In vivo Quantification of Human Brain Metabolites using ^{1}H-MRS for the Study of Patients with Juvenile Myoclonic Epilepsy

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Abstract

The technique of magnetic resonance spectroscopy (MRS) allows the detection of specific chemical compounds in the scanned sample. Given that it is a non-invasive technique, it has been used in the clinical environment for the in vivo study of the human brain in normal and pathological conditions, since several neurological disorders have been associated to variations in the concentrations of MRS detectable metabolites. The aim of this work was to quantify in vivo ^{1}H-MRS brain data of patients with Juvenile Myoclonic Epilepsy (JME) and of control subjects, to verify if there is a variation in the metabolic pattern of JME patients. For this, a study encompassing the physical principles underlying the MRS technique, the jMRUI software for pre-processing and quantification of MRS signals, and the AMARES quantification method (implemented in jMRUI) was performed. A sample containing 10 JME patients and 10 control subjects was quantified and statistically analyzed using the Systat 12 software. The groups quantification results were compared using the Kruskal-Wallis test, and this did not show significant differences among the relative concentrations of any peaks, what contradicts the results obtained in the pilot study performed by Mory et al. [Mory03]. This discrepancy may be due to the low signal-to-noise ratio of our data and to the small sample size. Next we intend to increase the number of subjects in both groups, and see whether the results change with a larger sample.

Generation of the MR signal and Formation of the MRS spectra

The phenomenon of MR occurs when a sample containing atoms with non-null nuclear spin is placed in a magnetic field $\mathbf{B_0}$. The spins of these atoms will precess around $\mathbf{B_0}$ (Fig. 1) with a frequency known as the Larmor frequency ω_0, given by:

$$\omega_0 = \gamma \cdot B_0 \qquad (1)$$

where γ is the gyromagnetic ratio, characteristic of each type of nucleus (γ = 42,58MHz/T for the hydrogen atom). Actually, this is a Quantum Mechanics phenomenon, and when the sample is placed in B_0 its spin energy state unfolds into 2 states (case of spin ½) separated by an energy proportional to ω_0. The sample can thus be excited by an electromagnetic wave with the Larmor frequency (which is in the range of radiofrequency), and when the spins relax the sample will emit a signal with this same frequency, which is the measured MR signal. This signal is proportional to the total magnetization of the sample, and is modulated by 2 relaxation processes: the spin-lattice (or longitudinal) relaxation (characterized by a time T1) and the spin-spin (or transversal) relaxation (characterized by a time T2). The MR signal is detected by a coil, where it induces a decaying voltage, and it is therefore known as FID (Free Induction Decay).

Chemical Shift

Although it would be expected that the FID contained a unique frequency component (the Larmor frequency), this is not the case. The FID consists of a superposition of many signals with different frequencies (Fig.2). This happens due to the different electronic environments that surround nuclei in different molecules (or in different positions inside the same molecule), which result in that each spin experiences a different local magnetic field, and therefore precesses with a different Larmor frequency. This is the basis of the technique of Magnetic Resonance Spectroscopy, since the different emitted frequencies can be related to specific chemical compounds in the sample. The MRS spectrum, which relates the amplitude of each signal to its frequency, is obtained through Fourier Transformation (FT) of the FID (Fig.3).

AMARES

AMARES is a quantification method that models the MRS data in the time domain, and attempts to fit this model to the measured data, following an iterative minimization procedure. For this, it uses initial values supplied by the user for the resonance frequencies and peak widths of the metabolites of interest. The result is an estimate of the concentrations of these metabolites in the scanned sample. The model function used by AMARES is:

$$y_n = \hat{y}_n + e_n = \sum_{k=1}^{K} a_k \exp\{j\phi_k + 2\pi f_k t_n) \exp[j(-d_k(1 - g_k + g_k t_n)t_n)] + e_n, \quad n = 0,\ldots,N-1 \qquad (2)$$

where $j=(-1)^{1/2}$, a_k is the amplitude, ϕ_k is the phase, d_k is the damping factor, and f_k is the frequency of the k-th FID sinusoidal component ($k=1,\ldots,K$). $t_n = n\Delta t + t_0$, where Δt is the sampling interval and t_0 is the time instant of the first point of the temporal series to be included into the analysis. en is a complex white Gaussian noise term. $\hat{y}$ is the model function and y represents the measured data. And the g_k parameter allows the user to choose between a Lorenzian (g_k=0) or Gaussian (g_k=1) line form for each peak of the spectrum. The aim of the quantification is to find values for the parameters a_k, ϕ_k, d_k and f_k. This is achieved by minimizing the functional

$$G(\mathbf{a}, \mathbf{d}, \mathbf{f}, \boldsymbol{\phi}, t_0) = \sum_{n=0}^{N} |y_n - \hat{y}_n|^2$$

Juvenile Myoclonic Epilepsy (JME)

Epilepsy is a disorder of cerebral origin caused by the predisposition to generate seizures. A seizure is a symptom caused by large electrical instability of some brain cells leading to a disorder in which neurons signal in an abnormal way. It is estimated that about 2% of the world population has this disorder, and this number may be higher in developing countries, which concentrate 85% of the cases. The Juvenile Myoclonic Epilepsy (JME) represents 7% to 9% of all of the epilepsies. Nowadays it is considered the most frequent among the idiopathic generalized epilepsies. It corresponds to an epileptic syndrome related to age, with peak between 12 and 14 years, affecting both sexes in a similar way. This work is of great importance for the study of this pathology, since it proposes a new diagnosis form, where the metabolic pattern of specific brain structures is compared between patients and controls.

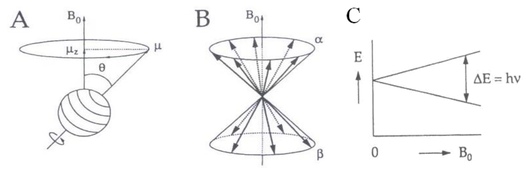

Fig.1: A) Spin precession around a magnetic field B_0. B) macroscopic sample of nuclear spins with s=½, that are distributed in 2 possible orientations (due to the Zeeman unfolding) : parallel or anti-parallel to B_0. C) Zeeman effect [Graaf07].

Systat 12 is a statistical analysis software. Kruskal Wallis is a non-parametric method for testing the equality of population medians among groups. It is an extension of the Mann-Whitney U test to 3 or more groups. Since it is a non-parametric method, the Kruskal-Wallis test does not assume a normal distribution.

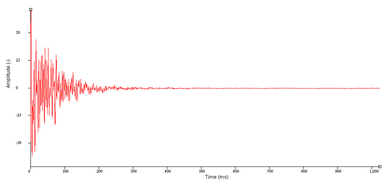

Fig.2: FID signal obtained from MR.

FT

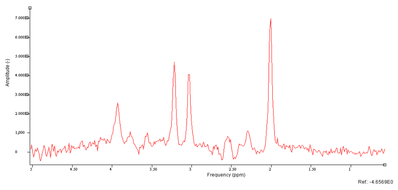

Fig.3: Typical MRS spectrum.

jMRUI

(http://www.mrui.uab.es) is a public domain software for pre-processing and quantification of MRS signals. It is a graphical interface that joins several programs developed for the processing of MRS signals in vivo obtained in clinical environments with magnetic fields of medium intensity (≤ 3T), among them the AMARES method used here. This software allows the representation of the data in the time (FID) and in the frequency (spectrum) domain.

Pre-processing of the MRS signal with the jMRUI software

To allow detection of metabolites which are present in the brain at tiny concentrations (about 10^{-5} the water concentration), the water signal is supressed during acquisition. The acquired signal is then digitized. To eliminate artifacts and to facilitate the quantification of the metabolites in the spectrum, some common processing steps must be performed, which can be done with the jMRUI software. These are:

- **Spectra averaging:** several spectra are collected (about hundreds of them) and summed to generate a single spectrum, increasing the SNR (Fig. 4).
- **Apodization:** the FID signal is multiplied by a function that decreases in time, normally a Lorentzian (Fig. 5) or Gaussian function, to eliminate the last points which contain mainly noise.
- **Filtering of the residual water signal:** inspite of supression there is always a residual water signal which can be filterd with the HLSVD (*Hankel Singular Lanczos Values Decomposition*) algorithm (Fig. 6). This models the peak in the time domain and subtracts it from the original FID.
- **Phasing:** corrections are made to the phase of the spectrum in order to make the peaks become the most symmetrical possible (Fig. 7).
- **Baseline correction** (Fig. 8)

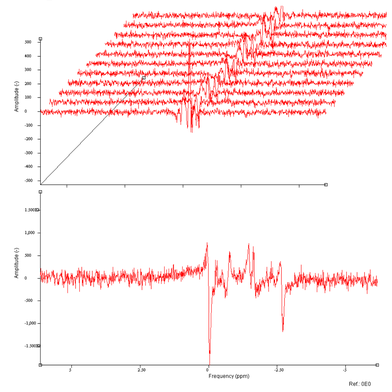

Fig.4: Average spectrum after summation of low SNR spectra.

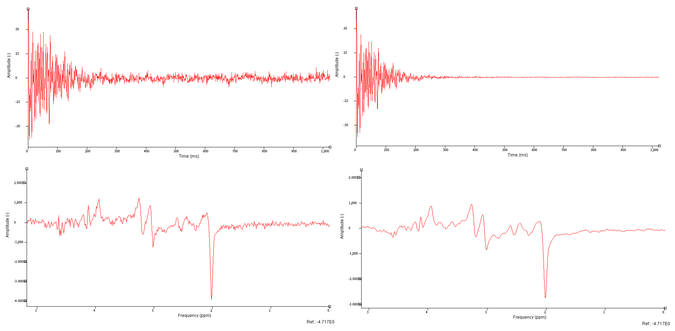

Fig.5: Apodization of the FID (top) and correspondent MRS spectra (bottom).

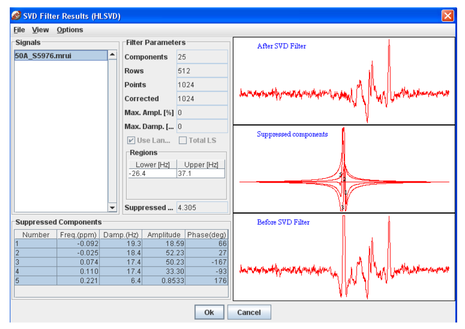

Fig.6: Residual water peak filtering.

Results

In the present work a sample of MR spectra containing 10 JME patients (60% women, mean age 30,1±7,9 years) and 10 control subjects (60% women, mean age 31,5±7,2 years) was quantified and statistically analyzed using the Systat 12 software. The MR spectra were obtained with an Elscint Prestige 2.0T scanner, using a PRESS sequence (TR=1500ms, TE=135ms, bandwidth=1000Hz, 1024 data points), from 2×2×2cm^3 voxels positioned on the thalami of all subjects.

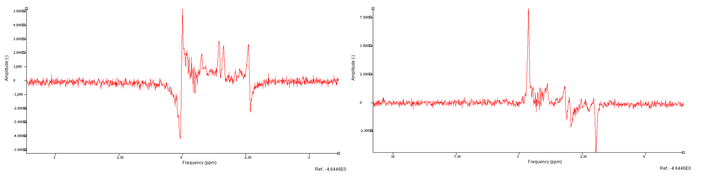

Fig.7: Spectrum phasing of the residual water peak.

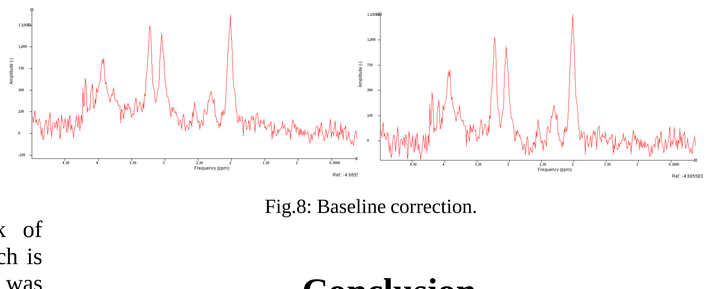

Fig.8: Baseline correction.

The quantifications were relative to the peak of Creatine plus Phosphocreatine (at 3.03ppm), which is known to stay constant for this pathology. There was not significant difference in age among the groups (T test, p=0,686). The groups quantification results were compared using the Kruskal-Wallis test, and this did not show significant differences among the relative concentrations of any peaks (the significance level used was 5%). The peaks quantified were: Lac (1,33ppm), Ala (1,47ppm), Lip (1,68ppm), NAA (2.01ppm), Glx1 (2,30ppm), Glx2 (2,39ppm), Glx3 (2,55ppm), Asp (2,80ppm), Cho (3,21ppm), Tau (3,38ppm), mIno (3,56ppm), Glx4 (3,76ppm) e Cre2 (3,96ppm) Lac (1.33ppm).

Conclusion

The study of JME is important given the prevalence of epilepsy in the world population. This work proposes an additional form of diagnosis of the disease through comparison of the cerebral metabolic pattern of JME patients and control subjects, obtained via MRS. Fig. 9 presents typical MRS spectra of those two types of individuals. Some problems of our data such as low SNR and small sample size did not allow to identify metabolic differences among the two groups. Next we intend to increase the number of subjects in both groups (to about 55 JME patients and 40 subject controls), and see whether the results change with a larger sample, which is already expected for this disease as seen in the pilot study performed by Mory et al. [Mory03].

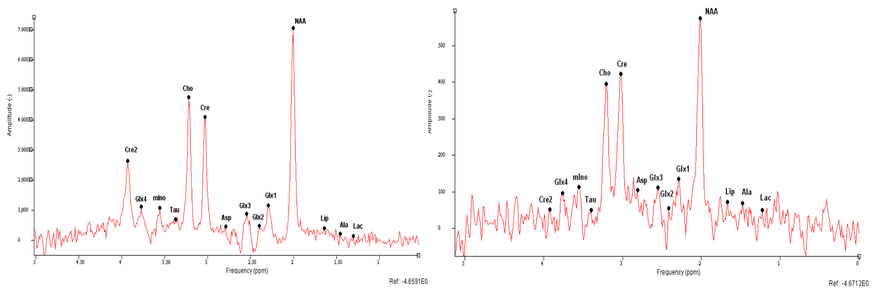

Fig.9: Spectrum from control subject (left) and JME patient (right).

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