Expression of Bone Morphogenetic Protein, Receptors, and Tissue Inhibitors in Human Fetal, Adult, and Osteoarthritic Articular Cartilage

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Introduction: Recent interest in the role of BMPs in development, growth, and fracture repair has led to the realization that coordinate expression of BMPs, BMP receptors, and BMP inhibitors may be necessary for physiologic BMP regulation and activity. The purpose of this investigation was to characterize the expression of BMPs, BMP receptors, and BMP inhibitors in human fetal and adult articular cartilage.

Methods: Adult articular cartilage was obtained from the knees of eight patients (average age 68.4 years; range 59-76 years) undergoing elective total knee arthroplasty for end-stage osteoarthritis. Fetal articular cartilage was obtained from the distal femora of four electively aborted fetuses (average gestational age, 21.2 weeks; range 19-23 weeks) without evidence of musculoskeletal abnormalities. As a control, normal adult articular cartilage was obtained from the femoral heads of three patients (average age 66.7 years; range 63-74 years) during hip hemiarthroplasty for proximal femoral fracture; these were intraoperatively without evidence of osteoarthritis. Total RNA extraction was performed by acid-guanidium thiocyanate-phenol-chloroform extraction followed by column purification. Following construction of cDNA by reverse transcription, PCR was performed using designed primers for BMPs 1-11, BMP receptors (IA, IB, and II), TGFβ1, TGFβ2, BMP inhibitors noggin and follistatin, CDMP-1, COMP, and GAPDH. The PCR-amplified sequences were then resolved by agarose-ethidium bromide gel electrophoresis. Cartilage protein extracts were separated by SDS-PAGE and Western Blotting using their specific antibodies examined the level of BMP3, BMP7 and BMP8.

Results: Systematic RT-PCR assay (Fig. 1) revealed that BMPs 1, 2, 4-6, and 11, BMPR-IA and II, noggin, follistatin, CDMP-1, COMP, and GAPDH mRNAs were expressed in relatively similar fashion in both the fetal and adult (normal and osteoarthritic) cartilage samples. BMPs 9 and 10 mRNAs were not expressed in either group, although control samples were positive, ensuring integrity of the experimental system. BMPs 7, and 8, and BMPR-IB mRNAs were consistently expressed in fetal but not adult (normal or osteoarthritic) articular cartilage. BMP 3 mRNA was expressed in all fetal and normal adult articular cartilage, but not osteoarthritic samples. TGF β1 was consistently expressed in both adult normal and osteoarthritic cartilage, but not in fetal samples. In line with the mRNA expression pattern, Western Blotting analysis (not shown) showed that BMP 7 and BMP 8 are positive in fetal articular cartilage but not adult samples (normal and osteoarthritic). In the case of BMP 3 protein, it was present in fetal cartilage and also detectable with lower level in adult normal tissues, but absent in osteoarthritic samples.

Discussion and Conclusions: The presence of BMPs 7 and 8 in fetal but not adult articular cartilage supports their roles in skeletal development and chondrogenesis. TGF β1 and β2 are known to be produced by chondrocytes of articular cartilage, serving as protective cytokines that inhibit the degradatory effects of IL-1 and enhance synthesis of matrix components, such as proteoglycan. Noggin and follistatin were highly expressed in both groups; this in addition to the consistent expression of BMPs 1, 2, 4-6, and 11, and BMPR-IA and II in both groups suggest that a major contributor to BMP regulation may occur at the extracellular level, with direct inhibition through specific binding of the receptor, BMP-receptor complex, or of the growth factor itself. Further investigation is necessary to elucidate the role of BMP-3, which was expressed in fetal and normal adult but not osteoarthritic samples, as well as the spatial and temporal patterns of expression that may influence the metabolic or pathogenic roles of the BMPs.