Soluble Factors Released from Articular Chondrocytes Reduce Hypertrophy of Mesenchymal Stem Cells and Matrix Calcification During Chondrogenesis

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Introduction
Bone marrow-derived mesenchymal stem cells (MSC) are a promising cell source for cell-based cartilage repair. A yet unsolved problem, however, is the unwanted up-regulation of hypertrophic markers such as alkaline phosphatase (ALP) and collagen type X during in vitro chondrogenesis of MSC. After ectopic transplantation into SCID mice MSC form only transient, calcified cartilage whereas articular chondrocytes form stable ectopic cartilage without calcification. Aim of this study was to address whether coculture with articular chondrocytes has the capacity to suppress undesired hypertrophy in differentiating MSC.

Materials and Methods
Differentiation of MSC was induced in chondrogenic medium which had or had not been conditioned by parallel chondrocyte pellet cultures. Alternatively, MSC were mixed with chondrocytes (1:1, 1:2) and cultured within the same pellet for 6 weeks. Following in vitro differentiation, pellets were transplanted into SCID mice.

Results
The gene expression ratio of COL10A1/COL2A1 and COL1A1/COL2A1 was significantly reduced by chondrocyte-conditioned medium while MMP13/COL2A1 remained unchanged. Western blot analysis of collagen lysates of MSC pellets cultured with conditioned medium revealed a lower collagen type X protein deposition relative to collagen type II compared to the control group. ALP-activity was significantly lower (1.9-fold, p<0.05) in the conditioned-medium group and semiquantitative histological scoring of respective transplants showed significantly reduced calcification in vivo. For mixed chondrocyte/MSC pellets, a dose-dependent suppression of ALP activity and an almost complete inhibition of in vivo calcification occurred.

Discussion and Conclusions
Soluble factors released by chondrocytes and direct co-culture suppressed hypertrophy of MSC during chondrogenesis and stabilized the chondrogenic phenotype of MSC in vivo.

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Disclosures
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