Title

Response of knee cartilage T1rho and T2 relaxation times to in vivo mechanical loading in individuals with and without knee osteoarthritis

Journal

Osteoarthritis and Cartilage, 22(10)

ISSN

1063-4584

Authors

Souza, RB
Kumar, D
Calixto, N
et al.

Publication Date

2014-10-01

DOI

10.1016/j.joca.2014.04.017

Peer reviewed

Published in final edited form as:
Osteoarthritis Cartilage. 2014 October ; 22(10): 1367–1376. doi:10.1016/j.joca.2014.04.017.

Response of Knee Cartilage T1rho and T2 Relaxation Times to *in vivo* Mechanical Loading in Individuals with and without Knee Osteoarthritis

Richard B. Souza, PhD, PT[1,2], **Deepak Kumar, PT, PhD**[1], **Nathan Calixto, BS**[1], **Justin Singh, BS**[1], **Joseph Schooler, MS**[1], **K. Subburaj, PhD**[1], **Xiaojuan Li, PhD**[1], **Thomas M. Link, MD**[1], and **Sharmila Majumdar, PhD**[1]

[1]Department of Radiology and Biomedical Imaging, University of California, San Francisco, San Francisco, CA.

[2]Department of Physical Therapy and Rehabilitation Science, University of California, San Francisco, San Francisco, CA.

Abstract

Objective—The objective of this study was to evaluate the effects of mechanical loading on knee articular cartilage $T_{1\rho}$ and T_2 relaxation times in patients with and without OA.

Design—MR images were acquired from 137 subjects with and without knee OA under two conditions: unloaded and loaded at 50% body weight. Three sequences were acquired: a high-resolution 3D-CUBE, a $T_{1\rho}$ relaxation time, and a T_2 relaxation time sequences. Cartilage regions of interest included: medial and lateral femur (MF, LF); medial and lateral tibia (MT, LT), laminar analysis (superficial and deep layers), and subcompartments. Changes in relaxation times in response to loading were evaluated using generalized estimating equations adjusting for age, gender, and BMI.

Results—In response to loading, we observed significant reductions in $T_{1\rho}$ relaxation times in the MT and LT. In both the MF and LF, loading resulted in significant decreases in the superficial layer and significant increases in the deep layer of the cartilage for $T_{1\rho}$ and T_2. All

Address correspondence to Dr. Richard B. Souza. Department of Physical Therapy and Rehabilitation Science, University of California, San Francisco. 185 Berry St. Suite 350. San Francisco, CA 94107. richard.souza@ucsf.edu.

CONTRIBUTIONS:

1. the conception and design of the study, or acquisition of data, or analysis and interpretation of data: RS, DK, NC, JSi, JSc, KS, XL, TL, SM
2. drafting the article or revising it critically for important intellectual content: RS, DK, NC, KS, XL, TL, SM
3. final approval of the version to be submitted: RS, DK, NC, JSi, JSc, KS, XL, TL, SM

COMPETING INTERESTS

None of the author have any financial and personal relationships with other people or organizations that could potentially and inappropriately influence (bias) this work and conclusions.

subcompartment of MT and LT showed significant reduction in $T_{1\rho}$ relaxation times. Reductions were larger for subjects with OA (range: 13–19% change) when compared to healthy controls (range: 3–13% change).

Conclusions—Loading of the cartilage resulted in significant changes in relaxation times in the femur and tibia, with novel findings regarding laminar and subcompartmental variations. In general, changes in relaxation times with loading were larger in the OA group suggesting that the collagen-proteoglycan matrix of subjects with OA is less capable of retaining water, and may reflect a reduced ability to dissipate loads.

Keywords

(3–6) magnetic resonance imaging; acute loading; osteoarthritis; cartilage

INTRODUCTION

Osteoarthritis (OA) is a degenerative disease that preferentially affects weight-bearing joints and results in disruption of the normal structure and load-bearing capacity of the articular cartilage. The composition of the cartilage extracellular matrix, consisting of proteoglycan, water, and collagen, is altered in OA, causing a disruption of the joint homeostasis. Finite element and mathematical models have explored the effects of disruption of the extracellular matrix on the joint biomechanical properties.[1] However, *in vivo* analysis of cartilage response to loads in healthy and diseased states remains understudied, mostly due to the challenges with quantifying cartilage composition non-invasively.

Several investigators have evaluated the effects of loading on changes in cartilage thickness and volume.[2–5] These studies have provided a wealth of information about the deformation behavior in healthy and OA knees. However, limitations in image resolution and relatively small deformation response have made these studies challenging. MRI relaxation time mapping is an established technique for the quantitative evaluation cartilage composition and structure. Specifically, $T_{1\rho}$ and T_2 relaxation time mapping have been extensively studied as imaging markers for early cartilage degeneration.[6–8] Studies evaluating the behavior of these metrics to mechanical loading have been performed previously by our group and others in healthy individuals, and in small cohorts of patients with OA.[9–11] These studies have revealed important initial findings. However, a comprehensive evaluation of the changes in relaxation times within the layers and subcompartments of the cartilage has not been performed and would be a valuable asset to understanding the in vivo response of cartilage to loading in healthy and OA knees.

It is therefore the purpose of this investigation to evaluate the effects of mechanical loading on tibiofemoral cartilage $T_{1\rho}$ and T_2 relaxation times in knees with varying degrees of OA.. We hypothesize that loading will have an effect on all knee compartments, with the greatest change being observed in the superficial layer of the cartilage, and in the weight-bearing subcompartments, and that changes will be larger in subjects with OA when compared to controls.

MATERIALS AND METHODS

Subjects

A total of 137 subjects (44 OA and 93 controls) recruited via posted flyers from the local community participated in the current investigation. To determine the presence and severity of OA, all subjects underwent bilateral weight-bearing, fixed-flexion postero-anterior knee X-ray with the aid of a Synaflexer device (Synarc, Newark, CA, USA).[12] A radiologist with more than 20 years of experience in musculoskeletal imaging (TML) performed the Kellgren-Lawrence (KL) scoring from these radiographs.[13] The inclusion criteria for OA patients were age > 35 years, knee pain, aching, or stiffness on most days per month during the past year, or use of medication for knee pain on most days per month during the past year, and definite radiographic evidence of knee OA (KL > 1). The inclusion criteria for controls were age > 35 years, no knee pain or stiffness in either knee or use of medications for knee pain in the last year, and no radiographic evidence of OA (KL ≤ 1) on either knee. The exclusion criteria for all subjects were 1) concurrent use of an investigational drug, 2) history fracture or surgical intervention in the study knee, and 3) contraindications to MRI. All subjects signed a written informed consent approved by the University of California, San Francisco Committee on Human Research. All subjects completed the Knee injury and Osteoarthritis Outcome Score (KOOS). The pain, symptoms, and activities of daily living (ADL) subscales of the KOOS were used to assess disability.

Imaging and Loading Procedures

All testing took place at the UCSF Department of Radiology and Biomedical Imaging. Knee images were acquired on a 3-Tesla GE MR 750w Scanner (General Electric, Milwaukee, WI, USA) using an eight-channel knee coil (Invivo, Orlando, FL, USA) and an MR-safe loading apparatus. All subjects were positioned in supine with their knee in neutral rotation and full extension. To reduce movement, the foot of the subject was secured in place, the study knee was stabilized with padding, and a belt was secured across the patient's waist. Images were acquired from one knee under two conditions: unloaded imaging (after a period of 45 minutes of non-weight-bearing), and loaded imaging at 50% body weight. For OA subjects, the knee with more severe findings on the radiographs was imaged. If the KL grade was same for both knees, the more symptomatic knee was imaged. The knee imaged for control subjects was determined randomly. All subjects were instructed to engage in typical physical activity behaviors during the week prior to the MRI session. Subjects arrived at the imaging center and were unloaded (seated in a chair) for a 45 minute period, after which the following sequences were acquired: (1) a high-resolution 3D fast spin-echo CUBE sequence for clinical grading and soft tissue segmentation- (TR/TE=1500/26.69 ms, field of view=16 cm, matrix=384 × 384, slice thickness=0.5 mm, echo train length=32, bandwidth=37.5 kHz, NEX=0.5, acquisition time=10.5 minutes) (2) the $T_{1\rho}$ relaxation time sequence (TR/TE=9/2.6 ms, time of recovery=1500 ms, field of view=14 cm, matrix=256 ×128, slice thickness=4 mm, bandwidth=62.5 kHz, time of spin-lock=0/2/4/8/12/20/40/80 ms, frequency of spin-lock=500 Hz, acquisition time=11 minutes) and (3) the T_2 relaxation time sequence (same as the $T_{1\rho}$ quantification except for magnetization preparation TE=1.8/3.67.3/14.5/29.1/43.6/58.2, acquisition time=11 minutes). Next, a load equivalent to 50% of the subject's body weight was applied to the foot using MRI compatible weights and

a pulley system built into the loading device in order to simulate static standing. The same three sequences described above were then acquired after a period of 10 minutes. Prospective registration algorithms were used to ensure similar field of view between the unloaded and loaded scans.[14]

MR analysis

Major Compartment Analysis—Sagittal high-resolution CUBE images were rigidly registered to the $T_{1\rho}$ relaxation time maps images and used for cartilage segmentation. Medial femoral condyle (MF), medial tibia (MT), lateral femoral condyle (LF), and lateral tibia (LT) cartilage compartments were segmented semi-automatically using in-house software developed with Matlab (Mathworks, Natick, MA, USA) based on edge detection and Bezier splines.[15]

Laminar Analysis—The segmented cartilage regions were then partitioned into two equal laminae: the deep layer (closer to the subchondral bone) and superficial layer (closer to articular surface; Figure 1).[16]

Subcompartment Analysis—Next, the major compartments were divided into subcompartments (Figure 1). The posterior boundary of the posterior meniscal horn was used to divide the MF and LF into the central femoral condyle (cMF/cLF) and posterior femoral condyle (pMF/pLF). The cMF and cLF were further partitioned into 3 weight-bearing subcompartments: anterior (cMF-a/cLF-a), central (cMF-c/cLF-c), and posterior (cMF-p/cLF-p) using the mesial edges of the meniscal horns as landmarks (Figure 1). Similarly, the MT and LT were each partitioned into 3 subcompartments: anterior (MT-a/LT-a), central (MT-c/LT-c), and posterior (MT-p/LT-p).

$T_{1\rho}$ and T_2 Relaxation Time Maps—To account for small movement during acquisition, echos 2–8 were each registered to the first echo of both the $T_{1\rho}$ and T_2 sequences. Additionally, all echos from the T_2 map sequence were registered to the first $T_{1\rho}$ echo. Relaxation time maps for $T_{1\rho}$ and T_2 were constructed by 3-parameter fitting of all eight of the $T_{1\rho}$- and T_2- weighted images pixel-by-pixel to the equations below using in-house developed software:

$$S(TSL) \propto A(\exp\left(-\frac{TSL}{T1\rho}\right))+Bfor\,T1\rho \; S(TE) \propto A(\exp\left(-\frac{TE}{T2}\right))+Bfor\,T2$$

where S is the image signal at a given time point – time of spin-lock (TSL) for $T_{1\rho}$ maps or echo time (TE) for T_2 maps, A = M0 or initial magnetization, and B = constant.

The cartilage regions of interest were overlaid onto the $T_{1\rho}$ and T_2 maps. The cartilage splines were adjusted manually in order to avoid synovial fluid or surrounding anatomy. To eliminate artifacts due to partial volume effects with synovial fluid, voxels with relaxation time ≥ 130 ms for $T_{1\rho}$ or 100 ms for T_2 maps were excluded. Mean $T_{1\rho}$ and T_2 values were calculated for the defined cartilage regions.

Statistical Analysis

All complete datasets, where subjects were able to tolerate the loaded and unloaded imaging free of movement artifact, were used for statistical analysis. Independent samples Student's t-tests were used to compare the age, and BMI between the 2 groups; chi-square test was used to compare distribution of males and females, and one-way ANOVA (with age and BMI as covariates) was used to compare KOOS scores between the 2 groups. Levene's test for homogeneity of variance was used to ensure homogenous variance in the two groups. Natural log transformations were used in case of non-homogenous variances in the two groups for any variable. Homogeneity of regression slopes for the covariates in the model was evaluated by including an interaction term in the model (group × covariate) and ensuring that the resulting significance was > 0.05. To compare differences with between unloaded and loaded conditions, and between the groups, a repeated measures analysis was performed using Generalized Estimating Equations (GEE), while adjusting for age, gender, and BMI. The GEE technique accounts for correlation of responses within subject for response variables.[17–18] Age and BMI were included because these variables were significantly different between the groups and are known to be related to cartilage MR relaxation times.[19–20] Gender was included because prevalence of knee OA is higher in women than men. The GEE model was first run with the interaction between group (Control vs. OA) and Condition (repeated measure). If the interaction term was not found to be statistically significant ($p > 0.05$), it was removed from the model and the analysis re-run. The 2 cartilage layers were analyzed separately since it was not an aim of the paper to compare the relaxation times between the two layers. All analyses were performed in IBM SPSS 20.0 (IBM Corporation, Armonk, NY, USA) with an alpha level of $P < 0.05$.

RESULTS

Subjects Characteristics

There were 93 controls and 44 subjects with knee OA with complete datasets free of movement artifact (Table 1). The OA group was older and heavier compared to controls but the distribution of males and females was not different between the 2 groups. The OA group had worse symptoms, pain, and limitations in their ADLs. The control group had 49 subjects with KL=0, and 44 with KL=1. The OA group had 19, 20, and 5 subjects each with KL 2, 3 and 4. Whole cartilage compartment changes are reported in Table 2.

Cartilage Laminar Analysis

Significant changes in $T_{1\rho}$ relaxation times of the deep and superficial layers in response to loading were noted in all four compartments (Table 3). However, the pattern of change differed between the femoral and tibial compartments. In both the MF and LF, loading resulted in significant decreases (range: 5.3–9.6%) in the superficial layer of the cartilage, and significant increases (range: 8.5–10.7%) in the deep layer of the cartilage (Figure 2). This pattern was not identified in the MT and LT. In both tibial compartments, both the superficial layer and the deep layer displayed significant reductions in $T_{1\rho}$ times with loading although the magnitudes were quite variable (range: 1.0–12.3%). Finally, in the deep layer of the LT, a significant group by condition interaction was identified in which a

greater $T_{1\rho}$ decrease occurred in the OA group compared to controls (17.7% vs. 7.4% for OA and controls, respectively; p = 0.008; Table 3).

Laminar analysis of T_2 relaxation times showed a very similar pattern to the one described above for $T_{1\rho}$ relaxation times. In both femoral compartments, loading resulted in significant decreases in the superficial layer (range: 3.9–6.3%) of the cartilage and significant increases (range: 6.6–12.9%) in the deep layer of the cartilage. However, in the tibial compartments, only the deep layer of the MT (p =0.001) showed significant change in response to loading and was similar between OA and control groups (Table 4). No other significant T2 changes or interactions were identified.

Cartilage Subcompartments

When analyzing subcompartments, changes in $T_{1\rho}$ relaxation times in response to loading were isolated to the MT and LT (Table 5). All subcompartments of MT and LT showed significant reduction in response to loading (range: 3.1–18.5%). In addition, significant interactions were identified in the LT-c and LT-p, where the subjects with OA demonstrated a greater reduction in $T_{1\rho}$ relaxation times in response to loading when compared to controls (13–14% vs. 3–4%, respectively; Table 5). No main effects or interactions were noted in the femoral condyle subcompartments in response to loading.

T_2 relaxation times of all subcompartments of the MT, and anterior subcompartment of the LT (LT-a) demonstrated significant changes in response to loading (Table 6). In all except for the anterior MT (MT-a) this was noted as a significant decrease in T_2 relaxation times in response to loading. However, in the MT-a, significant increases were noted with loading (11.5% in OA subjects). In addition, a significant interaction was identified in the MT-a, whereby the subjects with OA were observed to have a greater increase in T_2 times in response to loading with the control group showing minimal change (Figure 3). In the femoral cartilage, T_2 relaxation times were significantly increased in response to loading in both the pMF and pLF subcompartments (Table 6). Finally, a significant interaction was noted in the pMF, with a greater increase found in the subjects with OA group compared to controls (8.5% vs. 1.7%, respectively; Figure 4).

DISCUSSION

This study quantified the changes in tibiofemoral cartilage $T_{1\rho}$ and T_2 times to mechanical loading in healthy and OA knees. At the whole cartilage level, the largest reductions in relaxation times were localized to the MT and LT. Additionally, we observed large reductions in both $T_{1\rho}$ and T_2 times of the superficial layer of the femoral cartilage with concurrent increases in the deep layer, suggesting a transport of cartilage water from superficial to deeper regions. In general, changes in relaxation times due to loading were larger in the OA group suggesting that the collagen-proteoglycan matrix of subjects with OA is less capable of retaining water, and may reflect a reduced ability to dissipate loads.

A consistent reduction in tibial cartilage relaxation times was observed in response to loading in all subjects. This was observed across both compartments, and in both $T_{1\rho}$ and T_2 relaxation times (although it failed to reach statistical significance for T_2 of the LT).

Consistent with previous studies, we observed between 2%–15% reductions in $T_{1\rho}$ and T_2 times across the tibial compartments with loading.[9–10] The largest changes were observed in $T_{1\rho}$ relaxation times of the MT in subjects with OA (15%) and healthy controls (12%). Previous literature has linked $T_{1\rho}$ times to both glycosaminoglycan content and tissue hydration.[21–23] The reduction in $T_{1\rho}$ times observed in the tibia with loading may be the result of reduced hydration as water is squeezed out of the matrix and into the joint, or as a relative increase in glycosaminoglycan content as the cartilage thickness is reduced due to loading. Similarly, the reduction in T_2 times is likely reflecting the reduction of water content and increased collagen concentration.

We did not observe significant changes in $T_{1\rho}$ or T_2 values of the femoral cartilage in response to loading. In fact, all loaded femoral cartilage relaxation times were within 4% of unloaded relaxation times. This is in contrast to literature that reported significant reductions in the femoral cartilage T_2 times in response to loading, and no differences in the tibial cartilage.[10] However, these authors used coronal slice acquisition and evaluated subregions divided into six medial-to-lateral subcompartments. Ultimately, the regions reported cannot be directly compared to those evaluated in the current study.

A striking difference was observed in the cartilage layers of the tibial and femoral compartments in response loading. Specifically, the tibia cartilage layers showed a corresponding response, with both the superficial and deep layers demonstrating reductions in response to loading. However, the superficial and deep layers of the femoral cartilage demonstrated a different behavior – while the superficial layer revealed a reduction in response to loading, the deep layer was observed to increase in relaxation times with loading. This phenomenon was observed for $T_{1\rho}$ and T_2 of both groups. These data reveal important differences in biomechanical behavior of tibial and femoral cartilage in response to loading. The stress-resistance of the superficial cartilage is related to water content, permeability of fluid within the matrix, and integrity of the collagen matrix.[9] While the tibia appear to lose water content as the joint surface is loaded, the femoral cartilage appears to transport fluid to deeper regions of the cartilage. It has been previously reported that there is a clear depth-dependent variation in the biochemical and biomechanical properties of cartilage.[24–28] For example, Chen and colleagues used epiflourescent micrographs and osmotic compression to reveal differences in tissue displacement, compressive modulus and fixed charge density as a function of distance from the superficial surface.[24] Lower stiffness and fixed charge density were noted in the most superficial layer with a non-linear increase in deeper layers. This is consistent with the current observation of changes in relaxation time of the femoral cartilage, but somewhat in contrast with findings in tibiae where the largest changes were observed in the deep layer. However, it should be noted that the relationship between changes in cartilage relaxation times in response to loading and dynamic tissue mechanics remains unclear and should be considered speculative at this time. These data are also in agreement with previous loading studies by Mosher and colleagues who reported significant reductions in T_2 times of the superficial layers of the femoral cartilage after dynamic loading (a 30 minute bout of running).[6]

With regard to subcompartments, we observed consistent $T_{1\rho}$ reductions in all regions of the MT and LT. All reductions were larger for subjects with OA when compared to healthy

controls, with a significant interaction revealing statistically larger reductions for the central and posterior regions of the LT (Table 4). For T_2, significant reductions with loading were observed in central and posterior MT and the anterior LT, but without significant differences between OA and control groups. These data are similar to that of Nishii and colleagues that reported significant reduction in T_2 times with loading in healthy individuals.[9] These results are of similar magnitude to those reported in dynamic loading studies of articular cartilage.[29] In contrast to previous literature, we observed several subcompartments demonstrating significant increases in relaxation times in response to loading. These regions were generally non-weight-bearing regions and the larger increases were observed in the OA cohort. For example, in the posterior regions of the femoral condyles, we observed higher T_2 times with loading (Figure 3). This may suggest that cartilage water content is being squeezed into the non-weight-bearing regions as the primary loading sites become compressed. The increased change scores observed in the non-weight-bearing regions, coupled with the higher reductions in the primary weight-bearing regions, suggest that the collagen-proteoglycan matrix of subjects with OA is less capable of retaining water, and may reflect a reduced ability to dissipate loads. We also observe this phenomenon in the $T_{1\rho}$ of the anterior MT where the control cohort shows almost no change (0.6%) while the OA cohort demonstrates a large increase (11.5%; Figure 3). Contrary to our stated hypothesis, we did not observe significant reductions in relaxation times of the weight-bearing subcompartments of the MF and LF. It is likely that the laminar behavior discussed above of opposing changes in the superficial and deep layers is responsible for the minimal net change in femoral subcompartments with loading.

The results of this study highlight important biomechanical variations in both healthy and diseased cartilage. First, we observed significant difference in the behavior of the femoral and tibia cartilage layers. While the tibial deep and superficial layers both showed reductions in relaxation times in response to loading, the femoral layers showed opposing changes (deep layer increased while the superficial layer decreased). This was observed across all subjects. These data highlight the variability of articular cartilage even within a single joint. These differences are likely related to the biomechanical demands of the joint. As the knee flexes, the contact points undergo greater excursion on the femoral condyle when compared to the tibial plateau by virtue of the difference in shapes of the two articular surfaces.[1, 30–31] As such, the convex femur undergoes both compressive and shear forces. In contrast, the tibia is relatively flat and experiences less shear.[32–33]

This study also revealed important differences between healthy and arthritic cartilage. Larger changes in relaxation times (both increases and decreases) in response to loading were observed in subjects with knee OA compared to controls. Again, this likely reflects increased permeability and reductions in stress-resistance abilities in OA cartilage. It is possible that the change in relaxation times in response to loading may be a reflection of the load bearing capability of the articular cartilage and may be used in the future as a measure of tissue function *in vivo*. To investigate this further we performed additional *post-hoc* analyses (results not shown) stratifying the subjects into controls (KL = 0,1), early OA (KL =2), and advanced OA (KL = 3,4). The GEE models were re-run with this stratification. We observed that for the major compartment, the advanced OA group showed a greater decrease

compared to the control and early OA groups for $T_{1\rho}$ times in the MT. For laminar analyses, the advanced OA and control groups showed a greater increase in the deep layer compared to the early OA group for $T_{1\rho}$ of the LF. For subcompartment analyses, the OA groups were not significantly different from each other. These results further highlight the differences in the load bearing capacity of healthy vs. diseased articular cartilage.

The results of this study need to be viewed in light of their limitations. The final cohort analyzed incurred a 13% loss in data due to load intolerance/pain or movement artifact in acquired images. However, the dropped subjects included a similar number of OA and control subjects (lost data: 11 OA and 10 controls). The current study included a loading protocol that applied a 50% body-weight load for approximately 45 minutes. However, some of the imaging series were performed as early as 20 minutes after application of the load. Therefore, the total effects of loading may not have been fully realized, and the changes observed in the current study may be smaller than those observed after longer periods of loading. The timing and loading protocol was developed to optimize imaging data while limiting movement artifact and accounting for subject tolerance. Additionally, the unloading timing of 45 minutes may also have been insufficient to fully unload the cartilage. And the protocol used with subjects seated in a chair during unloading may be considered a minimally-loaded state rather than a complete unloaded state. Another issue that must be considered is the magnitude of changes in relaxation times in light of the inter-subject variability. The standard deviation of relaxation times in both groups was between 2.2 and 5.9 msec, a relatively large amount of variability. Thus, it remains clear that there does not exist a definitive threshold of $T_{1\rho}$ or T_2 relaxation that is indicative of knee OA. Finally, the significance of the change in relaxation times remains speculative. The group differences observed in the current study suggest that these may be related to biomechanical and biochemical deficits in osteoarthritic cartilage. However, further research is needed to confirm this hypothesis.

In conclusion, we observed the largest reductions in relaxation times in both of the tibial compartments in response to loading. Additionally, observed large reductions in both $T_{1\rho}$ and T_2 times of the superficial layer of the femoral cartilage with concurrent increases in the deep layer, suggesting a transport of cartilage water from superficial to deeper regions. In general, changes in relaxation times due to loading were larger in the OA group suggesting that the collagen-proteoglycan matrix of subjects with OA is less capable of retaining water, and may reflect a reduced ability to dissipate loads. This variable has received limited attention and should be further evaluated for its relationship to both biochemical and biomechanical predictors of disease progression.

Acknowledgments

The project described was supported by Grant Numbers R01 AR062370, R01 AR46906, and P50 AR060752, from the National Institute of Arthritis and Musculoskeletal and Skin Diseases.

ROLE OF THE FUNDING SOURCE

The funding source had no role in the study design, collection, analysis and interpretation of data; in the writing of the manuscript; and in the decision to submit the manuscript for publication.

REFERENCES

1. Li JS, Hosseini A, Cancre L, Ryan N, Rubash HE, Li G. Kinematic characteristics of the tibiofemoral joint during a step-up activity. Gait Posture. 2013; 38:712–716. [PubMed: 23541765]
2. Andreisek G, White LM, Sussman MS, Kunz M, Hurtig M, Weller I, et al. Quantitative MR imaging evaluation of the cartilage thickness and subchondral bone area in patients with ACL-reconstructions 7 years after surgery. Osteoarthritis Cartilage. 2009; 17:871–878. [PubMed: 19230721]
3. Eckstein F, Cicuttini F, Raynauld JP, Waterton JC, Peterfy C. Magnetic resonance imaging (MRI) of articular cartilage in knee osteoarthritis (OA): morphological assessment. Osteoarthritis Cartilage. 2006; 14(Suppl A):A46–A75. [PubMed: 16713720]
4. Subburaj K, Souza RB, Stehling C, Wyman BT, Le Graverand-Gastineau MP, Link TM, et al. Association of MR relaxation and cartilage deformation in knee osteoarthritis. J Orthop Res. 2012; 30:919–926. [PubMed: 22161783]
5. Cotofana S, Eckstein F, Wirth W, Souza RB, Li X, Wyman B, et al. In vivo measures of cartilage deformation: patterns in healthy and osteoarthritic female knees using 3T MR imaging. Eur Radiol. 2011; 21:1127–1135. [PubMed: 21448616]
6. Mosher TJ, Smith HE, Collins C, Liu Y, Hancy J, Dardzinski BJ, et al. Change in knee cartilage T2 at MR imaging after running: a feasibility study. Radiology. 2005; 234:245–249. [PubMed: 15550376]
7. Souza RB, Baum T, Wu S, Feeley BT, Kadel N, Li X, et al. Effects of unloading on knee articular cartilage T1rho and T2 magnetic resonance imaging relaxation times: a case series. J Orthop Sports Phys Ther. 2012; 42:511–520. [PubMed: 22402583]
8. Li X, Benjamin Ma C, Link TM, Castillo DD, Blumenkrantz G, Lozano J, et al. In vivo T(1rho) and T(2) mapping of articular cartilage in osteoarthritis of the knee using 3 T MRI. Osteoarthritis Cartilage. 2007; 15:789–797. [PubMed: 17307365]
9. Nishii T, Kuroda K, Matsuoka Y, Sahara T, Yoshikawa H. Change in knee cartilage T2 in response to mechanical loading. J Magn Reson Imaging. 2008; 28:175–180. [PubMed: 18581338]
10. Nag D, Liney GP, Gillespie P, Sherman KP. Quantification of T(2) relaxation changes in articular cartilage with in situ mechanical loading of the knee. J Magn Reson Imaging. 2004; 19:317–322. [PubMed: 14994300]
11. Souza R, Stehling C, Wyman BT, Hellio Le Graverand MP, Li X, Link TM, et al. The effects of acute loading on T1rho and T2 relaxation times of tibiofemoral articular cartilage. Osteoarthritis Cartilage. 2010; 18:1557–1563. [PubMed: 20950693]
12. Charles HC, Kraus VB, Ainslie M, Hellio Le Graverand-Gastineau MP. Optimization of the fixed-flexion knee radiograph. Osteoarthritis Cartilage. 2007; 15:1221–1224. [PubMed: 17977754]
13. Kellgren JH, Lawrence JS. Radiological assessment of osteo-arthrosis. Ann Rheum Dis. 1957; 16:494–502. [PubMed: 13498604]
14. Goldenstein J, Schooler J, Crane JC, Ozhinsky E, Pialat JB, Carballido-Gamio J, et al. Prospective image registration for automated scan prescription of follow-up knee images in quantitative studies. Magn Reson Imaging. 2011; 29:693–700. [PubMed: 21546186]
15. Carballido-Gamio J, Bauer JS, Stahl R, Lee KY, Krause S, Link TM, et al. Inter-subject comparison of MRI knee cartilage thickness. Med Image Anal. 2008; 12:120–135. [PubMed: 17923429]
16. Carballido-Gamio J, Stahl R, Blumenkrantz G, Romero A, Majumdar S, Link TM. Spatial analysis of magnetic resonance T1rho and T2 relaxation times improves classification between subjects with and without osteoarthritis. Med Phys. 2009; 36:4059–4067. [PubMed: 19810478]
17. McCullagh, P.; Nelder, JA. Generalized linear models. 2nd Edition. London: Chapman and Hall; 1989.
18. Nedler JA, Wedderburn RW. Generalized linear models. Journal of the Royal Statistical Society. 1972:370–384. Series A.
19. Baum T, Joseph GB, Nardo L, Virayavanich W, Arulanandan A, Alizai H, et al. Correlation of magnetic resonance imaging-based knee cartilage T2 measurements and focal knee lesions with

body mass index: thirty-six-month followup data from a longitudinal, observational multicenter study. Arthritis Care Res (Hoboken). 2013; 65:23–33. [PubMed: 22623435]

20. Mosher TJ, Liu Y, Yang QX, Yao J, Smith R, Dardzinski BJ, et al. Age dependency of cartilage magnetic resonance imaging T2 relaxation times in asymptomatic women. Arthritis Rheum. 2004; 50:2820–2828. [PubMed: 15457450]
21. Li X, Cheng J, Lin K, Saadat E, Bolbos RI, Jobke B, et al. Quantitative MRI using T1rho and T2 in human osteoarthritic cartilage specimens: correlation with biochemical measurements and histology. Magn Reson Imaging. 2011; 29:324–334. [PubMed: 21130590]
22. Wheaton AJ, Dodge GR, Borthakur A, Kneeland JB, Schumacher HR, Reddy R. Detection of changes in articular cartilage proteoglycan by T(1rho) magnetic resonance imaging. J Orthop Res. 2005; 23:102–108. [PubMed: 15607881]
23. Regatte RR, Akella SV, Lonner JH, Kneeland JB, Reddy R. T1rho relaxation mapping in human osteoarthritis (OA) cartilage: comparison of T1rho with T2. J Magn Reson Imaging. 2006; 23:547–553. [PubMed: 16523468]
24. Chen SS, Falcovitz YH, Schneiderman R, Maroudas A, Sah RL. Depth-dependent compressive properties of normal aged human femoral head articular cartilage: relationship to fixed charge density. Osteoarthritis Cartilage. 2001; 9:561–569. [PubMed: 11520170]
25. Erne OK, Reid JB, Ehmke LW, Sommers MB, Madey SM, Bottlang M. Depth-dependent strain of patellofemoral articular cartilage in unconfined compression. J Biomech. 2005; 38:667–672. [PubMed: 15713286]
26. Lai JH, Levenston ME. Meniscus and cartilage exhibit distinct intra-tissue strain distributions under unconfined compression. Osteoarthritis Cartilage. 2010; 18:1291–1299. [PubMed: 20633686]
27. Saarakkala S, Julkunen P, Kiviranta P, Makitalo J, Jurvelin JS, Korhonen RK. Depth- wise progression of osteoarthritis in human articular cartilage: investigation of composition, structure and biomechanics. Osteoarthritis Cartilage. 2010; 18:73–81. [PubMed: 19733642]
28. Lotz MK, Carames B. Autophagy and cartilage homeostasis mechanisms in joint health, aging and OA. Nat Rev Rheumatol. 2011; 7:579–587. [PubMed: 21808292]
29. Subburaj K, Kumar D, Souza RB, Alizai H, Li X, Link TM, et al. The acute effect of running on knee articular cartilage and meniscus magnetic resonance relaxation times in young healthy adults. Am J Sports Med. 2012; 40:2134–2141. [PubMed: 22729505]
30. Li G, DeFrate LE, Park SE, Gill TJ, Rubash HE. In vivo articular cartilage contact kinematics of the knee: an investigation using dual-orthogonal fluoroscopy and magnetic resonance image-based computer models. Am J Sports Med. 2005; 33:102–107. [PubMed: 15611005]
31. Kozanek M, Hosseini A, Liu F, Van de Velde SK, Gill TJ, Rubash HE, et al. Tibiofemoral kinematics and condylar motion during the stance phase of gait. J Biomech. 2009; 42:1877–1884. [PubMed: 19497573]
32. Stone JJ, Yu H. Computational contact analysis of joint congruency. Biomed Sci Instrum. 1997; 34:368–373. [PubMed: 9603068]
33. Plochocki JH, Ward CV, Smith DE. Evaluation of the chondral modeling theory using fe-simulation and numeric shape optimization. J Anat. 2009; 214:768–777. [PubMed: 19438771]

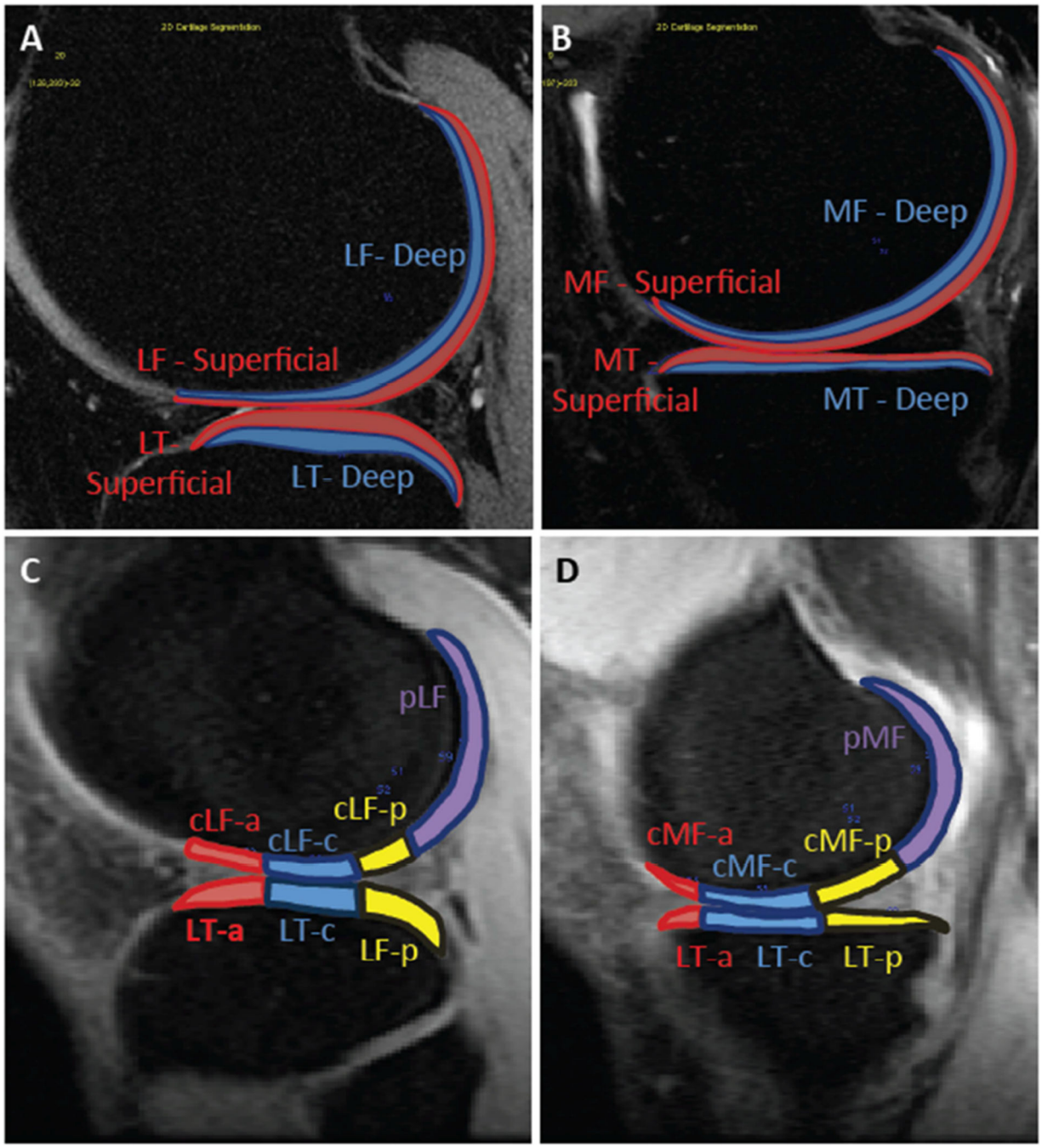
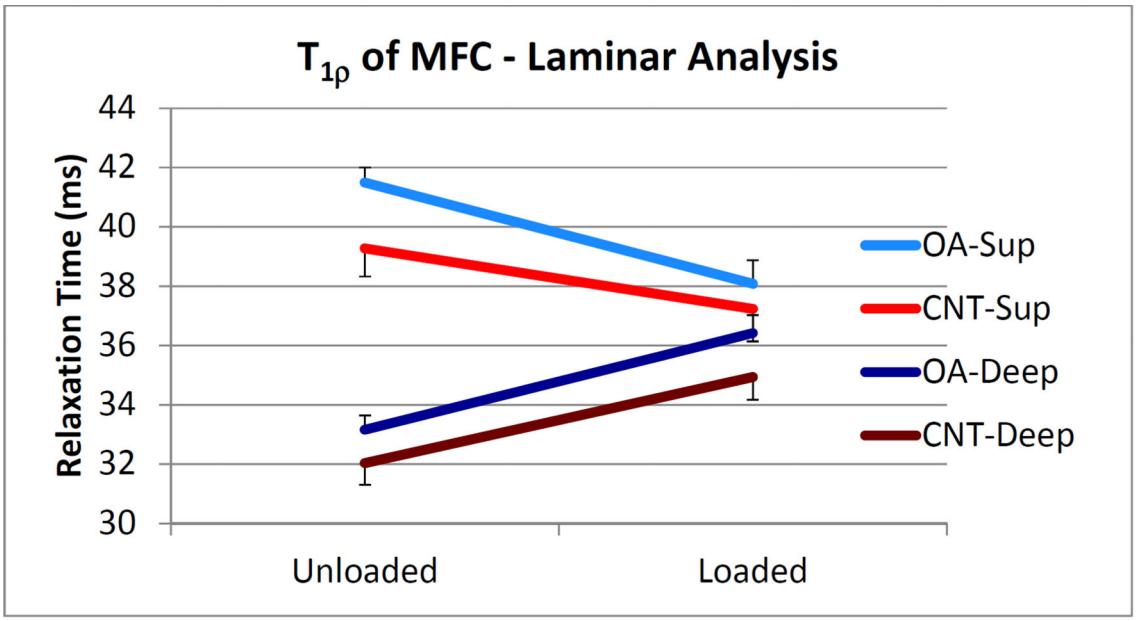


Figure 1.

Cartilage regions of interest for laminar analysis (A & B) showing deep and superficial layers; and subcompartments (C & D) showing weight-bearing and non-weight-bearing regions: LF: lateral femur; LT: lateral tibia; MF: medial femur; MT: medial tibia; cXF-x refers to the portion of the central femoral condyle that is either anterior (a), central (c), or posterior (p).



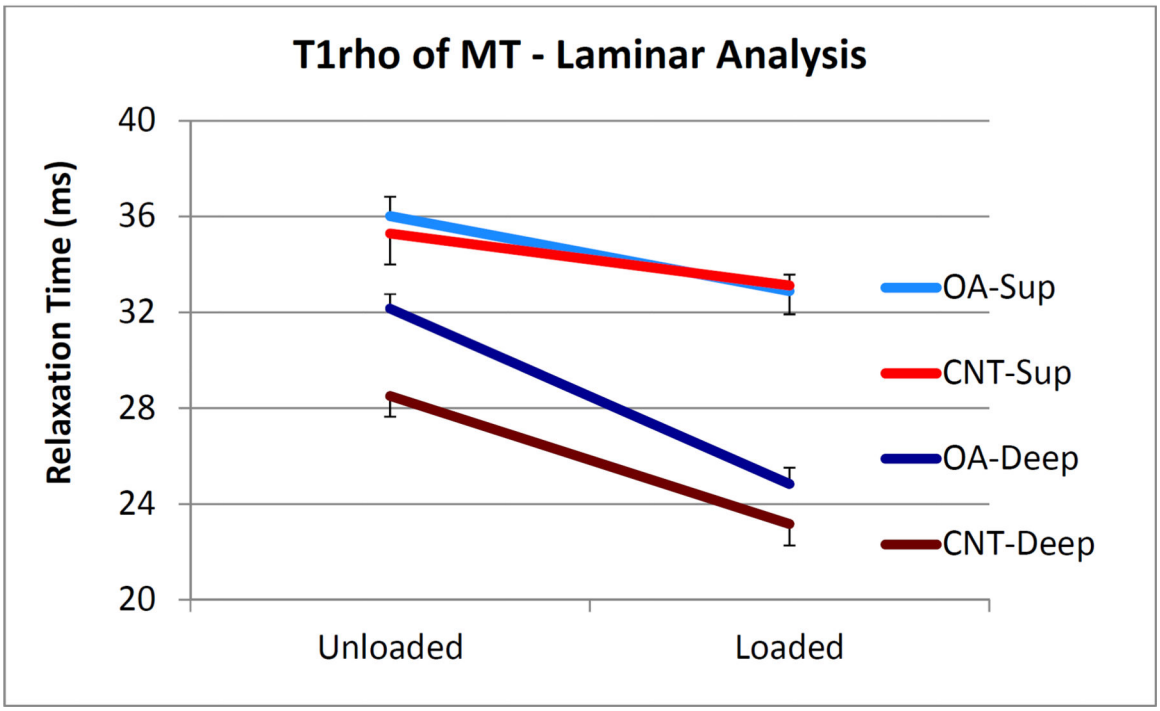


Figure 2.

$T_{1\rho}$ laminar behavior with loading. MF = medial femur; MT = medial tibia. OA-Sup = superficial layer in subjects with OA; CNT-Sup = superficial layer in control subjects; OA-Deep = deep layer in subjects with OA; and CNT-Deep = deep layer in control subjects. **Bars represent standard error bars**.

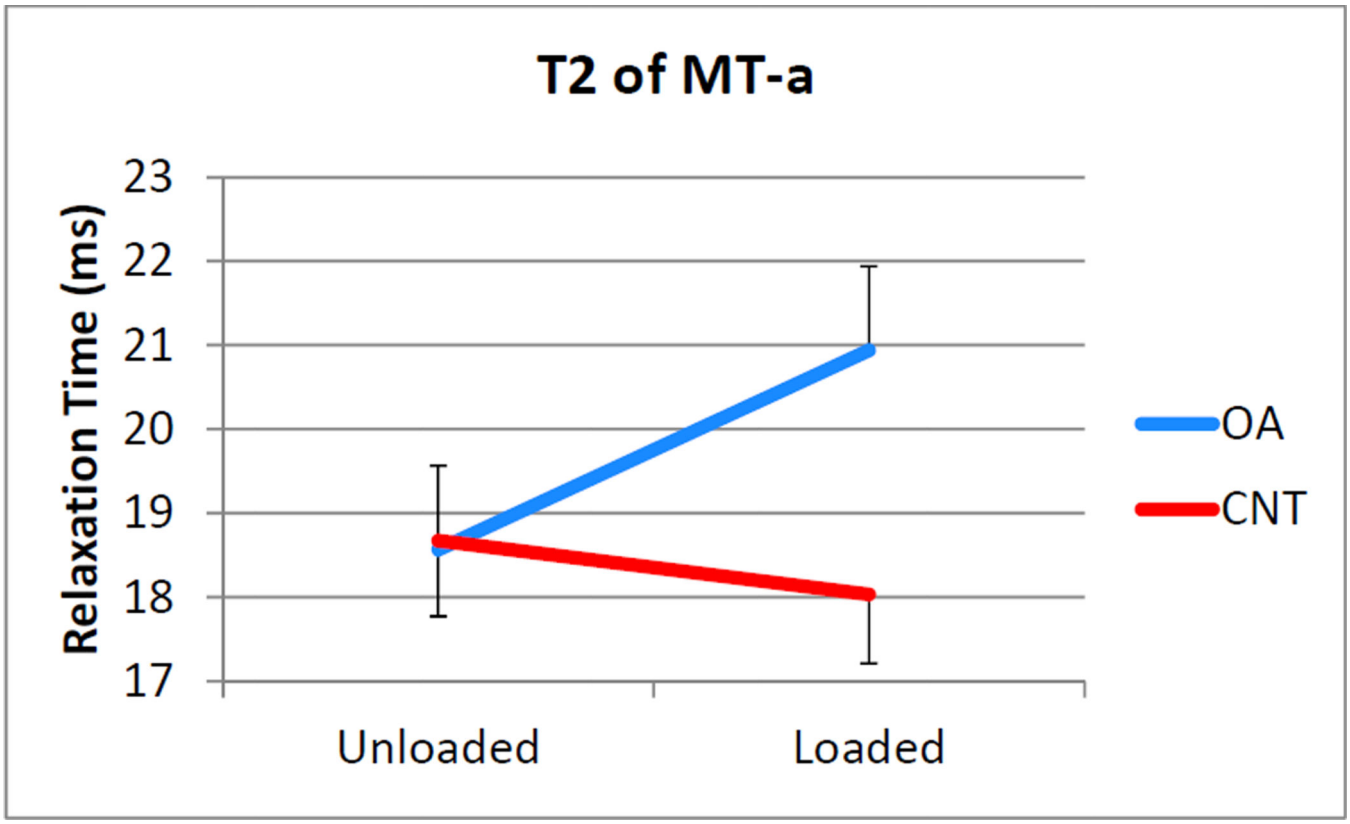


Figure 3.
T_2 of the anterior medial tibia (MT-a) displaying a significant difference in response to loading between OA and controls. **Bars represent standard error bars**.

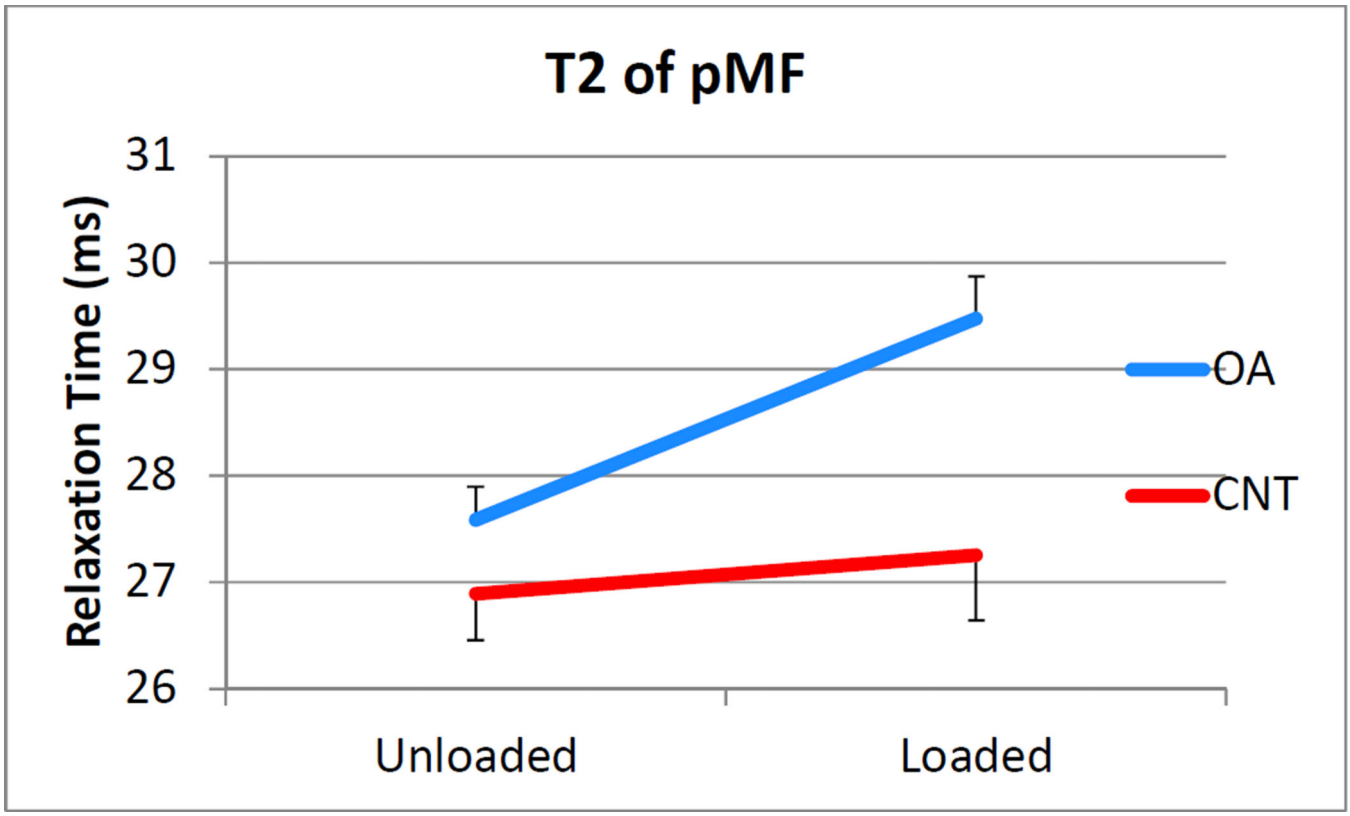


Figure 4.

T_2 of the posterior medial femur (pMF) displaying a significant difference in response to loading between OA and controls. **Bars represent standard error bars**.

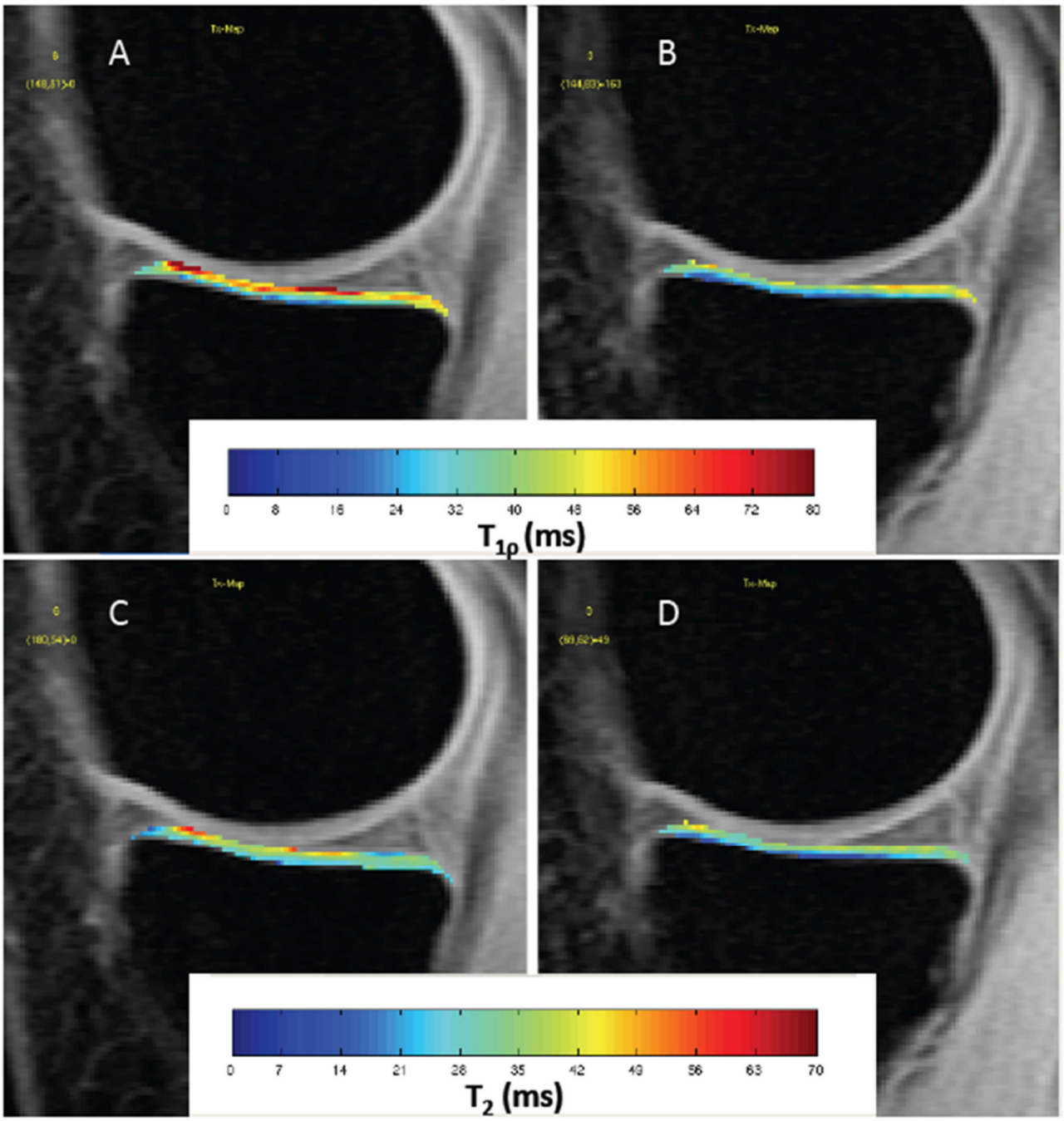


Figure 5.
Representative $T_{1\rho}$ (top row) and T_2 (bottom row) relaxation color maps of the medial femoral condyle in the unloaded (A & C), and loaded (B & D) conditions.

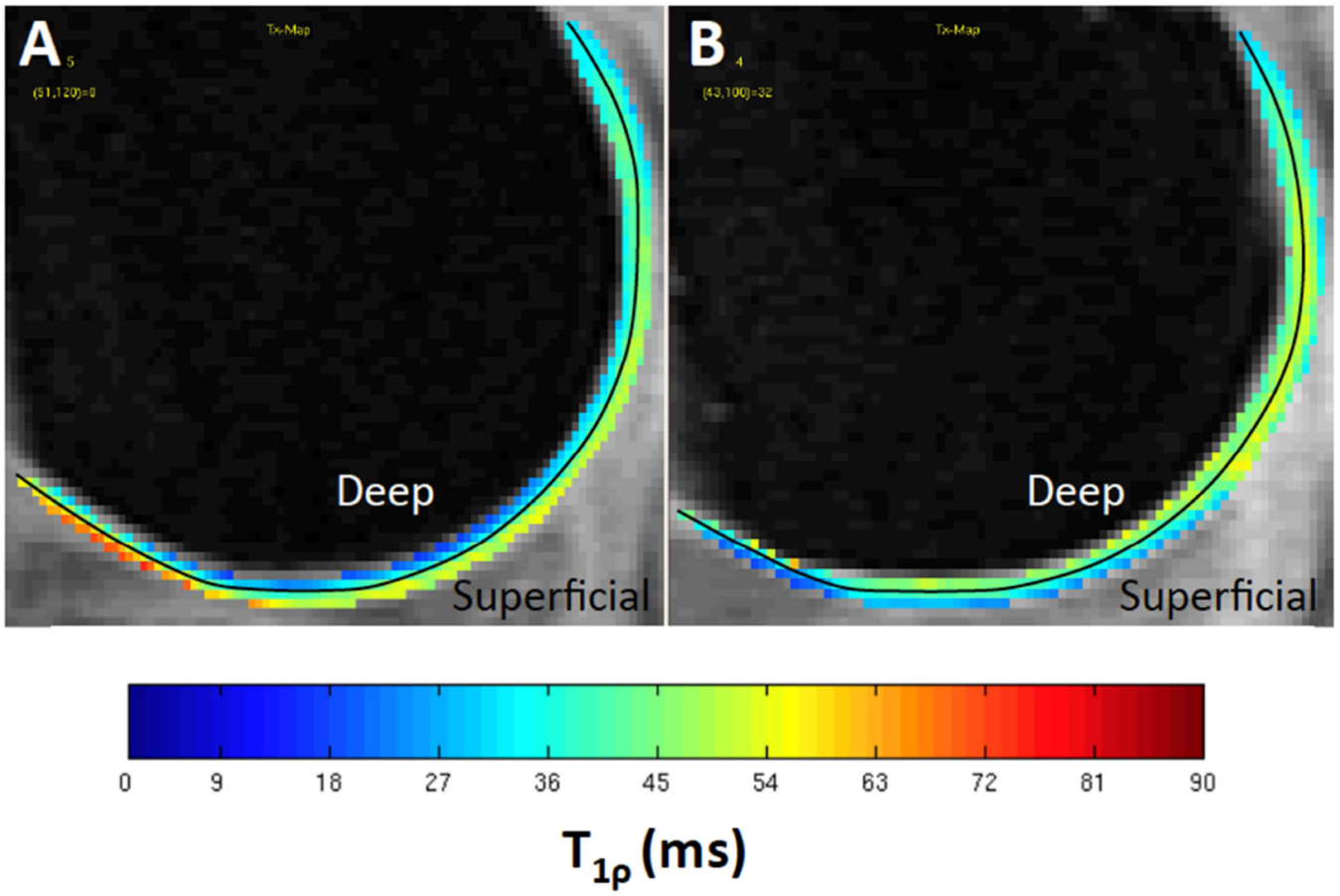


Figure 6.

Representative $T_{1\rho}$ relaxation color maps of the medial femoral condyle in the unloaded (A), and loaded (B) conditions.

Table 1

Subject Characteristics

		Control (n = 93) Mean (95% confidence intervals)	Osteoarthritis (n = 44) Mean (95% confidence intervals)	*P*
Age (years)		49.5 (47.6, 51.4)	57.4 (54.4, 60.3)	**<0.0001**
BMI (kg/m^2)		24.0 (23.3, 24.7)	26.4 (24.0, 28.7)	**0.016**
Gender (M:F)		39:54	17:27	0.714*
KOOS	**Symptoms**	89.7 (87.4, 92.0)	80.0 (74.4, 85.6)	**0.002**†
	Pain	90.8 (88.1, 93.6)	78. 5 (72.9, 84.1)	**<0.0001**†
	Activities of Daily Living	94.2 (92.0, 96.5)	83.1 (77.3 ,88.8)	**0.001**†

*P value from the Chi-Square test

†adjusted for age, BMI

Table 2

T1ρ and T2 Relaxation Times

		Control	Osteoarthritis
		Mean (95% Confidence Intervals)	**Mean (95% Confidence Intervals)**
T1ρ Relaxation Times			
Medial Femur	Unloaded	35.7 (35.0, 36.5)	37.3 (35.7, 38.8)
	Loaded	36.0 (34.9, 37.2)	37.2 (35.7, 38.6)
Medial Tibia*	**Unloaded**	**31.9 (30.8, 32.9)**	**33.9 (32.2, 35.6)**
	Loaded	**28.1 (26.9, 29.3)**	**28.8 (27.1, 30.5)**
Lateral Femur	Unloaded	33.7 (33.0, 34.5)	35.6 (34.2, 37.0)
	Loaded	34.1 (32.9, 35.2)	35.1 (33.5, 36.6)
Lateral Tibia*	**Unloaded**	**32.1 (31.2, 33.0)**	**32.9 (31.5, 34.2)**
	Loaded	**30.7 (29.5, 32.0)**	**29.0 (27.3, 30.7)**
T2 Relaxation Times			
Medial Femur	Unloaded	25.6 (25.1, 26.2)	26.7 (25.7, 27.7)
	Loaded	25.8 (25.1, 26.4)	27.7 (26.7, 28.8)
Medial Tibia*	**Unloaded**	**22.4 (21.6, 23.2)**	**23.0 (21.6, 24.4)**
	Loaded	**21.0 (20.2, 21.9)**	**22.2 (20.8, 23.5)**
Lateral Femur	Unloaded	24.4 (23.9, 25.0)	25.6 (24.3, 26.8)
	Loaded	25.1 (24.4, 25.7)	26.1 (24.9, 27.3)
Lateral Tibia	Unloaded	22.4 (21.6, 23.2)	22.6 (21.4, 23.8)
	Loaded	22.1 (21.2, 23.0)	22.0 (20.8, 23.2)

* indicates significant main effect for loading.

Table 3

Laminar T1ρ

			Control	Osteoarthritis	Group by Condition Interaction
			Mean (95% Confidence Intervals)	Mean (95% Confidence Intervals)	
Medial Femur	**Superficial***	**Unloaded**	**39.3 (38.3, 40.2)**	**41.5 (39.6, 43.4)**	n.s.
		Loaded	**37.2 (36.0, 38.4)**	**38.1 (36.5, 39.6)**	
	Deep*	**Unloaded**	**32.0 (31.0, 33.0)**	**33.2 (31.2, 35.1)**	n.s.
		Loaded	**34.9 (33.3, 36.5)**	**36.4 (34.2, 38.6)**	
Medial Tibia	**Superficial***	**Unloaded**	**35.3 (34.1, 36.5)**	**36.0 (34.3, 37.8)**	n.s.
		Loaded	**33.1 (31.8, 34.5)**	**32.9 (31.1, 34.7)**	
	Deep*	**Unloaded**	**28.5 (26.9, 30.1)**	**32.2 (29.6, 34.8)**	n.s.
		Loaded	**23.2 (21.8, 24.5)**	**24.8 (22.4, 27.3)**	
Lateral Femur	**Superficial***	**Unloaded**	**37.9 (36.9, 38.9)**	**40.0 (38.1, 41.9)**	n.s.
		Loaded	**35.8 (34.6, 37.0)**	**36.0 (34.3, 37.7)**	
	Deep*	**Unloaded**	**29.4 (28.5, 30.3)**	**31.1 (29.6, 32.6)**	n.s.
		Loaded	**32.4 (31.0, 33.7)**	**34.4 (32.5, 36.3)**	
Lateral Tibia	**Superficial***	**Unloaded**	**37.0 (36.0, 37.9)**	**36.7 (35.2, 38.2)**	n.s.
		Loaded	**36.3 (34.9, 37.8)**	**33.8 (31.9, 35.7)**	
	Deep*	**Unloaded**	**26.9 (25.7, 28.0)**	**29.0 (27.2, 30.8)**	**p = 0.008**
		Loaded	**24.9 (23.7, 26.1)**	**24.1 (22.3, 25.8)**	

* indicates loaded condition is significantly different from unloaded condition across both groups at $p<0.05$

Table 4

Laminar T2

			Control	Osteoarthritis	Group by Condition Interaction
			Mean (95% Confidence Intervals)	Mean (95% Confidence Intervals)	
Medial Femur	**Superficial***	**Unloaded**	**28.1 (27.5, 28.8)**	**29.3 (28.0, 30.6)**	n.s.
		Loaded	**26.6 (25.9, 27.4)**	**28.3 (27.0, 29.6)**	
	Deep*	**Unloaded**	**23.3 (22.4, 24.1)**	**24.3 (22.8, 25.8)**	n.s.
		Loaded	**24.9 (24.0, 25.7)**	**27.2 (25.9, 28.6)**	
Medial Tibia	Superficial	Unloaded	24.3 (23.4, 25.2)	24.6 (23.0, 26.2)	n.s.
		Loaded	23.7 (22.9, 24.5)	24.7 (23.2, 26.2)	
	Deep*	**Unloaded**	**20.7 (19.5, 21.8)**	**21.6 (20.0, 23.1)**	n.s.
		Loaded	**18.3 (17.2, 19.4)**	**19.6 (17.9, 21.3)**	
Lateral Femur	**Superficial***	**Unloaded**	**27.7 (27.0, 28.4)**	**28.7 (27.1, 30.2)**	n.s.
		Loaded	**26.2 (25.4, 26.9)**	**27.2 (26.1, 28.4)**	
	Deep*	**Unloaded**	**21.2 (20.4, 21.9)**	**22.7 (21.2, 24.2)**	n.s.
		Loaded	**23.9 (23.1, 24.7)**	**25.0 (23.3, 26.7)**	
Lateral Tibia	Superficial	Unloaded	25.8 (25.0, 26.6)	25.7 (24.4, 27.1)	n.s.
		Loaded	25.6 (24.7, 26.5)	25.4 (24.0, 26.7)	
	Deep	Unloaded	18.9 (18.0, 19.7)	19.5 (18.2, 20.8)	n.s.
		Loaded	18.3 (17.3, 19.4)	18.6 (17.3, 19.9)	

* indicates loaded condition is significantly different from unloaded condition across both groups at $p<0.05$

Table 5

Sub-compartment T1ρ

			Control	Osteoarthritis	Group by Condition Interaction
			Mean (95% Confidence Intervals)	Mean (95% Confidence Intervals)	
Medial Femur	cMF-a	Unloaded	35.2 (33.9, 36.4)	36.5 (34.5, 38.5)	n.s.
		Loaded	35.6 (34.3, 36.9)	36.0 (33.4, 38.6)	
	cMF-c	Unloaded	32.8 (31.7, 33.8)	34.8 (32.5, 37.1)	n.s.
		Loaded	33.0 (31.7, 34.3)	34.2 (31.9, 36.5)	
	cMF-p	Unloaded	34.1 (33.0, 35.3)	34.4 (32.3, 36.6)	n.s.
		Loaded	34.4 (32.9, 35.9)	35.2 (32.9, 37.5)	
	pMF	Unloaded	37.0 (36.2, 37.8)	38.1 (36.6, 39.6)	n.s.
		Loaded	37.8 (36.4, 39.1)	39.3 (37.7, 40.8)	
Medial Tibia	**MT-a***	**Unloaded**	**28.7 (26.8, 30.7)**	**31.6 (28.3, 34.9)**	n.s.
		Loaded	**24.9 (23.4, 26.3)**	**26.9 (24.1, 29.6)**	
	MT-c*	**Unloaded**	**30.9 (29.7, 32.1)**	**32.6 (30.6, 34.6)**	n.s.
		Loaded	**26.9 (25.6, 28.2)**	**27.6 (25.7, 29.4)**	
	MT-p*	**Unloaded**	**33.5 (32.4, 34.6)**	**35.3 (33.0, 37.7)**	n.s.
		Loaded	**30.4 (29.0, 31.8)**	**31.1 (28.9, 33.3)**	
Lateral Femur	cLF-a	Unloaded	31.2 (30.0, 32.3)	33.7 (31.8, 35.6)	n.s.
		Loaded	30.8 (29.3, 32.3)	31.0 (29.0, 33.0)	
	cLF-c	Unloaded	33.3 (32.1, 34.4)	35.0 (33.0, 37.0)	n.s.
		Loaded	34.4 (32.9, 35.9)	35.1 (32.7, 37.5)	
	cLF-p	Unloaded	35.1 (34.1, 36.1)	36.1 (34.1, 38.1)	n.s.
		Loaded	36.2 (34.7, 37.7)	37.3 (35.2, 39.4)	
	pLF	Unloaded	33.9 (33.1, 34.7)	36.1 (34.8, 37.5)	n.s.
		Loaded	34.0 (32.6, 35.3)	35.7 (34.0, 37.4)	
Lateral Tibia	**LT-a***	**Unloaded**	**31.6 (30.3, 32.8)**	**34.7 (32.5, 36.9)**	n.s.
		Loaded	**29.2 (27.7, 30.7)**	**30.0 (27.7, 32.3)**	
	LT-c*	**Unloaded**	**29.0 (28.0, 30.0)**	**31.4 (29.6, 33.2)**	**p = 0.023**
		Loaded	**28.0 (26.5, 29.4)**	**27.5 (25.6, 29.5)**	

			Control	Osteoarthritis	Group by Condition Interaction
			Mean (95% Confidence Intervals)	Mean (95% Confidence Intervals)	
	LT-p*	Unloaded	34.7 (33.7, 35.7)	35.1 (33.7, 36.5)	p = 0.009
		Loaded	33.7 (32.4, 35.0)	31.0 (29.3, 32.7)	

* indicates loaded condition is significantly different from unloaded condition across both groups at p<0.05. MF: medial femur; LF: lateral femur; MT: medial tibia; LT: lateral tibia; cXF-x refers to the portion of the central femoral condyle that is anterior (a), central (c), or posterior (p).

Table 6

Sub-compartment T2

			Control	Osteoarthritis	Group by Condition Interaction
			Mean (95% Confidence Intervals)	Mean (95% Confidence Intervals)	
Medial Femur	cMF-a	Unloaded	25.4 (24.5, 26.3)	27.0 (24.9, 29.2)	n.s.
		Loaded	24.7 (23.7, 25.7)	26.7 (25.0, 28.5)	
	cMF-c	Unloaded	23.1 (22.3, 23.9)	24.6 (22.8, 26.3)	n.s.
		Loaded	23.3 (22.4, 24.1)	24.6 (22.9, 26.3)	
	cMF-p	Unloaded	24.3 (23.4, 25.1)	24.2 (22.5, 25.9)	n.s.
		Loaded	24.3 (23.4, 25.3)	25.5 (23.8, 27.3)	
	pMF	Unloaded	26.9 (26.3, 27.5)	27.6 (26.7, 28.5)	**p = 0.027**
		Loaded	27.3 (26.5, 28.0)	29.5 (28.2, 30.7)	
Medial Tibia	**MT-a***	**Unloaded**	**18.7 (17.7, 19.6)**	**18.6 (16.7, 20.4)**	**p = 0.010**
		Loaded	**18.0 (17.2, 18.9)**	**20.9 (19.3, 22.6)**	
	MT-c*	**Unloaded**	**22.2 (21.3, 23.0)**	**22.3 (20.8, 23.8)**	n.s.
		Loaded	**20.2 (19.3, 21.0)**	**21.7 (20.2, 23.3)**	
	MT-p*	**Unloaded**	**23.3 (22.4, 24.3)**	**23.5 (21.7, 25.3)**	n.s.
		Loaded	**21.6 (20.5, 22.6)**	**22.5 (20.8, 24.3)**	
Lateral Femur	cLF-a	Unloaded	22.0 (21.1, 22.9)	23.5 (21.8, 25.2)	n.s.
		Loaded	21.5 (20.7, 22.4)	22.9 (20.9, 24.9)	
	cLF-c	Unloaded	24.4 (23.6, 25.2)	26.2 (24.7, 27.8)	n.s.
		Loaded	24.7 (23.8, 25.6)	25.5 (23.4, 27.6)	
	cLF-p	Unloaded	26.0 (25.2, 26.8)	25.9 (24.3, 27.6)	n.s.
		Loaded	26.4 (25.5, 27.4)	27.1 (25.4, 28.8)	
	pLF	Unloaded	24.4 (23.7, 25.0)	25.6 (24.4, 26.9)	n.s.
		Loaded	25.3 (24.5, 26.1)	26.9 (25.3, 28.5)	
Lateral Tibia	**LT-a***	**Unloaded**	**21.1 20.2, 22.0)**	**22.9 (21.5, 24.3)**	n.s.
		Loaded	**20.1 (19.1, 21.1)**	**21.7 (20.3, 23.0)**	
	LT-c	Unloaded	20.3 (19.4, 21.1)	21.5 (19.9, 23.0)	n.s.
		Loaded	19.9 (18.8, 20.9)	20.5 (19.1, 21.9)	

			Control	Osteoarthritis	Group by Condition Interaction
			Mean (95% Confidence Intervals)	Mean (95% Confidence Intervals)	
	LT-p	Loaded	24.5 (23.7, 25.3)	24.0 (22.7, 25.3)	n.s.
		Loaded	24.2 (23.3, 25.2)	23.9 (22.2, 25.6)	

* indicates loaded condition is significantly different from unloaded condition across both groups at $p<0.05$. MF: medial femur; LF: lateral femur; MT: medial tibia; LT: lateral tibia; cXF-x refers to the portion of the central femoral condyle that is anterior (a), central (c), or posterior (p).