Correlation between different stages of hepatic fibrosis and in vivo metabolic profile by 1H Magentic Resonante Spectroscopy

B. Martínez-Granados[1], M. C. Martínez-Bisbal[1,2], D. Monleón[3], L. Martí-Bonmatí[4], J. M. Rodrigo[5], J. del Olmo[5], A. Ferrández[6], and B. Celda[1,2]

[1]Physical Chemistry, Universitat de Valencia, Burjassot, Valencia, Spain, [2]CIBER Bioengineering, Biomaterials and Nanomedicine, ISC III, Burjassot, Valencia, Spain, [3]Fundación para la Investigación, Hospital Clínico Universitario, Valencia, Spain, [4]Radiology, Hospital Universitario Dr. Peset, Valencia, Spain, [5]Hepatology, Hospital Clínico Universitario, Valencia, Spain, [6]Pathology, Hospital Clínico Universitario, Valencia, Spain

Introduction: The extent of fibrosis and degree of inflammatory activity of hepatic pathology is nowadays established by histological evaluation of liver biopsy. This method has several disadvantages including that is an invasive examination with risk of serious complications. In vivo MR spectroscopy (^{1}H MRS) is a non-invasive technique that allows the direct study of hepatic tissue molecular composition. The aim of this study is to verify the potential clinical applications of in vivo liver metabolic profiling by ^{1}H MRS for supporting the diagnosis of different stage of liver diffuse disease.

Subjects and Methods: In vivo liver ^{1}H MRS studies were performed in 50 male patients (51±11 years) with clinic diagnosis of chronic liver disease and with different stages (Batts-Ludwig classification). All MRS spectra were recorded in a clinical MR unit operating at 1.5 T. The protocol of MRS data acquisition included SV on liver with dimensions of 45x45x45 mm (91.1cm^3) (figure 1) with TR 1800 ms and TE 136 ms. jMRUI program was used for processing and analyzing all the spectra. SPSS 14.0 program was used for statistical analysis.

Results: An important number of in vivo ^{1}H MRS resonances were assigned by using ex vivo HR-MAS (11 T) spectra [1] and with in vivo MRS literature data. This allowed the identification of weak and overlapped signals (figure 2). Different types of lipids, compounds of Choline and TMAO, glucose and glycogen signals were identified (figure 2). Their chemical shifts are gathered in table 1. Quantitative MRS spectra analysis has allowed obtaining statistical correlation between different metabolite/H_2O ratios and liver fibrosis stages. A direct relationship between the four liver fibrosis stages and two metabolite ratios has been observed: A/H_2O ratio (A = -CH_3 lipid) and E/H_2O (E = peak including glucose, lipids -CH=CHCH$_2$- and glycogen) with statistical values of Kruskal-Wallis of $\chi^2_{(3)}$=8.134, p=0.043 and $\chi^2_{(3)}$=10.766, p=0.013, respectively. A/H_2O ratio Tukey post-hoc test indicated that the statistical differences were observed between patients with third stage of fibrosis and those with first stage (p=0.006), second stage (p=0.001) and fourth stage (p=0.001) with highest A/H_2O values for the third stage of fibrosis. Differences in E/H_2O ratio was observed between third stage patients and those with second (p=0.042) and fourth stage of fibrosis (p=0.002), again with highest E/H_2O values of this ratio for third stage. Interesting metabolic differences, although without statistical significance, were also observed in the peak integrated by Choline components and TMAO relative to water signal (D/H_2O ratio) with the highest value to third stage of fibrosis.

Conclusion: Interesting and significant differences were found between stages of fibrosis and specific metabolic profiles by in vivo ^{1}H MRS. The metabolic alterations observed may be associated to the fact that the affected liver shows stromal collapse and fibrosis of many portal triads with distortion of lobular architectures, including hemodynamic derangement, to result in disturbed excretion of the intracellular metabolites. The in vivo liver metabolic profiles and their correlation with fibrosis stages may increase the clinical application of ^{1}H MRS adding non-invasively quantitative metabolic information relevant to liver pathology.

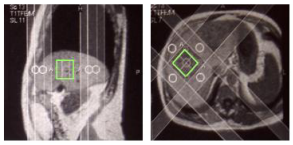

Figure 1: Volume of interest at the liver (45x45x45 mm).

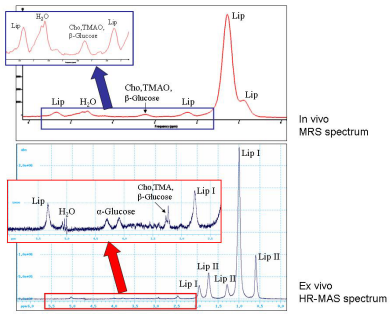

Figure 2: In vivo and ex vivo spectra in a liver of patient with chronic hepatitis. HR-MAS spectrum helped in the assignation of in vivo resonances.

Table 1. In vivo^{1}H MRS liver chemical shifts and metabolite assignment and identification.

Metabolite type	chemical group	chemical shift range (ppm)
Lipids (A)	-CH_3	0,90 (0,8-1,1)
Lipids (B)	(-CH_2-)n	1,20 (1,1-1,5)
Lipids (C)	CH_2=CH-CH_2-	2,15 (2,0-2,2)
Cho components and TMAO (D)	-CH_3	3,20
Glucose (E)	C1H	5,22
Lipids (E)	-CH=CHCH_2-	5,33
Glycogen (E)	C1H	5,42

Reference: [1] B. Martínez-Granados, D. Monleón, M.C. Martínez-Bisbal, J.M. Rodrigo, J. del Olmo, P. Lluch, A. Ferrández, L. Martí-Bonmatí, B. Celda "Metabolite identification in human liver needle biopsies by high-resolution magic angle spinning 1H NMR spectroscopy" NMR in biomedicine 2006; 19:90-100

Acknowledgements: eTUMOUR (contract no. FP6-2002-LIFESCIHEALTH 503094), Healthagents (contract no. FP6-2005-IST 027214). This work was also partially funded by National Grants from the Science and Education Ministery (SAF2004-06297, SAF2004-20971-E and SAF2005-23986-E) and Health Ministery (ISCIII2003-G03/185 and ISCIII2005-PI2695) of the Spanish Government and by ADIRM.