Figure 1

Figure 2

Figure 3

Conclusions: In this first study of Gd-DTPA²⁻ transport into meniscal tissue, the diffusion pattern was very similar between the meniscus and articular cartilage, with a continuous wash-in until approximately 90 minutes after injection. This is beneficial in a clinical perspective because both structures can be analyzed with dGEMRIC at the same time after contrast injection. Notably, the concentration of Gd-DTPA²⁻ was twice as high in the meniscus as in the cartilage, as shown from the Delta R¹ values. This may reflect that the meniscus contains less GAG than articular cartilage, a fact supported by several in vitro studies of canine and human meniscus. The higher Gd-DTPA²⁻ concentration in the meniscus could also be explained by a larger contact area with synovial fluid, i.e., that the diffusion occurs from both sides of the meniscus. In vitro experiments with higher resolution (higher field strength) are needed to confirm this hypothesis. Previously, a linear dose-response relationship has been shown in femoral knee cartilage [4]. The present study indicates that this may be true also for the meniscus. However, the drop in T¹ after contrast injection was highly significant already with the double dose. Our T¹ data at 90 minutes after the double dose are similar to what has been previously reported in human menisci by Krishnan et al. [5]. Future studies need to investigate the clinical relevance of dGEMRIC of the meniscus. However, the preliminary data presented by us and others [5] indicate that the method is clinically feasible. The lower T¹ in lateral compared to medial meniscus was unexpected but may reflect structural differences.

487

EXPRESSION PROFILING OF SKELETOGENIC-RELEVANT PATHWAYS REVEALS CONTRASTING PATHOGENETIC MECHANISMS IN MENISCAL RNA FROM OSTEOARTHRITIS VERSUS CHONDROCALCINOSIS PATIENTS
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Purpose: The purpose of this study was to assess the expression of ANK, and other skeletogenic transcripts, in the tissue that most often harbors crystals in individuals affected with idiopathic CPPD disease - the meniscus of the knee - and to contrast our findings with those in menisci from osteoarthritis patients without CPPD disease.

Methods: Menisci from total knee replacement (TKR) patients were obtained post surgery. Tissues sections were subjected to alizarin red staining to identify mineralized specimens and the chemical nature of the mineral was confirmed by FTIR analysis. For gene expression profiling, osteoarthritis patient samples included 3 with CPPD deposition and 3 without CPPD deposition. RNA was isolated from menisci by pulverization under liquid nitrogen followed by RNA extraction using the RNeasy Lipid tissue kit (Qiagen). cDNA was synthesized from the isolated RNA and used in qRT-PCR analyses using macroarrays specific for pathway analyses of osteogenesis and chondrogenesis transcripts (SA Biosciences). Immunohistochemical staining for proteins whose transcripts showed differences among OA/CPPD vs. OA menisci was performed on tissue sections from 12 TKR patients, 6 of whom suffered from FTIR- confirmed CPPD deposition.

Results: Contrary to our expectation, the expression of ANK, a gene whose over-expression and mutation is associated with CPPD mineralization, was not elevated in OA/CPPD menisci. Array transcripts included those for cell growth and differentiation, extracellular matrix and cell adhesion molecules, bone mineral metabolism, and mineralization. Among those transcripts that displayed elevated levels of expression in OA/CPPD menisci compared to OA menisci was osteopontin (OPN), MEPE, and MMP14. Expression of the matrix molecules osteonectin and decorin were significantly depressed in OA/CPPD menisci vs. OA specimens, as were transcripts important to chondrocyte differentiation, including SOX9 and CDH11. We performed immunohistochemical analyses of OPN expression in tissue sections from 12 TKR patients, 6 of whom suffered from FTIR- confirmed CPPD deposition.

Conclusions: Significant differences in the expression of several skeletogenic genes and their products were observed in OA/CPPD menisci compared to OA menisci was osteopontin (OPN), MEPE, and MMP14. Expression of the matrix molecules osteonectin and decorin were significantly depressed in OA/CPPD menisci vs. OA specimens, as were transcripts important to chondrocyte differentiation, including SOX9 and CDH11. We performed immunohistochemical analyses of OPN expression in tissue sections from OA/CPPD vs. OA menisci. In a blind screen of OPN staining of the sections, menisci containing CPPD crystals demonstrated significantly elevated expression of OPN (p<0.0001).

Conclusions: Significant differences in the expression of several skeletogenic genes and their products were observed in OA/CPPD menisci vs. OA menisci, suggestive of distinct pathogenetic etiologies between the two conditions. Furthermore, the observed expression profile was consistent with the non-endochondral ossification nature of CPPD deposition. Of particular interest was the fact that OA/CPPD meniscal specimens did not express aberrant levels of ANK, but rather displayed striking differences in the expression of OPN, a secreted phosphoprotein whose expression has previously been shown to facilitate CPPD deposition in vitro.