Applications of magnetic resonance spectroscopy for noninvasive assessment of hepatic steatosis
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HEPATIC UNSATURATED FATTY ACIDS IN PATIENTS WITH NONALCOHOLIC FATTY LIVER DISEASE ASSESSED BY 3.0T MR SPECTROSCOPY

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ABSTRACT

Purpose:
Nonalcoholic fatty liver disease (NAFLD) is related to the metabolic syndrome and obesity. Proton Magnetic Resonance Spectroscopy ($^1$H-MRS) is a noninvasive technique to assess hepatic triglyceride content (HTGC) and allows assessment of unsaturated fatty acids (UFA). There is increasing evidence that hepatic UFA are associated with the development of NAFLD. Therefore the objective of this study was to assess hepatic UFA in patients with NAFLD by using $^1$H-MRS.

Materials and Methods:
We included consecutive patients with deranged liver enzymes, with and without type 2 diabetes mellitus (DM2), suspected for NAFLD. Liver function and metabolic parameters were assessed. $^1$H-MRS measurements were performed at 3.0 Tesla. From the $^1$H-MR spectra two ratios were calculated: Ratio 1 (UFA); unsaturated fatty acid peak vs. reference water peak and ratio 2 (HTGC); total fatty acid peak vs. reference water peak.

Results:
Twenty-six patients were included. In these patients hepatic UFA (ratio 1) correlated with AST/ALT ratio (r=-0.46, p=0.02), glucose levels (r=0.46, p=0.018), HOMA-IR (r= 0.59, p=0.004) and HTGC (r=0.81, p<0.001). In diabetic patients (n=12) hepatic UFA correlated with alkaline phosphatase levels (r=0.72, p=0.01), HOMA-IR (r=0.73, p=0.01) and HTGC (r=0.83, p=0.002). Compared to non-diabetic patients with NAFLD, hepatic UFA levels were increased in patients with DM2 and NAFLD (0.032 vs. 0.014, p=0.03).

Conclusion:
Hepatic UFA can be assessed with $^1$H-MRS. $^1$H-MRS determined hepatic UFA correlate with clinical and metabolic parameters associated with NAFLD. Hepatic UFA are increased in patients with DM2. This study provides evidence for the use of noninvasive $^1$H-MRS to assess hepatic UFA in vivo.
Chapter 8: 3.0T $^1$H-MRS to assess unsaturated fatty acids in NAFLD

INTRODUCTION

Fatty liver disease is characterized by lipid accumulation in the liver. Causes are obesity, diabetes and dyslipidemia (metabolic syndrome), excessive alcohol consumption, and a variety of drugs and toxins [1]. Nonalcoholic fatty liver disease (NAFLD) includes a spectrum of diseases ranging from simple steatosis through steatohepatitis (NASH) to end-stage liver disease (cirrhosis). Insulin resistance, the metabolic syndrome and obesity are all strongly related to NAFLD. Due to the widespread obesity epidemic [1], NAFLD is recognized as the most common chronic liver disease in the Western world (estimated prevalence of 20-30%). The prevalence of NASH is estimated to be approximately 20% among the obese and 3% of the lean population [2-4]. Furthermore, it is estimated that 20-30% of patients with NASH will develop cirrhosis [5-8]. Consequently, the number of patients with NAFLD-associated cirrhosis who are in need for liver transplantation is increasing [9,10]. Together with increased cardiovascular morbidity and mortality [11], NAFLD is becoming a large burden on health care systems and the economy in general.

Percutaneous liver biopsy is the reference standard for grading hepatic steatosis, inflammation (NASH) and fibrosis. Unfortunately, due to the risk of complications, interobserver variability, sampling error and patients’ discomfort, this procedure is not applicable for population screening [12-14]. Noninvasive imaging techniques like abdominal ultrasonography and computed tomography lack sensitivity and specificity and are incapable of differentiating between simple steatosis and NASH [15-22]. Also conventional magnetic resonance imaging, considered as an accurate noninvasive technique, can not discriminate between simple steatosis and NASH [21,22].

In vivo proton magnetic resonance spectroscopy ($^1$H-MRS) is a safe, noninvasive diagnostic tool which has proven to be a sensitive and reproducible method to quantify hepatic triglyceride content (HTGC) in situ [23,24]. $^1$H-MRS is nowadays considered as the noninvasive reference standard for liver fat quantification [25-27]. It enables the quantitative evaluation of hepatic lipid content by measuring the nuclear chemical shift generated by hydrogen atoms of fatty acid chains in a magnetic field. The various fatty acids can be differentiated by the small shifts in nuclear resonance of the hydrogen atoms bound to different structured carbon chains. This technique has shown to correlate well with histological data from liver biopsies in healthy individuals and patients with hepatic steatosis ($r=0.9$, $p<0.001$) [28-30]. The largest study determining the prevalence of hepatic steatosis with $^1$H-MRS was performed by Szczepaniak et al. [9].
In this population of 2287 individuals over 30% was found to have some degree of hepatic steatosis. $^1$H-MRS is also suitable for grading and following up patients with NAFLD who participate in clinical trails [31]. Clinical experience with this technique is limited to 1.5 Tesla magnets [29-32]. There is sparse literature on addressing hepatic steatosis in humans at higher magnetic field strengths (3.0 Tesla or higher) [24,33,34].

Due to the higher spectral resolution, the use of increased magnetic fields allow quantification of various fatty acid components in more detail, e.g. differentiation of resonances from unsaturated fatty acid chains (UFA) and polyunsaturated fatty acids (PUFA) in steatotic liver triglycerides [35-38]. There is increasing evidence that hepatic UFA play an important role in the development of NAFLD. More precise, a decrease in omega-3 / omega-6 UFA results in uncontrolled hepatic lipid metabolism [39-44]. Omega-6 UFA are precursors to pro-inflammatory eicosanoids. The excessive caloric intake of current Western dietary behaviour is characterized by an increased intake of omega-6 UFA. This may be directly related to the development of nonalcoholic steatohepatitis and progression to NASH. Therefore, the objective of this study was the feasibility of hepatic UFA assessment in relation to HTGC by 3.0 Tesla $^1$H-MRS in patients suspected for NAFLD.

**MATERIAL AND METHODS**

**Study design**

This study is a single center, non-randomized pilot study in consecutive patients with well defined features consistent with NAFLD and ultrasonography suspected hepatic steatosis. This study was approved by the Medical Ethics Committee of our institution. All participants gave written informed consent.

**Patients**

Consecutive patients with deranged liver enzymes (defined as plasma alanine aminotransferase (ALT) > 1.5 x upper limit of normal) referred to our outpatient clinic, received a standardized workup. By exclusion of other causes of chronic liver diseases (negative viral (hepatitis B and C) and autoimmune (antinuclear antibodies and smooth muscle antigens) serology), alcoholic fatty liver disease (alcohol use not exceeding 140 g per week for males, 70 g per week for females) and normal iron and copper studies, the diagnosis of NAFLD was strongly suspected. Other exclusion criteria in this study were MRI related: pacemakers, pregnancy or a history of claustrophobia.
Medical history and physical examination focussed on features of the metabolic syndrome (e.g. body mass index (BMI) and waist-to-hip-ratio (WHR)) and signs of chronic liver disease were collected. BMI >27 kg/m$^2$ indicated overweight; BMI >30 kg/m$^2$ indicated obesity. In male, a WHR >1.0 and in female, a WHR >0.9 indicates increased risk for cardiovascular disease and insulin resistance [45]. Patients were divided into two groups according to the presence or absence of type 2 diabetes mellitus (DM2) as defined by international criteria [46].

In all included patients blood sampling and $^1$H-MRS was performed. Liver function (protrombin time, albumin and AST/ALT ratio [47]) and metabolic parameters (homeostasis model of assessment (HOMA-IR), glucose, insulin and lipid profile) were measured. Insulin resistance was defined as HOMA-IR >2.5.

**MR spectroscopy**

All measurements were performed in supine position on a 3.0T MRI scanner (Philips Intera, Philips Healthcare, Best, the Netherlands). A voxel of 20 x 20 x 20 mm was positioned in the right hepatic lobe (liver segments V - VIII by Couinaud), avoiding the diaphragm and edges of the liver, but also vascular and biliary structures (Figure 1). Voxel size and time for acquisition were standardized for all subjects. Spectra were acquired using a PRESS sequence with TE/TR=35/2000 msec and 64 signal acquisitions during free breathing without water-supression. Quality of $^1$H-MRS measurements was inspected visually, and when an experienced MR physicist was unsatisfied with the $^1$H-MR spectrum, the acquisition was repeated.

We evaluated the liver $^1$H-MR spectra by using jMRUI software. jMRUI is a graphical interface to several programs for processing magnetic resonance spectra and is specially designed for dealing with in-vivo MR spectra obtained in clinical MR spectrometers. Peak areas of the spectra were integrated using AMARES quantification technique. AMARES is a quantification method that estimates parameters for spectra by fitting them in the time domain. AMARES allows the inclusion of prior knowledge about the signal parameters of the model function [48]. Three peaks (at 5.4, 4.65 and 1.3 ppm) were analyzed and prior knowledge was used for all peak localizations by using soft constraints. In some cases constraints were used for peak bandwidths. All peaks were fitted using Lorentzian lineshapes. Phase variation was allowed (up to 40 degrees) around a manually found optimum.
Chapter 8: 3.0T ¹H-MRS to assess unsaturated fatty acids in NAFLD

From the ¹H-MR spectra two ratios were calculated (Table 1) and defined as:

Ratio 1: the unsaturated fatty acid peak area (methene) at 5.4 ppm versus the reference peak area of water at 4.65 ppm plus the unsaturated fatty acid peak area at 5.4 ppm. This represents total hepatic UFA.

Ratio 2: the total fatty acid peak area (methylene) at 1.3 ppm versus the reference peak area of water at 4.65 ppm plus the total fatty acid peak area at 1.3 ppm. Calculated peak areas of water and total fat were corrected for T2 relaxation (T2 water= 34msec, T2 fat= 68msec) [49]. HTGC was calculated according to methods described by Szczepaniak.

Normal hepatic fat content was defined as less than 5.6% measured by ¹H-MRS [9]. Total scanning time including positioning of the patient and acquisition of localizers amounted to 40 minutes.

Table 1: Calculated peak area ratios from ¹H-MR spectra

<table>
<thead>
<tr>
<th>Fatty acid component</th>
<th>Peak area ratio</th>
<th>index</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total unsaturated fatty acids (−CH=CH−) / (−CH=CH− + H₂O)</td>
<td>5.4/(5.4 + 4.65) ppm</td>
<td>Ratio 1</td>
</tr>
<tr>
<td>Hepatic triglyceride content (CH₂)n / ((CH₂)n + H₂O)</td>
<td>1.3/(1.3 + 4.65) ppm</td>
<td>Ratio 2</td>
</tr>
</tbody>
</table>

Statistical analysis

As this is a pilot exploratory study no formal sample size calculations could be carried out. The aim was to study the feasibility of hepatic UFA assessment in relation to HTGC with ¹H-MRS and correlate the measurements with clinical and biochemical data. To study correlations non-parametric Spearman correlation coefficients were used. For differences between groups non-parametric test for unrelated samples was used (Mann Whitney U analysis). A p-value <0.05 was considered significant. For statistical analysis SPSS (version 16.0, SPSS Inc, Chicago, Ill, USA) was used.
RESULTS

Patients suspected for NAFLD

Twenty-six patients were included (15 male and 11 female) with a mean age of 50.4 years (range 27 - 69). Of these patients, 12 had characteristics consistent with DM2. A history of DM2 was present for at least three years. Demographic data are shown in table 2. Mean body mass index (BMI) was 30.9 kg/m$^2$ (range 22.2 - 38.9). Twelve patients had a BMI of 30 kg/m$^2$ or less, whereas 14 patients had a BMI higher than 30 kg/m$^2$. Mean WHR in male patients was 1.06 (range 1.03 - 1.1) and in female 0.99 (range 0.9 - 1.1). As expected, patients with DM2 had higher HOMA-IR and thus were significantly more insulin resistant than those without DM2, and derangements of liver enzymes were comparable between both groups (Table 2).

![Figure 2: Three different spectra with increasing degree of steatosis. In spectrum A no UFA are visible at 5.4 ppm. In spectra B and C the amount of UFA is increasing and could be reliably quantified using the AMARES routine in jRMUI. In the upper panels the residues are shown for the peaks at 5.4, 4.65 and 1.3 ppm. The maximum of all spectra is scaled to 1.](image-url)
Chapter 8: 3.0T ¹H-MRS to assess unsaturated fatty acids in NAFLD

All ¹H-MRS measurements were of sufficient quality for analysis. Mean HTGC measured by ¹H-MRS in the entire group was 13.4% (range 3.3% – 29.7%). Eight patients had hepatic fat weight defined as less than 5.6% [9]. Sixty-nine percent (n=18) of patients had ¹H-MRS-proven NAFLD by the exclusion of other causes for chronic liver disease.

Figure 2 shows an example of three jMRUI fitted MR spectra. On the left a spectrum of a patient with 3.3% HTGC is shown. There is no peak visible at 5.4 ppm (hepatic UFA). As the amount of HTGC increases in the middle and far right spectrum, the 5.4 ppm peak representing hepatic UFA becomes visible and increases. In 24 out of 26 patients (92.3%) in this study the 5.4 ppm peak is detectable and used for quantification. All spectra acquired were of sufficient quality to assess hepatic UFA in vivo.

Hepatic UFA (ratio 1) showed a statistically significant correlation with the serum AST/ALT ratio (r=-0.46, p=0.02), HOMA-IR and plasma glucose levels (r=0.59, p=0.004 and r=0.46, p=0.018, resp.), (Figure 3, Table 3). There was a statistically significant correlation between hepatic UFA (ratio 1) and HTGC (ratio 2) (r=0.81, p<0.001).

### Table 2: Demographics, BMI and laboratory findings

<table>
<thead>
<tr>
<th>Parameters</th>
<th>DM2 (n= 12)</th>
<th>NO DM2 (n= 14)</th>
<th>p-value</th>
<th>Total (n= 26)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, yrs</td>
<td>52.1 ± 12.1</td>
<td>48.9 ± 9.5</td>
<td>ns</td>
<td>50.4 ± 10.7</td>
</tr>
<tr>
<td>Gender, male/female</td>
<td>08-apr</td>
<td>07-jul</td>
<td>ns</td>
<td>15-nov</td>
</tr>
<tr>
<td>BMI*, kg/m²</td>
<td>31.1 ± 4.5</td>
<td>30.8 ± 5.3</td>
<td>ns</td>
<td>30.9 ± 4.8</td>
</tr>
<tr>
<td>AST, U/L</td>
<td>47.9 ± 25.9</td>
<td>49.6 ± 27.2</td>
<td>ns</td>
<td>48.9 ± 26.2</td>
</tr>
<tr>
<td>ALT, U/L</td>
<td>75.5 ± 36.7</td>
<td>67.4 ± 39.6</td>
<td>ns</td>
<td>71.1 ± 37.7</td>
</tr>
<tr>
<td>AP, U/L</td>
<td>79.5 ± 27.7</td>
<td>86.8 ± 27.2</td>
<td>ns</td>
<td>83.3 ± 27.1</td>
</tr>
<tr>
<td>GGT, U/L</td>
<td>70.7 ± 37.5</td>
<td>97.1 ± 70.5</td>
<td>ns</td>
<td>84.9 ± 58.2</td>
</tr>
<tr>
<td>Glucose, mmol/L</td>
<td>7.4 ± 3.0</td>
<td>5.5 ± 0.7</td>
<td>ns</td>
<td>6.3 ± 2.2</td>
</tr>
<tr>
<td>HOMA-IR*</td>
<td>8.1 ± 7.0</td>
<td>5.2 ± 2.3</td>
<td>p= 0.05</td>
<td>6.6 ± 5.3</td>
</tr>
<tr>
<td>Cholesterol, mmol/L</td>
<td>4.5 ± 0.7</td>
<td>5.0 ± 1.4</td>
<td>ns</td>
<td>4.7 ± 1.1</td>
</tr>
<tr>
<td>LDL, mmol/L</td>
<td>2.5 ± 0.8</td>
<td>3.1 ± 1.2</td>
<td>p=0.02</td>
<td>2.8 ± 1.0</td>
</tr>
<tr>
<td>TG, mmol/L</td>
<td>1.6 ± 0.8</td>
<td>1.4 ± 0.7</td>
<td>ns</td>
<td>1.5 ± 0.7</td>
</tr>
</tbody>
</table>

*BMI = body mass index
HOMA-IR = homeostasis model of assessment
ns = not statistical significant
Chapter 8: 3.0T $^1$H-MRS to assess unsaturated fatty acids in NAFLD

Figure 3: Correlation between UFA (ratio 1) and insulin resistance (HOMA-IR) in all patients.

Table 3: Unsaturated fatty acids (UFA, ratio 1) and total hepatic fat content (HTGC, ratio 2); correlation with clinical and metabolic parameters

<table>
<thead>
<tr>
<th>Parameters</th>
<th>$^1$H-MRS Ratio 1 (UFA)</th>
<th>$^1$H-MRS Ratio 2 (HTGC)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total, n= 26</td>
<td>r = 0.48, p= 0.023</td>
<td>r = 0.43, p= 0.038</td>
</tr>
<tr>
<td>Glucose</td>
<td>r = 0.59, p= 0.004</td>
<td>ns</td>
</tr>
<tr>
<td>HOMA-IR</td>
<td>r =-0.46, p= 0.024</td>
<td>ns</td>
</tr>
<tr>
<td>AST/ ALT ratio</td>
<td>r = 0.81, p&lt; 0.001</td>
<td>ns</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>With DM II, n= 12</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>AF</td>
<td>r = 0.72, p= 0.013</td>
<td>r = 0.76, p= 0.004</td>
</tr>
<tr>
<td>HOMA-IR</td>
<td>r = 0.73, p= 0.011</td>
<td>ns</td>
</tr>
<tr>
<td>HTGC, Ratio 2</td>
<td>r = 0.83, p= 0.002</td>
<td>ns</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Without DM II, n= 14</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>HDL</td>
<td>r = 0.71, p= 0.010</td>
<td>ns</td>
</tr>
<tr>
<td>HTGC, Ratio 2</td>
<td>r = 0.82, p= 0.010</td>
<td>ns</td>
</tr>
</tbody>
</table>
Patients with and without DM2

Patients with and without DM2 had an average age of respectively 52.1 years (range 31 – 69) and 48.9 years (range 27 - 61) and a BMI of respectively 30.8 kg/m$^2$ (range 22.2 – 38.9) and 31.1 kg/m$^2$ (range 23.4 – 37.7). In patients without DM2 hepatic UFA (ratio 1) showed significant correlation with serum high density lipoprotein (HDL) cholesterol concentration ($r=0.71$, $p=0.010$) and HTGC (ratio 2) ($r=0.82$, $p=0.010$). In patients with DM2 hepatic UFA (ratio 1) showed significant correlation with serum alkaline phosphatase ($r=0.72$, $p=0.01$) and HOMA-IR ($r=0.73$, $p=0.01$). HTGC (ratio 2) correlated with serum alkaline phosphatase ($r=0.76$, $p=0.004$) and hepatic UFA ($r=0.83$, $p=0.002$), (Figure 4). Ratio 1, representing hepatic UFA, was significantly higher in patients with DM2 than in those without DM2 (0.032 vs. 0.014, $p=0.030$), (Figure 5). HTCG (ratio 2) was not different in both groups (0.47 vs. 0.37, $p=0.237$).

![Figure 4: Correlation between UFA (ratio 1) and HTGC (ratio 2) in patients with DM2.](image)

![Figure 5: Differences in $1H$-MRS measured total hepatic UFA in patients with and without DM2.](image)
DISCUSSION

In this study we investigated total hepatic UFA by 3.0T \(^1\)H-MRS in diabetic and non-diabetic patients with NAFLD and its correlations with clinical and metabolic parameters. Previous studies using noninvasive \(^1\)H-MRS measurements in NAFLD demonstrated significant correlations between HTGC and insulin resistance, diabetes and liver histology [29,30,50,51]. Other studies focussed on UFA in adipose tissue, marrow fat and skeletal muscle using 7.0 and 3.0 Tesla \(^1\)H-MRS [36,37]. Corbin et al. investigated hepatic UFA in a murine NAFLD model using \(^1\)H-MRS at 4.7 Tesla [35]. However, the assessment of hepatic UFA in humans at 3.0 Tesla has not been investigated yet.

Fatty acids in general; Fatty acids are carboxylic acids with a long aliphatic tail which are either saturated or unsaturated. Depending on one or more double bonds, UFA can be separated into mono (MUFA) - and polyunsaturated (PUFA) fatty acids. Omega-3, or n-3 PUFA, are UFA with a double bond at the third carbon-carbon bond from the terminal methyl end (n) of the carbon chain. They are important in human nutrition, e.g. \(\alpha\)-linolenic acid. UFA with a double bond at the sixth carbon-carbon bond from the end of the chain, are named omega-6, or n-6 PUFA. These are precursors of pro-inflammatory eicosanoids, n-6 prostaglandines and n-6 leukotrienes.

Hepatic UFA and NAFLD/NASH; 3.0T \(^1\)H-MRS is able to measure total hepatic UFA (i.e. the combination of MUFA and PUFA) at 5.4 ppm and total fatty acids (unsaturated and saturated fatty acids) at 1.3 ppm. In the liver, omega-3 and omega-6 PUFA exert opposite activities. Omega-3 PUFA activate peroxisome proliferator activated receptor-\(\alpha\) (PPAR-\(\alpha\)), a nuclear hormone receptor for the transcriptional regulation of genes responsible for fatty acid oxidation. Omega-3 PUFA cause a down-regulation of sterol regulatory element binding protein-1c (SREBP-1c), responsible for gene transcription in fatty acid synthesis [52]. Net result is a decrease of oxidative stress, in HTGC and inflammation. In contrast, omega-6 PUFA (e.g. arachidonic acid) are precursors of pro-inflammatory eicosanoids [53]. Hepatic UFA could be an important biomarker in this process because NASH induced in animal models show increased UFA [35]. Furthermore, in recent clinical trials omega-3 PUFA significantly improved metabolic and inflammatory markers in patients with NASH [42].

In this study, total hepatic UFA were measured with 3.0T \(^1\)H-MRS and correlations with clinical and metabolic parameters associated with NAFLD/NASH could be demonstrated, e.g. HOMA-IR, serum AST/ALT-ratio and total HTGC. Significant correlations were found between UFA and these parameters.
In DM2, ratio 1 representing total hepatic UFA, was significantly higher than in patients without DM2, whereas for total HTGC no differences were detected. From larger studies it is known that the presence of NASH is more frequent in patients with DM2 [54-56]. Hypothetically, this increase in UFA in patients with DM2 may be attributed to a decreased omega-3/omega-6 ratio with a substantial hepatic accumulation of omega-6 PUFA. Corbin et al. already showed that $^1$H-MRS can detect fat composition in the liver. In the steatotic liver hepatic fat content is sufficiently high enough for additional resonances like UFA at 5.4 ppm to become detectable. Our study supports these results, and show that UFA is also detectable in human livers at 3.0 Tesla. To our knowledge this is the first study assessing hepatic UFA by means of noninvasive 3.0T $^1$H-MRS in patients with NAFLD.

Limitations; As this was an exploratory pilot study to investigate the role of total hepatic UFA assessment, the number of included patients is small. No formal sample size calculation was carried out. We chose a sample size that was considered appropriate to address the study aims. We did not perform a reference standard liver biopsy due its invasiveness. This is the main limitation of this study. Diagnosis of NAFLD was made based on the combination of exclusion of other chronic liver diseases and the presence of enhanced echogenicity on hepatic ultrasonography.

Standardization and lifestyle control was not implemented in this study. We think this is not strictly necessary since standardization and lifestyle control can be implemented in many different ways. In daily clinical practice patients are not standardized as well, and no lifestyle control is performed.

In this study $^1$H-MRS was performed during free breathing. This is a potential limitation because the volume interrogated by $^1$H-MRS is blurred in the longitudinal direction by two to three centimetres respiratory excursions of the liver. In this study we did not expect major $^1$H-MRS acquisition problems caused by respiratory excursions. Voxels were carefully positioned in the right liver, avoiding the diaphragm by at least four centimetres. In this way $^1$H-MRS was always performed in liver tissue.

Finally, in this study differentiation between MUFA and PUFA and assessment of omega-3 and omega-6 PUFA individually was not possible using 3.0T $^1$H-MRS measurements.

In conclusion, 3.0T $^1$H-MRS represents a noninvasive technique to assess total hepatic UFA and HTGC. Hepatic UFA correlates well with clinical and metabolic parameters associated with the metabolic syndrome and NAFLD/NASH. Hepatic UFA are increased in patients with DM2, possibly due to an unequal increase of omega-6 PUFA. The presence of NASH is more frequent in patients with DM2.
Therefore hepatic UFA could be a predictive parameter to discriminate simple steatosis from NASH. This pilot study provides evidence for the use of 3.0T $^1$H-MRS as a noninvasive diagnostic tool to assess UFA in vivo in human livers. Further research in UFA and identification of omega-3 and omega-6 PUFA in order to diagnose patients at risk for NAFLD and to distinguish simple steatosis from NASH is needed.
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