Quantification of Glutamate and Glutamine using CT-PRESS at 3T

M. Gu[1], N. M. Zahr[2,3], D. M. Spielman[1], E. V. Sullivan[2,3], A. Pfefferbaum[2,3], and D. Mayer[1,3]
[1]Radiology, Stanford University, Stanford, California, United States, [2]Psychiatry & Behavioral Sciences, Stanford University, Stanford, California, United States,
[3]Neuroscience Program, SRI International, Menlo Park, California, United States

Introduction

Glutamate (Glu) and glutamine (Gln) are two major neurochemicals in the central nervous system. Quantifying Glu and Gln *in vivo* using magnetic resonance spectroscopy (MRS) has been of particular interest to basic science for understanding the Glu/Gln component of the Krebs cycle and its alteration by neuropsychiatric disorders such as Alzheimer's disease, epilepsy and alcoholism. However, quantification of Glu and Gln using conventional *in vivo* MRS techniques such as PRESS or STEAM at clinically available field strength, e.g. 1.5-3T, is difficult because of the multiplet structure of the coupled resonances and signal overlap.

Constant time point resolved spectroscopy (CT-PRESS) [1] has been developed to reduce signal overlap by applying effective homonuclear decoupling, and the method has been optimized to detect the Glu C4 resonance at 2.35 ppm [2]. However, the Gln C4 resonance is not well resolved as it overlaps with the resonance from the aspartate moiety of NAA at 2.5 ppm. Because the decoupled CT-PRESS spectra are generated by integrating the 2D spectra along the diagonal in magnitude mode for the purpose of SNR, linear least square fitting techniques with prior knowledge, e.g., LCModel [3], are not directly applicable to quantify overlapping resonances. In this work, we developed a method that achieves quantification of both Glu and Gln using CT-PRESS, validated it in phantom experiments, and applied it to data from an *in vivo* study on the effects of ethanol (EtOH) on rat brain chemistry.

Methods

At 3T, a CT-PRESS sequence was optimized for detection of Glu C4 resonance (tc=139ms, n1=129, $\Delta t1/2$=0.8ms) [2]. 2D Fourier transform was performed for reconstruction after apodization in both t1 and t2 dimensions and the 1D-diagonal spectrum was generated by integrating the 2D magnitude spectrum along F2 within a ±13Hz interval. To reference the quantification to tissue water concentration, another acquisition was performed without water suppression (n1=17, $\Delta t1/2$=6.4ms).

The quantification started with simulating time-domain data set for each metabolite of interest using GAMMA [4] with the full density matrix and the same timing parameters as in the CT-PRESS. Chemical shifts and J-coupling constants used in the simulations were taken from [5]. Simulated time-domain data of each metabolite were weighted by an exponential with corresponding T2 relaxation time constant. The T2s for the singlets of NAA, Cho and Cre were estimated from the CT-PRESS data by mono-exponential fitting of their peak areas at different echo times. To account for the dephasing caused by B0 inhomogeneity, each time-domain data set was further weighted by another exponential with time constant T2' that was estimated from the corresponding data set acquired without water suppression. The full basis set was generated by summation of each metabolite's time-domain data set multiplied by its concentration weight. The full basis set was reconstructed in the same way as measured CT-PRESS data for effectively decoupled magnitude spectrum.

The procedure to determine the concentration weights comprised the following steps: first, the initial basis magnitude spectrum was reconstructed from a basis set with equal concentration weights for all metabolites. Concentration of the NAA singlet was estimated by setting its concentration weights in time domain to match the basis spectrum to the measured spectrum at 2ppm. Using the coupled NAA resonance at 2.67ppm, its T2 was determined by equating its estimated concentration with that of the NAA singlet. With effectively decoupled spectra and given the T2 of the Glu C4 resonance, Glu concentration was estimated by fitting the resolved resonance at 2.35ppm. For a validation phantom with known concentrations, the T2 of the Glu C4 resonance was estimated by adjusting it until actual and estimated NAA/Glu concentration ratio agreed. For *in vivo* studies, the T2 of the Glu C4 resonance was set at 125ms [2]. The T2 of the Gln C4 resonance was set to be the same as for Glu for both phantom and *in vivo* studies due to the similarity of their chemical structures. Given the estimated concentration and the T2 of the coupled NAA resonance, the contribution from the coupled NAA resonance to the peak at 2.45ppm was determined and the Gln concentration was estimated by adjusting its concentration weight until the basis and measured resonance fit at 2.45ppm. Concentrations of the Cho and Cre singlets were estimated in the same way as for the NAA singlet.

Results and Discussion

To validate the quantification method, the CT-PRESS sequence was performed on 3 custom-built spherical phantoms with different Glu/Gln concentrations using a GE 3T whole-body MR scanner (GE Healthcare, Waukesha, WI, USA). The concentration of NAA(12.5mM), Cho(3mM), Cre(10mM) and mI(7.5ppm) were kept constant across the phantoms while the Glu/Gln concentrations differed by plan: 12.5/6mM, 12.5/12.5mM and 6/12.5mM. Figure 1 shows the 1D-diagonal spectra reconstructed from the 3 phantoms. Variation in the Glu/Gln ratio was clearly demonstrated from their C4 resonances at 2.35 and 2.45ppm. Using the quantification method with the corresponding metabolite basis set, actual and estimated concentration ratios of Gln to Glu for 3 phantoms were 0.48/0.52, 1/0.81 and 2.08/2, demonstrating accurate concentration ratio tracking (Figure. 2).

The method was then used to reanalyze data from a study in which 10 wild-type Wistar rats were exposed to EtOH vapor over a period of 24 weeks (10 littermates breathed air without EtOH for control) [6]. MRS was performed prior to EtOH exposure (MRS1) and at weeks 16 (MRS2) and 24 (MRS2). The original analysis using peak integration had revealed higher Glu for the EtOH group at MRS3 and an increasing trend for the peak at 3.75 ppm, labeled Glx for combined Glu and Gln. However, this peak also has significant contributions from the Glutathione (GSH) singlet at 3.77 ppm. Analysis with the proposed method confirmed the finding for Glu concentration, but also revealed significantly higher Gln concentration in the EtOH group at MRS2 compared to controls (Figure. 3).

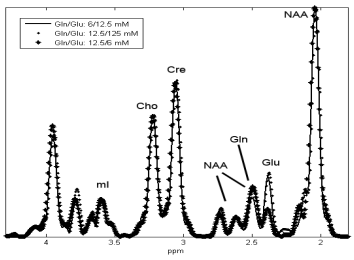


Fig. 1. CT-PRESS spectra of 3 phantoms.

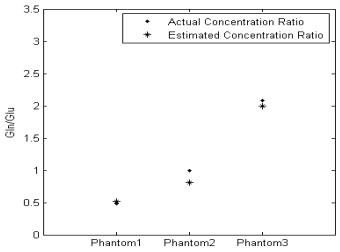
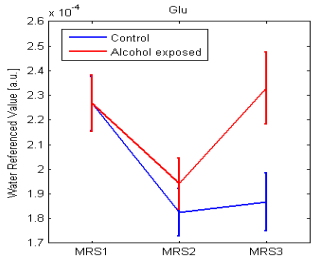


Fig. 2. Actual and estimated Gln/Glu ratios.



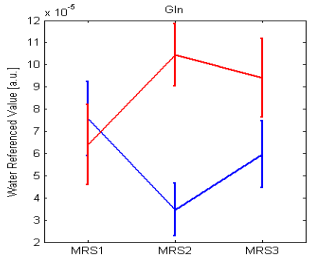


Fig. 3. Quantified Glu and Gln from control and EtOH exposed rat brain.

Conclusions

We have developed and validated a quantification method for Glu and Gln using CT-PRESS at 3T. It can also be used to quantify both singlets and other coupled resonances if they are sufficiently separated. In future work, the method will be extended similar to ProFit [7] to use the full 2D information in CT-PRESS.

Acknowledgements Lucas foundation, GE Healthcare, NIH grants RR 09784, AA005965, AA012388 and AA013521-INIA.

References

[1] W. Dreher, et al, MRI 17 :141-150, 1999. [2] D. Mayer, et al, MRM 54:439-442, 2005. [3] S. Provencher, et al., MRM 30:672-679, 1993. [4] SA. Smith et al, JMR A 106 :75-105, 1994. [5] V. Govindaraju, et al, NMR Biomed. 13 :129-153, 2000. [6] NM. Zahr et al., Neuropsychopharmacology 34(6) : 1427-42, 2009. [7] R. Schulte et al, NMR Biomed 19(2) :255-263, 2006.