Michael T. McMahon
Kennedy Krieger Institute

Syllabus for Thursday educational course (Preclinical & Clinical Applications of CEST)

Chemical Exchange Saturation Transfer (CEST) is an emerging MRI contrast mechanism with unique features that have sparked the interest of numerous researchers worldwide. Since the first reports of this contrast by and the Balaban group(1-3), the past decade has resulted in the development of numerous CEST contrast agents, many advances in the imaging sequences used to detect this contrast, and demonstrations of multiple pre-clinical and clinical applications which should benefit from CEST imaging. In this talk I will discuss applications of this technology which appear particularly promising, and group these according to whether the contrast is generated by a compound which is expressed by the body (endogenous CEST agent) or generated after a compound/particle is administered (exogenous CEST agent).

Endogenous CEST contrast agent Applications

The first *in vivo* images produced using this technology relied on the presence of endogenous biomolecules with suitable exchangeable protons to allow the generation of CEST contrast. The Balaban group showed that fluids rich in urea could be highlighted using saturation imaging(4). Certain peptides possess amide protons in peptides suitable for CEST imaging(5), and one observation that sparked a great deal of interest was that CEST contrast could be applied to the detection of strokes(6,7) and brain tumors(8) due to perturbation of exchange rates/concentration of endogenous amide protons. Very recently, it has been demonstrated that glycosaminoglycans can be detected using CEST contrast (GAG-CEST), allowing the monitoring of their concentration in cartilage(9). In addition, reporter genes have been developed to allow the use of CEST contrast to detect cells transfected with this gene. One of the first successful genes was termed the Lysine Rich reporter gene(10) due to the large amounts of lysine in the amino acid sequence of this artificial gene. Additional peptide sequences were designed which possess many copies of protons with suitable exchange rates(11), and recently 9L cells xenografted in the brains of mice have been successfully detected in vivo using a second CEST reporter gene based on human protamine.

CEST imaging technology has evolved quite a bit since these early studies, with the application of saturation transfer imaging to patients requiring numerous refinements which provide corrections for B_0 field inhomogeneities(12-15) and speed acquisition to allow the collection of full 3D CEST images. After collecting data on numerous patients, it is clear that CEST imaging can provide additional information for high-grade gliomas(16), and recent studies indicate that this contrast might allow better monitoring of brain tumors after radiation treatment than diffusion weighted images or gadolinium(17).

Exogenous CEST contrast agent Applications

Many compounds have been developed which possess protons suitable for CEST imaging. Both DIAmagnetic and PARAmagnetic CEST agents have been developed (DIACEST, PARACEST). The initial agents were designed to maximize the detectability

of the compounds(18-21), and more recent efforts have been geared towards tailoring the exchange properties with an application in mind. As an exogenous contrast agent, the sensitivity of this contrast is expected to be particularly useful for monitoring pH(22-24), concentrations of metabolites(25), peptides(26), ions(27), in addition to monitoring temperature(28). In addition, the specificity of the contrast to the saturation frequency provides the unique opportunity to discriminate between multiple CEST agents after administration, allowing similar Multi-color imaging studies to those performed using fluorescent compounds(11,29,30).

More recently it has been shown that CEST agents can be integrated into particles allowing the detection of low concentrations(31,32). This has sparked interest in using CEST particles for three different types of applications. Multiple groups are interested in using CEST contrast to monitor the liposomal delivery and release of drugs (33-35). Targeted imaging studies have been performed to detect the presence of antibodies in clots using perfluorocarbon emulsions. In addition, immunoprotective alginate cell carriers have been developed, with the hope of using CEST contrast to locate these carriers and monitor the encapsulated cell viabilityafter transplantation(36,37).

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