sonography. Fat-suppressed 3D spoiled gradient-echo MRI was also used to get the reference value. The joint space width (JSW) and Kellgren and Lawrence (K-L) grade were measured in weight-bearing anteroposterior knee radiograph. The Kappa and intraclass correlation coefficient (ICC) were used to determine inter- and intra-observer agreement of the US measurements.

Results: In medial femoral condyle, the opportunity to obtain cartilage thickness was increased significantly using the longitudinal US scan as compared with transverse scan (48 cases vs. 36 cases, \( p < 0.05 \)). There was a good correlation between longitudinal US scan and MRI in the maximum and minimum cartilage thicknesses of medial condyle (\( r = 0.568 \); \( r = 0.844 \), respectively, \( p < 0.01 \)). However, there was no correlation between suprapatellar transverse US scan and MRI in medial condyle. In lateral condyle, both US scans showed good correlations with MRI. In Bland-Altman analysis, longitudinal US scan showed good agreement with MRI except in the minimal cartilage thickness of lateral condyle. There was high overall intra- and inter-observer agreement in US scan.

Conclusions: US scan in the longitudinal plane is a more feasible method than suprapatellar transverse scan for measuring cartilages thickness of medial femoral condyle in knee OA patient.

387 SECOND HARMONIC GENERATION IMAGING AND COLLAGENOUS MATRIX MODIFICATION IN OSTEOARTHRITIS DISEASE

E. Werkmeister\(^1\), N. de Isla\(^1\), D. Bensousan\(^2\), D. Mainard\(^3\), D. Dumas\(^1\), J-F. Stoltz\(^1\), 1Nancy-Université-UHP Groupe d’Ingénierie Cellulaire et Tissulaire, Vandœuvre-lès-Nancy, FRANCE, 2CHU Nancy, Unité de Thérapie Cellulaire Brabois, Vandœuvre-lès-Nancy, FRANCE, 3CHU Nancy, Unité Département de Chirurgie Orthopédique, Vandœuvre-lès-Nancy, FRANCE.

Purpose: In healthy cartilage, collagen fibers are pseudo-randomly distributed and interact with a gel of Proteoglycans. Degenerative disease such as osteoarthritis can affect the organization of the extracellular matrix (ECM) surrounding chondrocytes leading to a modification or even degradation of the collagen network. The aim of this work was to characterize the remodelling of the collagen network under mechanical or biochemical stress.

Methods: Near infrared tomography (Multiphoton Excitation, Second Harmonic Generation SHG, Fluorescence Lifetime imaging Microscopy FLIM) represents an appropriate tool for cartilage imaging due to its advantages in terms of depth penetration. In the ECM collagen fibers give rise to a strong SHG signal (high non linear susceptibility) and Proteoglycans show a high level of autofluorescent after multiphoton excitation.

Under mechanical stress, a remodeling of the collagen network occurs and can be comparable to disturbance occurring in disease. To characterize structural modification on the arrangement of collagen fibers in the ECM, we used image analysis based on co-occurrence matrix (Haralick). To characterize structural modification on the arrangement of collagen fibers in the ECM, we used image analysis based on co-occurrence matrix (Haralick). Textural parameters can give information like homogeneity (‘Angular Second Moment’) or size of textural elements (‘Inverse Difference Moment’, ‘Correlation’). We followed their evolution when samples were submitted to mechanical (compression) or biochemical (Collagenase) stress.

Results: It came out that the behavior of the collagen network was different under compression or enzymatic action. Enzymatic action of Collagenase lead to a loss of SHG signal according to time of incubation: this evolution can either be attributed to a loss of collagen content or to a modification of collagen molecules affecting their non linear susceptibility. By this way, we proved that the SHG signal came specifically from collagen in cartilage samples. Samples submitted to compression were characterized by higher ‘Correlation’, associated with a decrease of ‘IDM’ and ‘ASM’. Those evolutions suggest the presence of linear structures, an effect of packing of collagen fibrils and the apparition of nodes where the density of collagen is important versus areas showing a lack of molecules. Moreover the ECM seemed more dense and compact and SHG signal was even more intense. We also were interested in the pericellular matrix of chondrocytes containing type VI Collagen. This molecule acts as a transducer of biochemical or biomechanical signals and hypothesis have been emitted about its protective role. Moreover, during osteoarthritis, its content in the pericellular area increases when compared to healthy specimens. Thus Collagen VI can be considered as a biomarker characterising disease states. FLIM associated to Spectral and SHG analysis confirmed the presence of Collagen VI in the pericellular matrix of chondrocytes.

Conclusions: SHG, FLIM and Spectral Imaging combined with multiphoton excitation enable tissue imaging at deep penetration. The association of all this imaging modalities represents a potential diagnostic tool for cartilage disease, since it enables to detect local modification of the collagen network of the ECM without any labelling (SHG) and the presence of collagen VI in the lacunae around cells. Moreover these imaging techniques can be used to validate the well functionality of bioconstructs by following synthesis of collagen for instance.

This work was partly supported by a grant of CG54 and Region de Lorraine.

388 COMPARISON OF ONE YEAR CHANGE IN MINIMUM JOINT SPACE WIDTH TO FIXED LOCATION JOINT SPACE MEASUREMENTS IN LYON SCHUSS X-RAYS FROM THE A9001140 STUDY

B. Wyman\(^1\), R. Buck\(^1\), E. Vignon\(^2\), A. Brett\(^1\), M-P. Gastineau Le-Graverand\(^1\), A9001140 Study Investigators. \(^1\)Pfizer, Inc., New London, CT, USA, \(^2\)Universite Claude Bernard, Lyon, FRANCE, \(^3\)Optasia Inc., Manchester, UNITED KINGDOM

Purpose: Joint space narrowing (JSN) calculated from the change in minimum joint space width (mJSW) from x-ray has become a valuable tool for the monitoring of progression in osteoarthritis (OA). The goal of this analysis was to investigate the relative sensitivity of JSN when measured at fixed locations in Lyon schuss x-rays.

Methods: Lyon schuss x-rays from a subset of 67 subjects from the A9001140 study acquired at 7 sites at baseline and 12 months were analyzed using KneeAnalyzer (Optasia Medical), proprietytistical model-based analysis software. The femoral and tibial margins of the medial compartment were segmented and JSW was measured along a normalized distance across the medial compartment from 0% at the tibial spine to 100% at the medial margin of the tibia. 31 subjects had Non-OA defined as Kellgren and Lawrence (KL) grades of 0 or 1. The OA subjects had KL = 2 (n = 17) or KL = 3 (n = 19). The JSN at the mJSW location and the average JSN between 51%-90% (aJSN 51−90%) were calculated along with the standard deviation (Std Dev) and the standard response mean (SRM). Significance was determined by \( p < 0.05 \).

Results: Figure 1 shows the mean (top) and standard deviation (bottom) of the JSN for each KL group across the medial compartment. The squares are the location of the mJSW measurement (± one Std Dev). Between 50 and about 85% the profiles are relatively flat indicating a consistent difference in JSN. However, around 90% (the approximate