CEST Imaging

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Abstract Chemical exchange saturation transfer (CEST) is a technique that allows generating a contrast in magnetic resonance images based on chemical exchange of free water protons and protons of solute molecules. From the many exogenous and endogenous molecules that have been introduced as CEST contrast agents, CEST contrast mediated by glycosaminoglycans (GAG) in cartilage has been suggested to potentially provide novel avenues for non-invasive investigation of cartilage health. Initial applications showed that the gagCEST contrast is sensitive to the GAG content in cartilage and can detect cartilage defects in patients after cartilage repair surgery in the knee joint.

Keywords CEST · gagCEST · Glycosaminoglycan · GAG · Cartilage · Clinical MRI · 3 Tesla · 7 Tesla

Introduction

Hyaline cartilage tissue consists of a small cellular fraction and an extracellular matrix, which is mainly composed of oriented collagen fibers and an abundant ground substance

rich in proteoglycan (PG) content [1]. PGs are made up of a variety of glycosaminoglycans (GAGs), i.e., long unbranched oligosaccharides that are bound to a protein [2, 3]. Via a strong negative charge, GAGs mediate the attractive potential of PG for cations and their capability to bind water molecules [4]. Since loss of proteoglycans is thought to mark the beginning of osteoarthritis [5, 6], the search for, preferably non-invasive, options to quantify PG content in vivo has emerged as a focus of research.

Several MRI methods such as sodium imaging [7, 8], delayed gadolinium-enhanced MRI of cartilage (dGEMRIC [9]), measurements of the longitudinal relaxation time in the rotating frame ($T_{1\rho}$ [10, 11]), and chemical exchange saturation transfer (CEST [12]) imaging have since been presented as techniques with the potential to measure PG content in cartilage. These techniques exploit the biochemical properties of GAG, i.e., their fixed charge density (sodium and dGEMRIC) or chemical exchange of labile protons with bulk water ($T_{1\rho}$ and gagCEST), respectively. However, a broader use of these techniques in the clinical routine has so far been limited by the low specificity ($T_{1\rho}$ [13]), low signal-to-noise ratio (SNR) at clinical field strengths (sodium imaging [14]) or a complicated measurement protocol involving the administration of a double dose of a gadolinium-based contrast agent and a significant delay between contrast agent administration and MR examination (dGEMRIC [15]).

Since the introduction of the gagCEST method, studies have been performed on patient and volunteer subjects at field strengths of 3.0 and 7.0 T, respectively, to explore the potential of the technique for clinical use. These studies clearly demonstrated the usefulness of the technique to assess the cartilage GAG content at 7.0 T and to evaluate intervertebral disc (IVD) degeneration at 3.0 T [16, 17, 18, 19, 20, 21]. Further studies aimed mostly at general optimization of gagCEST imaging techniques [22–24].

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Especially because gagCEST imaging is entirely non-invasive by using MRI without requiring administration of a contrast agent, and it could provide contrast that is very selective for GAG content in connective tissues, it seems very appealing for clinical use. This is reflected in the growing number of applications of the technique, which is still restricted to research use as the remaining technical challenges, such as scan time and reproducibility, still have to be addressed in greater depth to make the technique available as a commercial clinical product.

Technical Background

CEST Principle

Chemical exchange saturation transfer is a technique in ^1H MR imaging (MRI) and spectroscopy (MRS) that enables quantification of chemical exchange processes between protons bound to solutes and surrounding bulk water molecules [25–27]. A precondition for measuring CEST effects is a chemical system, e.g., a body tissue or fluid, that contains water and solute molecules. These solute molecules must then be sufficiently mobile and carry labile protons; labile in this context means that the protons of the chemical end groups are in constant chemical exchange with the surrounding bulk water protons at a specific rate, which is described by the rate constant k . Due to chemical shifts, the resonance frequency of the labile protons will be different from bulk water, i.e., they are off-resonant; this is similar to the chemical shift between lipid protons and water protons. The CEST effect is then initially induced by labeling or saturating the off-resonant solute protons with a radio-frequency (RF) pulse. Subsequently, the label is transferred to bulk water by chemical exchange. The magnitude of this subsequent reduction of bulk water signal consequently depends on the dynamics of chemical exchange as well as the ratio of exchangeable solute protons to bulk water protons. The rate constant of chemical exchange (k) itself is influenced by the pH value and the temperature within the exchange environment. If the latter two parameters can be assumed to be distributed homogeneously in tissue, such as in connective tissues, the CEST effect will be a surrogate marker for the concentration of a certain species of solute molecule in the tissue.

In order for a solute molecule to be considered suitable as an endogenous CEST agent, it must carry labile protons that exchange with bulk water at exchange rates that fulfill the condition $k \leq \Delta\omega$, where $\Delta\omega$ is the resonance offset of the solute protons to the water protons measured in s^{-1} [28].

Acquisition Techniques

The most common method for acquisition of a CEST data set is to acquire multiple image data sets with pre-saturation at different offset frequencies (S_{sat}) around the water resonance, and one reference data set without saturation (S_0) or with saturation at a very large offset frequency [28, 29•]. The normalized signal as a function of the pre-saturation offset (z -spectrum) can then be used to determine and quantify CEST effects, which are asymmetric with respect to the water resonance, i.e., a CEST effect appears either up- or down-field from water and can hence be extracted from the z -spectrum via analysis of its asymmetry with respect to the water resonance. The measure of a CEST effect obtained through such an asymmetry analysis is regularly referred to as MTR_{asym} . This magnetization transfer ratio is always expressed as the relative signal change in bulk water signal. Nevertheless, in vivo z -spectra are inherently asymmetric because of conventional magnetization transfer (MT) effects. Additionally, CEST effects are masked by concomitant direct water saturation as a result of the bandwidth of the saturation pulses. Hence, it is necessary to use tailored CEST acquisition and saturation strategies to limit these confounding factors. Although multiple imaging strategies, such as fast spin echo (FSE), gradient spin echo (GRASE) and echo-planar imaging (EPI), etc., have been introduced for acquisition of the water signal in CEST experiments [30–34], a clear suggestion toward a particular imaging technique is not straightforward. Especially because the choice of the imaging technique for CEST depends on many additional factors compared to standard MR image contrasts, e.g., the reliability of CEST quantification, the tolerated scan time, desired SNR, SAR load of the saturation scheme, etc. Typical imaging times for gagCEST examinations of a full knee joint have durations around 9–12 min for resolutions on the order of $0.8 \times 0.8 \times 3 \text{ mm}^3$ at 7.0 T, while the resolutions at 3.0 T would be slightly lower to maintain similar scan times.

The employed saturation scheme should always be parameterized to provide highly selective saturation of off-resonant protons and at the same time enable a highly efficient reduction of the bulk water signal. Since the most intuitive approach for obtaining narrowband saturation, i.e., continuous-wave (CW) irradiation, is often not possible on clinical MRI scanners because of technical and SAR constraints, a considerable amount of CEST research is focused on optimizing pulsed saturation schemes [34–37]. Since multiple data sets are acquired for one z -spectrum, it is also mandatory to compensate for patient motion between acquisitions in order to correctly align the data sets with different saturation offsets for proper analysis of z -spectra. Lipid resonances have to be suppressed as they

can interfere with the CEST effects of interest. Correction of inhomogeneities of the static magnetic field, which can be severe in many body regions, has to be performed before analysis of z -spectra [38•]. In general, multi-channel RF coils, enabling acceleration of the acquisition via application of parallel imaging strategies, are absolutely necessary.

gagCEST

GAG molecules were shown to be equipped with labile $-NH$ ($\Delta\omega = 3.2$ ppm offset from the water resonance) and $-OH$ ($\Delta\omega \approx 0.8$ – 1.9 ppm) protons that can be used as CEST agents through selective saturation of their resonance signals [39]. The signal of the hydroxyl protons at $\Delta\omega = 1.0$ ppm [12, 17, 19, 21] or the average effect over the entire range of $-OH$ signals [16, 18•, 20•], i.e., from ≈ 0.8 – 1.9 ppm, is commonly used to quantify gagCEST effects in tissue. The latter method has the potential advantage of automatically partly compensating for insufficient selectivity of the saturation scheme, residual inhomogeneities of the static magnetic field and altered resonance signals depending on different magnetic environments. This potentially contributes to a higher signal yield and quantification confidence compared to single-point evaluation [20•]. The MTR_{asym} in gagCEST experiments can also be referred to as gagCEST signal intensity, but it is important to differentiate between the respective offsets that were used to derive the gagCEST signal intensities. The exchange rate of GAG protons was shown to range from 100 up to $1,000\text{ s}^{-1}$. The upper end of this range would be considered rapid chemical exchange, which is less favorable for CEST experiments as it violates the exchange condition ($k \leq \Delta\omega$), especially at 3.0 T, where $\Delta\omega = 0.8$ ppm corresponds to roughly 100 s^{-1} . Thus, although the actual exchange rate of the GAG hydroxyl protons strongly depends on the geometry of the GAG molecules and the viscosity of the chemical environment, it is obvious that higher magnetic field strengths provide better conditions to perform CEST experiments. This is not only due to the larger chemical shift dispersion, which avoids violation of the exchange condition, but is also due to longer T_1 relaxation times and the intrinsically higher proton signal. Longer T_1 is directly reflected by a longer lifetime of saturation after protons have been transferred from GAG molecules to bulk water. This increased lifetime, in turn, reduces the decay of the saturation signal during image readout and hence enables greater accuracy for gagCEST quantification compared to lower fields.

The rather rapid chemical exchange of GAG protons compared to other labile proton species also has advantages, which lie in the fact that saturation is transported rapidly to bulk water. This means that a fast buildup of

gagCEST effects can be obtained. This reduces the required saturation duration and potentially enables gagCEST to be sensitive to lower concentrations of solute molecules than CEST contrasts using slower exchanging solutes [29].

Changes in the T_2 relaxation time of tissue induce changes in the shape of resonance signals. If T_2 becomes longer, resonance lines will generally become narrower. Such an increase of T_2 has been repeatedly reported for defects in cartilage tissue [40, 41•, 42, 43] and can potentially impair the quantification of gagCEST signals in cartilage regions with prominently elevated T_2 values. The effect will lead to an artificial increase of gagCEST signals in regions with increased T_2 values and needs to be considered in gagCEST exams at 3.0 T. Otherwise, this effect may lead to misinterpretation of results and be especially problematic as damaged cartilage would usually exhibit low GAG contents. If the damage coincides with increased T_2 , falsely increased or normal gagCEST values could be measured. Due to the larger chemical shift dispersion at 7.0 T, a bias of gagCEST quantification due to T_2 -induced changes of the linewidth of bulk water can be assumed to be negligible.

Although the maximum gagCEST signal, which can be obtained at 3.0 T, is lower compared with 7.0 T, it can be shown in human knee joints that there is a linear correlation of the gagCEST signals measured at the two field strengths (Fig. 1). To obtain such a result, it is necessary to adapt the acquisition parameters to the field strength and make sure the optimum saturation efficiency can be obtained at both field strengths. At 3.0 T, gagCEST signals were reported to range between 8.5 and 9 % in the nucleus pulposus of healthy IVDs [16, 17], while the values reported for the

Fig. 1 Relationship between gagCEST signals (MTR_{asym}) measured in four different regions of knee cartilage in ten healthy volunteers at 3.0 and 7.0 T. The correlation coefficient indicated a strong linear correlation between both field strengths; $r = 0.82$ ($P < 0.001$)

healthy cartilage in the knee joint vary between -2 and 5% [21]. In contrast, values reported for the healthy knee cartilage at 7.0 T range between 4 and 15% [18•, 20•].

Clinical Application of gagCEST

Validation

Due to the fact that, for gagCEST, CEST effects are evaluated at the offset frequency of the exchangeable protons of GAG molecules, the derived contrast is intrinsically sensitive to the underlying content of GAG protons in the tissue. However, GAGs are saccharides, and their chemical structure is similar to other saccharides, which leads to the fact that molecules with similar structures will have protons with similar exchange properties. Thus, it is not possible to attribute the gagCEST signal with perfect accuracy exclusively to GAG. However, in connective tissues such as cartilage, the concentration of GAG hydroxyl protons is on the order of 200–300 mM [4],

which is considerably higher than the concentration of protons with similar resonance footprints from other saccharides. Thus, it is reasonable to assume that gagCEST effects and changes observed in such connective tissue can predominantly be attributed to the GAG content in the tissue. Validation of this assumption is possible by comparing gagCEST results to other GAG-sensitive imaging techniques, such as sodium MRI [18•, 20•] (Fig. 2) and dGEMRIC, or by comparison to results from histobiochemical analysis of tissue. Previous studies have shown that gagCEST signals measured in knee cartilage at 7.0 T exhibit linear correlations with sodium MRI [18•, 20•], which is well in line with theory as both should have linear relationships with GAG content in cartilage.

Preliminary data obtained through gagCEST examination of cartilage from human ankle cadavers at 3.0 T and subsequent histobiochemical quantification of absolute GAG content also clearly demonstrate a linear relationship between the gagCEST signal and the underlying GAG content in cartilage (Fig. 3). These results were confirmed for IVD samples in a similar way at 7.0 T [19].

Fig. 2 Sagittal knee images of a patient (age: 55 years) 6.7 years after autologous osteochondral transplantation in the medial femoral condyle. From *left to right*, PDw image, gagCEST overlay and sodium overlay. Note the good agreement between the signal patterns

between the sodium image and gagCEST, confirming that both techniques are similarly sensitive to the GAG content of cartilage. Also note the higher granularity of the gagCEST overlay, which can help to detect smaller lesions

Fig. 3 **a** Axial ankle PDw image with fatsat overlaid with the gagCEST signal measured at 3.0 T. This shows that ankle cartilage can well be examined with gagCEST using appropriate clinical equipment. **b** Correlation between gagCEST signal intensities and biochemically determined GAG content. The strong linear correlation ($r^2 = 0.82$, $P < 0.001$) confirms that gagCEST is sensitive to the

cartilage GAG content over a wide range of signal values from -5 to 15% . Please note that ankle cartilage on average has higher GAG content than knee cartilage, which gives rise to higher gagCEST signals. **c** z-Spectrum and corresponding asymmetry curves from the tibia and talus in **(a)**

Fig. 4 **a** PDw image with fatsat and sodium overlay; **b** with CEST overlay. **c** Morphological PDw image with fatsat. **d** T1w image with T_2 map overlay. All images were taken from a healthy volunteer.

Morphological imaging, CEST and T_2 mapping were performed at 3 T, sodium MRI at 7 T. While the signal patterns of sodium and gagCEST consistently indicate a small area of degeneration in the center of the tibial plateau through signal loss, the morphological image and T_2 map do not exhibit changes. This emphasizes the finding that T_2 mapping and GAG-sensitive techniques provide different information, while the latter techniques can be especially useful for investigation of early stages of cartilage degeneration

A study that evaluates the correlation between dGEMRIC and gagCEST appears to be feasible, but might raise ethical concerns because of the required double dose of contrast agent for dGEMRIC and has not been published so far.

Comparison with T_2 mapping and $T_{1\rho}$

It has been reported that T_2 values correlate with the level of degeneration in cartilage and IVDs [16, 44, 45]. Similar observations were reported for $T_{1\rho}$ [13, 46, 47], although the underlying reasons for these contrast variations are assumed to be of different origin. While T_2 relaxation is thought to reflect changes in the water content and the geometric order of the collagen fiber architecture, $T_{1\rho}$ also seems to reflect the GAG content in cartilage as $T_{1\rho}$ values are correlated with the GAG concentration in vitro [11, 13, 48]. However, recent results indicate that $T_{1\rho}$ does not exclusively reflect the GAG content, but is also closely connected unspecifically to changes that alter T_2 relaxation times because of the strong correlation of results from both techniques in cartilage evaluations [44, 46].

A recent study showed no correlation between gagCEST at 7.0 T and T_2 mapping in cartilage degeneration, although gagCEST detected reduced GAG content and was

confirmed by sodium MRI [18]. This indicates a clear difference in the information content of gagCEST signals and T_2 measurements. Additionally, since high-grade cartilage defects exhibit T_2 changes while very early degeneration does not, gagCEST seems to be especially useful to detect early pathological changes in cartilage that coincide purely with a primary loss of cartilage GAG content (Fig. 4). At later stages of degeneration, when T_2 increases and morphological changes become more pronounced, gagCEST may become gradually biased by T_2 increases and hence be less useful. Compared with $T_{1\rho}$ mapping, gagCEST signals may reflect changes in GAG content more specifically because of the lower influence of other cross-relaxation effects, but thorough studies on investigating this question still need to be conducted.

Conclusion

Glycosaminoglycan-dependent CEST imaging provides a powerful tool for non-invasive assessment of changes in the GAG content of connective tissues. The technique can be applied well with clinical 3.0-T MRI systems, which is not possible for sodium MRI of cartilage, although

Fig. 5 Histogram of CEST signal intensities in ROIs from different cartilage areas in the knee of a healthy volunteer [trochlea, patella, central and posterior lateral femoral condyle (LFC)]. All regions exhibit normally distributed values. The distribution with the highest mean value was found in the central, weight-bearing area of the LFC

gagCEST also benefits from being applied at higher magnetic fields. Compared with T_2 and $T_1\rho$ mapping, it seems to reflect the cartilage GAG content more specifically, which is advantageous for detecting early stages of cartilage degeneration in osteoarthritis, where isolated GAG loss occurs without further structural changes [5, 6]. It also does not require complicated imaging protocols or the administration of a contrast agent like in dGEMRIC [49].

However, the acquisition and evaluation of CEST data sets bear a significant level of complexity. A successful gagCEST experiment relies on the correct parameterization of the saturation scheme to ensure that CEST effects are generated efficiently. Additionally, concomitant effects, such as RF spillover and homogeneity of the magnetic field, need to be carefully taken into account or corrected to avoid errors in the interpretation of data. Additionally, scan times on the order of 9–12 min for a full knee, ankle or L-spine may be difficult to realize in a routine clinical setting, although it is similar to protocols for dGEMRIC or $T_1\rho$ mapping.

A further limitation of the clinical applicability may be the compromise that needs to be made between the resolution and reliability of CEST signal. This may lead to the fact that joints with thin cartilage areas, which are difficult to image with MRI, such as the hip and shoulder, may in the short term be exempt from gagCEST imaging at 3.0 T.

Even though retrospective correction of patient motion and inhomogeneities of the static magnetic field are possible, it is necessary to limit the maximum extent of these factors by adequate measures, which should include careful extremity fixation and manual shimming before the

measurement. Moreover, large inhomogeneities of the static magnetic field induced by, e.g., metallic particles can lead to severe distortions of CEST effects, which cannot be compensated retrospectively.

Since maximum CEST signals are typically only a few percent of the bulk water signal and the nature of a z -spectrum is very complex, automation of signal evaluation and adequate correction is not trivial. Additionally, the magnitude of gagCEST signals varies not only with the field strength of the MR system, but also with the GAG content in different tissues, different joints and even within multiple regions in one joint (Fig. 5). These aspects have to be taken into account to be able to work with quantitative numbers in CEST imaging. CEST values in cartilage can also become negative. This does not appear intuitive, but comes from nuclear Overhauser effects that are competing effects for CEST and occur asymmetrically on the opposite side of gagCEST effects in the z -spectrum [12]. Once the relative GAG content in cartilage is below a certain threshold, which is tissue-specific, these effects dominate the z -spectrum and induce negative gagCEST signals. This can be perfectly physiological, for example, in the posterior non-loading area of the femoral condyle at 3.0 T, but also needs to be understood to avoid misinterpretation of results.

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Compliance with Ethics Guidelines

Conflict of Interest Benjamin Schmitt, Martin Brix and Stephan Domayer declare that they have no conflicts of interest.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by the authors.

References

Papers of particular interest, published recently, have been highlighted as:

- Of importance,
- Of major importance

1. Ayad S. The extracellular matrix factsbook, factsbook series. 2nd ed. San Diego: Academic Press; 1998. p. 301.
2. Wong M, Hunziker EB. Articular cartilage biology and biomechanics. In: Insall JN, Scott WN, editors. Surgery of the knee. Philadelphia: Churchill Livingstone; 2000. p. 317–25.
3. Roughley PJ, Lee ER. Cartilage proteoglycans—structure and potential functions. Microsc Res Tech. 1994;28(5):385–97.
4. Roughley PJ. The structure and function of cartilage proteoglycans. Eur Cell Mater. 2006;12:92–101.
5. • De Ceuninck F, Sabatini M, Pastoureaux P. Recent progress toward biomarker identification in osteoarthritis. Drug Discov Today. 2011;16(9–10):443–9. *Major overview article of OA research especially focusing on actual and potential biomarkers.*
6. Brown MP, et al. Exercise and injury increase chondroitin sulfate chain length and decrease hyaluronan chain length in synovial fluid. Osteoarthritis Cartilage. 2007;15(11):1318–25.
7. Borthakur A, et al. Sodium and T-1 rho MRI for molecular and diagnostic imaging of articular cartilage. NMR Biomed. 2006;19(7):781–821.
8. Trattnig S, et al. ²³Na MR Imaging at 7 T after knee matrix-associated autologous chondrocyte transplantation: preliminary results. Radiology. 2010;257:175–84.
9. Eckstein F, Burstein D, Link TM. Quantitative MRI of cartilage and bone: degenerative changes in osteoarthritis. NMR Biomed. 2006;19(7):822–54.
10. Keenan KE, et al. Prediction of glycosaminoglycan content in human cartilage by age, T1rho and T2 MRI. Osteoarthritis Cartilage. 2011;19(2):171–9.
11. Li X, et al. In vivo T-1 rho and T-2 mapping of articular cartilage in osteoarthritis of the knee using 3 T MRI. Osteoarthritis Cartilage. 2007;15(7):789–97.
12. Ling W, et al. Assessment of glycosaminoglycan concentration in vivo by chemical exchange-dependent saturation transfer (gagCEST). Proc Natl Acad Sci USA. 2008;105(7):2266–70.
13. Stahl R, et al. T1rho, T-2 and focal knee cartilage abnormalities in physically active and sedentary healthy subjects versus early OA patients—a 3.0-Tesla MRI study. Eur Radiol. 2009;19(1):132–43.
14. Staroswiecki E, et al. In vivo sodium imaging of human patellar cartilage with a 3D cones sequence at 3 and 7 T. J Magn Reson Imaging. 2010;32(2):446–51.
15. Bashir A, et al. Glycosaminoglycan in articular cartilage: in vivo assessment with delayed Gd(DTPA)(2-)-enhanced MR imaging. Radiology. 1997;205(2):551–8.
16. Haneder S, et al. Glycosaminoglycan content in intervertebral discs using chemical exchange saturation transfer at 3T MRI: preliminary results in patients with low back pain. Eur Radiol. 2013;23(3):861–8.
17. Kim M, et al. Assessment of glycosaminoglycan distribution in human lumbar intervertebral discs using chemical exchange saturation transfer at 3 T: feasibility and initial experience. NMR Biomed. 2011;24(9):1137–44.
18. •• Krusche-Mandl I, et al. Long-term results 8 years after autologous osteochondral transplantation: 7 T gagCEST and sodium magnetic resonance imaging with morphological and clinical correlation. Osteoarthritis Cartilage. 2012;20(5):357–63. *First publication correlating the clinical outcome of patients after AOT with GAG-sensitive MR techniques.*
19. Saar G, et al. Assessment of glycosaminoglycan concentration changes in the intervertebral disc via chemical exchange saturation transfer. NMR Biomed. 2012;25(2):255–61.
20. •• Schmitt B, et al. Cartilage quality assessment by using glycosaminoglycan chemical exchange saturation transfer and ²³Na MR imaging at 7 T. Radiology. 2011;260(1):257–64. *First clinical test of gagCEST imaging on patients after cartilage repair surgery at 7 T.*
21. Singh A, et al. Chemical exchange saturation transfer magnetic resonance imaging of human knee cartilage at 3 and 7 T. Magn Reson Med. 2011;68(2):588–94.
22. Varma G, et al. Optimization of pulsed-gagCEST at 3.0 T. In: 19th Annual Meeting of the ISMRM. ISMRM, Montreal; 2011.
23. Wei W, et al. Examining the accuracy of dual echo B0 map for field inhomogeneity correction with the application of gagCEST in articular cartilage at 3 T. In: 19th Annual Meeting of the ISMRM. ISMRM, Montreal; 2011.
24. Zhang B, et al. Center-corrected gagCEST assessment of intervertebral disc degeneration. In: 19th Annual Meeting of the ISMRM. ISMRM, Montreal; 2011.
25. Guivel-Scharen V, et al. Detection of proton chemical exchange between metabolites and water in biological tissues. J Magn Reson. 1998;133(1):36–45.
26. Ward KM, Aletras AH, Balaban RS. A new class of contrast agents for MRI based on proton chemical exchange dependent saturation transfer (CEST). J Magn Reson. 2000;143(1):79–87.
27. Ward KM, Balaban RS. Determination of pH using water protons and chemical exchange dependent saturation transfer (CEST). Magn Reson Med. 2000;44(5):799–802.
28. Zhou JY, van Zijl PCM. Chemical exchange saturation transfer imaging and spectroscopy. Prog Nucl Magn Reson Spectrosc. 2006;48(2–3):109–36.
29. •• van Zijl PC, Yadav NN. Chemical exchange saturation transfer (CEST): what is in a name and what isn't? Magn Reson Med. 2011;65(4):927–48. *Major overview article on the principles of CEST with good focus on potential clinical pitfalls.*
30. Dixon WT, et al. A multislice gradient echo pulse sequence for CEST imaging. Magn Reson Med. 2010;63(1):253–6.
31. Park SH, Duong TQ. Alternate ascending/descending directional navigation approach for imaging magnetization transfer asymmetry. Magn Reson Med. 2011;65(6):1702–10.
32. Shah T, et al. CEST-FISP: a novel technique for rapid chemical exchange saturation transfer MRI at 7 T. Magn Reson Med. 2011;65(2):432–7.
33. Sun PZ, et al. Fast multislice pH-weighted chemical exchange saturation transfer (CEST) MRI with unevenly segmented RF irradiation. Magn Reson Med. 2011;65(2):588–94.
34. Zhu H, et al. Fast 3D chemical exchange saturation transfer (CEST) imaging of the human brain. Magn Reson Med. 2010;64(3):638–44.
35. Desmond KL, Stanisz GJ. Understanding quantitative pulsed CEST in the presence of MT. Magn Reson Med. 2011;67(4):979–90.
36. Schmitt B, et al. Optimization of pulse train presaturation for CEST imaging in clinical scanners. Magn Reson Med. 2011;65(6):1620–9.
37. Sun PZ, et al. Simulation and optimization of pulsed radio frequency irradiation scheme for chemical exchange saturation transfer (CEST) MRI—demonstration of pH-weighted pulsed-

- amide proton CEST MRI in an animal model of acute cerebral ischemia. *Magn Reson Med*. 2011;66(4):1042–8.
38. • Kim M, et al. Water saturation shift referencing (WASSR) for chemical exchange saturation transfer (CEST) experiments. *Magn Reson Med*. 2009;61(6):1441–50. *Important method for correction of the static magnetic field inhomogeneities*.
  39. Ling W, et al. Characterization of bovine patellar cartilage by NMR. *NMR Biomed*. 2008;21(3):289–95.
  40. Baum T, et al. Association of magnetic resonance imaging-based knee cartilage T2 measurements and focal knee lesions with knee pain: data from the Osteoarthritis Initiative. *Arthritis Care Res (Hoboken)*. 2012;64(2):248–55.
  41. • Baum T, et al. Changes in knee cartilage T2 values over 24 months in subjects with and without risk factors for knee osteoarthritis and their association with focal knee lesions at baseline: Data from the osteoarthritis initiative. *J Magn Reson Imaging*. 2012;35(2):370–8. *Analysis of risk factors in OA with biochemical imaging techniques in a longitudinal follow-up setting over 24 months*.
  42. Welsch GH, et al. Quantitative T2 mapping during follow-up after matrix-associated autologous chondrocyte transplantation (MACT): full-thickness and zonal evaluation to visualize the maturation of cartilage repair tissue. *J Orthop Res*. 2009;27(7):957–63.
  43. Welsch GH, et al. T2 and T2* mapping in patients after matrix-associated autologous chondrocyte transplantation: initial results on clinical use with 3.0-Tesla MRI. *Eur Radiol*. 2010;20(6):1515–23.
  44. Mlynarik V, et al. The role of relaxation times in monitoring proteoglycan depletion in articular cartilage. *J Magn Reson Imaging*. 1999;10(4):497–502.
  45. Mosher TJ, Dardzinski BJ. Cartilage MRI T2 relaxation time mapping: overview and applications. *Semin Musculoskelet Radiol*. 2004;8(4):355–68.
  46. Nishioka H, et al. T(1rho) and T(2) mapping reveal the in vivo extracellular matrix of articular cartilage. *J Magn Reson Imaging*. 2012;35(1):147–55.
  47. Zarins ZA, et al. Cartilage and meniscus assessment using T1rho and T2 measurements in healthy subjects and patients with osteoarthritis. *Osteoarthritis Cartilage*. 2010;18(11):1408–16.
  48. • Stikov N, et al. Cross-relaxation imaging of human articular cartilage. *Magn Reson Med*. 2011;66(3):725–34. *Evaluation and comparison of different cross-relaxation measurement techniques and their application in cartilage*.
  49. Burstein D, et al. Protocol issues for delayed Gd(DTPA)(2)-enhanced MRI (dGEMRIC) for clinical evaluation of articular cartilage. *Magn Reson Med*. 2001;45(1):36–41.