In Vivo Magnetic Resonance Imaging of Renal pH Using GdDOTA-4AmP

1Natarajan Raghunand, 1Christine Howison, 2A. Dean Sherry, 2Shanrong Zhang, 1Robert J Gillies
1University of Arizona HSC, Cancer Center Division, Tucson, AZ 85724-5024, USA
2University of Texas at Dallas, Richardson, TX 75083-0688, USA

Abstract. Pathologically altered renal physiology can manifest with perturbations in systemic and renal pH. The ability to image tissue pH would have considerable biomedical and clinical relevance, by enabling the non-invasive assessment of renal disease extent, progression, and response to therapy. The pH-sensitive contrast agent GdDOTA-4AmP and its pH-insensitive analog GdDOTP were administered 1 h apart to the same animal. T1-weighted MR images of the kidneys were collected dynamically for 60 min following each agent. The pharmacokinetics of GdDOTP were used as a surrogate for the pharmacokinetics of GdDOTA-4AmP, and renal pH images were calculated for control and acetazolamide-treated mice.

Introduction. The pH of bodily fluids affects the organism in many ways, especially through its effects on cellular and plasma proteins. Maintenance of acid-base homeostasis is therefore critical, and occurs at several levels. The most immediate and local response to an acid or alkali load is through chemical processes, including intracellular and extracellular buffers. However, buffering capacity is typically limited, and must be backed up by physiologic responses to the acid-base imbalance. These physiologic processes can be at the cellular level, such as feed-back changes in metabolism, and at the systemic level, involving adaptive changes to the excretion of volatile acids by the lungs and fixed acids by the kidneys [1]. Pathologically altered renal physiology can manifest with perturbations in systemic and renal pH. Hereditary defects and acquired deficiencies in renal tubular ion transport systems can result in systemic metabolic acidosis or alkalosis [2,3]. Novel gene therapies have shown promise for the treatment of some of these diseases, but there are still problems with heterogeneous gene delivery to and loss of corrective gene from the target tissue over time [4,5]. Methodologies to image the spatial distribution of renal pH would have considerable biomedical and clinical relevance in such cases, by enabling the non-invasive assessment of disease extent, progression, and response to therapy. Recently, a pH-sensitive gadolinium-based contrast agent, GdDOTA-4AmP, was introduced [6]. We demonstrate the use of this agent to measure tissue pH by contrast-enhanced MRI.

Methods. MRI was performed on female SCID mice on a Bruker Biospec 4.7 T system with 14 G/cm self-shielded gradients, using a 25 mm Helmholtz coil. Mice were anesthetized by inhaled isoflurane (1.5%, rest O2 at 1 L/min), and circulating warm water jackets were used to keep animals warm in the magnet. Body temperature was monitored during all MRI experiments using a rectal fluoroptic temperature probe (Luxtron Corporation, Santa Clara, CA, USA). Mice were cannulated via the tail vein prior to positioning in the magnet, and contrast agent (0.05 mmole/Kg) was administered immediately chased with 0.15 mL saline via this catheter at the appropriate time during each experiment. T1 maps were obtained in all cases prior to administration of contrast agent. The contrast-enhanced portion of each imaging experiment consisted of three consecutive phases: fat-suppressed images were acquired every 40 s for 1 hour following the injection of first GdDOTP, then GdDOTA-4AmP (or GdDTPA), and then GdDOTP again, with the following imaging parameters: in-plane spatial resolution = 273 um, slice-thickness = 1-1.3 mm, number of slices = 2, recycle time = 80 ms, echo time = 6 ms, number of averages = 3. The renal pharmacokinetics of the two injections of GdDOTP in a given animal were compared to each other, and also to the pharmacokinetics of GdDOTA-4AmP in some animals. Data were discarded for experiments in which the area-under-the-curve pharmacokinetics at 25 min post-injection, of the second injection of GdDOTP, were not within 10% of the first injection. The pharmacokinetics of GdDOTA-4AmP were assumed to be identical to those of GdDOTP in a given animal, given their similar molecular size and charge. pH was calculated for each pixel from these pharmacokinetics, the measured T1 maps, and a titration of the relaxivity of GdDOTA-4AmP that was obtained in vitro in phosphate-buffered saline at 37°C and 4.7 T. Some mice were treated with acetazolamide (30 mg/Kg/day orally for 7 days prior to imaging).

Results. The renal cortical pH in control mice was calculated to be 6.9-7.5, while medullary pH ranged from 6.6-7.0. Acetazolamide treatment increased medullary pH by about 0.7 pH units compared to control mice. The higher contrast-enhancement by GdDOTA-4AmP in the more acid regions tended to increase the precision of pH measurement with decrease in pH. Precision of pH measurement was also significantly lower in non-renal tissues compared to the kidney, due to the higher concentration of the agents in the kidney. All these factors result in a widely varying estimated precision of measurement of 0.2-0.6 pH units in control animals, with the higher precision being in the collecting ducts, and the least precision in the non-renal tissues. Acetazolamide treatment increases the pH associated with renal pixels, with a somewhat decreased precision.

Conclusions. We have demonstrated the feasibility of pH imaging in mice by contrast-enhanced MRI using a dual contrast agent method. The calculated renal pH values in acetazolamide-treated mice were in consonance with the expected activity of the drug on renal tubular ion transport in mice, and also with the measured urine pH.

Acknowledgements. This work was funded by grants from the American Cancer Society (IRG7400124-451224 to NR) & the National Institutes of Health (R21-CA84697 to ADS; ROI-CA77575 & R24-CA83148 to RJJ).