Published in final edited form as:

Magn Reson Med. 2017 March ; 77(3): 1276–1283. doi:10.1002/mrm.26214.

Quantitative susceptibility mapping (QSM) detects abnormalities in cartilage canals in a goat model of preclinical osteochondritis dissecans (OCD)

Luning Wang^{1,2,*}, Mikko J. Nissi^{3,4}, Ferenc Toth^{2,5}, Casey P. Johnson¹, Michael Garwood¹, Cathy S. Carlson^{2,5}, and Jutta Ellermann¹

¹Center for Magnetic Resonance Research (CMRR), Department of Radiology, University of Minnesota, Minneapolis, MN, United States ²Department of Orthopaedic Surgery, University of Minnesota, Minneapolis, MN, United States ³Department of Applied Physics, University of Eastern Finland, Kuopio, Finland ⁴Diagnostic Imaging Center, Kuopio University Hospital, Kuopio, Finland ⁵Department of Veterinary Population Medicine, College of Veterinary Medicine, University of Minnesota, Saint Paul, MN, United States

Abstract

Purpose—To use quantitative susceptibility mapping (QSM) to investigate changes in cartilage canals in the distal femur of juvenile goats after their surgical transection.

Methods—Chondronecrosis was surgically induced in the right medial femoral condyles of four 4-day-old goats. Both the operated and control knees were harvested at 2, 3, 5, and 10 weeks after the surgeries. Ex vivo MRI scans were conducted at 9.4T using T_{RAFF}-weighted fast spin echo imaging and QSM to detect areas of chondronecrosis and investigate cartilage canal abnormalities. Hematoxylin & eosin (H&E) and safranin O stained histological sections from these same areas were evaluated to assess the affected tissues.

Results—Both the histological sections and the T_{RAFF}-weighted images of the femoral condyles demonstrated focal areas of chondronecrosis, evidenced by pyknotic chondrocyte nuclei, loss of matrix staining, and altered MR image contrast. At increasing time-points after surgery, progressive changes and eventual disappearance of abnormal cartilage canals were observed in areas of chondronecrosis by using QSM.

Conclusion—Abnormal cartilage canals were directly visualized in areas of surgically-induced chondronecrosis. QSM enabled investigation of the vascular changes accompanying chondronecrosis in juvenile goats.

Keywords

osteochondritis dissecans; osteochondrosis; epiphyseal cartilage; chondronecrosis; vascular canal abnormality; cartilage canals; quantitative susceptibility mapping; high field MRI; OCD; QSM

*Corresponding Author: Luning Wang, Ph.D., Center for Magnetic Resonance Research (CMRR), University of Minnesota, Twin Cities, 2021 6th Street SE, Minneapolis, MN 55455 USA, wang5517@umn.edu.

Introduction

Osteochondritis dissecans (OCD) is a developmental orthopedic disease that may cause premature osteoarthritis later in life (1). OCD most commonly occurs in the medial femoral condyle of the knee and is characterized by the presence of a cartilaginous or osseocartilaginous flap extending into the joint causing pain, swelling, and locking (2). The etiology of OCD is unknown, but one theory implicates ischemic insult to the richly vascularized epiphyseal cartilage. The epiphyseal cartilage is located subjacent to the avascular articular cartilage and is gradually replaced by bone during development. The ischemic theory is based on animal studies, which have demonstrated that surgical interruption of cartilage canal vessels (minute structures providing vascular supply to the epiphyseal cartilage) causes ischemic chondronecrosis, leading to a focal failure of enchondral ossification, a lesion that is pathognomonic for osteochondrosis (OC). OC is known to precede OCD in domestic animals, although this has not yet been proven in humans (3-5). Areas of ischemic chondronecrosis are characterized by proteoglycan loss and collagen matrix disruption (6-9), and the resulting lesions can be detected using MRI parametric mapping of relaxation times such as T_2 , $T_{1\rho}$, and T_{RAFF} (relaxation time along a fictitious field) (10-18).

While histological techniques can be used for ex vivo visualization of epiphyseal cartilage canals, new imaging methods are needed to evaluate these structures in vivo for longitudinal studies and potentially for diagnostic purposes. Toward this end, we recently demonstrated the feasibility of visualizing cartilage canals noninvasively using susceptibility-weighted imaging (SWI) and quantitative susceptibility mapping (QSM) (19,20). Both methods take advantage of MR phase, which demonstrates dipolar field patterns that are a few times larger than the size of the small blood vessels that are the source of the field disturbance. Although multiple MRI methods have been proposed for noninvasive detection of OC lesions (14,17,21-23), and the visualization of cartilage canals has been demonstrated (19,20), direct visualization of abnormalities in the epiphyseal cartilage canals using phase sensitive MRI has not been reported.

The purpose of this work was to use QSM to evaluate changes involving cartilage canals after their surgical transection. To achieve this goal we have utilized a goat model of OC (3,4,24) in which ischemic chondronecrosis of an area of the medial femoral condyle is surgically induced. Harvested goat knees were scanned ex vivo at increasing time points after surgically-induced chondronecrosis using a 9.4 T MRI scanner. We hypothesized that (i) surgical transection of cartilage canal vessels would result in morphological changes to these vessels in and adjacent to areas of chondronecrosis and (ii) these changes could be detected noninvasively using QSM.

Methods

Animal model

Details of the animal model have been reported previously in detail (4). Briefly, to create chondronecrosis in the epiphyseal cartilage, four 4-day-old goats underwent a surgical procedure in the right femorotibial joint to interrupt the blood supply to a focal area of the

epiphyseal cartilage in the central (axial) aspect of the right medial femoral condyle (MFC). The left, non-operated femorotibial joints were used as controls. Both distal femora of the juvenile goats were harvested at 2, 3, 5, and 10 weeks after the surgeries for ex vivo MRI scans. This study was approved by the Institutional Animal Care and Use Committee (IACUC) of the University of Minnesota.

Ex vivo MRI

Individual distal femora were suspended in flexible latex containers filled with perfluoropolyether oil for a proton-free and susceptibility-matched background. MRI experiments were conducted using a 9.4 T Varian scanner for small animal studies (Agilent Technologies, Santa Clara, CA, USA). The specimens were placed at the center of a shielded quadrature volume coil (Millipede, Varian NMR Systems, Palo Alto, CA), which was used for both RF transmission and signal acquisition. B_0 field shimming and RF pulse calibration were conducted for each imaging session.

Based on our previous study showing high sensitivity of T_{RAFF} to cartilage necrosis (17), a T_{RAFF} -weighted 2D fast spin echo (FSE) sequence was utilized to identify the area of chondronecrosis in each sample. Specifically, the T_{RAFF} contrast was created by magnetization preparation consisting of 24 RAFF pulses of 4.53 ms RF duration (15,16). The first echo time (TE) was set to the shortest possible value of 5.38 ms and used as the effective TE of the FSE sequence to minimize T_2 contrast, and a long repetition time (TR) of 5.0 s was used to allow full recovery of longitudinal magnetization. The echo train length (ETL) was set to 8. The matrix size was set to 256^2 with 1.0 mm thickness in the coronal plane. The acquisition bandwidth was set to 500 Hz/pixel. A square field-of-view (FOV) of 40 mm was adjusted to the sample size, providing a high in-plane resolution of approximately 150 μm . The number of averages was one. The scan time per specimen was about 3 minutes.

A three dimensional (3D) gradient-recalled echo (GRE) sequence was utilized for QSM to visualize the blood vessels in epiphyseal cartilage. Specifically, a cubic 40 mm FOV was adjusted to the physical size of individual samples with a constant matrix size of 384^3 , resulting in an isotropic resolution of approximately 100 μm . A 15° flip angle was used for all scans. TR and TE were 40 ms and 15 ms, respectively. In order to enhance the susceptibility-induced phase contrast, the acquisition bandwidth was reduced to 43 Hz/pixel, the lowest value allowable for the selected TE and matrix size. The number of averages was one. The scan time per specimen was about 98 minutes.

Histology

Following the MRI scans, the distal femora were fixed in 10% neutral-buffered formalin for 48 hours and then decalcified by immersion in 10% ethylenediaminetetraacetic acid for several weeks. After decalcification, the femoral condyles were serially sectioned in the coronal plane to match the MRI studies, into 2 to 3 mm thick slabs, which were routinely processed into paraffin blocks. Sections of 5 μm thickness were obtained from the surface of each block and stained with hematoxylin & eosin (H&E). If cartilage necrosis was present in the H&E-stained sections, additional sections were obtained 200 to 500 μm intervals deep to

the original block face, until the lesion was no longer apparent, to ensure that its entire extent was sampled. Selected sections were then stained with safranin O to qualitatively evaluate the associated proteoglycan loss.

Quantitative susceptibility mapping (QSM)

The 3D GRE raw data were read into Matlab (Matlab 2013b, Mathworks, Natick, MA, USA) and Fourier transformed to produce the magnitude and phase images. The cartilage was segmented based on the magnitude images using ITK-SNAP (www.itksnap.org) to create a binary cartilage mask for the further processing (25). Prior to the QSM processing, the raw phase was unwrapped using the Laplacian unwrapping method (26), followed by the SHARP filtering method to remove the background field (27). Because the epiphyseal cartilage was thin (less than 3 mm for all goat knees, with decreasing thickness as the goats aged), a kernel size of $3 \times 3 \times 3$ and a truncation threshold of 0.05 were selected for the SHARP filtering based on visual assessment of the quality of the filtering (28). This kernel size resulted in a filter length of approximately 0.3 mm. The susceptibility maps were then derived from the pre-processed phase based on the regularized QSM method with automatic parameter selection (28). 2D maximum intensity projection (MIP) images of the GRE magnitude images as well as the obtained 3D susceptibility maps were generated in the coronal and sagittal planes with about 1 mm thickness (about 10 slices), and the coronal MIP images were selected to match the slice positions of the T_{RAFF} -weighted FSE images showing regions of chondronecrosis. Based on the susceptibility images, cartilage canals in areas of chondronecrosis and the contralateral condyle were manually segmented, and the means of the estimated susceptibility values of abnormal and normal cartilage canals were calculated. To further validate the progressive changes, 3D volumetric MIP images were generated using OsiriX (Pixmeo, Geneva, Switzerland) (29) to illustrate the cartilage canals in the epiphyseal cartilage of the operated condyles at different time points.

Results

Both H&E and safranin O stained sections of the femoral condyles, as well as T_{RAFF} -weighted FSE images, identified areas of ischemic chondronecrosis characteristic for various stages of OC. Specifically, in H&E stained sections, nuclei of necrotic chondrocytes were pyknotic and the surrounding matrix exhibited progressive loss of extracellular matrix staining with increased time after surgery (Figures 1a-d) (3,4,24). Progressive loss of safranin O staining, consistent with continuous reduction of proteoglycan content was also observed (Figure 1e-h). Soon after the vascular insult (within days) endothelial necrosis in the cartilage canal vessels was apparent and by 1-2 weeks after surgery the vessels became empty of contents and dilated. By 2-3 weeks, chondrified cartilage canals intensely staining with safranin O were present (lumina filled with cartilage) distal to the surgical incisions (Figures 1e and 1f). At 10 weeks post-surgery, new blood vessels (arrows, Figure 1h) appeared at the interface between the necrotic and viable epiphyseal cartilage. At this late stage, necrotic cartilage was partially incorporated in the bone due to the progression of the ossification front (Figure 1h). In the T_{RAFF} -weighted FSE images, lesions of chondronecrosis were visible as areas of positive contrast (arrows, Figures 1i-l), and could be clearly distinguished from the normal epiphyseal cartilage at all time points. At 10 weeks

post-surgery, the T_{RAFF} -weighted FSE images also demonstrated that the area of chondronecrosis was partially surrounded by bone (Figure 1l) consistent with the histological results (Figures 1d and 1h).

Intermediate QSM processing results are provided in Figure 2 to evaluate the SHARP filtering and the QSM method. The background fields (Figure 2b) were successfully removed by the SHARP filtering (Figure 2c) with the empirically chosen optimal parameters of $3 \times 3 \times 3$ kernel size and 0.05 truncation level. The cartilage canals can be clearly observed in Figure 2d. Phase in the area of bone marrow appeared as noisy as the background (Figure 2a), and was thus removed with the binary mask. No artifacts from chemical shift between water and fat were observed in the cartilage area of the phase images. The relative susceptibility distribution was successfully estimated using the regularized QSM method, enabling direct visualization of the cartilage canals.

Figures 3 to 6 show 1 mm thick 2D MIPs of the GRE and QSM data at different stages of the chondronecrosis process to demonstrate progressive changes in the operated distal femora. The slice positions were adjusted to closely match the positions of the T_{RAFF} -weighted FSE images (Figures 1i to 1l). At 2 weeks post-surgery, the area of chondronecrosis was not clearly demarcated in the GRE image (Figure 3a) compared to the T_{RAFF} -weighted image (Figure 1i). Both the susceptibility and the morphology of the cartilage canals in the area of chondronecrosis (arrows, Figures 3b and d) appeared similar to those of the normal epiphyseal cartilage (Figures 3f and h) at this early stage. Both the operated and control knees were visually similar, with blood vessels visible in coronal and sagittal planes extending from the ossification front toward the articular cartilage.

At 3 weeks post-surgery, the area of chondronecrosis showed slightly increased signal intensity in the GRE images (Figures 4a and c). Failed ossification of the area of chondronecrosis was demonstrated in the sagittal plane (Figure 4c). Disintegration of the affected cartilage canals continued from 2 weeks post-surgery to 3 weeks post-surgery (Figures 1e and 1f), and was identified by QSM in the area of chondronecrosis (Figures 4b and 3d, Figure 7b). The average susceptibility of the abnormal cartilage canals was approximately 0.032 ppm (arrow, Figures 4b and d, Table 1), which was lower than that of the normal vascular canals in the contralateral condyle (approximately 0.054 ppm, Table 1). For the control site, the transition from cartilage to bone at the ossification front was smooth (curvilinear) (Figures 4e and g), and no cartilage canal abnormalities were seen (Figures 4f and h).

At 5 weeks post-surgery, areas of chondronecrosis were detectable with positive contrast on the GRE images (Figures 5a and c). Unlike the previous two time points, the abnormal cartilage canals in the area of chondronecrosis had ill-defined margins in the QSM images (arrows, Figures 5b and d, Figure 7c), indicating continued disintegration of the cartilage canal structures. The observation was consistent with histological results (Figure 1g), in which cartilage canals were difficult to identify. A decreased susceptibility value consistently was observed in the operated site compared to the normal cartilage canals in the contralateral condyle (Table 1). In the control knee, all blood vessels that were present had sharply-defined margins (Figures 5f and h).

At 10 weeks post-surgery, the region of chondronecrosis had resulted in an area of delayed endochondral ossification due to failure of ossification of the necrotic cartilage (Figures 6a and c), while unaffected epiphyseal cartilage was replaced by subchondral bone. The disintegrated vascular canals had vanished or were poorly visible in the area of chondronecrosis (Figures 6b, 6d, and 7d). However, new cartilage canals were visible using QSM at the interface of healthy and necrotic epiphyseal cartilage (arrows, Figures 6b, 6d, and 7d). The presence of new vessels was confirmed histologically (Figure 1h). In contrast, the control knee had a curvilinear ossification front (Figures 6e and g) and normal blood vessels distributed throughout the epiphyseal cartilage (Figures 6f and h).

The estimated susceptibility values of the normal cartilage canals in the contralateral condyles of the operated knees appeared stable with age, as indicated by the smaller standard deviation for the mean value, while the susceptibility values were reduced at later time points in the operated knees (Table 1). In addition to the 2D MIPs (Figures 3 to 6), the 3D MIPs at different time points further demonstrated the progressive changes in the cartilage canals in the epiphyseal cartilage of the operated condyle (Figure 7), including reduced susceptibility and the ill-defined margins caused by disintegration. Eventually, these abnormal cartilage canals vanished in areas of chondronecrosis (Figure 7d), and new cartilage canals appeared at the peripheral regions of the chondronecrosis (arrow, Figure 7d).

Discussion

Progressive changes in the morphology of epiphyseal cartilage canal vessels in regions of ischemia-induced chondronecrosis was demonstrated in this study using QSM. This technique is complementary to imaging methods that are sensitive to cartilage matrix changes associated with OC lesions, such as T_{RAFF} parametric mapping (17), and may provide valuable insight into the etiology and pathogenesis of OCD. Furthermore, the direct visualization of abnormal cartilage canals in epiphyseal cartilage may be clinically useful, as it provides a potential noninvasive method to identify subclinical lesions of OCD in pediatric patients (3,4,24,30). Although cartilage matrix changes occurring subsequent to ischemia have been identified using 3T and higher field magnets, both in vivo and ex vivo, it is still challenging to visualize progressive changes of the affected cartilage canals using non-invasive MRI techniques. Our results suggest that QSM is a good candidate for this task.

The abnormalities in vascular canals identified with QSM are consistent with the histological findings, and provide evidence of a pattern of vascular decline and regeneration in OC. At the very early stage of ischemic chondronecrosis (2 weeks post-surgery), QSM identified blood vessels undergoing necrosis or chondrification, which appeared similar to those in normal epiphyseal cartilage in ex vivo MRI. As the cartilage necrosis progressed (3 weeks post-surgery), susceptibility changes of cartilage canals became more apparent by QSM. Whether these were due to vascular necrosis or to subsequent chondrification of the affected vessels is unclear. Regardless, the affected vascular canals demonstrated lower relative susceptibility compared to the unaffected vascular canals in the susceptibility maps, even though the morphology of the abnormal cartilage canals still appeared similar to the normal cartilage canals. At 5 weeks post-surgery, vascular canals were poorly visible in or adjacent to the area of necrosis in the safranin O stained section of femoral condyle.

Disintegration of the abnormal cartilage canals is a possible explanation for the ill-defined vascular canals seen with QSM at this time point. 10 weeks post-surgery, the area of chondronecrosis was partially surrounded by bone due to the advancement of the ossification front. This process was delayed in the area of chondronecrosis but progressed normally in adjacent healthy cartilage. Eventually, abnormal vascular canals vanished, and revascularization commenced as new blood vessels appeared at the interface of normal epiphyseal cartilage and areas of chondronecrosis. This finding has been reported previously in histological preparations and suggests that the healing mechanism of OC involves neovascularization at the margins of the lesion (30,31).

QSM imaging of the epiphyseal cartilage vasculature is technically challenging. During skeletal maturation, the epiphyseal cartilage volume decreases rapidly as the cartilage is replaced by the advancing ossification front. Therefore, high-resolution 3D images with sufficient SNR are needed. In order to apply the QSM method, the phase images are typically filtered so that the background field in the original phase image is eliminated. In the present study, the SHARP filtering method was implemented for this purpose (27). A large kernel size, typically used in brain applications, could have resulted in undesirable removal of voxels at the margins of the epiphyseal cartilage. Therefore, a small kernel of $3 \times 3 \times 3$ was selected to avoid this artifact during the SHARP filtering, while ensuring that no significant errors would be introduced to the reconstructed susceptibility maps. The progressively diminishing thickness of the epiphyseal cartilage that occurs naturally with age is particularly problematic for QSM. If the epiphyseal cartilage becomes too thin it may not be possible to reconstruct susceptibility maps of the tissue. Strategies such as spatial interpolation or zero filling of the k-space could then be utilized for increasing the apparent spatial resolution.

A recent review article summarized the basic technical concepts, clinical applications and challenges of QSM (32). It is noted that chemical shift between water and fat may result in an inaccurate estimation of susceptibility; therefore, QSM with chemical shift correction is desirable in applications such as breast imaging (33). The epiphyseal cartilage, mainly composed of water, type II collagen and proteoglycan, is free of fat, and the bone-cartilage boundary is the only location where the chemical shift may be present. However, the MR phase in the area of the bone marrow (containing fat) appeared similar to the background noise (Figure 2a). Therefore, it is assumed that the chemical shift would act more like the background noise and reduce the phase SNR at the edge of the cartilage. In the unwrapped phase images (Figures 2b and 2c), due to the high susceptibility, strong phase perturbation from cartilage canals was observed, but no obvious phase perturbation or artifact from bone marrow was evident in the unwrapped, SHARP filtered phase (Figure 2c) that was used for QSM. Furthermore, the SHARP filtering removed a few voxels at the bone-cartilage boundary, which further eliminated the influence from chemical shift between water and fat. Thus, chemical shift between water and fat is not considered a major issue in this study. However, the chemical QSM method (33) should be explored and compared with the regularized QSM methods (28,34) in future in vivo studies in humans.

Although important information regarding the imaging of epiphyseal vasculature was obtained in this study, there were several limitations. First, abnormal cartilage canals were

evaluated in only four specimens and imaging was done only ex vivo. Additional animals will be included in future studies to assess how statistically robust these preliminary findings are. However, the evidence provided herein clearly demonstrates a distinct longitudinal pattern of cartilage canal degeneration and revascularization in areas of ischemic chondronecrosis, and the MRI results closely matched the histological results. Second, this study was conducted at 9.4 T and, although this is suitable for ex vivo studies of small joints, future work will need to determine the utility of the QSM method in vivo, for potential clinical imaging of the epiphyseal cartilage. Third, although a precise biological explanation of the susceptibility changes associated with the abnormal cartilage canals is important, it is beyond the scope of this preliminary work.

Conclusion

Quantitative susceptibility mapping (QSM) enabled direct visualization of progressive degeneration of epiphyseal cartilage canals in an animal model of OC. This method may provide important insights into the etiology and pathogenesis of juvenile OC dissecans and other developmental joint diseases, particularly if it can be applied in vivo at lower magnetic field strengths. Furthermore, this noninvasive method has the potential to be useful in the clinical evaluation of pediatric patients with suspected joint disease.

Acknowledgments

This research was supported in part by NIH/NIAMS R21AR065385, NIH T32OD10993, NIH/NCRR K18RR033297, NIH P41EB015894, the WM KECK Foundation, the Comparative Medicine Signature Program at the University of Minnesota, and the Academy of Finland grant #285909.

References

1. Pascual-Garrido C, Moran CJ, Green DW, Cole BJ. Osteochondritis dissecans of the knee in children and adolescents. *Curr Opin Pediatr*. 2013; 25(1):46–51. [PubMed: 23128839]
2. Bates JT, Jacobs JC Jr, Shea KG, Oxford JT. Emerging genetic basis of osteochondritis dissecans. *Clin Sports Med*. 2014; 33(2):199–220. [PubMed: 24698039]
3. Toth F, Nissi MJ, Zhang J, Benson M, Schmitter S, Ellermann JM, Carlson CS. Histological confirmation and biological significance of cartilage canals demonstrated using high field MRI in swine at predilection sites of osteochondrosis. *J Orthop Res*. 2013; 31(12):2006–2012. [PubMed: 23939946]
4. Toth F, Nissi MJ, Wang L, Ellermann JM, Carlson CS. Surgical induction, histological evaluation, and MRI identification of cartilage necrosis in the distal femur in goats to model early lesions of osteochondrosis. *Osteoarthritis and Cartilage*. 2015; 23(2):300–307. [PubMed: 25463443]
5. Carlson CS, Hilley HD, Meuten DJ. Degeneration of cartilage canal vessels associated with lesions of osteochondrosis in swine. *Vet Pathol*. 1989; 26(1):47–54. [PubMed: 2913703]
6. Poole AR, Kobayashi M, Yasuda T, Lavery S, Mwale F, Kojima T, Sakai T, Wahl C, El-Maadawy S, Webb G, Tchetina E, Wu W. Type II collagen degradation and its regulation in articular cartilage in osteoarthritis. *Ann Rheum Dis*. 2002; 61(2):ii78–81. [PubMed: 12379630]
7. Nakano T, Thompson JR, Aherne FX. Cartilage proteoglycans from normal and osteochondrotic porcine joints. *Can J Comp Med*. 1985; 49(2):219–226. [PubMed: 3926288]
8. Ekman S, Heinegard D. Immunohistochemical localization of matrix proteins in the femoral joint cartilage of growing commercial pigs. *Vet Pathol*. 1992; 29(6):514–520. [PubMed: 1448898]
9. Carlson CS, Hilley HD, Henrikson CK, Meuten DJ. The ultrastructure of osteochondrosis of the articular-epiphyseal cartilage complex in growing swine. *Calcif Tissue Int*. 1986; 38(1):44–51. [PubMed: 3079652]

10. Nieminen MT, Rieppo J, Toyras J, Hakumaki JM, Silvennoinen J, Hyttinen MM, Helminen HJ, Jurvelin JS. T2 relaxation reveals spatial collagen architecture in articular cartilage: a comparative quantitative MRI and polarized light microscopic study. *Magn Reson Med*. 2001; 46(3):487–493. [PubMed: 11550240]
11. Menezes NM, Gray ML, Hartke JR, Burstein D. T2 and T1rho MRI in articular cartilage systems. *Magn Reson Med*. 2004; 51(3):503–509. [PubMed: 15004791]
12. Mosher TJ, Dardzinski BJ. Cartilage MRI T2 relaxation time mapping: overview and applications. *Semin Musculoskeletal Radiol*. 2004; 8(4):355–368.
13. Duvvuri U, Reddy R, Patel SD, Kaufman JH, Kneeland JB, Leigh JS. T1rho-relaxation in articular cartilage: effects of enzymatic degradation. *Magn Reson Med*. 1997; 38(6):863–867. [PubMed: 9402184]
14. Regatte RR, Akella SV, Borthakur A, Kneeland JB, Reddy R. Proteoglycan depletion-induced changes in transverse relaxation maps of cartilage: comparison of T2 and T1rho. *Acad Radiol*. 2002; 9(12):1388–1394. [PubMed: 12553350]
15. Liimatainen T, Sorce DJ, O'Connell R, Garwood M, Michaeli S. MRI contrast from relaxation along a fictitious field (RAFF). *Magn Reson Med*. 2010; 64(4):983–994. [PubMed: 20740665]
16. Liimatainen T, Hakkarainen H, Mangia S, Huttunen JM, Storino C, Idiyatullin D, Sorce D, Garwood M, Michaeli S. MRI contrasts in high rank rotating frames. *Magn Reson Med*. 2015; 73(1):254–262. [PubMed: 24523028]
17. Wang L, Nissi MJ, Toth F, Shaver J, Johnson CP, Zhang J, Garwood M, Carlson CS, Ellermann JM. Multiparametric MRI of Epiphyseal Cartilage Necrosis (Osteochondrosis) with Histological Validation in a Goat Model. *PloS one*. 2015; 10(10):e0140400. [PubMed: 26473611]
18. Nissi MJ, Rieppo J, Toyras J, Laasanen MS, Kiviranta I, Jurvelin JS, Nieminen MT. T(2) relaxation time mapping reveals age- and species-related diversity of collagen network architecture in articular cartilage. *Osteoarthritis and Cartilage*. 2006; 14(12):1265–1271. [PubMed: 16843689]
19. Nissi MJ, Toth F, Zhang J, Schmitter S, Benson M, Carlson CS, Ellermann JM. Susceptibility weighted imaging of cartilage canals in porcine epiphyseal growth cartilage ex vivo and in vivo. *Magn Reson Med*. 2014; 71(6):2197–2205. [PubMed: 23857631]
20. Nissi MJ, Toth F, Wang L, Carlson CS, Ellermann JM. Improved Visualization of Cartilage Canals Using Quantitative Susceptibility Mapping. *PloS one*. 2015; 10(7):e0132167. [PubMed: 26168296]
21. Rautiainen J, Nissi MJ, Salo EN, Tiitu V, Finnla MA, Aho OM, Saarakkala S, Lehenkari P, Ellermann J, Nieminen MT. Multiparametric MRI assessment of human articular cartilage degeneration: Correlation with quantitative histology and mechanical properties. *Magn Reson Med*. 2015; 74:249–259.
22. Ellermann J, Ling W, Nissi MJ, Arendt E, Carlson CS, Garwood M, Michaeli S, Mangia S. MRI rotating frame relaxation measurements for articular cartilage assessment. *Magn Reson Imaging*. 2013; 31(9):1537–1543. [PubMed: 23993794]
23. Regatte RR, Akella SV, Borthakur A, Kneeland JB, Reddy R. In vivo proton MR three-dimensional T1rho mapping of human articular cartilage: initial experience. *Radiology*. 2003; 229(1):269–274. [PubMed: 14519880]
24. Toth F, Nissi J, Ellermann J, Wang L, Shea KG, Polousky J, Carlson CS. Novel Application of Magnetic Resonance Imaging Demonstrates Characteristic Differences in Vasculature at Predilection Sites of Osteochondritis Dissecans. *Am J Sports Med*. 2015; 43(10):2522–2527. [PubMed: 26286878]
25. Yushkevich PA, Piven J, Hazlett HC, Smith RG, Ho S, Gee JC, Gerig G. User-guided 3D active contour segmentation of anatomical structures: significantly improved efficiency and reliability. *NeuroImage*. 2006; 31(3):1116–1128. [PubMed: 16545965]
26. Li W, Wu B, Liu C. Quantitative susceptibility mapping of human brain reflects spatial variation in tissue composition. *NeuroImage*. 2011; 55(4):1645–1656. [PubMed: 21224002]
27. Schweser F, Deistung A, Lehr BW, Reichenbach JR. Quantitative imaging of intrinsic magnetic tissue properties using MRI signal phase: an approach to in vivo brain iron metabolism? *NeuroImage*. 2011; 54(4):2789–2807. [PubMed: 21040794]

28. Bilgic B, Fan AP, Polimeni JR, Cauley SF, Bianciardi M, Adalsteinsson E, Wald LL, Setsompop K. Fast quantitative susceptibility mapping with L1-regularization and automatic parameter selection. *Magn Reson Med*. 2014; 72(5):1444–1459. [PubMed: 24259479]
29. Rosset A, Spadola L, Ratib O. OsiriX: an open-source software for navigating in multidimensional DICOM images. *J Digit Imaging*. 2004; 17(3):205–216. [PubMed: 15534753]
30. McCoy AM, Toth F, Dolvik NI, Ekman S, Ellermann J, Olstad K, Ytrehus B, Carlson CS. Articular osteochondrosis: a comparison of naturally-occurring human and animal disease. *Osteoarthritis and Cartilage*. 2013; 21(11):1638–1647. [PubMed: 23954774]
31. Ytrehus B, Carlson CS, Ekman S. Etiology and pathogenesis of osteochondrosis. *Vet Pathol*. 2007; 44(4):429–448. [PubMed: 17606505]
32. Wang Y, Liu T. Quantitative susceptibility mapping (QSM): Decoding MRI data for a tissue magnetic biomarker. *Magn Reson Med*. 2015; 73(1):82–101. [PubMed: 25044035]
33. Dimov AV, Liu T, Spincemaille P, Ecanow JS, Tan H, Edelman RR, Wang Y. Joint estimation of chemical shift and quantitative susceptibility mapping (chemical QSM). *Magn Reson Med*. 2015; 73(6):2100–2110. [PubMed: 24947227]
34. de Rochefort L, Liu T, Kressler B, Liu J, Spincemaille P, Lebon V, Wu J, Wang Y. Quantitative susceptibility map reconstruction from MR phase data using bayesian regularization: validation and application to brain imaging. *Magn Reson Med*. 2010; 63(1):194–206. [PubMed: 19953507]

Fig. 1.

The H&E (top row) and safranin O (middle row) stained histological sections of femoral condyles illustrate the lesions of induced ischemic chondronecrosis inside the dotted curves at 2, 3, 5, and 10 weeks post-surgery (left to right). Areas of chondronecrosis are characterized by pallor of the matrix in both the H&E and safranin O stained sections that increases with time after surgery. Chondrified cartilage canals at the margins of the lesion (e) 2 weeks and (f) 3 weeks post-surgery are indicated by long arrows and enlarged in insets. Cartilage canal vessels were indistinguishable (g) 5 weeks post-surgery. A cluster of viable blood vessels was identified at (h) 10 weeks post-surgery at the margin of the lesion (arrow). The area of chondronecrosis is partially surrounded by bone at (h) 10 weeks post-surgery due to progression of the ossification front. 2D T_{RAFF}-weighted FSE images (bottom row) illustrate the area of chondronecrosis (arrows) at the same time points with increased MR image contrast. Insets show higher magnification of the medial femoral condyle.

Fig. 2. Intermediate results of the operated goat knee at 2 weeks post-surgery to demonstrate (a) raw phase, (b) unwrapped phase before SHARP filtering and (c) after SHARP filtering, and (d) the derived susceptibility map.

Fig. 3.

Comparison of the GRE and QSM images of the (a-d) operated and (e-h) control goat knees at 2 weeks post-surgery. The GRE images of the operated condyle are enlarged and illustrated in the (a) coronal and (c) sagittal planes with an arrow pointing to the region of chondronecrosis. The corresponding cartilage canals reconstructed by QSM are also shown with a slight magnification in the (b) coronal and (d) sagittal planes, with an arrow pointing to the cartilage canals within the area of chondronecrosis. These vascular canals appear similar to those in normal epiphyseal cartilage. The GRE images of the control condyle are enlarged and illustrated on the (e) coronal and (g) sagittal planes. The corresponding cartilage canals are also shown on the (f) coronal and (h) sagittal planes. No cartilage canal abnormalities are identified. The dynamic range of the susceptibility MIP images is from -0.05 ppm to 0.12 ppm.

Fig. 4.

Comparison of the GRE and QSM images of the operated and control condyles at 3 weeks post-surgery displayed in the same manner as described in Figure 3. The area of chondronecrosis in the operated knee has resulted in abnormal cartilage canals with reduced susceptibility compared to surrounding normal cartilage canals were identified in this region (arrows, b and d). A curvilinear ossification front and no cartilage canal abnormalities were seen in the control knee (e-h). The dynamic range of the susceptibility MIP images is from -0.05 ppm to 0.12 ppm.

Fig. 5.

Comparison of the GRE and QSM images of the operated and control goat knees at 5 weeks post-surgery displayed in the same manner as described in Figures 3 and 4. Areas of chondronecrosis are present in the GRE images (arrows, a and c), and the corresponding cartilage canals (arrows, b and d) appear ill-defined morphologically compared to the cartilage canals outside the region of chondronecrosis. In contrast, no cartilage canal abnormalities were identified in the control knee (e-h). The dynamic range of the susceptibility MIP images is from -0.05 ppm to 0.12 ppm.

Fig. 6.

Comparison of the GRE and QSM images of the operated and control goat knees at 10 weeks post-surgery displayed in the same manner as described in Figures 3-5. The region of chondronecrosis is partially surrounded by bone due to a delay in endochondral ossification at the site of the lesion (arrows, a and c). In the area of chondronecrosis, no abnormal cartilage canals are identified, but blood vessels are present at the margins of the lesion (arrows, b and d). In the control knee images (h-e), a curvilinear ossification front was present and no cartilage canal abnormalities were identified. The dynamic range of the susceptibility MIP images is from -0.02 ppm to 0.12 ppm.

Fig. 7.

Three dimensional (3D) visualization of the cartilage canals in the epiphyseal cartilage of the operated condyles at (a) 2 weeks, (b) 3 weeks, (c) 5 weeks, and (d) 10 weeks post-surgery. No obvious abnormality of cartilage canals was observed at (a) 2 weeks post surgery. Abnormal cartilage canals with decreased susceptibility were observed at (b) 3 weeks post surgery, pointed by arrows. Disintegrated cartilage canals with ill-defined margins were observed at (c) 5 weeks post surgery, pointed by arrows. The abnormal cartilage canals vanished at (d) 10 weeks post surgery, but new viable cartilage canals, pointed by arrows, appeared at the margin between normal epiphyseal cartilage and the area of chondronecrosis.

Table 1
Susceptibility of cartilage canals in the areas of chondronecrosis and the contralateral condyles of the operated knees at different time points
(unit in [ppm])

Condyles	2 weeks	3 weeks	5 weeks	10 weeks	Mean values
Operated	0.047 ± 0.010	0.032 ± 0.008	0.038 ± 0.010	-	0.039 ± 0.007
Contralateral	0.049 ± 0.011	0.051 ± 0.010	0.045 ± 0.010	0.048 ± 0.012	0.048 ± 0.002
% difference	4.2%	45.8%	16.9%	-	20.7%

- at 10 weeks post surgery, no abnormal cartilage canals were observable by MRI within the area of chondronecrosis