Australia; ‖ Repatriation Gen. Hosp., Adelaide, Australia; ¶ Univ. of Adelaide, Adelaide, Australia

Purpose: Increasing evidence suggests associations between osteoarthritis (OA) and the metabolic syndrome and its components. However, these associations have been questioned with respect to their independence from obesity. Using a prospective cohort study, we examined whether individual components of the metabolic syndrome, either singly or additively, were associated with the incidence of total knee and hip replacement due to severe OA, and whether the associations were independent of obesity.
Methods: Eligible participants were selected from 21,837 Melbourne Collaborative Cohort Study (MCCS) participants who were recruited in 1990-1994 and had fasting blood lipids measured during 2003-2007. Waist circumference, height, weight and blood pressure were also measured during 2003-2007. Metabolic syndrome was defined using the International Diabetes Federation definition: central obesity (defined by waist circumference) and any two of the four factors - raised serum triglyceride level, reduced serum high-density lipoprotein cholesterol level, hypertension and impaired fasting glycaemia. Primary knee and hip replacements for OA from the date of lipid measurement until 2011 were determined by linking the MCCS records to the Australian Orthopaedic Association National Joint Replacement Registry.
Results: 685 participants had total knee replacement and 580 participants had total hip replacement. Each of the 5 components of the metabolic syndrome and the metabolic syndrome itself were associated with increased incidence of knee OA, adjusted for age, gender, country of birth, and level of education. After including body mass index in the regression models, the magnitude of the HRs attenuated, and central obesity (hazard ratio (HR) 1.51, 95% confidence interval (CI) 1.20-1.91], hypertension (HR 1.25, 95% CI 1.05-1.50) and the metabolic syndrome (HR 1.25, 95% CI 1.01-1.54) remained statistically significant. There was a dose-response relationship between each additional component of the metabolic syndrome and the incidence of knee OA, independent of body mass index: one component HR 2.11 (95% CI 1.11-4.01), two components HR 2.94 (95% CI 1.57-5.52), three or more components HR 3.13 (95% CI 1.66-5.91), p for trend <0.001. Only central obesity was associated with the incidence of hip OA when adjusted for age, gender, country of birth, and level of education, but this association disappeared when body mass index was added to the model. Individual components of the metabolic syndrome were not additively associated with the incidence of hip OA.
Conclusion: Our results extend previous studies by relating the individual components of the metabolic syndrome and their total to the risk of knee and hip OA in the same large cohort independent of obesity. The risk of knee OA was associated singly and additively with components of the metabolic syndrome and the metabolic syndrome as a whole. In contrast, no relationship was seen for hip OA. The findings suggest different pathogenesis for OA of the knee and hip and that management of the metabolic syndrome has the potential to reduce the risk of knee OA.

436
SOCS-3 EXPRESSION IN THE CARTILAGE IS REDUCED IN OBESE PATIENTS WITH OSTEOARTHRITIS AND REGULATES LEPTIN RESPONSES IN CHONDROCYTES

A. Koskinen [†], K. Vuolteenaho [†], R. Korhonen [†], T. Moilanen [‡,†], E. Moilanen [†]. [†] *The Immunopharmacology Res. Group, Univ. of Tampere Sch. of Med. and Tampere Univ. Hosp., Tampere, Finland;* [‡] *Coxa Hosp. for Joint Replacement, Tampere, Finland*

Background and Purpose: Adipocytokine leptin was originally discovered in white adipose tissue and found to regulate energy metabolism and appetite. More recently it has been recognized as an effector and regulator in inflammation and arthritis. We and others have shown that leptin has detrimental effects on cartilage metabolism including upregulation of proinflammatory and catabolic factors in OA cartilage. However, there are significant differences in the leptin responsiveness in the cartilage between different donor patients. Suppressor of cytokine signaling 3 (SOCS-3) is an intracellular regulator of inflammatory response and a negative regulator of leptin signaling in the hypothalamus. It has also been shown to be expressed in chondrocytes and overexpression of SOCS-3 has been reported to reduce severity of arthritis in mice models. The aim of the present study was to investigate the regulation of leptin responses in the OA cartilage, especially the role of SOCS-3.
Methods: Synovial fluid and cartilage samples were collected from 91 OA patients [age 70.2 (9.6) years, BMI 30.8 (5.8) kg/m2; mean (sd); females 66%] undergoing knee replacement surgery. Leptin, MMP-1 and MMP-3 in synoval fluid were measured by immunoassay. SOCS-3 expression in cartilage was determined in a sub-group of the patients with Western blotting. The results were analyzed in the whole group and in non-obese (BMI less than 30, n=45) and obese (BMI over 30, n=46) patients separately. In addition, the leptin effects were studied in chondrocyte cultures. SOCS-3 expression was down-regulated with siRNA and interleukin-6 (IL-6), MMP-1, MMP-3 and inducible nitric oxide synthase (iNOS) expression was measured by RT-PCR, ELISA and Western blotting.
Results: Leptin concentrations in synovial fluid were higher ($p < 0.001$) and SOCS-3 expression in cartilage samples was lower ($p = 0.032$) in obese than in non-obese patients. Leptin correlated positively with MMP-1 and MMP-3 levels in synovial fluid from obese ($r = 0.49$, $p = 0.001$; $r = 0.48$, $p = 0.001$, respectively) but not from non-obese patients. Also, SOCS-3 levels in the cartilage correlated negatively with synovial fluid MMP-1 and MMP-3 (r=-0.49, p=0.013; r=-0.44, p=0.024, respectively). Leptin enhanced MMP-1, MMP-3, IL-6 and iNOS expression in chondrocyte cultures. Interestingly, when SOCS-3 was down-regulated by small interfering RNA, chondrocytes' response to leptin was enhanced.
Conclusion: The results show, for the first time, that SOCS-3 is associated with and regulates leptin-induced responses in the cartilage; When SOCS-3 expression was down-regulated, leptin-induced effects were enhanced. In OA patients high leptin levels and low SOCS-3 levels were associated with increased concentrations of cartilage degrading MMPs and obesity. Assuming that SOCS-3 is a factor that inhibits the effects of leptin in cartilage, obese patients are possibly more susceptible to detrimental effects of leptin not only because of its elevated levels in joint, but also because of the disturbed regulation mechanism.

437
PATTERN IN FEMORAL CARTILAGE THICKNESS MAP ALLOWS SUBTLE SCORING OF MEDIAL COMPARTMENT KNEE OSTEOARTHRITIS SEVERITY

J. Favre [†], B. Fasel [†], T.P. Andriacchi [†,‡]. [†] *Stanford Univ., Stanford, CA, USA;* [‡] *VA Bone and Joint Ctr., Palo Alto, CA, USA*

Purpose: Although change in cartilage thickness is a hallmark of knee osteoarthritis (OA), the low sensitivity of mean thickness measures to disease state remains a barrier to the evaluation of new interventions for OA. There is potentially important information in the spatial variations in thickness over the entire articular surface (ie. thickness map) that could enhance detection and understanding of OA progression. Computer vision and artificial intelligence algorithms offer opportunities to analyze thickness maps in their entirety and separate patterns that are characteristic of OA severity from subject-specific features. The purpose of this study was to develop a pattern-based method to score femoral cartilage thickness maps according to their disease severity. To demonstrate the application of the method, the score attributed to medial OA knees were compared to Kellgren/Lawrence grades (KLG).
Methods: 140 knees, namely 60 asymptomatic knees and four groups of 20 medial OA knees with KLG of 1, 2, 3, and 4, were scanned at 1.5 T sing a 3D-SPRG sequence after IRB-approved consent (67 male; 60±9 yrs; 1.7±0.1 m; 78±14 kg). 3D models of the femoral cartilage were built from segmentation of MR images. The models were then converted into 2D anatomically-standardized thickness maps (images) using a shape matching routine. A pattern recognition technique was developed to calculate a statistical model that links all the pixels of a thickness map to a single score and vice versa, based on a "learning" dataset of thickness maps with defined OA stage. The continuous pattern-based score was designed to increase with worsening of OA severity (0= typical asymptomatic knee and 4= typical end-stage medial OA knee). In this study, knees were scored using a leave-one-out procedure, consisting of scoring each knee using a statistical model calculated based on all the other knees. For comparison, the mean thickness over the medial compartment was also calculated for each knee. ANOVA and Tukey tests were performed to compare pattern-based score and mean thickness between knee groups, while Spearman correlations were calculated across groups.
Results: The method isolated a characteristic progression in thickness with increasing medial knee OA over the entire plate (Fig.1). The progression notably involved differences in the medial compartment

that evolved from the medial anterior boundary to the central region. The scoring method showed excellent discrimination between disease stages: the pattern-based score was significantly different between all knee groups and the correlation across groups indicated a strong monotonic score increase with worsening of disease severity (Fig.2). In contrast, medial compartment thickness did not show significant differences until an advanced disease state.

Conclusions: This study identified a characteristic progression in thickness map with increasing medial knee OA and the scoring method allowed for excellent differentiation between disease severities. The fact that thickness patterns can be statistically associated with increased OA severity is not trivial as thickness is known to vary among asymptomatic individuals and response to OA is partially subject-specific. In fact the novel method introduced in this study might constitute a shift in the analysis of cartilage thickness. While a discrete reference (KLG) was used to assess the pattern-based score, the method provides continuous scoring and thus has the potential to detect more subtle differences. Clearly the method will need to be tested with larger cohorts and other OA conditions. However, the robust statistical findings here are very promising given the limited size of the training datasets used for this study.

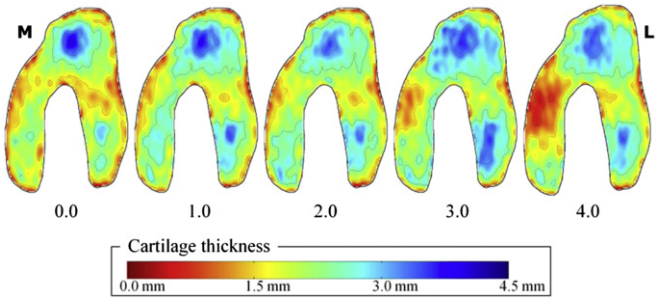

Figure 1. Characteristic progression in cartilage thickness with increasing medial knee OA (left to right) identified using the method introduced in this study.

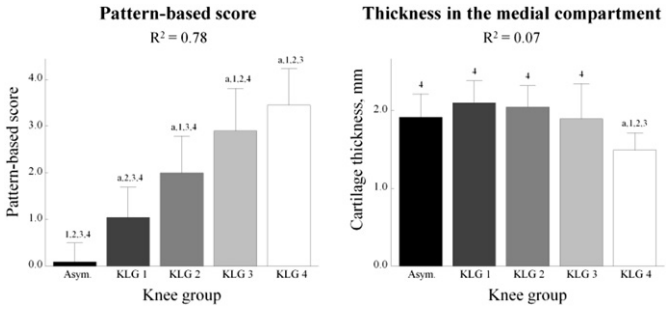

Figure 2. Pattern-based score was significantly different between all knee groups (left plot), whereas mean compartment thickness was less sensitive to increases in OA severity (right plot). Data are presented as mean ± SD, Superscripts indicate significant differences ([a]significantly different from asymptomatic; [1]significantly different from KLG 1; [2]significantly different from KLG 2; [3]significantly different from KLG 3; [4]significantly different from KLG 4).

438

BONE MECHANOTRANSDUCTION VIA T-TYPE VOLTAGE SENSITIVE CALCIUM CHANNEL PLAYS A KEY ROLE IN INDUCTION OF THE OSTEOARTHRITIC PHENOTYPE

P. Srinivasan, A. Kronbergs, R.L. Duncan, C. Kirn-Safran. *Univ. of Delaware, Newark, DE, USA*

Purpose: Mechanotransduction in bone is partially mediated through activation of voltage sensitive calcium channels (VSCC). Mice deficient in $Ca_v3.2$ (alpha1H), the pore forming subunit of T type-VSCC (T-VSCC), exhibit reduced new bone formation response to mechanical loading. It is believed that soluble factors released by mechanically stressed subchondral osteoblasts can activate catabolic pathways in adjacent cartilage tissue. The objective of this study is to understand the role of T-VSCC in mediating the mechanotransduction signals in osteoblasts and test whether the selective inhibition of T-VSCC in mechanically stimulated osteoblasts prevents the development of an osteoarthritic phenotype in chondrocytes.

Methods: The response of murine pre-osteoblastic cells (MC3T3) subjected to either static or fluid shear stress (FSS) (~3.5 dynes/cm^2 for 2 hours) conditions was compared using real-time PCR and Western blot analyses. T-VSCCs were inhibited by NNC 55-0396 in cells treated under the same conditions. Phenotypic changes of primary mouse chondrocytes grown in 3D micromass cultures and treated for one week with conditioned media (CM) collected from MC3T3 (sheared or static) were assessed by real-time PCR, immunolabeling, alcian blue and alizarin red staining.

Results: While cyclooxygenase2 (COX2) and osteopontin are significantly increased following FSS in MC3T3 cells, responses to FSS are reduced by half in the presence of NNC 55-0396. Additionally, treatment of primary chondrocytes with CM from sheared MC3T3 cells induced expression of hypertrophy markers (Collagen X and Alkaline Phosphatase). This catabolic effect was nearly abolished when primary chondrocytes were treated with CM obtained from MC3T3 sheared in the presence of NNC 55-0396.

Conclusions: Inhibition of T-VSCC's reduces the response of osteoblasts to FSS. Sheared osteoblasts secrete factors capable of inducing hypertrophy and potentially pro-inflammatory pathways in chondrocytes. The observed inhibitory effect of NNC 55-0396 indicates that T-VSCC is upstream to the release of soluble catabolic factors from osteoblasts and therefore can be a potential target for blocking subchondral bone response to abnormal mechanical loading and subsequent cartilage degeneration in OA. To test this hypothesis, we are currently comparing OA progression and subchondral bone response between $Ca_v3.2$ knockout and wild type mice using a noninvasive in vivo knee loading model.

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439

AUTOMATIC CARTILAGE SEGMENTATION AND MEASUREMENT FOR DIAGNOSIS OF OA USING 3D BOX AND GAUSSIAN FILTERS

X. Cui [†], S. Li [†], M. Yu [†], H. Kim [†], K.-S. Kwack [‡], B.-H. Min [§]. [†] *Sch. of Information and Communication Engineering, INHA Univ., incheon, Republic of Korea;* [‡] *Dept. of Radiology, Ajou Univ. Sch. of Med., Suwon, Republic of Korea;* [§] *Dept. of Orthopaedic Surgery, Ajou Univ. Sch. of Med., Suwon, Republic of Korea*

Purpose: This paper presents an automatic cartilage segmentation using 3D Box filter and multi-level Gaussian filters for diagnosis of Osteoarthritis (OA) with gradient multi-echo Magnetic Resonance Images (MRI). The 3D Box filter is applied for automatically detecting the Volume of Interest (VOI) of bone, and the multi-level Gaussian filters are applied for accurately computing the 3D normal vectors of cartilage. Then, the measurement of cartilage thickness and volume is made along the direction of the normal vectors in the VOI. Experimental results demonstrate the robustness and efficiency of the proposed approach to 3D cartilage segmentation in the gradient multi-echo MRI.

Methods: In the gradient multi-echo MR images, separating cartilage and muscle based on intensity is a difficult job because they are very similar in intensity. This paper firstly introduces a 3D box filter for rough selection of the bone VOI where cartilage is attached. In order to speed up the processing time, a 3D integral image is computed. Secondly, a 3D hybrid level set segmentation algorithm is applied to the detected bone VOI for fine segmentation of bone from other tissues. Traditional hybrid level set algorithms need to manually set an initial seed volume. However, this paper proposes an automatic initial position estimation method by computing the minimum energy in the 3D space, which corresponds to the center position of the bone. Then, the 3D multi-level Gaussian filters are developed for computing the Bone Cartilage Interface (BCI) by finding 3D normal vectors from the 3D bone surface. Finally, the border between cartilage and other tissues such as muscle or fat is determined based on local and global histogram information of intensity from the bone up to 5mm region along the normal vectors (The maximum cartilage thickness is assumed 5mm). The thickness and volume of local cartilage in the 3D VOI are measured along the 3D normal vectors.

Results: In this research, six gradient multi-echo MRI's from three different patients are used in order to demonstrate the performance of the proposed method. It is able to segment the cartilage and measure the thickness efficiently for all the test data. Fig. 1 and Fig.2 show the segmentation and measurement results with gray and color types. The