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BRIEF ARTICLE

3.0 T proton magnetic resonance spectroscopy of the liver: Quantification of choline

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Abstract

AIM: To investigate the normal hepatic magnetic resonance spectroscopy findings choline/lipid2 (Cho/Lip2) associated with age and body mass index (BMI).

METHODS: A total of 58 single-voxel proton spectra of the liver were acquired at 3.0 T using the eight-channel phased array abdominal coil as the receiver coil. Consecutive stacks of breath-hold spectra were acquired using the point resolved spectroscopy technique at a short echo time of 30 ms and a repetition time of 1500 ms. The spectra were processed with the SAGE software package. Areas and heights for metabolite resonance were obtained. Student's *t* test for unpaired data was used for comparisons of shimming, Cho/Lip2, and lipid content.

RESULTS: There were significant negative correlations between the Cho/Lip2 peak height ratios and BMI (r = -0.615) and age (r = -0.398) (all $P < 0.01$). Compared with the high-BMI group, the low-BMI group was younger (39.1 ± 13.0 years *vs* 47.6 ± 8.5 years, t = -2.954, P = 0.005); had better water suppression (93.4% ± 1.4% *vs* 85.6% ± 11.6%, t = 2.741, P = 0.014); had higher Cho/Lip2 peak heights ratio (0.2 ± 0.14 *vs* 0.05 ± 0.04, t = 6.033, $P < 0.000$); and had lower lipid content (0.03 ± 0.08 *vs* 0.29 ± 0.31, t = -3.309, P = 0.004). Compared with the older group, the younger group had better shimming effects (17.1 ± 3.6 Hz *vs* 22.0 ± 6.8 Hz, t = -2.919, P = 0.008); higher Cho/Lip2 peak heights ratios (0.03 ± 0.05 *vs* 0.09 ± 0.12, t = 2.4, P = 0.020); and lower lipid content (0.05 ± 0.11 *vs* 0.23 ± 0.32, t = -2.337, P = 0.031). Compared with the low-choline peak group, the high-choline peak group had lower lipid content (0.005 ± 0.002 *vs* 0.13 ± 0.23, t = -3.796, $P < 0.000$); lower BMI (19.6 ± 2.4 *vs* 23.9 ± 3.0, t = -4.410, $P < 0.000$); and younger age (34.7 ± 10.0 years *vs* 43.2 ± 12.5 years, t = -2.088, P = 0.041).

CONCLUSION: Lipid accumulation could result from the increased fat in the body depending on age and BMI. Lipid can mask the resonance signal of choline.

Key words: Magnetic resonance spectroscopy; High-field imaging; Choline

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INTRODUCTION

Magnetic resonance spectroscopy (MRS) is a non-invasive technique that is being increasingly applied to describe biochemical changes in the liver[1-5]. The recent installation of higher field strength (3 T) clinical magnets with multicoil arrays offers new opportunities for performing whole-body MRS. The improved signal-to-noise ratio (SNR) can reduce acquisition times, and the higher field strength also provides better separation of resonances[6,7].

Choline is a precursor of acetylcholine and a component of the phospholipid metabolism of cell membranes. It is known that MRS may be used to diagnose malignancy; usually by measuring the choline peak. Absolute quantification of hepatic metabolite concentrations offers several advantages for the evaluation of *in vivo* MRS data. Unfortunately, absolute quantification of choline-containing compounds is impractical for most clinical applications. A few studies of *in vivo* MRS have reported an increase in choline-containing compounds relative to lipids within tumors such as hepatocellular carcinoma, and a reduction in the lipid-to-choline ratio after transarterial embolization for hepatocellular carcinoma. However, the ability to distinguish reliably benign and malignant tumors from normal liver parenchyma has yet to be established. A major limitation is the observation that relatively large amounts of choline-containing compounds may occur even in normal liver[8-13].

Knowledge of the normal findings associated with age and body mass index (BMI) is valuable. Therefore, the central questions of our study were: (1) does choline/lipid2 (Cho/Lip2) have a relationship with BMI? and (2) does Cho/Lip2 have a relationship with age? Finally, we try to explain why no observable choline peak was detected on liver MRS in obese and elderly individuals.

MATERIALS AND METHODS

Patients

The study was approved by our institutional review board, and written informed consent was obtained from all patients. We evaluated non-hepatic disease and fatty liver in 58 patients (29 men, 29 women; age range, 19-65 years; median age, 42 years) with no history of liver disease and with normal liver function test results. The mean height of the cohort was 164.4 ± 7.4 cm (range, 150-178 cm), mean body weight was 62.7 ± 11.3 kg (range, 39-93 kg), and mean BMI was 23.1 ± 3.3 (range, 16.9-30.4).

MRS protocol

MRS was performed using a GE Signa 3.0 T whole-body system (GE Signa Excite HD; GE Medical Systems, Milwaukee, WI, United States) with the standard proton MRS acquisition software provided by the manufacturer. The body coil was used as the transmitter, and a torso phased-array coil (eight coils, four anterior and four posterior coils; Waukesha, WI, United States) was used as the receiver. Single-volume spin-echo point-resolved spectroscopy was used with parameters of 1500/30/64 [repetition time (TR)/echo time (TE)/excitations] in all patients. The patients entered the magnetic field in the supine position with their feet first. The total acquisition time (2 min 12 s per scan) was split into consecutive blocks to match the length of a breath-hold period of about 15-40 s, while data acquisition was performed at end expiration. Volume of interest of 2 cm × 2 cm × 2 cm was localized in the middle portion of the right hepatic lobe, on the basis of T2-weighted single-shot fast spin-echo pictures in the transverse planes with a TR of 935 ms, TE of 83 ms, a matrix of 288 × 192, field of view of 40 cm, section thickness of 8 mm, an intersection gap of 1.5 mm, and a number of excitations of 0.62. The voxel was localized in such a way as to avoid large vessels, bile ducts, and fatty tissue (Figure 1).

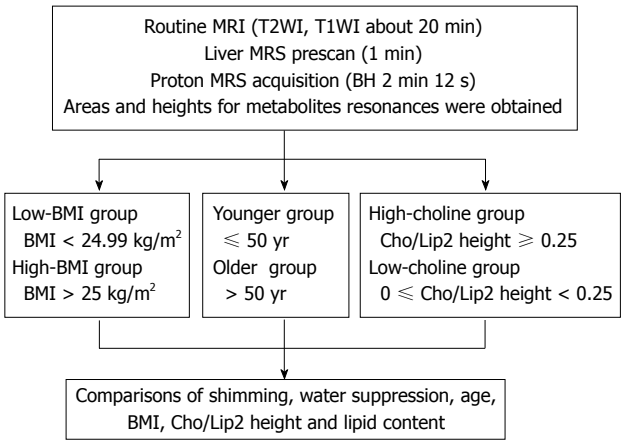

Figure 1 Detailed scanning protocols and statistical analysis. BMI: Body mass index; MRS: Magnetic resonance spectroscopy; Cho/Lip2: Choline/lipid2; MRI: Magnetic resonance imaging.

After acquisition, data were processed by using the MR spectroscopic analysis package provided by the manufacturer (SAGE 7.1; GE Medical Systems). The raw data were zero-filled once, apodized with a 5-Hz Gaussian filter, Fourier transformed, and phase and baseline corrected. Marquardt curve fitting was performed by using a Gaussian line shape to calculate the area under the peak. MR spectroscopic data were analyzed by a single medical physicist (Xu L) with > 7 years experience in MRS analysis. For each MRS measurement of unsuppressed water, we normalized the amplitude of the lipid signal to the sum of the lipid plus water signals to obtain the percentage lipid within the liver.

For all data acquisition, water suppression was performed using a series of three chemical-shift-selective pulses with predefined flip angles to leave a significant amount of residual water in the spectrum, and high-order

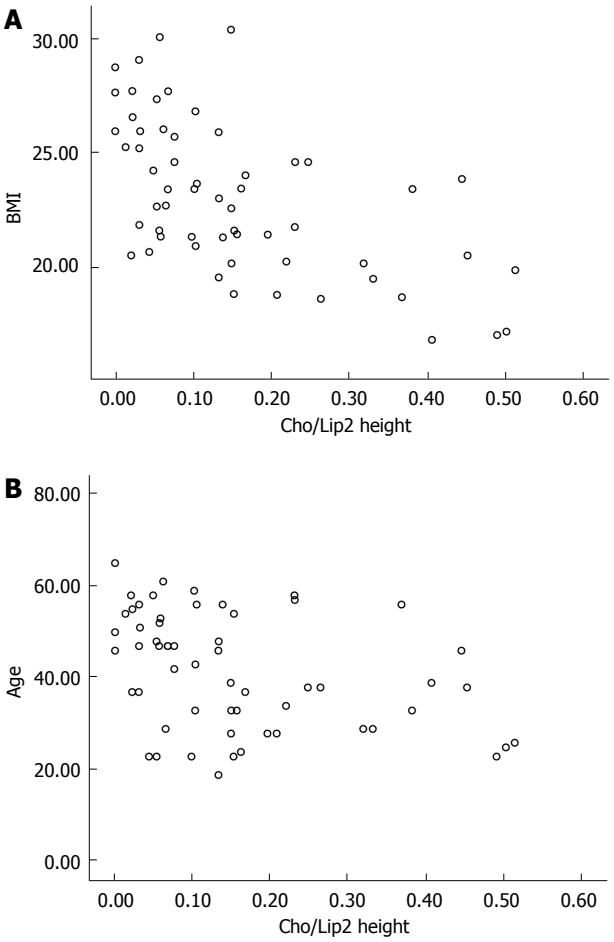

Figure 2 Scatter plots of relationships. A: Between choline/lipid2 (Cho/Lip2) and body mass index (BMI), a good inverse correlation was observed; B: Between Cho/Lip2 and age, a moderate inverse correlation was observed.

shimming followed by automatic local shimming adjustment was used. Line widths (full-width half-maximum) and water suppression were obtained.

Statistical analysis

The study group was divided into two subgroups: low BMI (< 24.99 kg/m^2) and high BMI group (> 25 kg/m^2). The participants were divided into two subgroups according to age: younger (≤ 50 years) and older (> 50 years). The participants were divided into two subgroups based on the Cho/Lip2 ratio: high choline (Cho/Lip2 ≥ 0.25) and low/non-choline (0 ≤ Cho/Lip2 < 0.25) (Figure 1).

For all tests, $P < 0.05$ was considered to indicate statistically significant differences. Statistical analysis was performed using SPSS version 10.0.1 (SPSS, Chicago, IL, United States). Using Spearman's correlation, we determined the relationship between Cho/Lip2 and BMI, and between Cho/Lip2 and age.

Student's *t* test for unpaired data was used for comparison of shimming, Cho/Lip2 and lipid content between the low-BMI and high-BMI groups, and between the younger and older groups. Student's *t* test for unpaired data was also used for comparison of shimming, BMI and age between the high-choline and non-choline groups.

RESULTS

Scatter plots and Spearman's correlations

The scatter plots were used to reveal the relationships between the variables (Figure 2). There were significant negative correlations between the Cho/Lip2 peak heights ratios and BMI ($r = -0.615$, $P < 0.000$), and age ($r = -0.398$, $P = 0.002$).

Comparison between low-BMI and high-BMI group

Compared with the high-BMI group, the low-BMI group was younger (39.1 ± 13.0 years *vs* 47.6 ± 8.5 years, $t = -2.954$, $P = 0.005$); had better water suppression (93.4% ± 1.4% *vs* 85.6% ± 11.6%, $t = 2.741$, $P = 0.014$); had higher Cho/Lip2 peak heights ratios (0.20 ± 0.14 *vs* 0.05 ± 0.04, $t = 6.033$, $P < 0.000$); and had lower lipid content (0.03 ± 0.08 *vs* 0.29 ± 0.31, $t = -3.309$, $P = 0.004$) (Figure 3).

Comparison between younger and older groups

Compared with the older group, the younger group had better shimming effects (17.1 ± 3.6 Hz *vs* 22.0 ± 6.8 Hz, $t = -2.919$, $P = 0.008$); higher Cho/Lip2 peak heights ratios (0.03 ± 0.05 *vs* 0.09 ± 0.12, $t = 2.4$, $P = 0.020$); and lower lipid content (0.05 ± 0.11 *vs* 0.23 ± 0.32, $t = -2.337$, $P = 0.031$) (Figure 3).

Comparison between high-choline and low-choline groups

Compared with the low-choline group, the high-choline group had lower lipid content (0.005 ± 0.002 *vs* 0.13 ± 0.23, $t = -3.796$, $P < 0.000$); lower BMI (19.6 ± 2.4 *vs* 23.9 ± 3.0, $t = -4.410$, $P < 0.000$); and younger age (34.7 ± 10.0 years *vs* 43.2 ± 12.5 years, $t = -2.088$, $P = 0.041$) (Figure 4).

DISCUSSION

MRS at 3.0 T provides improved SNR and spectral resolution compared with 1.5 T MRI scanners, therefore, it is expected to yield more reliable measurements of metabolite concentrations[6,8,14]. The principal metabolite that has been targeted in focal liver disease is choline. In general, choline is elevated in tumors, because choline is a cell membrane component and increased cell turnover is associated with malignancy. *In vivo* ^{1}H MRS has proved valuable in the diagnosis of tumors in the brain, prostate, breast, and uterine cervix. ^{1}H MRS is also useful for evaluation of treatment responses in malignant tumors of the head and neck, as well as in breast cancer[1,13,15-17].

However, the ability to distinguish reliably benign and malignant tumors from normal liver parenchyma has yet to be established. According to the data from our study, the lipid content in liver parenchyma increased with both age and BMI; there were significant negative correlations between Cho/Lip2 and age; and there was no observ-

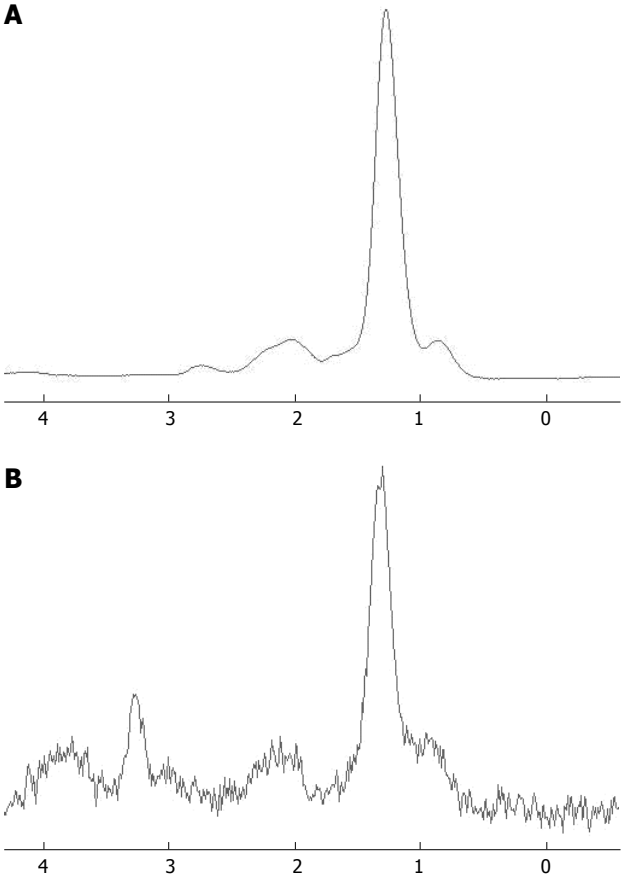

Figure 3 Point resolved spectroscopy-localized single voxel ^{1}H magnetic resonance spectrum. A: Originating from liver parenchyma of an obese and elderly (body mass index = 30.04, 52 years) volunteer. No observable choline peak was detected at 3.2 ppm; B: Originating from liver parenchyma of a lean and young volunteer (body mass index = 18.87, 23 years). High-choline peak was detected at 3.2 ppm.

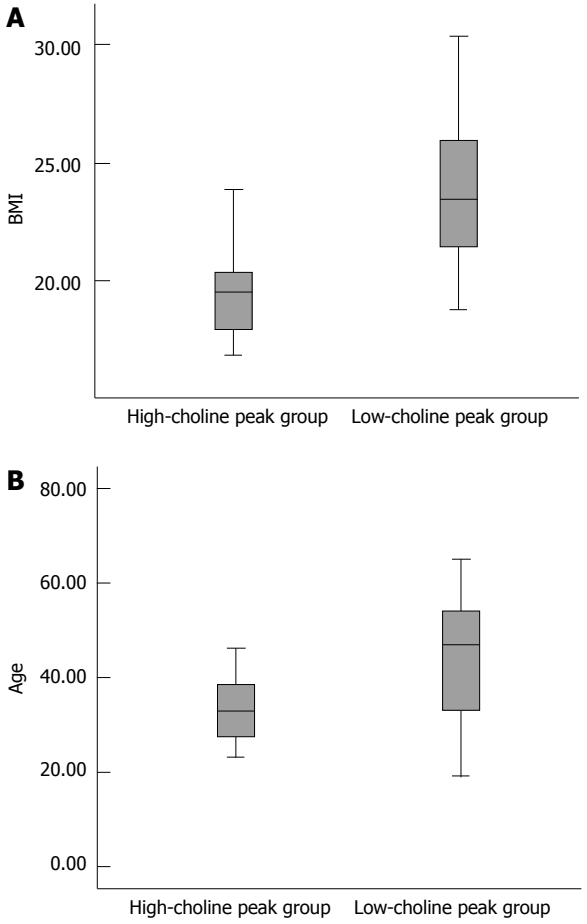

Figure 4 High-choline peak group. A: Had lower body mass index than the low-choline group; B: Was younger than the low-choline group. BMI: Body mass index.

able choline peak in obese and elderly individuals. Possible reasons for the above conclusions include: (1) the lipid content in liver parenchyma increased with both age and BMI; and (2) the choline level in liver parenchyma changed with both age and BMI.

The signals of the corresponding lipid groups can be observed at 1.3 ppm for (-CH2) n and 0.9 ppm for (-CH3), and between 2.0 and 2.3 ppm for (-CH=CH-CH2-), with signals of markedly lower intensity. In this study, the analysis of the spectra only focused on the concentrations of (-CH2) n. The lipid content in liver parenchyma increased with both age and BMI. This conclusion has been confirmed by Müller *et al*[1] and Fischbach *et al*[18]. Assuming that all of the metabolites that contribute to the choline peak remain constant, when the lipid volume fraction is high, the choline fraction should decrease in signal intensity. In other words, the hydrogen found in lipid can produce very strong resonance signals that can mask the resonance signal of lower concentration compounds of interest, usually metabolites such as choline[18-23].

Choline is a nutrient essential for normal function of all cells. It is a precursor not only for acetylcholine but also for phospholipids that are found in intracellular membranes and in the cell membrane. The liver parenchyma already contains large choline metabolite pools. Although catabolic and anabolic reactions are predominant in the liver, leading to elevated choline-containing compounds (CCC) levels. Aging and high BMI may decrease hepatic metabolism, resulting in lower choline concentration. Unfortunately, this conclusion has not been confirmed by previous studies. Fischbach *et al*[18] have acquired 113 spectra in normal-appearing parenchyma. The mean ± SD of the normalized measurements of CCC for the younger group (≤ 40 years) and the older group (> 40 years) was 8.14 ± 6.0 and 7.20 ± 4.32, respectively. The researchers found that no significant differences were observed.

The presence of a large lipid peak should in principle not influence the fitting of the choline peak. It is known that the concentration of lipid in human liver is 10-1000 times greater than that of most tissue metabolites such as choline. Consequently, the signal of lipid is dominant in ^{1}H-MRS in some cases. Experimental evidence suggests that spectroscopic data, or metabolic measurements, may be affected by a dominant lipid peak, which makes visualization of the metabolites of interest difficult. This is because the large lipid peak overlaps with adjacent small peaks, and scaling the signal intensity is difficult. This is

the reason why no observable choline peak was detected on liver MRS in obese and elderly individuals. To compensate, fat-suppression techniques are recommended for hepatic MRS to distinguish benign and malignant tumors from normal liver parenchyma[23-26].

This technique has its limitations in methodology. Although we used a 3.0 T MR imager and shorter TE to increase SNR, spectra containing only noise without any identifiable choline metabolite peaks still existed in a few cases. High-field MRI equipment and/or advanced techniques, such as nuclear Overhauser effect enhancement and proton decoupling, may demonstrate improved SNRs and spectral resolution between MRS peaks. The application of those new techniques may be necessary to answer this question.

In conclusion, Lipid accumulation in the liver could result from increased fat in the body, depending on age and BMI. Hydrogen found in subjects with lipid accumulation can produce very strong resonance signals that can mask the resonance signal of lower concentration compounds such as choline. This is the reason why no observable choline peak was detected on liver MRS in obese and elderly individuals. Fat suppression techniques are recommended for hepatic MRS to distinguish benign and malignant tumors from normal liver parenchyma.

COMMENTS

Background

It is known that magnetic resonance spectroscopy (MRS) can be used to diagnose malignancy; usually by measuring the choline peak. A major limitation is the observation that relatively large amounts of choline-containing compounds may occur even in normal liver. Knowledge of the normal findings associated with age and body mass index (BMI) is valuable.

Research frontiers

A few studies of *in vivo* MRS have reported an increase in choline-containing compounds relative to lipids within tumors such as hepatocellular carcinoma, and a reduction in the lipid-to-choline ratio after transarterial embolization for hepatocellular carcinoma. However, the ability to distinguish reliably benign and malignant tumors from normal liver parenchyma has yet to be established.

Innovations and breakthroughs

The ability to distinguish reliably benign and malignant tumors from normal liver parenchyma has yet to be established. According to the data from this study, the lipid content in liver parenchyma increased with both age and BMI, there was a significant negative correlation between choline/lipid2 (Cho/Lip2) and age, and there was no observable choline peak in obese and elderly individuals.

Applications

Fat suppression techniques are recommended for hepatic MRS to distinguish benign and malignant tumors from normal liver parenchyma.

Terminology

Breathholding either eliminated or markedly reduced phase and frequency shifts and outer voxel contamination that were associated with the motion of the abdomen during breathing.

Peer review

The authors investigated the normal hepatic MRS findings (Cho/Lip2) associated with age and BMI. The study was well designed, and this paper is important and interesting.

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