# Localized *in vivo* $^{13}$C MRS of brain glycogen at 9.4 and 14.1 T: a comparison

**R. B. van Heeswijk[1], Y. Pilloud[1], F. D. Morgenthaler[1], and R. Gruetter[1,2]**
[1]Laboratory for Functional and Metabolic Imaging (LIFMET), Ecole Polytechnique Fédérale de Lausanne (EPFL), Lausanne, VD, Switzerland, [2]Radiology, Universities of Lausanne & Geneva, Lausanne & Geneva, Switzerland

## Introduction

Glycogen (Glyc) is the main energy store in the brain, but its precise metabolic role remains to be fully elucidated [1]. Currently, the only method to study brain glycogen non-invasively *in vivo* is $^{13}$C NMR together with $^{13}$C label incorporation through a substrate such as [1-$^{13}$C] glucose (Glc). Nevertheless, this method remains challenging and time consuming, not only due to the low amount of signal per unit of time, but also due to the broad line shape of the glycogen resonances. The main cause of this detection challenge are its short $T_1$ and $T_2$ relaxation times (on the order of a few hundred and 5-10 ms respectively), caused by its branched polysaccharide structure. The aim of this study was to follow the improvement of the Glyc C1 resonance with a magnetic field strength $B_0$ increase from 9.4 to 14.1 T, and to compare it to the adjacent Glc C1β resonance. Since the Glyc $T_1$ relaxation time is known to increase proportionally with $B_0$ [2,3] and might have an influence on the signal-to-noise ratio (SNR) at short relaxation times, it was also determined at both field strengths.

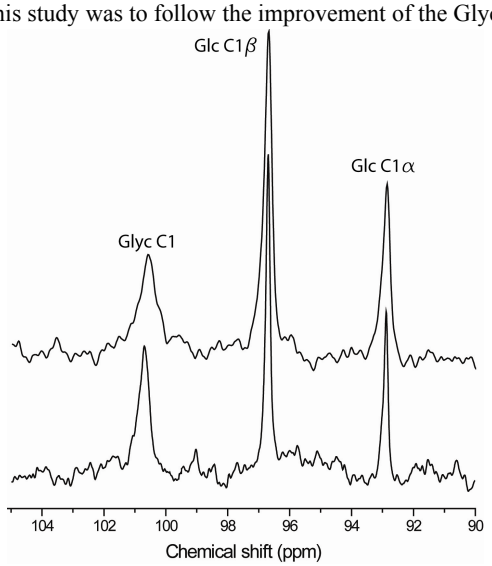


***Figure 1.*** *A comparison of the signals generated at 9.4 T (upper spectrum) and 14.1 T (lower spectrum).The spectra are equalized to the height of the Glc C1β peak. 15 Hz line broadening and two times zero filling have been applied. Note the relatively narrow linewidth at 14.1 T, which leaves a clearer baseline.*

## Materials and Methods

All methods and equipment were used as similar as possible for the two magnetic field strengths. Male Sprague-Dawley rats (n=4 at 9.4 T, n=3 at 14.1 T) were pre-labeled at the Glyc C1 position as previously described, such that the Glyc C1 isotopic enrichment (IE) was high and constant [4]. A femoral vein was then catheterized for infusion of α-chloralose (26.7 mg/kg/hr) and [1-$^{13}$C] Glc (~0.2 mg/hr), while a femoral artery was catheterized for blood sampling. A quadrature $^{1}$H coil with a single 3-loop 10 mm diameter $^{13}$C coil was positioned on top of the rat head. The animal was then inserted into a Varian 9.4 T or 14.1 T horizontal bore animal spectrometer. Blood samples were taken each 30 min for blood gas and glucose analysis and according adjustments.

The IE of NAA was determined with a carbon-edited $^{1}$H STEAM sequence, which was used to calculate the IE of brain Glyc C1 [4]. Localized $^{13}$C spectroscopy was then performed on a 6x8x10 mm$^3$ voxel with a modified SIRENE pulse sequence, which consisted of 3D OVS, 1D ISIS and 1D inversion nulling [4]. The SNR of this volume was determined for batches of 4096 acquisitions (TR=1 s; 1hr9min) by comparing the average of a 20 ppm noise region to the peak height. Waltz-16 NOE and decoupling were applied at the Glyc/Glc $^{1}$H frequency. Next, the $T_1$ relaxation time was determined by fitting the equation $S(t)=S(1-\beta\cdot\exp(t/T_1))$ to the glycogen signal integrals $S$ of an unlocalized adiabatic inversion recovery sequence with 7 inversion times. The rat was then replaced with a phantom containing Glc and Glyc to calculate their absolute concentrations as detailed in [4].

## Results and Discussion

Glyc could be detected with good SNR at both field strengths, and had ~1.7 times the SNR at 14.1 T compared to 9.4 T, while Glc showed a ~1.5 times increase (Table 1). Since the linewidth of both Glyc and Glc did not change significantly with the field increase, this was a net signal increase.

***Table 1.*** *Results of the Glyc and Glc measurements at 9.4 and 14.1 T. All errors are standard deviations of the mean.*

| $B_0$ | $SNR_{Glyc}$ (-) | $lw_{Glyc}$ (Hz) | $T_{1,Glyc}$ (ms) | IEGlyc (%) | [Glyc] (mM) | $SNR_{Glcβ}$ (-) | $lw_{Glcβ}$ (Hz) | $T_{1,Glcβ}$ (s) |
|---|---|---|---|---|---|---|---|---|
| **9.4 T** | 7.4 ± 1.8 | 48±8 | 332 ± 15 | 39 ± 5 | 5.8 ± 0.9 | 16.4 ± 2.5 | 11.7±1.1 | 1.9±0.1 |
| **14.1 T** | 12.3 ± 0.8 | 53±9 | 521 ± 34 | 36 ± 4 | 6.0 ± 0.4 | 23.8 ± 2.4 | 11.3±1.2 | 2.0±0.2 |

The constant linewidths together with the increased spectral dispersion further resulted in a more defined baseline at 14.1 T (Fig. 1), which allowed better spectral peak fitting and quantification, as evidenced in the halving of the error of the calculated absolute concentration of glycogen. The Glyc $T_1$ relaxation increased proportionally with the field strength, as expected from literature [3]. This increase together with the relatively short TR of 1 s means that the ~1.7 times signal increase underestimated the full SNR increase by 10% due to the incomplete recovery of the longitudinal magnetization (95% at $T_1$=332 ms vs 85% at $T_1$=521 ms).

To our best knowledge, this is the first report of *in vivo* $^{13}$C MRS of glycogen at a field strength higher than 9.4 T. In conclusion, the increase to 14.1 T offers not only a clear increase in SNR for $^{13}$C Glyc spectroscopy, but also a better defined spectrum, despite the increase in $T_1$ relaxation times.

## Acknowledgements

Supported by the CIBM of the UNIL, UNIGE, HUG, CHUV, EPFL and the Leenaards and Jeantet Foundations, as well as FNS grant #122498.

## References

1. Gruetter, *J Neurosci Res*, 74(2), p179-83 (2003) 2. Overloop et al., *Magn Reson Med* 36(1), p45-51 (1996)
3. Zang et al., *Biochemistry* 29(29), p6815-20 (1990) 4. Morgenthaler et al., *J Neurochem* 107(5), p1414-23 (2008)