Comparison of $^1$H MR Spectroscopy, 3-point DIXON, and Multi-echo Gradient Echo for Measuring Hepatic Fat Fraction

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Purpose: We evaluated and compared the reproducibility, diagnostic accuracy, and sequence dependency of the fat fraction (FF) determined by 3-point Dixon (DIXON) and multi-echo gradient-echo (MEGE) methods to that of the FF determined by magnetic resonance spectroscopy (MRS).

Methods: Our study included 98 volunteers, ten of whom underwent scanning twice to evaluate sequence reproducibility. We compared the FFs determined by the DIXON and MEGE methods to that by MRS as the gold standard, calculated sensitivity and specificity for each image analysis method at a threshold value of 6.25% of spectroscopic value, and used Pearson’s correlation coefficient and Bland-Altman analysis to compare agreement among the repeated measurements and FF values with the 3 methods, in 98 volunteers.

Results: There was no significant difference in repeated scans in any sequence with Wilcoxon’s t-test. Each correlation coefficient ($r$) exceeded 0.930 for the repeated measurements of all 3 sequences. Sensitivity of DIXON was 82% and specificity, 96%; sensitivity of MEGE was 70% and specificity, 99%. The FFs determined by DIXON and MEGE correlated well with that by MRS ($r=0.920$) but showed significant difference (paired t-test, $P<0.001$). The mean difference between the FF determined by DIXON and that by MEGE were 0.93 and −1.16, respectively. The slope of the regression lines as determined by DIXON was $-0.655$ ($P<0.001$) and that by MEGE was $-0.527$ ($P<0.001$). When the FF by MRS was less than 6.25%, the FF values by DIXON and MEGE were significantly higher; when the spectroscopic value was greater than 6.25%, their values were significantly lower.

Conclusion: We demonstrated the high reproducibility of each FF measurement using MRS, DIXON, and MEGE. Compared to MRS, both DIXON and MEGE showed high sensitivity and specificity for determining FF. The FFs by DIXON and MEGE showed sequence dependency because DIXON had proportional and additional errors, and MEGE had a proportional error.

Keywords: DIXON, hepatic fat fraction, multi-echo gradient echo, $^1$H-MRS

Introduction

Non-alcoholic fatty liver disease (NAFLD) is a common chronic liver disease primarily caused by accumulation of triglycerides in hepatocytes. Fatty deposition in the liver has been associated with metabolic syndrome, and NAFLD can progress to nonalcoholic steatohepatitis (NASH), cirrhosis, and end-stage liver disease.¹,² NAFLD is currently assessed by histologic visualization of hepatocellular fat vacuoles,³,⁴ but biopsy is invasive and prone to sampling error.⁵,⁶ A noninvasive, objective, and reproducible assessment to replace biopsy is needed in clinical settings.

Ultrasonography (US), computed tomography (CT), and magnetic resonance (MR) imaging have been used to evaluate fatty liver.⁷–¹⁰ US does not allow quantitative assessment of fat fraction.¹¹ In addition, with US, the threshold of the detectable fat fraction (FF) for a fatty liver exceeds 30%, which may be insufficient to detect a mildly fatty liver.¹²
Multi-echo gradient echo (MEGE) can correct T2* in the case of a fatty liver with iron deposition, whereas MRS cannot give T2*-corrected FF values in the lack of significant medical histories. We then acquired an MEGE scan at the center of the location used for MRS.

Materials and Methods

Subjects
Our institutional review board approved the study design, and we obtained informed consent from all volunteers. The study included 98 randomly recruited healthy volunteers (80 men, 18 women; aged 21 to 60 years; body mass index [BMI], 23.3 ± 3.1 [range, 17.3 to 33.7]). None of the volunteers had been checked for NASH, NAFLD, or iron deposition because they had no significant medical histories.

Scans and data processing
All volunteers underwent 1H MR spectroscopy and MR imaging using a whole-body 1.5-tesla magnet (Achieva A-series, PHILIPS Medical Systems, Best, The Netherlands) equipped with a 16-channel XL-torso phased-array coil.

We first scanned the 3-plane T2-weighted image and DIXON and used those images to set the volume of interest (VOI) for the MRS scan we then performed, placing the VOI on the right hepatic lobe to avoid the portal vein, biliary structures, and inhomogeneous fat accumulation as much as possible. We then acquired an MEGE scan at the center of the location used for MRS.

To evaluate reproducibility, we randomly selected 10 volunteers (9 men, 1 woman; aged 29 to 53 years; BMI, 24.2 ± 1.8 [range, 22.3 to 27.6]) from the 98, to undergo repositioning and rescanning after the first scan. Two trained technologists (KI and SM, both with 3 years’ experience) performed MRS and MR imaging. Figure 1 shows the representative fat and water images of DIXON, FF map of MEGE, and MRS spectra.

1H MR spectroscopy
MRS was performed with a single-voxel, point-resolved spectroscopy sequence (PRESS) using parameters: repetition time (TR), 2000 ms; echo time (TE), 40 ms; voxel size, 30 × 30 × 30 mm; 8 averages; 2048 points over 2500 Hz; and scan time of 18 ms on a single breath-hold. We performed iterative shimming of the VOI in the automatic mode. After scanning the liver using the automatic fitting software on the console, we measured the signal integrals of fats (CH2 and CH3 at 1.3 ppm) and water (H2O at 4.7 ppm). Hepatic fat content was expressed as FF [Ifat/(Iwater + Ifat) × 100], where Ifat was the signal integral of fat and Iwater was the signal integral of water.

Three-point DIXON
We performed DIXON with a 3-dimensional (3D) fast-field echo sequence and parameters: TR, 6.2 ms; TE1/TE2/TE3, 1.6/3.2/4.8 ms; flip angle (FA), 10 degrees; matrix size, 128 × 95; field of view (FOV), 360 × 258 mm; pixel size, 2.8 × 2.7 mm; one average; slice thickness, 8 mm; scan range of global liver; and scan time, 18 ms on a single breath-hold. T2* was not corrected. Using the reconstructed fat and water image, one of the authors (KI) placed a region of interest (ROI) 30 × 30 mm at the location used for MRS, and the signal intensities of fat and water were measured. Hepatic fat content was expressed as FF [Sfat/(Swater + Sfat) × 100], where Sfat was the signal intensity of fat and Swater was the signal intensity of water.

Multi-echo gradient echo
We performed MEGE using a 2D fast-field echo sequence and parameters: TR, 48 ms; TEs, 2.3/4.6/6.9/9.2 ms; 4 echoes; FA, 20 degrees; matrix size, 128 × 128; FOV, 360 × 360 mm; pixel size, 2.8 × 2.7 mm; one average; slice thickness, 10 mm; single slice; and scan time, 20 ms on a single breath-hold. To correct the T2* value, we obtained 4 echoes at serial opposed-phase and in-phase echo times (2.3, 4.6, 6.9, 9.2 ms). We used PRIDE research software (PHILIPS Medical Systems, Best, The Netherlands) to obtain the fat fraction image and T2*-corrected FF. First, we calculated the initial values of the T2* decay-corrected water signal integral of water. We then performed 3-point Dixon (DIXON) can scan the whole liver in one breath-hold and detect inhomogeneous fat accumulation but is also unable to correct the T2* effect.

We investigated the reproducibility, diagnostic accuracy, and sequence dependency of the FFs determined by the DIXON and MEGE methods compared to that by MRS.
Fig. 1. A 34-year-old man. (A) Fat and (B) water image of DIXON; (C) fat fraction (FF) map of multi-echo gradient echo (MEGE); and (D) magnetic resonance (MR) spectra. FF value of DIXON was 13.1% in A and B. FF value of MEGE was 14.1% in C. FF value of MR spectroscopy (MRS) was 19.6% in D.

\[ S(TE) = \exp \left( -\frac{TE}{T_{2}^*} \right) \cdot \left( (S_{w0})^2 + (S_{f0})^2 + 2 \cdot S_{w0} \cdot S_{f0} \cdot \cos \Delta \omega \cdot TE \right)^{1/2}, \]  

where \( T_{2}^* \) is the \( T_{2}^* \) value of tissue, and \( \Delta \omega \) is the difference in resonance frequency of water and fat. In this model, the \( T_{2}^* \) of water and fat were compartmentalized, but it was too difficult to perform fitting. Therefore, the \( T_{2}^* \) decay terms of water and fat were conflated by fixing the phase differences of water and fat. We obtained a \( T_{2}^* \)-corrected FF image using \( S_{w0} \) and \( S_{f0} \) obtained by fitting. Hepatic fat content was expressed as FF \( \left[ S_{f0}/(S_{w0} + S_{f0}) \times 100 \right] \). Using ImageJ software (available at http://rsb.info.nih.gov/ij/), one of the authors (KI) placed an ROI of 30 \( \times \) 30 mm at the MRS location, and FF values were measured.

**Data analysis**

We summarized all continuous variables using means and standard deviations.

**Reproducibility**

Ten of the 98 volunteers were underwent scanning twice with all 3 methods, and we analyzed each data set for sequence reproducibility.

**Diagnostic accuracy**

In 98 volunteers, we calculated performance statistics (sensitivity, specificity) for DIXON and MEGE methods using a threshold value that was 6.25% of the spectroscopic FF. This value was equivalent to an FF of 5.56% by wet weight, which previous studies identified as a classification threshold for fatty liver.

**Sequence dependency**

In 98 volunteers, we compared the FF obtained by DIXON and MEGE methods to that by MRS as the gold standard.

**Statistical analysis**

We evaluated the degree of agreement between the 2 repeated studies in terms of the mean absolute difference, 95% confidence intervals (CI) of the mean difference, and slope of the regression line. We used Pearson's correlation coefficient and Bland-Altman analysis to compare agreement among the repeated studies and the FF values by the 3 methods. We tested statistical significance
using Wilcoxon's t-test for repeated studies and paired t-test for sequence dependency, with statistical significance defined as $P<0.05$.

**Results**

FF measurements of all 3 methods were fully implemented in all subjects. In 98 volunteers, the spectroscopic FF ranged from 0.0% to 32.5% (mean, 5.1%).

**Reproducibility**

Correlation coefficients that exceeded 0.93 ($P<0.001$) in 10 volunteers for each repeated measurement demonstrated the high reproducibility of each FF (Fig. 2). The Bland-Altman plots showed mean differences between the repeated studies with regard to FF measurements as $-1.27$ (MRS), $-0.16$ (DIXON), and $-0.06$ (MEGE); 95% confidence intervals of the mean difference as $-3.40$ to $0.82$ (MRS), $-1.85$ to $1.35$ (DIXON), and $-1.05$ to $0.65$ (MEGE); and slopes of the regression lines of 0.199 ($P=0.071$) (MRS), 0.171 ($P=0.212$) (DIXON), and $-0.054$ ($P=0.481$) (MEGE) (Fig. 3). Wilcoxon’s t-test revealed no significant difference between repeated scans in MRS ($P=0.193$), DIXON ($P=0.922$), and MEGE ($P=0.901$).

**Diagnostic accuracy**

Table shows performance statistics of DIXON and MEGE methods at the threshold value of 6.25% for spectroscopic FF. Sensitivity of DIXON was 82% and specificity, 96%; sensitivity of MEGE was 70% and specificity, 99%.

**Sequence dependency**

In 98 volunteers, the FFs determined by DIXON and MEGE correlated well with that by MRS ($r=0.920$, $P<0.001$) for each sequence (Fig. 4). The Bland-Altman plots showed that the mean differences between FFs determined by DIXON and MEGE, with regard to FF by MRS, were 0.93 and $-1.16$, respectively. The 95% confidence intervals of the mean difference between FFs determined by

![Graphs showing reproducibility of fat fraction (FF) by magnetic resonance spectroscopy (MRS), DIXON, and multi-echo gradient-echo (MEGE) methods.](image)
Fig. 3. Bland-Altman plots of the repeated studies by magnetic resonance spectroscopy (MRS) (a), DIXON (b), and multi-echo gradient-echo (MEGE) (c) methods. The mean differences between the repeated studies with each sequence were $-1.27$, $-0.16$ and $-0.06$, for MRS, DIXON and MEGE, respectively. The 95% confidence intervals of the mean difference were $-3.40$ to $0.82$, $-1.85$ to $1.35$, and $-1.05$ to $0.65$; and the slopes of the regression lines were $0.199$ ($P = 0.071$), $0.171$ ($P = 0.212$), and $-0.054$ ($P = 0.481$), for MRS, DIXON and MEGE, respectively.

Table. Diagnostic sensitivity and specificity of DIXON and multi-echo gradient-echo (MEGE) methods at a threshold value of 6.25% for spectroscopic fat fraction.

|     | DIXON       | MEGE       |
|-----|-------------|------------|
| Sensitivity | 0.815 (22/27) | 0.704 (19/27) |
| [0.620, 0.935] | [0.495, 0.870] |
| Specificity  | 0.958 (68/71) | 0.986 (70/71) |
| [0.880, 0.990] | [0.955, 1.000] |

Notes. We used numbers in parentheses to calculate sensitivities and specificities. Brackets enclose 95% confidence intervals.

DIXON and MRS were $0.80 \pm 2.30$ and those determined by MEGE and MRS were $1.30 \pm 0.05$. The slopes of the regression lines as determined by DIXON and MEGE, with regard to MRS, were $-0.655$ ($P<0.001$) and $-0.527$ ($P<0.001$), respectively (Fig. 5). Paired t-test revealed that FFs determined by DIXON and MEGE were significantly different from that by MRS ($P<0.001$).

When FFs exceeded 6.25% by MRS ($N=27$), strong correlation was observed between FFs by DIXON and MRS ($r=0.917$, $P<0.001$), and between FFs by MEGE and MRS ($r=0.949$, $P<0.001$). The Bland-Altman plots showed that the mean differences between FFs determined by DIXON and MEGE, with regard to FF by MRS, were $2.79$ and $0.47$, respectively. The 95% confidence intervals of the mean difference between FFs determined by DIXON and MRS were $2.40$ to $3.10$ and those determined by MEGE and MRS were $0.10$ to $0.75$. The slopes of the regression lines as determined by DIXON and MEGE, with regard to MRS, were $-0.344$ ($P = 0.027$) and $-0.343$ ($P = 0.017$), respectively. Paired t-test revealed that FFs by DIXON and MEGE were significantly different from FF by MRS ($P<0.001$).

When FFs exceeded 6.25% by MRS ($N=27$), strong correlation was observed between FFs by DIXON and MRS ($r=0.928$, $P<0.001$), and between FFs by MEGE and MRS ($r=0.936$, $P<0.001$). The Bland-Altman plots showed that the mean differences between FFs determined by DIXON and MEGE, with regard to MRS, were $-3.52$ and $-5.05$, respectively. The 95% confidence intervals of the mean difference between FFs determined by
**Fig. 4.** Graphs showing sequence dependency of DIXON (a) and multi-echo gradient-echo (MEGE) (b) methods compared to magnetic resonance spectroscopy (MRS). Correlation coefficients of fat fractions (FFs) determined by DIXON and MEGE with the FF determined by MRS exceeded 0.920 ($P < 0.001$) in 98 volunteers for each sequence.

**Fig. 5.** Bland-Altman plots of fat fraction (FF) by DIXON (a) and multi-echo gradient-echo (MEGE) (b) methods compared to that by magnetic resonance spectroscopy (MRS). The Bland-Altman plots showed that the mean differences between FFs determined by DIXON and MEGE, with regard to FF by MRS, were 0.93 and $-1.16$, respectively. The 95% confidence intervals of the mean difference between FFs determined by DIXON and MRS were $0.80$ to $2.30$ and those determined by MEGE and MRS were $-1.30$ to $-0.05$. The slopes of the regression lines as determined by DIXON and MEGE, with regard to MRS, were $-0.655$ ($P < 0.001$) and $-0.527$ ($P < 0.001$), respectively.

DIXON and MRS were $-5.55$ to $-2.20$ and those determined by MEGE and MRS were $-6.85$ to $-3.80$. The slopes of the regression lines as determined by DIXON and MEGE, with regard to MRS, were $-0.525$ ($P < 0.001$) and $-0.368$ ($P = 0.006$), respectively. Paired t-test revealed that FFs determined by DIXON and MEGE were significantly different from that by MRS ($P < 0.001$).

**Discussion**

We investigated the sequence reproducibility of 3 methods and evaluated the diagnostic accuracy and sequence dependency of FFs determined by DIXON and MEGE compared to those by MRS. Each of the 3 MR methods showed high reproducibility. FFs determined by DIXON and MEGE systematically resulted in overestimation when fat content was less than 6.25%, probably because of relatively large background noise effects. When fat content was low, background noise increased the FF determined by DIXON. Similarly, fitting errors caused by background noise likely increased the FF determined by MEGE. However, FFs determined by both DIXON and MEGE resulted in improved diagnostic specificity (0.958 [DIXON]; 0.986 [MEGE]) at the 6.25% threshold. Therefore, FFs by DIXON and MEGE can be used for detect-
ing mild fatty liver.

In contrast, when the FF by MRS exceeded 6.25%, it was significantly higher than those by DIXON and MEGE, possibly because of MRS scan parameters (TR, 2000 ms; TE, 40 ms). At 1.5-tesla, the T1 value of a normal liver is approximately 586 ms and the T2 value, 40 ms, and the T1 value of fat is approximately 343 ms and the T2 value, 58 ms.23 Because the signal intensity of water is more greatly affected than that of fat, non-corrected T1 and T2 values with MRS might tend to be overrated. This might cause the lower diagnostic sensitivity above the 6.25% threshold. However, there was high correlation of both the DIXON and MEGE methods with MRS when FF values exceeded 6.25%, which suggests the small impact of this factor on FF evaluation.

Our study was limited because we did not know if volunteers with mild fatty liver were affected by NASH and NAFLD. In the case of NASH or cirrhosis, measurement values with T2* non-corrected DIXON might not give correct values. Further investigation is needed that includes cases with known iron deposition. Nevertheless, DIXON can overcome the limitation of restricted ROI with single-voxel MRS, so it would be the most useful sequence to evaluate the distribution of fat accumulation in diffuse fatty liver disease. At this time, we recommend using DIXON first. In the case with iron deposition, a more accurate FF could be measured by adding an MEGE sequence for T2* correction.

Conclusion

We demonstrated high reproducibility of FF measurements using MRS, DIXON, and MEGE. Both DIXON and MEGE showed high specificities of more than 95%, with relatively low sensitivities, 82% (DIXON) and 70% (MEGE) compared to those by MRS. Compared to the FF determined by MRS, those by DIXON and MEGE showed sequence dependency because of proportional and additional errors with DIXON and a proportional error with MEGE.

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