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Title: Identification of TGF $\beta$ -related genes regulated in murine osteoarthritis and chondrocyte hypertrophy by comparison of multiple microarray datasets

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Abstract: Objective: Osteoarthritis (OA) is a joint disease characterized by progressive degeneration of articular cartilage. Some features of OA, including chondrocyte hypertrophy and focal calcification of articular cartilage, resemble the endochondral ossification processes. Alterations in transforming growth factor  $\beta$  (TGF $\beta$ ) signaling have been associated with OA as well as with chondrocyte hypertrophy. Our aim was to identify novel candidate genes implicated in chondrocyte hypertrophy during OA pathogenesis by determining which TGF $\beta$ -related genes are regulated during murine OA and endochondral ossification.

Methods: A list of 580 TGF $\beta$ -related genes, including TGF $\beta$  signaling pathway components and TGF $\beta$ -target genes, was generated. Regulation of these TGF $\beta$ -related genes was assessed in a microarray of murine OA cartilage: 1, 2 and 6 weeks after destabilization of the medial meniscus (DMM). Subsequently, genes regulated in the DMM model were studied in two independent murine microarray datasets on endochondral ossification: the growth plate and transient embryonic cartilage (joint development).

Results: A total of 106 TGF $\beta$ -related genes were differentially expressed in articular cartilage of DMM-operated mice compared to sham-control. From these genes, 43 were similarly regulated during chondrocyte hypertrophy in the growth plate or embryonic joint development. Among these 43 genes, 18 genes have already been associated with OA. The remaining 25 genes were considered as novel candidate genes involved in OA pathogenesis and endochondral ossification. In supplementary data of published human OA microarrays we found indications that 15 of the 25 novel genes are indeed regulated in articular cartilage of human OA patients.

Conclusion: By focusing on TGF $\beta$ -related genes during OA and chondrocyte hypertrophy in mice, we identified 18 known and 25 new candidate genes potentially implicated in phenotypical changes in chondrocytes leading to OA. We propose that 15 of these candidates warrant further investigation