angioCEST: using TmDOTMA liposomes and chemical exchange saturation transfer for MR angiography

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Target audience: This information will benefit Scientists and Physicians interested in the chemical engineering, development, and preclinical *in vivo* imaging of novel, highly sensitive MRI contrast agents. Specifically, those used for angiography, molecular targeting, functional imaging, and chemical shift imaging.

Purpose: Liposomal encapsulation of paramagnetic chemical exchange saturation transfer (paraCEST) agents creates enhanced contrast in MRI by shifting the resonance frequency of the water molecule protons inside the liposome away from the bulk water frequency by 2 ppm to 10 ppm (lipoCEST) (1). Radio-frequency saturation at this shifted proton resonance, along with water molecule exchange across the liposome membrane, causes enhanced negative contrast (image darkening) in areas of liposome uptake. LipoCEST greatly increases the relative sensitivity of paraCEST contrast agents by using the approximately 10^6 to 10^9 shifted protons contained within the liposome for CEST. In this research, we investigate the idea of utilizing the high CEST sensitivity and the long blood circulation half-life (~18 hours) (2,3) of polyethylene glycol (PEG) coated "stealth" liposomes encapsulating TmDOTMA to perform CEST-based MRI angiography for vascular space imaging and quantitative blood volume measurements (angioCEST).

Methods: A 67 mM aqueous solution of TmDOTMA was encapsulated inside of 135 nm diameter liposomes. The total intra-liposome water was 27% of the stock solution volume, giving an effective Tm^{3+} concentration of 18 mM. *In vitro* z-spectrum of the stock solution were measured at 37 °C on an Agilent 9.4 T small animal MRI system using a 38 mm diameter birdcage coil and the gradient-echo pulse sequence with centric k-space ordering. A 5 second long saturation pulse was used with varied power (0.3, 0.6, 1.3, 2.5, 5, and 10 μT) in order to find the optimal power and saturation frequency settings. These ideal settings were then used to image the abdominal vasculature of healthy female Black-6 mice that were tail vein injected with 500 μL of the 18 mM TmDOTMA liposome solution (i.e., a 0.4 mmol[Tm^{3+}]/kg dose).

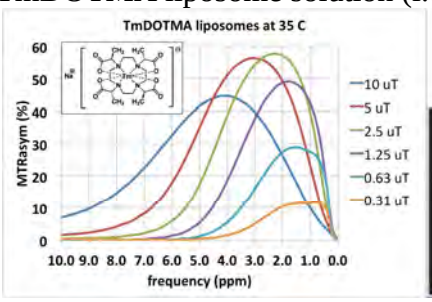

Fig. 1

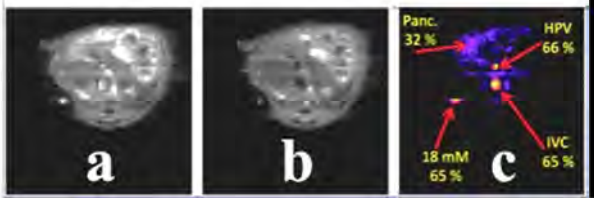

Fig. 2

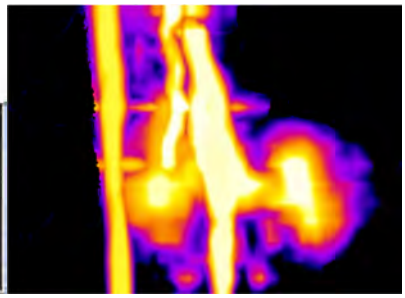

Fig. 3

Results: **Fig 1:** *In vitro* z-spectrum of the stock 18 mM TmDOTMA liposomes showing a large MTR_{asym} of 58% at 2.2 ppm using a 5 second, 2.5 μT saturation pulse (green line). **Fig 2:** Gradient-echo mouse abdominal axial CEST images with saturation at **(a)** -2.2 ppm (Off), and **(b)** +2.2 ppm (On); **(c)** Off-On CEST image showing an average MTR_{asym} of 66% in the inferior vena cava (IVC) and hepatic portal vein (HPV). **Fig 3:** A coronal 3D angioCEST maximum intensity projection of the mouse abdominal vasculature made using stacked gradient-echo axial slices where the kidneys, IVC, HPV, and capillary containing the stock TmDOTMA liposomes can be clearly seen. These data were taken approximately one hour after injection.

Conclusion: These preliminary data show that liposomal TmDOTMA angioCEST can image the vascular space with zero signal from background tissue using low-power saturation (2.5 μT). This technique could be used to evaluate therapeutic efficacy of cancer agents by measuring tumor blood volume before and after treatment. Further work includes incorporating B_o corrections using the WASSR method (4), speeding up acquisition using 3D gradient-echo and shorter saturation pulses, and improving chemical shift and sensitivity by increasing the Tm^{3+} concentration inside the liposome through chelate polymerization techniques.

References:

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