

Simultaneously Measuring Glycogen and Lipid Levels Using Localized CEST Spectroscopy at 3T

Stephen J Bawden¹, Olivier Mougin¹, Karl Hunter², Luca Marciani³, and Penny Gowland¹

¹SPMMRC, University of Nottingham, Nottingham, United Kingdom, ²Unilever Discover, Bedford, United Kingdom, ³NDDC Biomedical Research Unit, University of Nottingham, Nottingham, United Kingdom

Introduction: Following meal consumption carbohydrates are transported to the liver or muscles, and stored as glycogen or lipids. Understanding the basic mechanism of glycogen and lipid turnover is therefore of much interest since glycogen provides energy between meals, whilst lipids are stored as a long term energy supply [1]. ¹H MRS is widely used for non-invasively assessing lipid levels *in vivo* [2]. Likewise ¹³C MRS provides the only validated and widely used method of non-invasively monitoring glycogen levels [3,4]. However, labelled ¹³C MRS is expensive and natural abundance ¹³C MRS provides very low sensitivity. Long fat carbon chains in spectra also make analysis difficult. GlycoCEST provides an alternative new method of measuring glycogen using the chemical exchange of saturation magnetization from the glycogen hydroxyl group to water [5]. The CEST measurement involves suppression of the water signal, which is often used to detect lipids in ¹H MRS [2] and opens an interesting possibility for simultaneous measurements.

Aim: To develop a method of simultaneously monitoring lipid and glycogen levels using localised GlycoCEST ¹H MRS for improved sensitivity.

Method: All scans were performed on a Philips Achieva 3T and XL Torso Coil. CEST sequence used ten 100ms block pulses with 0.5ms spacing for gradient dephasing, followed by PRESS localization. Three phantoms were made with coconut milk to simulate liver fat profiles and glucose (100mM+24g fat and 200mM+24g fat) and also glycogen (200mM). Phantoms were imaged together and 30mm³ VOIs for PRESS-CEST placed over each phantom to acquire ¹H spectra for lipid analysis (BW=2000Hz) and Z Spectra for CEST analysis ($\Delta\omega = -1000$ to +1000 Hz in 20Hz steps, $B_1 = 2\mu\text{T}$). Individual spectra were phase corrected and water peak height calculated (S_0 is unsaturated peak height). For fat analysis, spectra with high water saturation (<2%,

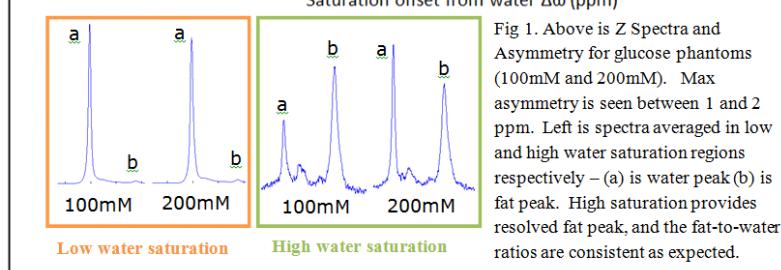


Fig 1. Above is Z Spectra and Asymmetry for glucose phantoms (100mM and 200mM). Max asymmetry is seen between 1 and 2 ppm. Left is spectra averaged in low and high water saturation regions respectively – (a) is water peak (b) is fat peak. High saturation provides resolved fat peak, and the fat-to-water ratios are consistent as expected.

high water saturation region on fig 1) and low water saturation (>98%, low water saturation region on fig 1) were averaged separately and Lorenzian curves fitted to determine the area ratios of fat (peak **b** in high water saturation spectrum, fig 1) to water (peak **a** in low water saturation spectrum, fig 1). Intra-hepatocellular lipid fraction (%IHCL) was quantified using equation defined previously [2] and CEST asymmetry was quantified using $MTR_{asym}(\Delta\omega) = S(-\Delta\omega)/S_0 - S(\Delta\omega)/S_0$. An *in vivo* Z spectrum was also acquired (post breakfast) during 4 breath-holds ($\omega_0=0$ and $\pm\Delta\omega$ Hz for each breath hold, where $\Delta\omega=50, 100, 150$ and 1000 , $B_1=1\mu\text{T}$, NSA=3, VOI=65mm³). Further *in vivo* data was acquired in another subject (post lunch) to test the effect of varying pulse power ($\Delta\omega=-1, 0$ and 1ppm, $B_1=0.25$ to $4\mu\text{T}$, VOI=50mm³), with each B_1 acquired during a single breath hold.

Results: The PRESS-CEST spectra showed high SNR, and averaging the water saturated spectra provided highly resolved lipid peaks (peak **b** in high water saturation spectrum, fig 1, and fig 3b at 0ppm). Peak MTR_{asym} was 11% for 100mM and 17% for 200mM glucose (fig 1) and the fat-to-water ratios were 0.028 and 0.034 (equivalent %IHCL ~3). 200 mM glycogen phantom showed peak MTR_{asym} of 6% at $\Delta\omega=1.1$ ppm (not shown). The *in vivo* Z spectra (fig 2) showed a peak MTR_{asym} of 5.3% with a fat-to-water ratio of 0.036 (%IHCL ~3.5). *In vivo* data at varying saturation powers showed decreasing water peak height with increasing pulse power for both off resonance saturations (fig 3c). The difference between the curves in fig 3c represents the glycogen in the region of interest, with a peak MTR_{asym} at 1 μT (fig 3b) of 11%. Glycogen signal was higher post lunch compared post breakfast (for $B_1=1\mu\text{T}$) as might be expected.

Conclusions: PRESS-CEST offers a promising new technique of simultaneously measuring glycogen and lipid levels *in vivo* that would reduce the cost of metabolic studies and make it easier to investigate multiple metabolic pathways in the postprandial and fasted state.

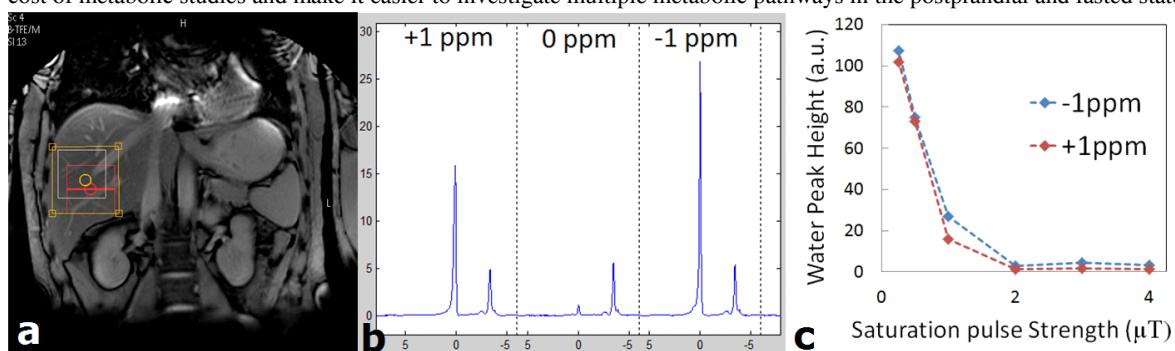


Fig 3. VOI for PRESS-CEST (a); Spectra showing saturation at -1, 0, 1ppm, $B_1 = 1\mu\text{T}$ (b); and plot of peak height at increasing saturation pulse power (c)

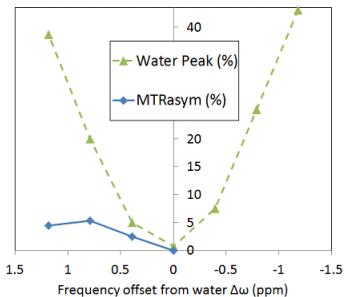


Fig 2. *In Vivo* Z spectra and MTR_{asym} for one subject during 4 breath holds ($B_1 = 1\mu\text{T}$)

Ref: [1] Lavoie et al, Can J Phys Pharm (1997) 1:26-35 [2] Stephenson MC et al, NMR in Biomed (2013) 26:1518-1526 [3] Taylor et al, Mag Res in Med (1992) 27:13-20 [4] Rode et al, Rec Prog Horm Res (2001) 56:219-37 [6] van Zijl et al, PNAS (2007) 104:4359-4364.

Acknowledgements: We thank Unilever and BBSRC for funding S Bawden in his industrial CASE studentship