Conclusions: Preliminary data of this study suggest that CS alone or in combination with GS is able to decrease significantly synovitis as measured by US. The decrease in synovitis after SYSADOA treatment has been correlated with an improvement in knee pain. The decrease in synovitis elicited by SYSADOA treatment has been correlated with an improvement in knee pain. The decrease in synovitis as measured by US. The decrease in synovitis after SYSADOA treatment has been correlated with an improvement in knee pain.

045

SERUM LEVELS OF INFLAMMATORY MARKERS, KNEE RADIOGRAPHIC OSTEOARTHRITIS, AND KNEE CARTILAGE LOSS IN OLDER ADULTS

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Purpose: To determine the associations between serum levels of inflammatory markers, radiographic osteoarthritis (OA) and knee cartilage loss over time in older adults.

Methods: A total of 193 randomly selected subjects (mean 63 years, range 51-78, 47% female) were studied at baseline and 2.9 years later. Serum high-sensitivity C-reactive protein (hs-CRP), interleukin 6 (IL-6) and tumor necrosis factor α (TNF-α) were assessed by radioimmunoassay. T1-weighted fat-suppressed MRI of the right knee was performed to determine knee cartilage volume. Kneec radiographic osteoarthritis (ROA) was also assessed.

Results: At baseline, quartiles of IL-6 and TNF-α were associated with increased prevalence of medial tibiofemoral joint space narrowing (grade ≥1) in multivariable analyses (IL-6: OR=1.41 and 1.49 per quartile; TNF-α: OR=1.62 and 1.49 per quartile for right and left knees, respectively; all P <0.05). Longitudinally, baseline IL-6 (not TNF-α) predicted loss in both medial and lateral tibial cartilage volume (β: -0.87% and -1.02% per annum per quartile, P<0.05 and P<0.01, respectively). An increase in IL-6 was associated with increased loss of medial and lateral tibial cartilage volume (β: -0.89% and -0.78% per annum per quartile, both P<0.05), and change in TNF-α was also negatively associated with change in medial and lateral tibial cartilage volume (β: -0.75% and -0.80% per annum per quartile, both P<0.05). The associations of IL-6 and TNF-α were dependent, in part, on each other except those between baseline IL-6 and cartilage loss. Serum hs-CRP was not associated with ROA and cartilage loss.

Conclusions: Serum levels of IL-6 and, to a lesser extent, TNF-α are associated with knee cartilage loss in older people suggesting low level inflammation plays a role in the pathogenesis of knee OA.

046

RACIAL DIFFERENCES IN OSTEOARTHRITIS PAIN AND FUNCTION: POTENTIAL EXPLANATORY FACTORS

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Purpose: African Americans with osteoarthritis (OA) report greater pain and functional limitations than Caucasians, but the reasons for these racial disparities are not well understood. This study examined factors underlying racial differences in pain and function among patients with hip and/or knee osteoarthritis (OA), including psychological variables that have not previously been examined in this context.

Methods: Participants were n=491 African Americans and Caucasians enrolled in a clinical trial of telephone-based OA self-management. All measures were obtained at baseline. Pain and function were assessed using the Arthritis Impact Measurement Scales-2 (AIMS2) subscales. Potential explanatory variables included demographic factors (age, gender, marital status, education, income, working status, health literacy), health-related factors (body mass index, self-reported health, joint(s) with OA, duration of OA symptoms, pain medication use, current exercise), and psychological factors (arthrits self-efficacy, problem- and emotion-focused pain coping, AIMS2 affect subscale). The AIMS2 pain subscale was also included in models of AIMS2 function. Potential explanatory variables that were associated with both race and pain or function, and that reduced the association of race with pain or function by ≥10%, were included in the final multivariable linear regression models where racial differences in AIMS2 pain and function scores were examined.

Results: In unadjusted linear regression models, African American race was associated with higher (worse) AIMS2 pain scores (unadjusted B = 0.65, p<0.01) and higher (worse) AIMS2 function scores (unadjusted B = 0.59, p<0.01). African American race was no longer significantly associated with AIMS2 pain scores in the final regression model (B = 0.03, p=0.874); variables significantly associated with worse AIMS2 pain scores in that model were: worse AIMS2 affect scores, greater emotion-focused coping, lower arthritis self-efficacy, and fair or poor self-reported health. African American race was also no longer significantly associated with AIMS2 function scores in the final regression model (B = 0.07, p=0.509). Variables significantly associated with worse AIMS2 function scores were the same as described above for AIMS2 pain scores. In addition, worse AIMS2 pain scores were significantly associated with worse AIMS2 function scores.

Conclusions: Factors explaining racial differences in pain and function were largely psychological, including AIMS2 affect (which comprises mood and level of tension), use of emotion-focused coping, and arthritis self-efficacy. Self-management and psychological/behavioral interventions can influence these factors, and greater dissemination of these programs among African Americans may be a key step toward reducing racial disparities in OA-related pain and function.

047

DIFFERENTIAL REGULATION OF THE INSULIN-LIKE GROWTH FACTOR BINDING PROTEIN-5 AND MATRIX METALLOPROTEASE-13 GENES BY THE MICRONAS MIR-140 AND MIR-27A IN HUMAN OSTEOARTHRITIC CHONDROCYTES

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Purpose: MicroRNAs are small non-coding RNAs that regulate target messenger RNA. We investigated the implication of two miRNAs, miR-140 and miR-27a, in MMP-13 and IGFBP-5 gene regulation in human osteoarthritic (OA) chondrocytes.

Methods: Gene expression was determined by real-time PCR. The effect of each miRNA on IGFBP-5 and MMP-13 expression/production was evaluated by transiently transfecting their precursors (pre-miRNAs) and inhibitors (anti-miRNAs) into human OA chondrocytes. Modulation of IGFBP-5, miR-140 and miR-27a
expression was determined upon treatment of OA chondrocytes with cytokines and growth factors.

**Results:** IGFBP-5 was expressed in human chondrocytes with levels significantly \( p<0.04 \) lower in OA. Computational algorithms identified miR-140 and miR-27a as possible regulators of MMP-13 and IGFBP-5 expression. Data showed that both miRNAs were expressed in chondrocytes. There was a significant (\( p<0.01 \)) reduction in miR-140 expression in OA, whereas miR-27a expression was only slightly decreased. Transfection with pre-miR-140 significantly \( p<0.005 \) decreased IGFBP-5 expression at 24 hours, while pre-miR-27a did not affect either MMP-13 or IGFBP-5. Treatment with anti-miR-27a, but not with anti-miR-140, significantly increased the expression of both MMP-13 \( p<0.05 \) and IGFBP-5 \( p<0.01 \) after 72 hours incubation. These data indicate that IGFBP-5 is a direct target of miR-140, whereas miR-27a indirectly down-regulates both MMP-13 and IGFBP-5. TGF-\( \beta \) down-regulates miR-140 and up-regulates IGFBP-5.

**Conclusions:** This study is the first to show the involvement of miRNAs in human OA chondrocytes. Their effect on two genes involved in OA pathophysiology adds another level of complexity to gene regulation, which could open up novel avenues in OA therapeutic strategy.

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**048**

**EVIDENCE FOR A ROLE OF DICKOPPFP-3 IN THE PATHOGENESIS OF OSTEOARTHRITIS**

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**Purpose:** To identify genes showing altered expression in osteoarthritic cartilage and synovium. Dkk3, a member of the Dickopp family of Wnt signalling antagonists was overexpressed and this work highlights the potential function of Dkk3 in OA.

**Methods:** Real-time PCR was used to compare expression of 270 cytokines, chemokines and their receptors in cartilage and synovium from OA and non-OA patients. Expression of Dkk3 during chondrogenic differentiation of ATDC5 cells was also assessed using real-time PCR and compared to ATDC5 cells not stimulated to undergo chondrogenesis. The effect of TGF\( \beta \) on Dkk3 expression in chondrocytes was measured following treatment of SW1353 cells with TGF\( \beta \). To assess the distribution of Dkk3 in OA cartilage immunohistochemistry was carried out on articular cartilage specimens. The level of Dkk3 in synovial fluid tricompartmental and unicompartamental cartilage lesions was measured using ELISA.

**Results:** Several components of the Wnt pathway showed altered expression between normal and OA tissue. Strikingly, the greatest and most significant change was in Dkk3 expression, with a 10-fold increase in OA cartilage \( p=0.0001 \) and a 3.5-fold increase in OA synovium \( p=0.007 \) when compared to respective control tissues. Dkk3 expression decreased during chondrogenic differentiation of ATDC5 cells and to be decreased following TGF\( \beta \) treatment of SW1353 cells.

Immunohistochemical analysis of specimens from individuals with articular cartilage degeneration (AMG, a subtype of knee OA), demonstrated Dkk3 protein in chondrocytes of the superficial zones of the cartilage, but not the deeper zones. Additionally there is an increase in Dkk3 expression in superficial zone chondrocytes in damaged when compared to undamaged cartilage from within the same knee. Using ELISA we have shown increased Dkk3 protein in the synovial fluid of individuals with tricompartmental OA \( n=4 \) versus those with unicompartamental cartilage lesions \( n=10 \) (182ng/ml v 116 ng/ml, \( p<0.01 \), a single non-OA control synovial fluid measured 43ng/ml.

**Conclusions:** Dkk3 is a molecule with poorly ascribed function, especially within the musculoskeletal system. In contrast to other members of the Dkk family, Dkk3 does not act consistently as a Wnt antagonist. Literature on a number of tumour-derived cells shows that Dkk3 can potentially regulate Wnt, TGF\( \beta \), BMP, FGF and Activin signalling and cell proliferation and apoptosis. These cellular processes are highly relevant to OA. In this preliminary study we have shown that Dkk3 is overexpressed in OA cartilage and synovium. Interestingly, the increased expression of Dkk3 is also detected at the protein level in the synovial fluid; with increased Dkk3 present as disease severity increases. Furthermore Dkk3 protein in cartilage is detected more highly in damaged articular cartilage when compared to macroscopically normal cartilage. The decreased expression of Dkk3 during chondrogenesis is suggestive of a role of Dkk3 not only in articular cartilage maintenance but also in development. We have also seen decreased expression of Dkk3 in response to TGF\( \beta \) treatment. Our current work aims to identify the function of Dkk3 in articular chondrocytes, the signaling pathways activated by Dkk3 and the mechanisms causing increased Dkk3 expression in OA.

Dkk3 is an intriguing molecule in the Wnt signalling field since the evidence for its function as a Wnt antagonist is at best equivocal. Evidence for a role for Dkk3 in OA is compelling. Dissecting its function in cartilage and OA will both shed light on its overall molecular mechanism of action, enhance our knowledge of the pathogenesis of OA and also uncover potential new therapeutic targets for chondroprotection.