A Single PARACEST MRI Contrast Agent for Accurate in vivo pH Measurements

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Introduction:

pH is a physiological indicator of tumor lesions and other pathological abnormalities in soft tissues. MRI has shown tremendous potential for noninvasive *in vivo* pH measurements, though formidable challenges exist regarding sensitivity, accuracy and/or the need of a second pH-unresponsive agent as control.[1] Endogenous Chemical Exchange Saturation Transfer (CEST)[2,3] and PARAmagnetic CEST (PARACEST)[4,5] contrast agents have been successfully applied to measure pH, although a second pH-unresponsive agent is still required for accurate measurements. A single contrast agent with a pH-responsive and pH-unresponsive PARACEST effect has been developed to achieve pH measurements with a single agent.[6] Herein we propose a single contrast agent with two PARACEST effects that have opposite dependencies on physiological pH, which possesses the ideal characteristics for MRI-based pH measurements to assess tumors and other pathologies.

Methods:

Yb(III)-1,4,7,10-tetraazacyclododecane-1,4,7,10-tetraacetic acid o-Aminoanilide (Yb-DO3A-oAA) was synthesized, and the product was confirmed by MS and NMR spectroscopy. CEST spectra were obtained by a modified presaturation pulse sequence with a 600 MHz Varian NMR scanner.[3] The range and accuracy of pH measurements were determined using a series of pH phantoms containing 20 mM Yb-DO3A-oAA in PBS with pH ranging from 6.0 to 8.0 at 37°C. Investigations were conducted to confirm that pH measurements were independent of concentration (5 mM to 80 mM), T_1 relaxation (0.5 s to 3 s) and temperature (8°C to 37°C). PARACEST MRI was performed with a 9.4T Bruker Biospec animal MRI scanner equipped with a 35 mm birdcage RF coil. The images were acquired after 50 uL of 60 mM Yb(III)_DO3A-oAA solution was directly injected into the tumor of a 6 week-old female athymic NCR nu/nu mouse with a MCF-7 subcutaneous flank tumor. Parametric maps of tissue pH were obtained by processing PARACEST images with the PARACEST-pH calibration achieved with pH phantoms.

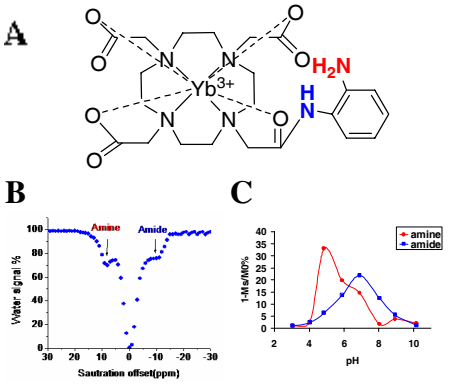

Figure 1. A) The structure of YBDO3A-oAA , B) the CEST spectrum, and C) dependencies of the CEST effects of the amine and amide relative to pH.

Results and Discussion:

The new PARACEST agent DO3A-oAA was synthesized and characterized (Figure 1A). Two PARACEST effects of Yb(III) DO3A-oAA were observed at +9 ppm and -11 ppm, and were assigned to the protons of the amine and amide groups respectively (Figure 1B). The PARACEST effects from the amide and amine showed different pH dependencies (Figure 1C). A ratiometric approach that uses (M_0/M_s-1) ratio of amide and amine PARACEST effects was used to measure pH in phantoms with pH values ranging from 6.0 to 8.0. This pH measurement was verified to be independent of concentration, T_1 relaxation, and temperature. The minimum concentration that can be detected during practical MRI studies (defined as the minimum concentration that can cause a 1% change in image contrast) was determined to be 3.5 mM, which is sufficient for most extracellular pH applications. The feasibility of in vivo applications was demonstrated with a mouse tumor model (Figure 2). The pH image of the tumor area accords with previous literature reports.

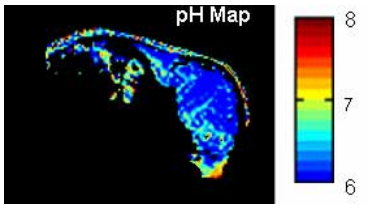

Figure 2. pH map of a mouse tumor measured by ratiometric approach as described in text part.

Conclusions:

We have developed a new PARACEST MRI contrast agent Yb(III) DO3A-oAA and demonstrated its capabilities of measuring extracellular pH within *in vivo* animal models without the need for a second "control" agent to account for agent concentrations. Furthermore, the two pH-responsive PARACEST effects of this new agent provides a greater range for pH measurements and improved measurement accuracy relative to a previously-reported PARACEST contrast agent with a pH-responsive and pH-unresponsive PARACEST effect.[6]

References:

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