedures (CP) in the haemarthrosis while 10 days after AAP most cells were countered. Their morphology changed from spindle-shaped fibroblast like cells to rounded chondrocyte-like-cells. Using FACS analysis haemarthrosis-derived cells 22 h after AAP and CP are positive for CD 44, 73, 90, 105 and negative for 34. In contrast to MSC after AAP, MSC after CP do not proliferate wich is negative for the proliferation marker Ki-67. In a high density culture, MSC differentiate to a chondrocyte-like cell type and produce an extracellular matrix which is positive for SOX-9, chondroitin-4-sulfate and collagene type II. Comparing the chondrogenic potential of different cell media after co-culturing with MSC no differences regarding their number and quality were observed.
Conclusions: Interestingly, mononuclear cells after solely chondral procedures display characteristic MSC markers but do not proliferate in a high density culture and thereby explain the poor clinical results after chondral procedures. This study provided evidence that haemarthrosis-derived cells after AAP and MF can differentiate to chondrogenic lineage cells in vitro. The morphological convergence of the cell culture to hyaline cartilage dependent on the kind of arthroscopic procedure underlines the benefit of bone marrow stimulating techniques in the arthroscopic treatment of cartilage defects.