Quantification of in vivo short echo-time proton magnetic resonance spectra at 14.1 T using two different approaches of modelling the macromolecule spectrum

Article *in* Measurement Science and Technology · October 2009

DOI: 10.1088/0957-0233/20/10/104034 · Source: OAI

4 authors, including:

Cristina Cudalbu
École Polytechnique Fédérale de Lausanne
75 PUBLICATIONS **1,277** CITATIONS

SEE PROFILE

Vladimir Mlynarik
Medical University of Vienna
213 PUBLICATIONS **4,861** CITATIONS

SEE PROFILE

Lijing Xin
École Polytechnique Fédérale de Lausanne
84 PUBLICATIONS **1,134** CITATIONS

SEE PROFILE

Meas. Sci. Technol. **20** (2009) 104034 (7pp) doi:10.1088/0957-0233/20/10/104034

Quantification of *in vivo* short echo-time proton magnetic resonance spectra at 14.1 T using two different approaches of modelling the macromolecule spectrum

C Cudalbu[1], V Mlynárik[1], L Xin[1] and Rolf Gruetter[1,2,3]

[1] Laboratory of Functional and Metabolic Imaging (LIFMET), Ecole Polytechnique Fédérale de Lausanne, Lausanne, Switzerland
[2] Department of Radiology, University of Lausanne, Lausanne, Switzerland
[3] Department of Radiology, University of Geneva, Geneva, Switzerland

E-mail: cristina.cudalbu@epfl.ch, vladimir.mlynarik@epfl.ch, lijing.xin@epfl.ch and rolf.gruetter@epfl.ch

Received 30 January 2009, in final form 23 March 2009
Published 4 September 2009
Online at stacks.iop.org/MST/20/104034

Abstract
Reliable quantification of the macromolecule signals in short echo-time ^{1}H MRS spectra is particularly important at high magnetic fields for an accurate quantification of metabolite concentrations (the neurochemical profile) due to effectively increased spectral resolution of the macromolecule components. The purpose of the present study was to assess two approaches of quantification, which take the contribution of macromolecules into account in the quantification step. ^{1}H spectra were acquired on a 14.1 T/26 cm horizontal scanner on five rats using the ultra-short echo-time SPECIAL (spin echo full intensity acquired localization) spectroscopy sequence. Metabolite concentrations were estimated using LCModel, combined with a simulated basis set of metabolites using published spectral parameters and either the spectrum of macromolecules measured *in vivo*, using an inversion recovery technique, or baseline simulated by the built-in spline function. The fitted spline function resulted in a smooth approximation of the *in vivo* macromolecules, but in accordance with previous studies using Subtract-QUEST could not reproduce completely all features of the *in vivo* spectrum of macromolecules at 14.1 T. As a consequence, the measured macromolecular 'baseline' led to a more accurate and reliable quantification at higher field strengths.

Keywords: *in vivo* short echo-time ^{1}H MRS, quantification of neurochemical profile, macromolecule contribution.

(Some figures in this article are in colour only in the electronic version)

1. Introduction

Localized proton magnetic resonance spectroscopy (^{1}H MRS) became an important tool for investigating brain metabolism non-invasively. In animal models, ^{1}H MRS allows the detection of almost 20 metabolites [1], also called the neurochemical profile: taurine (Tau), creatine (Cr), phosphocreatine (PCr), phosphocholine (PCho), glycerophosphocholine (GPC), glutamate (Glu), glutamine (Gln), myo-inositol (Ins), ascorbate (Asc), alanine (Ala), aspartate (Asp), γ-aminobutyrate (GABA), glutathione (GSH), *N*-acetylaspartate (NAA), *N*-acetylaspartylglutamate (NAAG), glucose (Glc), lactate (Lac), phosphoethanolamine (PE) and glycine (Gly). Since brain metabolite concentrations can vary depending on the type of pathology, the usefulness of ^{1}H MRS has been demonstrated in many brain disorders,

e.g., in hepatic encephalopathy; Alzheimer's, Huntington's and Parkinson's diseases; acute traumatic brain injury; cancer; dementia; etc [2, 3]. The metabolite changes help to understand metabolic processes related to the studied pathologies and/or enable to monitor the effect of treatment.

At longer echo times (TE above 20 ms), the number of spectral lines, which can be used for quantification, is reduced. As a result, in most *in vivo* ^{1}H MRS studies only changes of NAA, Cr+PCr, Cho and Lac have been detected. Short echo-time *in vivo* ^{1}H MRS spectra (TE = 1–20 ms) contain more information due to minimal distortions of multiplets of coupled spin systems such as Glu, Gln, Ins, Glc, Asp, Ala, GABA, Asc, PE, Gly and Tau. However, due to overlap of proton signals from brain metabolites, sophisticated approaches for the separation of the metabolite signals are required. Consequently, metabolite concentrations are usually determined by fitting the measured *in vivo* ^{1}H MRS spectrum to a linear combination of the spectra of individual metabolites (also called the metabolite basis set). The metabolite basis set can be obtained either by measuring aqueous solutions of pure metabolites or by quantum-mechanical simulations using known spectral parameters [4]. Well-known time- and frequency-domain algorithms [5–8], based on metabolite basis sets, are currently used for an accurate quantification. The most frequently used algorithms for proton spectra quantification are QUEST [7] from jMRUI software working in the time domain and LCModel [5, 6] working in the frequency domain.

In addition, the spectra measured at short echo times are further complicated by the presence of macromolecules (broad signals ascribed mainly to cytosolic proteins [9]), which further complicates the quantification. Reliable quantification of the macromolecule signals in short echo-time proton MRS spectra plays a key role in determining the neurochemical profile [10, 11]. Consequently, an error in the macromolecule estimation can lead to substantial errors in the obtained metabolite concentrations [10, 11]. In addition, it is well known that changes in the macromolecule concentrations can be considered as disease markers [12, 13].

Modelling the macromolecule contributions in an *in vivo* spectrum can be done using two main approaches. The macromolecule signal is approximated by a mathematical function and then included in the fit [5, 6, 13–16] or is separately estimated in a pre-processing step [7, 8, 14, 18–22]. It is well known that these procedures provide only a mathematical approximation of the real *in vivo* macromolecule spectra [11, 23]. This mathematical approach can be satisfactory at low magnetic fields. However, more prior knowledge is needed to obtain an accurate estimation of the metabolite concentrations at higher magnetic fields, where the macromolecule spectrum is better resolved.

In addition to the mathematical approximation of the macromolecules, a second approach was proposed, which provides the necessary prior knowledge. It is based on a separate acquisition of the *in vivo* macromolecule spectra using an inversion recovery method (a metabolite-nulled spectrum). Then, during the quantification step the *in vivo* acquired spectrum of macromolecules is either added to the basis set [10, 11, 24–29] or subtracted from the *in vivo* signal [23, 30, 31]. Small residuals attributed to metabolites are still observed in the metabolite-nulled spectra due to variability in longitudinal relaxation times of metabolites (T_1). Thus, knowledge of the T_1 relaxation times of metabolites is required for identification of these residual metabolite signals and subsequent removal by postprocessing.

The purpose of the present study was to assess the importance of the accurate estimation of the macromolecule contributions in short echo-time proton MRS spectra. Consequently, using LCModel [5, 6] we compared two different approaches of estimating macromolecule contributions in the quantification step at 14.1 T. In the first approach, the spectrum of macromolecules was measured *in vivo* using the inversion recovery technique and added to the metabolite basis set. In the second approach, the macromolecule spectrum was mathematically approximated using the built-in LCModel spline baseline. Preliminary results of this study have been reported in [32].

2. Methods

2.1. Animals

In vivo experiments were performed on adult female Sprague-Dawley rats (five animals, ∼250 g), which were anesthetized during the experiments with 1.5–2.5% isoflurane using a nose mask. Body temperature was maintained at 37.5 ± 1.0 °C by circulating warm water around the animals. All animal experiments were conducted according to federal and local ethical guidelines and the protocols were approved by the local regulatory body.

2.2. MRS measurments

Proton spectra were acquired using a Varian Inova (Palo Alto, CA, USA) console interfaced to a 14.1 T magnet (Magnex Scientific, Oxford, UK) with a 26 cm horizontal bore. The magnet was equipped with 12 cm inner-diameter actively shielded gradient sets giving a maximum gradient of 400 mT m $^{-1}$ in 120 μs. Home-built 14 mm diameter ^{1}H quadrature surface coils were used as transceivers. Eddy currents were minimized using time-dependent quantitative eddy current field mapping [33]. The static field homogeneity was adjusted by means of first- and second-order shims using an EPI version of FASTMAP [34]. Localized images were obtained in the coronal planes using a multislice fast spin echo protocol with TE/TR = 60/5000 ms, a slice thickness of 1 mm and an in-plane resolution of 94 μm. Spectra were acquired using an ultra-short echo-time spin echo full intensity acquired localization (SPECIAL) technique (TE/TR = 2.8/4000 ms, 4096 complex data points, SW = 7 kHz, 320 averages) [1, 35]. The size of the voxel for ^{1}H MRS was 3 × 4 × 4 mm^3 including frontal cortex, corpus callosum and striatum. After first- and second-order shimming, the typical linewidth of the water resonance at TE = 2.8 ms was 18–20 Hz. The water signal was suppressed by a series of seven 25 ms asymmetric variable power RF pulses with optimized relaxation delays (VAPOR) [36]. The water suppression pulses were interleaved

with three modules of outer volume saturation as described elsewhere [36]. To compensate for the magnetic field drift, spectra were collected in blocks of 16 averages, which were stored separately in the memory and were corrected for the relative shift in frequency.

For measuring the *in vivo* spectrum of macromolecules, the SPECIAL sequence was extended with a 2 ms nonselective hyperbolic secant inversion pulse (a bandwidth of 10 kHz), which was applied at a certain time period (inversion time, TI) before starting the localization part of the sequence, as described in [1]. A series of these inversion recovery spectra (TE of 2.8 ms or 40 ms, TR of 2500 ms and 640 averages) were measured with inversion times of 420, 600, 750, 850 and 1000 ms. Residual signals attributed to incompletely nulled metabolites were removed using HLSVD (Hankel-Lanczos singular value decomposition algorithm) [37] from the jMrui software [38].

2.3. Data analysis

Metabolite concentrations were estimated using the LCModel, combined with a simulated basis set of metabolites and either the spectrum of macromolecules measured *in vivo* using an inversion recovery technique or the spectrum of macromolecules mathematically approximated using the built-in LCModel spline baseline. The water signal was used as an internal reference and the average relative differences between the concentration estimates using the two approaches were compared. The basis set was created by quantum mechanics simulations, based on the density-matrix formalism [39], using published values of J-coupling constants and chemical shifts [4]. The Cramér–Rao lower bounds (CRBs) were calculated by LCModel as a measure of the reliability of the metabolite estimates. The metabolite concentrations were estimated for the following 19 neurochemical profiles: Ala, Asp, PCho, Cr, PCr, GABA, Gln, Glu, GSH, Gly, Ins, Lac, NAA, Tau, Asc, Glc, NAAG, GPC and PE.

To evaluate any possible differences between the metabolite concentrations obtained using the two approaches, the metabolite concentrations were statistically compared pair-wise using a paired two-tailed Student's *t*-test.

3. Results and discussion

Figure 1 displays the macromolecule spectrum acquired *in vivo* in the rat brain at 14.1 T. The main resonances of the macromolecules were well identified. However, since T_1 relaxation times of metabolite resonances are not identical [40], a macromolecule spectrum with completely suppressed metabolite resonances could not be obtained. Consequently, based on the evolution of the metabolite intensities over the series of inversion recovery spectra (changing from negative to positive), the spectrum at TI = 750 ms and TE = 2.8 ms (figure 1(*a*)) was taken as a basis for the spectrum of macromolecules, since it contained the smallest residual metabolite peaks (positive or negative). In addition, the residual metabolite peaks were confirmed by measuring an inversion recovery spectrum with the same TI = 750 ms,

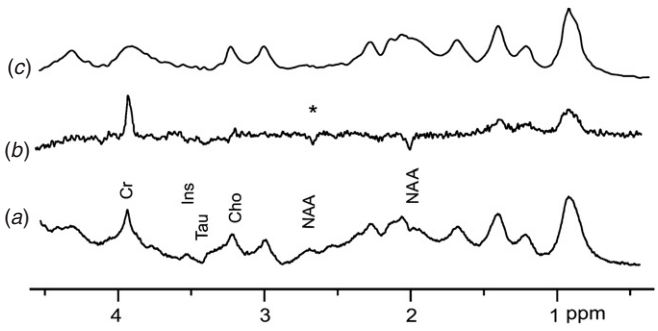

Figure 1. Estimation of the spectrum of macromolecules based on IR-SPECIAL spectra using the TI = 750 ms and TE = 2.8 ms. The spectrum (*a*) showing minimal residual peaks (negative peaks of NAA at 2 ppm and Tau at 3.4 ppm, and positive peaks of NAA at 2.7 ppm, GPC/PCho at 3.2, Ins at 3.6 ppm and Cr/PCr at 3.9 ppm) was taken as a basis for the spectrum of macromolecules. The spectrum (*b*) presents the final macromolecule spectrum measured with TI = 750 ms and TE = 40 ms. The spectrum (*c*) displays the final macromolecule spectrum after removal of the residual metabolite peaks using HLSVD. The residual signal of NAA at 2.7 ppm marked with an asterisk in the spectrum (*b*) is reduced and inverted relative to the spectrum from (*a*) due to J-evolution.

however, with a longer TE = 40 ms. In this spectrum (figure 1(*b*)) the residual signals of metabolites were visible due to longer T_2 relaxation times, while the signals of macromolecules were substantially reduced due to their short T_2 relaxation times. The residual peaks of NAA at 2.0 and 2.7 ppm, Tau at 3.4 ppm, total choline at 3.2 ppm, Ins at 3.6 ppm and total Cr at 3.9 ppm, identified in the spectrum in figure 1(*b*), were then removed from the macromolecule spectrum in figure 1(*a*) using the HLSVD algorithm. Since the residual peaks were visible in the *in vivo* spectrum (figure 1(*a*)), they were removed by using the option from the HLSVD algorithm, in which the user defines the frequency region to be removed in the spectrum. The final macromolecule spectrum is displayed in figure 1(*c*).

The LCModel analysis of a representative 14.1 T spectrum is shown in figure 2. Figure 2(*a*) presents the measured *in vivo* spectrum in the rat brain at 14.1 T. As can be seen, despite excellent spectral dispersion at 14.1 T, many spectral lines of metabolites remained overlapped among each other as well as with macromolecule signals. Figures 2(*b*) and (*c*) display the corresponding LCModel fits using the measured macromolecule spectrum (figure 2(*b*)) and the built-in LCModel spline baseline (figure 2(*c*)). The LCModel quantification using a basis set of simulated spectra of metabolites combined with the measured spectrum of the macromolecules provided an excellent fit to the experimental data. The spectrum fitted without the experimental macromolecule spectrum provided a poor match of the experimental spectrum, with residuals observed particularly in the spectral region from 0.5 to 2.0 ppm, compared to the LCModel fit with the macromolecule spectrum. The fits of the individual metabolites are plotted in figure 2(*d*).

Figure 3(*a*) shows the built-in LCModel spline baselines obtained from five rats and overlaid in different colours on a representative 14.1 T *in vivo* spectrum of rat brain, whereas figure 3(*b*) displays the *in vivo* measured macromolecule

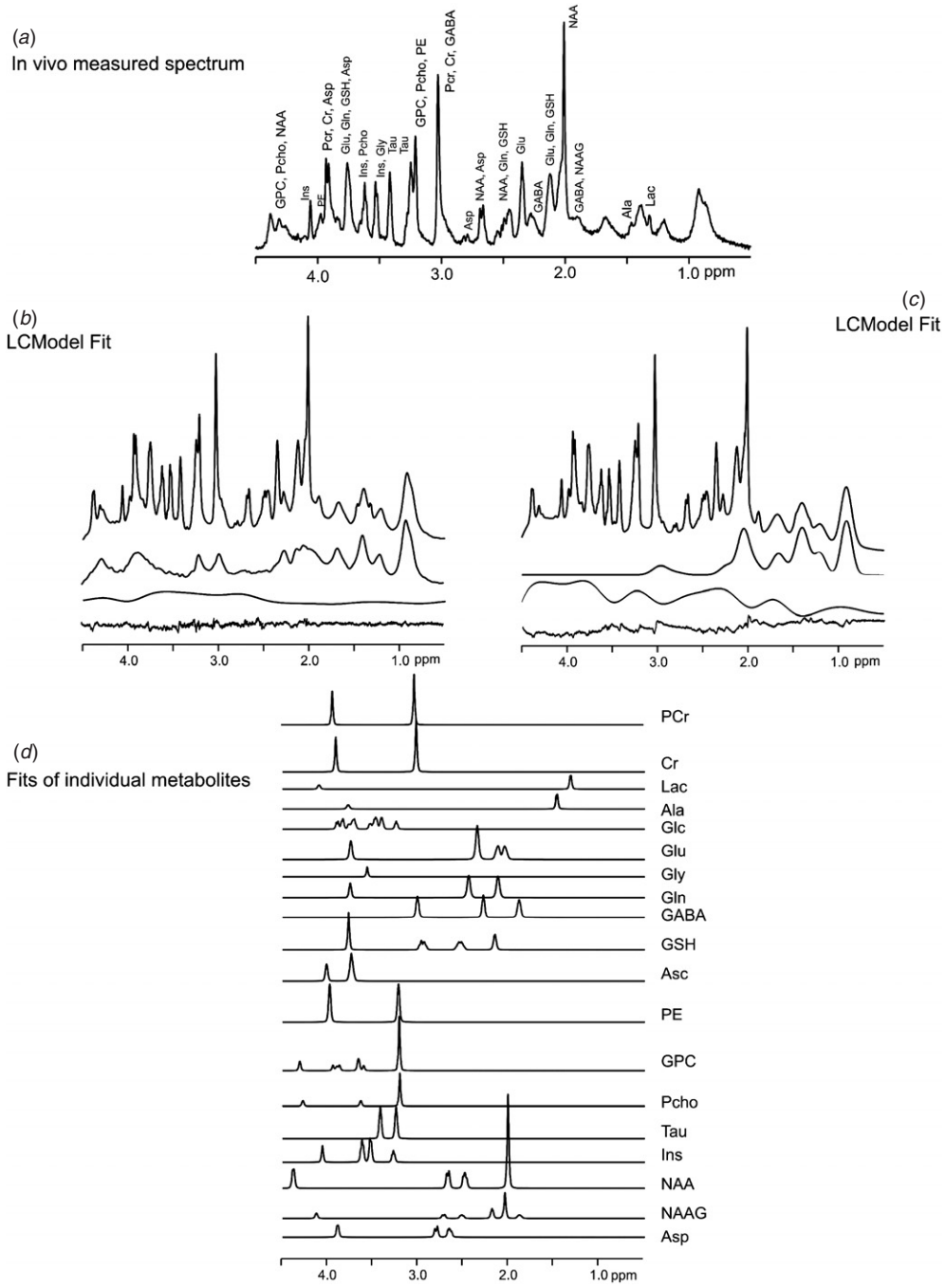

Figure 2. The LCModel analysis of a representative 14.1 T spectrum. The measured *in vivo* spectrum in the rat brain at 14.1 T is shown in (*a*). The corresponding LCModel fits using the measured macromolecule spectrum and the built-in LCModel spline baseline are also displayed in (*b*) and (*c*), respectively. The traces below represent from top to bottom, measured (*b*) or modelled (*c*) macromolecules, residual baseline and the difference between the measured and fitted data (also called residue of the quantification). The fits of the individual metabolites are plotted in (*d*).

spectrum combined with the same 14.1 T *in vivo* spectrum of the rat brain. The built-in LCModel spline baselines obtained from five rats fitted well the signals of macromolecules and they were highly consistent among the animals, except for the region close to H_2O at around 4.5 ppm (figure 3(*a*)). However, this smooth approximation of the *in vivo* macromolecules could not reproduce completely all features of the *in vivo* spectrum of macromolecules at 14.1 T as shown in figure 3(*b*). As can be seen in figure 3(*a*), compared to the *in vivo* measured spectrum of macromolecules, the built-in LCModel spline baseline was raised around 2.0 and 4.0 ppm and lowered around 3.0 and 3.2 ppm, leading to a decrease and an increase of concentrations of metabolites resonating in these regions, respectively.

The mean values and standard deviations of the metabolite concentrations obtained using the two approaches are shown in figure 4. Although there was an overall reasonable agreement in metabolite concentrations, NAA and Glu concentrations

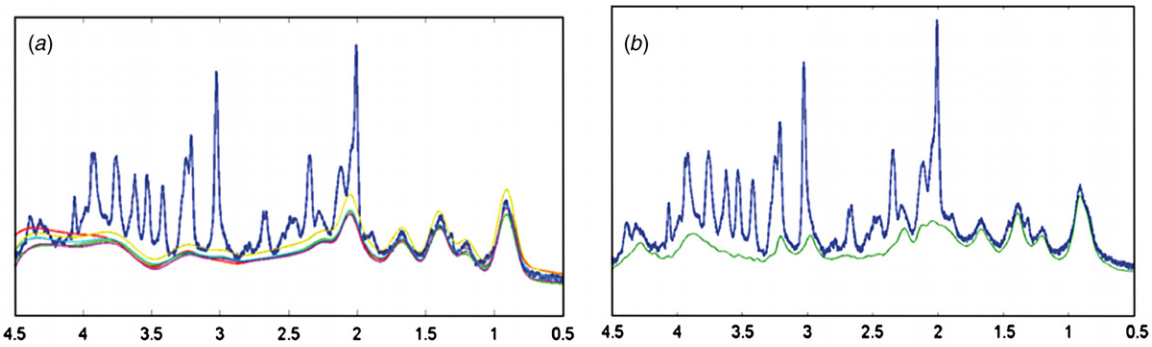

Figure 3. A representative 14.1 T spectrum (320 averages) of rat brain measured from a VOI = 3 × 4 × 4 mm^3 (blue line) combined with (*a*) the built-in LCModel spline baselines obtained from fitting spectra of five rats, which are plotted in different colours and with (*b*) the measured *in vivo* macromolecule spectrum (green line). Note the difference in the macromolecules estimation around 2 ppm, 3.0 ppm, 3.2 ppm and 4 ppm. Reproduced with permission from [32] Copyright © 2008 IEEE.

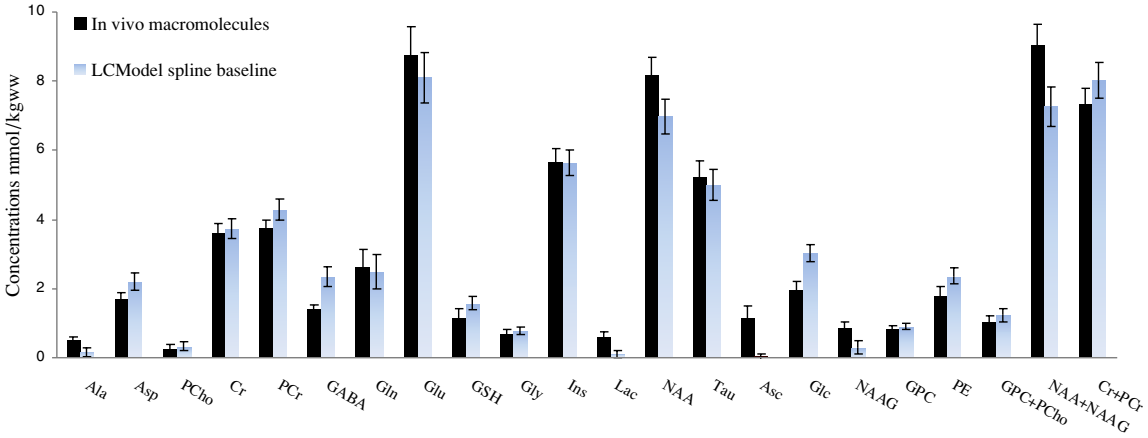

Figure 4. The mean values and standard deviations of the metabolite concentrations obtained using: (1) the spectrum of macromolecules measured *in vivo* (black bars) and (2) the built-in LCModel spline baseline (light blue bars). Reproduced with permission from [32] Copyright © 2008 IEEE.

were slightly lower (8–15%) and Cr+PCr, GPC+PCho and Gly concentrations were slightly higher (10–17%) when using the fitted splines. A systematic increase of Glc, GABA, PCho, GSH, PE and Asp concentrations (∼30–70%) and a decrease of Ala, Asc, NAAG and Lac concentrations (∼70–100%) were noted when using the fitted splines compared with the approach using measured macromolecules. For almost all metabolites, the standard deviations and CRBs of the metabolite concentrations were similar in the two approaches, except for a few low concentrated metabolites (Ala, Lac, Asc and NAAG), which were higher for the baseline fitted with splines. Statistical significant differences with p values in the range from 0.02 to 0.0001 were obtained for Ala, Asp, GABA, GSH, Lac, NAA, NAAG, Asc, Glc and PE. The metabolite concentrations obtained using the *in vivo* measured macromolecule spectrum, were in agreement with previous studies [1, 10, 28]. Figure 3 demonstrates that the bias in the metabolite concentrations is due to the raised or lowered baseline fitted with the spline functions.

Only two studies at lower magnetic fields of 3 T and 7 T [11, 23] compared the simulated and experimentally obtained spectra of macromolecules in the quantification of *in vivo* proton spectra using the Subtract-QUEST algorithm [7]. In this algorithm, the macromolecule spectrum is mathematically estimated from the first data points of the MRS signal. This approach exploits the fact that the macromolecule signals decay rapidly in the time domain due to shorter T_2 relaxation times. Significant differences in the determined concentrations were obtained by the authors when using the mathematical estimation of the macromolecules. These differences are in agreement with our study. Moreover, the authors also observed that the macromolecule spectrum estimated by Subtract-QUEST displayed only broader features, which disagrees with what is actually seen in the measured spectra.

Our results showed that an experimental estimation of the macromolecule spectrum can improve the quality of the fit at 14.1 T. The absence of the measured macromolecule spectrum in the database led to a large and unpredictable bias in the concentrations of many metabolites. Even at the highest magnetic fields available for *in vivo* experiments, spectral overlap of proton signals from brain metabolites and macromolecules still requires an experimental assessment of macromolecular contribution to the proton spectrum for an accurate metabolite quantification. The main reason for the poor mathematical estimation of macromolecules at high magnetic fields is due to the fact that the largely

field-independent linewidth of the signals of macromolecules increasingly approaches that of metabolites. As can also be seen from [1, 10, 11, 41], an effectively increased spectral resolution of the macromolecule components can be observed at higher magnetic field strengths, making the estimation of the macromolecules by means of mathematical approximations even more difficult.

4. Conclusion

To our knowledge, this is the first study demonstrating the importance of the accurate assessment of the macromolecule contributions in quantification of short echo-time proton MR spectra using LCModel. Even if the fitted splines reproduce the *in vivo* macromolecules very well, we can conclude that the measured macromolecular 'baseline' represents additional prior knowledge, which leads to a more accurate and reliable quantification. This kind of knowledge becomes important particularly at higher static magnetic fields.

Acknowledgments

This work was supported by Centre d'Imagerie BioMédicale (CIBM) of the UNIL, UNIGE, HUG, CHUV, EPFL, the Leenaards and Jeantet Foundations, and by the EU grant no MRTN-CT-2006-035801.

References

[1] Mlynárik V, Cudalbu C, Xin L and Gruetter R 2008 ^{1}H NMR spectroscopy of rat brain *in vivo* at 14.1 Tesla: improvements in quantification of the neurochemical profile *J. Magn. Reson.* **194** 163–8

[2] Thorsen F, Jirák D, Wang J, Syková E, Bjerkvig R, Øyvind Enger P, Van Der Kogel A and Hájek M 2008 Two distinct tumor phenotypes isolated from glioblastomas show different MRS characteristics *NMR Biomed.* **21** 830–8

[3] Ross B D and Michaelis T 1994 Clinical applications of magnetic resonance spectroscopy *Magn. Reson. Q.* **10** 191–247

[4] Govindaraju V, Young K and Maudsley A A 2000 Proton NMR chemical shifts and coupling constants for brain metabolites *NMR Biomed.* **13** 129–53

[5] Provencher S W 1993 Estimation of metabolite concentrations from localized *in vivo* proton NMR spectra *Magn. Reson. Med.* **30** 672–9

[6] Provencher S W 2001 Automatic quantitation of localized *in vivo* ^{1}H spectra with LCModel *NMR Biomed.* **14** 260–4

[7] Ratiney H, Coenradie Y, Cavassila S, van Ormondt D and Graveron-Demilly D 2004 Time-domain quantitation of ^{1}H short echo-time signals: background accommodation *MAGMA* **16** 284–96

[8] Poullet J B, Sima D M, Van Huffel S and Van Hecke P 2007 Frequency-selective quantitation of short-echo time ^{1}H magnetic resonance spectra *J. Magn. Reson.* **186** 293–304

[9] Behar K L and Ogino T 1993 Characterization of macromolecule resonances in the ^{1}H NMR spectrum of rat brain *Magn. Reson. Med.* **30** 38–44

[10] Pfeuffer J, Tkáč I, Provencher S and Gruetter R 1999 Toward an *in vivo* neurochemical profile: quantification of 18 metabolites in short echo-time ^{1}H spectra of the rat brain *J. Magn. Reson.* **141** 104–20

[11] Cudalbu C, Beuf O and Cavassila S 2009 *In vivo* short echo-time localized ^{1}H MRS of the rat brain at 7 T: influence of two strategies of background-accommodation on the metabolite concentration estimation using QUEST *J. VLSI Signal Process. Syst.* **55** 25–34

[12] Ratiney H, Srinivasan R, Henry R G, Okuda D, Nelson S J and Pelletier D 2007 Early and progressive disease marker in MS; results from a large cross-sectional spectroscopic imaging study at 3 T *American Academy of Neurology 59th Annual Meeting (Boston, USA, May 2007)* p 02.041

[13] Hakumäki J M, Gröhn O H J, Pirttilä T R M and Kauppinen R A 1996 Increased macromolecular resonances in the rat cerebral cortex during severe energy failure as detected by ^{1}H nuclear magnetic resonance spectroscopy *Neurosci. Lett.* **212** 151–4

[14] Soher B J, Young K and Maudsley A A 2001 Representation of strong baseline contributions in ^{1}H MR spectra *Magn. Reson. Med.* **45** 966–72

[15] Schubert F, Seifert F, Elster C, Link A, Walzel M, Mientusand S, Haas J and Rinneberg H 2002 Serial ^{1}H-MRS in relapsing–remitting multiple sclerosis: effects of interferon-beta therapy on absolute metabolite concentrations *MAGMA* **14** 213–22

[16] Lemmerling P, Vanhamme L, in't Zandt H J A, Van Huffel S and Van Hecke P 2002 Time-domain quantification of short-echo-time *in-vivo* proton MRS *MAGMA* **15** 178–9

[17] Weiland E, Roell S A, Leibfritz D and Krueger G 2003 Time-domain fitting of ^{1}H-MR spectra of the human brain: a model-free integration of the macromolecular baseline *Proc. Int. Society for Magnetic Resonance in Medicine (Toronto, Canada, May 2003)* p 1160

[18] Young K, Govindaraju V, Soher B J and Maudsley A A 1998 Automated spectral analysis II: application of wavelet shrinkage for characterization of non-parameterized signals *Magn. Reson. Med.* **40** 816–21

[19] Coenradie Y, de Beer R, van Ormondt D, Cavassila S, Ratiney H and Graveron-Demilly D 2002 Background-signal parametrization *in vivo* MR spectroscopy *MAGMA* **15** 369

[20] Coenradie Y, de Beer R, van Ormondt D, Ratiney H, Cavassila S and Graveron-Demilly D 2002 Background-signal parametrization *in vivo* MR spectroscopy *Proc. ProRISC, IEEE Benelux (Veldhoven, The Netherlands, 2002)* pp 248–54

[21] Stanley J A and Pettegrew J W 2001 Postprocessing method to segregate and quantify the broad components underlying the phosphodiester spectral region of *in vivo* ^{31}P brain spectra *Magn. Reson. Med.* **45** 390–6

[22] Cavassila S, van Ormondt D and Graveron-Demilly D 2002 Cramer–Rao bound analysis of spectroscopic signal processing methods *Signal Processing for Magnetic Resonance Imaging and Spectroscopy* ed Y Hong (New York: Dekker) chapter 22

[23] Gottschalk M, Lamalle L and Segebarth C 2008 Short-TE localized ^{1}H MRS of the human brain at 3 T: quantification of the metabolite signals using two approaches to account for macromolecular signal contributions *NMR Biomed.* **21** 507–17

[24] Hofmann L, Slotboom J, Boesch C and Kreis R 2001 Characterization of the macromolecular baseline in localized ^{1}H-MR spectra of human brain *Magn. Reson. Med.* **39** 855–63

[25] Hofmann L, Slotboom J, Jung B, Maloca P, Boesch C and Kreis R 2002 Quantitative ^{1}H magnetic resonance spectroscopy of human brain: influence of composition and parametrization of the basis-set in linear combination model-fitting *Magn. Reson. Med.* **48** 440–53

[26] Seeger U, Mader I, Nagele T, Grodd W, Lutz O and Klose U 2001 Reliable detection of macromolecules in single volume ^{1}H NMR spectra of the human brain *Magn. Reson. Med.* **45** 948–54

[27] Seeger U, Klose U, Mader I, Grodd W and Nagele T 2003 Parameterized evaluation of macromolecules and lipids in proton MR spectroscopy of brain diseases *Magn. Reson. Med.* **49** 19–28

[28] Tkáč I, Rao R, Georgieff M K and Gruetter R 2003 Developmental and regional changes in the neurochemical profile of the rat brain determined by *in vivo* ^{1}H NMR spectroscopy *Magn. Reson. Med.* **50** 24–32

[29] Cudalbu C, Bucur A, Graveron-Demilly D, Beuf O and Cavassila S 2007 Comparison of two strategies of background-accommodation: influence on the metabolite concentration estimation from *in vivo* magnetic resonance spectroscopy data *Conf. Proc. IEEE Eng. Med. Biol. Soc (Lyon, France, 2007)* pp 2077–80

[30] Kassem M N E and Bartha R 2003 Quantitative proton short-echo-time LASER spectroscopy of normal human white matter and hippocampus at 4 T incorporating macromolecule subtraction *Magn. Reson. Med.* **49** 918–27

[31] Rabeson H, Ratiney H, Cudalbu C, Cavassila S, Capobianco E, de Beer R, van Ormondt D and Graveron-Demilly D 2006 Signal disentanglement in *in vivo* MR spectroscopy: by semi-parametric processing or by measurement? *Proc. ProRISC, IEEE Benelux (Veldhoven, The Netherlands, 2006)* pp 176–83

[32] Cudalbu C, Mlynárik V, Xin L and Gruetter R 2008 Comparison of two approaches to model the macromolecule spectrum for the quantification of short TE ^{1}H MRS spectra *IST 2008 – IEEE Workshop on Imaging Systems and Techniques Proceedings,* pp 311–4 (art. no. 4659991)

[33] Terpstra M, Andersen P M and Gruetter R 1998 Localized eddy current compensation using quantitative field mapping *J. Magn. Reson* **131** 139–43

[34] Gruetter R and Tkáč I 2000 Field mapping without reference scan using asymmetric echo-planar technique *Magn. Reson. Med.* **43** 319–23

[35] Mlynárik V, Gambarota G, Frenkel H and Gruetter R 2006 Localized short echo-time proton MR spectroscopy with full signal-intensity acquisition *Magn. Reson. Med.* **56** 965–70

[36] Tkáč I, Starčuk Z, Choi I Y and Gruetter R 1999 *In vivo* ^{1}H NMR spectroscopy of rat brain at 1 ms echo time *Magn. Reson. Med.* **41** 649–56

[37] Pijnappel W W F, Vandenboogaart A, Debeer R and van Ormondt D 1992 SVD-based quantification of magnetic-resonance signals *J. Magn. Reson.* **97** 122–34

[38] Naressi A, Couturier C, Castang I, de Beer R and Graveron-Demilly D 2001 Java-based graphical user interface for MRUI, a software package for quantitation of *in vivo*/medical magnetic resonance spectroscopy signals *Comput. Biol. Med.* **31** 269–86

[39] Mulkern R and Bowers J 1994 Density matrix calculations of AB spectra from multipulse sequences: quantum mechanics meets *in vivo* spectroscopy *Concepts Magn. Reson.* **6** 1–23

[40] Cudalbu C, Mlynárik V, Xin L and Gruetter R 2008 ^{1}H NMR T_1 relaxation times of the neurochemical profiles in rat brain at 14.1 T *Proc. Int. Society for Magnetic Resonance in Medicine (Toronto, Canada, May 2008)* p 3174

[41] Otazo R, Mueller B, Ugurbil K, Wald L and Posse S 2006 Signal-to-noise ratio and spectral linewidth improvements between 1.5 and 7 Tesla in proton echo-planar spectroscopic imaging *Magn. Reson. Med.* **56** 1200–10