

# THE HANES-WOOLF LINEAR QUEST METHOD PROVIDES THE MOST ACCURATE DETERMINATION OF FAST CHEMICAL EXCHANGE RATES FOR CEST MRI CONTRAST AGENTS

Edward T Randtke<sup>1</sup>, Liu Qi Chen<sup>1</sup>, Rene Corrales<sup>1</sup>, and Marty Pagel<sup>2</sup>

<sup>1</sup>Chemistry and Biochemistry, University of Arizona, Tucson, AZ, United States, <sup>2</sup>Biomedical Engineering, University of Arizona, Tucson, AZ, United States

**INTRODUCTION:** The chemical exchange rate of CEST MRI contrast agents is often measured to validate CEST MRI signal properties, to select the MRI protocol for best detection, to evaluate new CEST MRI protocols, and to evaluate molecular biomarkers that are detected with responsive CEST agents. Fitting CEST spectra with the Bloch equations modified for chemical exchange [1], measuring CEST as a non-linear function of saturation time (QUEST) or saturation power (QUESP) [2], the linear omega-plot method [3], and measuring CEST as a linear function of concentration [4] have been used to determine chemical exchange rates. However, most methods fail to determine an accurate exchange rate when the exchange rate is fast (> 20% of the chemical shift difference), primarily due to incomplete saturation that is exacerbated by fast exchange. We have investigated the accuracies of each method using theoretical simulations and experimental measurements of iopromide, a DIACEST agent with slow and fast exchange rates. We have also developed new analysis methods and compared these methods with existing analysis methods.

**METHODS:** Theoretical analyses were conducted by simulating CEST spectra using Bloch equations, for a DIACEST agent that had chemical shift of 5.6 ppm and chemical exchange rates ranging from 0 to 1200 rad/sec, T1 relaxation times ranging from 0.1 to 2.0 sec, saturation times ranging from 0 to 6 sec, saturation powers ranging from 0 to 10  $\mu$ T, and concentrations ranging from 0 to 3 mM. Lorentzian lines were fit to the CEST spectra [5], and the CEST amplitudes were used to determine exchange rates with QUEST, QUESP, omega-plot, and concentration-dependent methods, and two new analysis methods. These determinations were compared with the exchange rates used to calculate the simulated CEST spectra to evaluate the accuracy of each method.

Experimental CEST spectra were acquired using iopromide, a DIACEST agent with a fast-exchanging proton that resonates at 5.6 ppm and a slow-exchanging proton that resonates at 4.2 ppm. The experimental spectra were fit with the Bloch equations using a matrix-based method [6] to determine chemical exchange rates. Lorentzian lines were fit to the experimental CEST spectra [5], and the CEST amplitudes were used to determine exchange rates with QUEST, QUESP, omega-plot, and concentration-dependent methods, and two new analysis methods. These determinations were compared with the exchange rates from Bloch fittings to evaluate the accuracy of each method.

**RESULTS:** The determination of slow and fast exchange rates using Bloch fitting produced the most accurate results, especially when multiple CEST spectra with a range of saturation times and powers were simultaneously fit. The QUEST and omega-plot methods produced accurate results when measuring slow exchange rates and with highest saturation powers [2] and shortest T1 relaxation times (and the T1 time does not have to be pre-determined to achieve good accuracy), while QUEST and concentration-based methods overestimated slow exchange rates. Except for Bloch fitting, all methods failed to accurately determine a fast exchange rate.

The omega-plot method is a linear version of QUESP that is analogous to the linear Lineweaver-Burk plot that is used to analyze non-linear Michaelis-Menten kinetics. The linear Eadie-Hoffstee and Hanes-Woolf plots can also analyze Michaelis-Menten kinetics. Therefore, we developed linear versions of QUESP, termed the Eadie-Hoffstee linear QUESP and Hanes-Woolf linear QUESP methods. Both of these new linear QUESP methods accurately determined fast exchange rates, because these methods cancel the systematic error caused by incomplete saturation. The Hanes-Woolf method has a better dynamic range than the Eadie-Hoffstee method, so that Hanes-Woolf is preferred for determining fast exchange rates.

**DISCUSSION:** The use of a new linear analysis method that can accurately determine fast exchange rates, the Hanes-Woolf linear QUESP method, improves the analysis of CEST MRI contrast agents. The omega-plot method (a.k.a., the Lineweaver-Burk linear QUESP method) is adequate for accurately determining slow exchange rates. Both of these methods are fast and relatively easy to use. Block fitting is the most accurate method for determining slow and fast exchange rates, especially when multiple CEST spectra are simultaneously fit, although this method is slower and more difficult to use relative to the linear fitting methods.

**REFERENCES:** 1. Woessner, et al., MRM, 2005. 2. McMahon, et al., Magn Reson Med, 2006. 3. Dixon, et al., Magn Reson Med, 2010. 4. Ali, et al., Acc Chem Res, 2009. 5. Sheth, et al., CMMI, 2012. 6. Murase and Tanki, MRI, 2011.

Chemical Exchange Rate Measurements (rad/sec) with conf. intervals		
Method	4.2 ppm	5.6 ppm
Bloch Fitting	55.9 (53.7 - 58.3)	1117.1 (1100.5 - 1134.0)
QUEST (4 $\mu$ T)	28.9 (17.5 - 47.8)	87.8 (57.6 - 133.9)
QUEST (10 $\mu$ T)	---	694.7 (453.8 - 1063.7)
QUESP	118.0 (65.2 - 213.6)	2826.3 (2059.7 - 3878.2)
Omega Plots (linear QUESP)		
LB-QUESP	101.2 (80.2 - 111.3)	922.3 (648.8 - >10000)
EH-QUESP	300.0 (0 - 416.1)	1225.2 (1224.6 - 1226.0)
HW-QUESP	-118.3 (-41.7 - -162.0)	1160.5 (1040.3 - 1269.3)
Concentration Plots		
LB-Conc	595.4 (537.2 - 653.5)	308.3 (210.1 - 406.4)
EH-Conc	655.6 (516.3 - 794.8)	198.5 (0 - 841.6)
HW-Conc	607.3 (494.4 - 720.2)	327.1 (162.2 - 492.0)