Molecular Mechanisms of the Difference in Osteoarthritis Susceptibility between Human Knee and Ankle Articular Cartilage

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Abstract

Clinically, the incidence of osteoarthritis (OA) differs greatly in knee cartilage compared to ankle cartilage. The knee joint is more susceptible to OA than the ankle, and several anatomical and clinical differences exist between the two tissues. For example, the thickness of articular cartilage is greater in the knee (~3-6.6mm) than the ankle (~1-1.7 mm). Further, different types of OA characterize human knee and ankle cartilage; in the knee joint, degenerative OA is the most common type, while in the ankle, traumatic arthritis predominates. However, the difference in OA prevalence between the two joints has not yet to be adequately addressed at a molecular level.

In OA, there is a disruption of matrix equilibrium leading to progressive loss of cartilage tissue and clonal expansion of cells in the depleted regions. Chondrocyte metabolism is unbalanced due to excessive production of catabolic factors, including matrix metalloproteinases (MMPs), aggrecanases (ADAMTS), and other cytokines and growth factors released by chondrocytes that aid in the destruction of proteoglycans and the ECM(2).

PGE2 and its specific receptors EP2/4 have been associated with degenerative cartilage via loss of proteoglycan and suppression of aggrecan expression(3). Asporin, a member of the small leucine-rich proteoglycans(SLRPs) binds to and inhibits transforming growth factor-β (TGF-β) and illustrates a possible pathogenic role in OA via inhibition of TGF-β-induced chondrogenesis(5).

Our findings demonstrate striking differences between knee and ankle cartilage at the molecular level. Several mediators of articular cartilage homeostasis are increased in knee compared to ankle cartilage, suggesting a link between these mediators and a faulty cartilage repair system, thus inducing cartilage degeneration and offering a biomolecular explanation for the difference in OA susceptibility between knee and ankle cartilage. Knee ability to repair is less than that of the ankle.

Materials and Methods

Adult articular cartilage was obtained from knee and ankle joints of asymptomatic human organ donors with no history of joint diseases (Gift of Hope Organ & Tissue Donor Network). Tissues were age and grade-matched. Human adult articular chondrocytes were isolated from tissue donor (grade 0-1) cartilage and were cultured in monolayer (short-term) or in alginate beads (long-term for 21 days; analyzed for PG accumulation by DMMB assay). Total tissue and cellular RNA isolation was performed using Trizol reagent and reverse transcription was carried out using ThermoScript system (Invitrogen). For real-time PCR, equal amount cDNA was amplified using Bio-Rad MyiQ Real-Time PCR Detection System. iQ5 Optical System Software was used for data analysis. Asporin, EP2, and ADAMTS5/4 gene expression levels were analyzed. Analysis of variance was performed using StatView 5.0 software. P values <0.05 were considered significant.

Results

Fig 1 Dimethylthylene Blue (DMMB) assay for proteoglycan production shows that PGE2 and EP2 receptor specific agonist (Butaprost) significantly suppress PG accumulation per cell in the cell-associated matrix of knee, but not ankle, articular chondrocytes.

Fig 2 The relative expression of ADAMTS4,5 in normal human adult knee chondrocytes is more predominant than in normal adult ankle chondrocytes.

Fig 3 PGE2 combined with IL-1 synergistically upregulates IL-6 mRNA levels in knee chondrocytes. Synergistic augmentation of iNOS was also observed when knee chondrocytes were stimulated with a combination of PGE2 and IL-1, albeit that iNOS mRNA expression was modestly reduced after treatment with PGE2 alone.

Fig 4 There is a significant difference in asporin gene expression between knee and ankle joint cartilage. Asporin expression in normal human adult knee cartilage is roughly 20-fold greater than normal adult ankle cartilage, suggesting that increased asporin levels in the knee may contribute to inhibition of TGF-β-induced chondrogenesis, thus enhancing cartilage loss with increasing age or degeneration in knee cartilage compared to ankle cartilage.

Conclusion

Our findings demonstrate striking differences between knee and ankle cartilage at the molecular level. Several mediators of articular cartilage homeostasis are increased in knee compared to ankle cartilage, suggesting a link between these mediators and a faulty cartilage repair system, thus inducing cartilage degeneration and offering a biomolecular explanation for the difference in OA susceptibility between knee and ankle cartilage. Knee ability to repair is less than that of the ankle.

Acknowledgements

This work was founded by NIH RO1 (AR0533220), NIH training grant 2T-AR-007590, Arthritis Foundation, National Arthritis Research Foundation. Special thanks to the Gift of Hope Organ and Tissue Donor Network and Dr. Margulis for donor tissues.

Reference


Paper No. 286 • 55th Annual Meeting of the Orthopaedic Research Society