In vivo quantitative MR spectroscopy using Relaxation Enhancement: unassigned brain metabolite resonances at 21T upon Stroke

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Target audience: Researchers and clinicians interested in stroke and *in vivo* quantitative localized MR spectroscopy techniques.

Purpose: To use Relaxation Enhancement for *in vivo* quantitative MRS; explore the downfield spectrum and search for novel stroke biomarkers.

Introduction

The effective T1 of macromolecular resonances may be shortened significantly using spectrally selective excitations[1]. By focusing the excitation on specific resonances, a large reservoir of magnetization remains unperturbed and can enhance MRS sensitivity by magnetization and chemical exchange[2]. Relaxation Enhanced (RE) ^{1}H MRS exploits this in *in vivo* studies, enhancing the global spectral quality and revealing unexpected apparent T1 reductions even for methyl resonances[3,4]. **This paper presents spatially localized spin-echo sequences based on RE and dedicated to *in vivo* quantitative MRS at 21.1T of metabolic resonating downfield from the water peak**. This downfield spectral region has been studied with care, but some resonances still remain unassigned[5,6].Most of the studies focused on specific metabolic resonance including Histidine, Homocarnosine, Phenylalanine, Glutathione (GSH) and recently NAD+[7-11].The resonances' intensities and chemical shift dependencies on pH, temperature and chemical exchange, make their assignment process very challenging. In this study we explored the downfield spectral region, quantified metabolite resonances such as ATP, Gln, GSH, NAA and PCr, and estimate the metabolites' changes in concentration upon stroke.

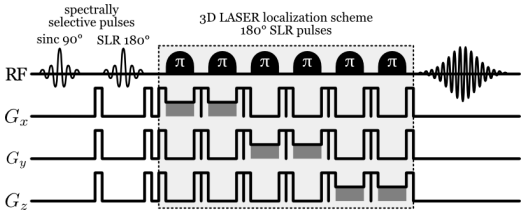

Fig. 1: Spatially localized RE spin-echo using 3D LASER and spectrally selective pulses.

Method

Pulse sequence. All experiments were performed at the National High Magnetic Field Laboratory using the homebuilt 21.1T MRI equipped with a Bruker Avance III spectrometer (Bruker Biospin, MA). The middle cerebral artery of Sprague-Dawley rats was occluded for 1.5h, followed by re-perfusion. Rats were scanned at 12h post ischemia. ^{1}H MRS were acquired from ~4.8x4.8x5.2mm voxel placed in the *contra-* and the *ipsi-lateral* (stroke) side of the brain using the localized RE spin-echo sequence, which targeted the 6-10ppm spectral region by means of 90° sinc pulse and SLR-designed[12] 180° pulses, respectively 5 and 4 ms long (TR=1.5s, NA=1024). Spatial localization was achieved using 3D LASER[13] (Fig. 1).

Quantification. Metabolites were quantified using a home-made algorithm based on GAMMA[14]. The model sums metabolic spectral signatures and fits them to the data by adjusting parameters such as concentrations, line-widths and frequency shifts. According to literature, most of the detectable metabolites in the downfield spectral region are in low concentration (≤100µM) and therefore undetectable. The metabolite basis set used here thus contained only ATP, Gln, GSH, NAA, PCr, present at ≥1mM concentrations and 4 broad Gaussian resonances dedicated to model the spectral baseline.

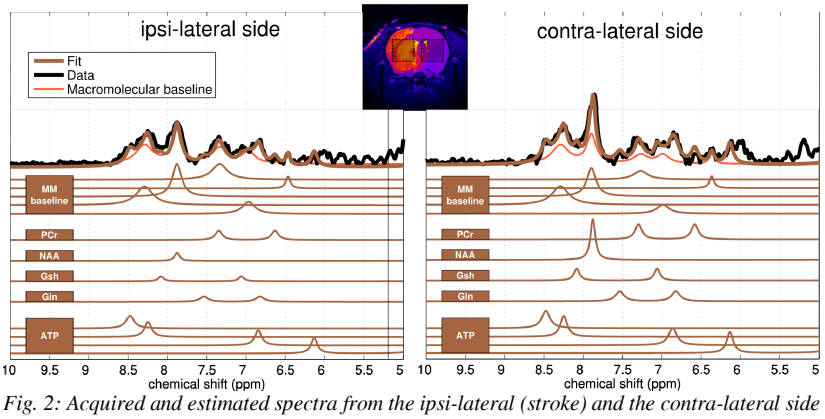

Fig. 2: Acquired and estimated spectra from the ipsi-lateral (stroke) and the contra-lateral side of the rat brain (water image on top).

Results & Discussion

Downfield spectra acquired on the *ipsi-lateral* and *contra-lateral* hemispheres of stroked brains show rich *in vivo* metabolic information (Fig. 2). The average SNR for the 7.8ppm NAA peak was 45. The estimated spectra fit the acquired data, showing intense macromolecular contributions especially around 8ppm. Resonance show substantial line-widths –presumable due to chemical exchange with water– leading to spectral overlapping even at 900MHz. All the quantified metabolites exhibit lower concentrations in the *ipsi-lateral* than in the *contra*-lateral side (Fig. 3); especially NAA, which is affected by a 5-fold difference (also observed in the up-field spectral region).

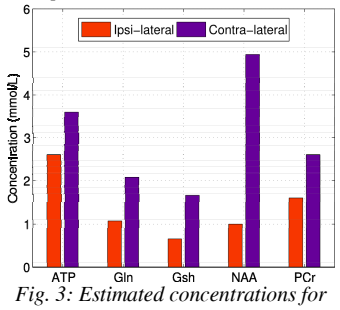

Fig. 3: Estimated concentrations for each metabolite.

Conclusion

For the first time, *in vivo* quantitative MRS of the downfield spectral region was successfully performed at 21.1T. RE significantly enhanced the global spectral quality while providing a commensurate increase in SNR per unit time. This allowed us to characterize hitherto poorly assigned resonances, including ATP. Quantification of such data remains challenging because of the strong dependences on environmental parameters (pH, temperature). However, this technique provides an alternative to conventional ***in vivo* 1H MRS and a new path to explore the little-known downfield spectral region which is potentially concealing important biomarkers.**

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