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Quantitative Susceptibility Mapping versus R2*-based Histogram Analysis for Evaluating Liver Fibrosis: Preliminary Results

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Purpose: The staging of liver fibrosis is clinically important, and a less invasive method is preferred. Quantitative susceptibility mapping (QSM) has shown a great potential in estimating liver fibrosis in addition to R2* relaxometry. However, few studies have compared QSM analysis and liver fibrosis. We aimed to evaluate the feasibility of estimating liver fibrosis by using QSM and R2*-based histogram analyses by comparing it with ultrasound-based transient elastography and the stage of histologic fibrosis.

Methods: Fourteen patients with liver disease were enrolled. Data sets of multi-echo gradient echo sequence with breath-holding were acquired on a 3-Tesla scanner. QSM and R2* were reconstructed by water-fat separation method, and ROIs were analyzed for these images. Quantitative parameters with histogram features (mean, variance, skewness, kurtosis, and 1st, 10th, 50th, 90th, and 99th percentiles) were extracted. These data were compared with the elasticity measured by ultrasound transient elastography and histological stage of liver fibrosis (F0 to F4, based on the new Inuyama classification) determined by biopsy or hepatectomy. The correlation of histogram parameters with intrahepatic elasticity and histologically confirmed fibrosis stage was examined. Texture parameters were compared between subgroups divided according to fibrosis stage. Receiver operating characteristic (ROC) analysis was also performed. $P < 0.05$ indicated statistical significance.

Results: The six histogram parameters of both QSM and R2* were significantly correlated with intrahepatic elasticity. In particular, three parameters (variance, percentiles [90th and 99th]) of QSM showed high correlation ($r = 0.818-0.844$), whereas R2* parameters showed a moderate correlation with elasticity. Four parameters of QSM were significantly correlated with fibrosis stage ($p = 0.637-0.723$) and differentiated F2-4 from F0-1 fibrosis and F3-4 from F0-2 fibrosis with areas under the ROC curve of > 0.8 , but those of R2* did not.

Conclusion: QSM may serve as a promising surrogate indicator in detecting liver fibrosis.

Keywords: liver fibrosis, quantitative susceptibility mapping, texture analysis

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Introduction

Chronic liver disease (CLD) is caused by various factors, such as alcoholic and nonalcoholic fatty liver disease (NAFLD) and viral infection. It can lead to progressive structural distortion of the entire liver by destruction and regeneration of the liver parenchyma, resulting in fibrosis, cirrhosis, and, eventually, end-stage liver disease.¹ An early diagnosis of liver fibrosis is important for the clinical management of viral hepatitis, portal hypertension, and hepatocellular carcinoma.² Liver biopsy has been considered the gold standard for staging liver fibrosis, using the new Inuyama classification or METAVIR score.^{3,4} However, liver biopsy is invasive, with the risk of rare, but critical, complications, such as infection and hemorrhage. In addition, the sampling result can have low reproducibility because of interobserver variability and sampling error.^{5,6}

Less invasive methods for the staging of liver fibrosis have been developed, including ultrasonography (US)-based transient elastography (UTE),⁷ MR elastography (MRE),^{8,9} and hepatocyte fraction (HeF).^{10,11} UTE is a widely used US-based elastographic method to evaluate elasticity by measuring the velocity of elastic shear waves in the parenchyma generated by a mechanical push. MRE is also used clinically as a MRI-based method to obtain the information on the stiffness of tissue by assessing the propagation of mechanical waves through the tissue using a special technique.⁸ Meanwhile, HeF is obtained by calculating changes to R1 in the liver and spleen after administration of gadolinium ethoxybenzyl diethylenetriamine pentaacetic acid. HeF has been reported to be a novel method for assessing liver function¹⁰ and fibrosis.¹¹

However, some techniques have their limitations. UTE requires expensive equipment and considerable expertise. Obesity and ascites reduce the diagnostic performance of UTE because of poor penetration, and UTE requires additional equipment for appropriate measurement in patients with obesity.¹² HeF requires the injection of a contrast agent, which is not suitable for patients with severe kidney failure. Therefore, there has been considerable interest in identifying other noninvasive surrogate markers of liver fibrosis, which are more reliable, faster, and provided without any additional devices.

CLD often causes intrahepatic iron deposition by dysregulation of iron metabolism, making liver iron accumulation a common observation in adult CLD patients.^{13–15} The evaluation of iron deposition is clinically important, as liver iron accumulation has been associated with increased fibrosis.¹⁶

Several serum markers for iron metabolism are associated with CLD, such as ferritin and hepcidin. However, ferritin is non-specific as an acute-phase protein affected by various elements, such as inflammation, infection, vitamin C levels, and liver damage.^{17,18} Likewise, hepcidin is also affected by many different stimuli, such as chronic kidney disease, inflammation, infection, and alcohol abuse.¹⁹ Therefore,

these markers are not appropriate for a more accurate diagnosis and monitoring of liver iron deposition.²⁰

There are several quantitative methods to measure liver iron deposition, such as biomagnetic susceptometry using a superconducting quantum interference device (SQUID) and MRI, which includes R2 and R2* relaxometry. However, there are very few SQUID devices available worldwide, which limit their use. On the other hand, R2 relaxometry is not practical in the setting of limited resources and time because it requires a long acquisition time of up to 20 min with free breathing, which makes it prone to respiratory motion artifacts.²¹

In addition to R2* relaxometry, quantitative susceptibility mapping (QSM) aims to evaluate magnetic susceptibility distribution and can be used to estimate iron concentration and the integrity of the collagen network.²² It was mainly developed for the brain, but several other applications have emerged.^{22–24} In QSM reconstruction of the trunk, fat tissue causes inaccurate estimation of magnetic susceptibilities because it has a different susceptibility and resonance frequency from water. In this regard, the water–fat (WF) separation method allows accurate QSM reconstruction.²⁵ It also improves the homogeneity and reproducibility of susceptibility values in the upper abdominal organs of normal subjects.²⁶ To the best of our knowledge, few studies have compared QSM analysis and the status of liver fibrosis. An *ex vivo* feasibility study exists,²⁷ but there is no *in vivo* study.

The aim of this study was to investigate the value of histogram parameters derived from QSM images and R2* images in estimating intrahepatic fibrosis, through comparison with US-based transient elastography and pathological fibrous stage.

Materials and Methods

This retrospective observational study was approved by the Institutional Review Board, and the need for informed consent was waived.

From July 2017 to October 2019, the electronic medical records of a tertiary hospital were searched for patients with liver fibrosis or disease who fulfilled the following criteria: 1) aged 20 years and above, 2) had both MRI and UTE of the liver performed within 8 months, and 3) had histopathological confirmation of liver fibrosis using the New Inuyama classification system (describing later) by biopsy or hepatectomy. In those patients in the F0–F3 stages, histopathological data were obtained within one year of the MRI examination. In the case of the F4 (cirrhosis) stage, histological data were obtained at any time before the MRI examination because cirrhosis represented the irreversible late stage of chronic progressive liver disease.²⁸ Sixteen patients with liver diseases were included in this study. Among them, two patients were excluded owing to the poor image quality: one patient was excluded because his or her QSM and R2* images had too many motion artifacts caused by poor breath-holding,

and another patient was excluded because QSM reconstruction failed owing to having a too large R2*, which led to insufficiency of intensity of magnitude image acquired with long TE for the development of an adequate phase map. Finally, 14 patients (10 men and 4 women; age, 33–85 years; median age, 73 years) were eligible for this study. Their diagnoses were hepatitis B ($n = 6$), hepatitis C ($n = 2$), NAFLD ($n = 3$), hepatic echinococcosis ($n = 2$), and metastatic tumor ($n = 1$). Five and nine subjects underwent liver biopsy and hepatectomy, respectively.

MRI examinations were performed using a 3-Tesla scanner (Hitachi, Tokyo, Japan). The 3D-gradient echo sequence with breath-holding was used for QSM data acquisition, with the following parameters: TR = 22.6 ms, six TEs = 3.1/6.6/10.1/13.6/17.1/20.6 ms, flip angle (FA) = 10°, FOV = 350 mm, section thickness = 3 mm, slab thickness = 192 mm, matrix = $160 \times 128 \times 32$ (reconstructed to $256 \times 256 \times 64$), and number of excitations (NEX) = 1. The scan duration was 19 s.

QSM and R2* reconstructions were performed using the WF separation method with the multipeak fat correction, as previously described.^{25,26} Briefly, the susceptibilities for the water and fat regions were calculated separately. Then, the two reconstructed images were combined to create the final QSM image, reducing shading artifacts at the edges of fat tissue.

UTE (FibroScan; Echosens, Paris, France) examinations were performed by six trained sonographers, the experience of whom in UTE was as follows: one had 7 months, one had one and a half year, and four had 5 years. The examination procedure has been previously described.^{29,30} All patients had at least 10 valid measurements with success rates higher than 60%. The median value of the 10 successful measurements was considered representative of liver elasticity, expressed in kilopascals (kPa).

Standard references were pathological fibrosis grades (F-grade) and elasticity measured by UTE (kPa). Histopathologic reports assessing the liver fibrosis stage were reviewed based on the New Inuyama classification system³ as follows: F0 = no fibrosis, F1 = fibrous portal expansion (mild fibrosis), F2 = bridging fibrosis (significant fibrosis), F3 = bridging fibrosis with architectural distortion (severe fibrosis), and F4 = cirrhosis. If the pathologists assigned several grades regarding fibrosis stage, the higher stage was adopted in this study.

ROIs were analyzed for QSM and R2* images. Eight round ROIs (diameter = 17 pixels) were manually placed in the liver parenchyma for each hepatic segment (S1–S8). These ROIs were carefully placed on the same places in QSM and R2* images, avoiding large vessels, cavities, mass lesions, gas-induced susceptibility artifacts, and ghost artifacts. ROIs were at least 5 pixels away from the liver boundary because edge of the liver often got blurred by respiratory artifact and might affect susceptibility and R2*. Segments excluded from measurement were those that had been resected by previous surgery and were occupied by huge masses.

The metrics derived from the gray-level histogram of susceptibility (ppb) and R2* (Hz) were mean, variance, skewness, kurtosis, and 1st, 10th, 50th, 90th, and 99th percentiles, which were calculated using MaZda software version 4.6 (Technical University of Lodz, Institute of Electronics, Poland).³¹ For each subject, these values were averaged across all ROIs in the liver parenchyma.

JMP software version 14.0 (SAS Institute, Cary, NC, USA) and Bellcurve for Excel version 3.20 (Social Survey Research Information, Tokyo, Japan) were used for statistical analyses. The correlation between the elasticity measured using transient elastography and histogram parameters of QSM and R2* was determined using Pearson correlation coefficient. A non-parametric Spearman's rank correlation test was performed to evaluate the correlation of elasticity measured by UTE and histogram parameters of QSM or R2* with F-grade. Unpaired student's t-test was used to compare all histogram parameters between the subgroups F0–1 (F0 + F1, non-significant fibrosis) and F2–4 (F2 + F3 + F4, significant fibrosis) of the fibrosis stage, and between the subgroups F0–2 (F0 + F1 + F2, mild fibrosis) and F3–4 (F3 + F4, severe fibrosis). A P value < 0.05 was considered statistically significant. Receiver operating characteristic (ROC) curve analysis was performed on the basis of binary logistic regression to assess the diagnostic utility of all measurements (UTE, QSM, and R2*) to differentiate significant from nonsignificant fibrosis (F0–1 versus F2–4), and to differentiate between mild and severe fibrosis (F0–2 versus F3–4). The value that maximized the Youden index was chosen as the optimal cutoff value. To evaluate intra- and interobserver reproducibility, reliability analysis was performed by calculating the intraclass correlation coefficient (ICC) on the basis of QSM measurements.

Results

The number of subjects with fibrosis stages F0, F1, F2, F3, and F4 were 4, 2, 3, 1, and 4, respectively. The median time between the MR examination and tissue sampling (biopsy or hepatectomy) was 23 days (interquartile range [IQR], 2–288 days) for F0–F3 cases and 4195 days (IQR, 652–5699 days) for F4.

The intrahepatic texture showed higher susceptibilities in patients with higher elasticities and severe fibrosis (Figs. 1 and 2). No serious artifacts appeared near the heart or intestinal tract (Fig. 3).

In the histogram analysis, hepatic elasticity significantly ($P < 0.05$) correlated with the mean, variance, and 1st, 50th, 90th, and 99th percentiles of intrahepatic susceptibility (Fig. 4). The correlation coefficients were 0.643, 0.844, -0.675, 0.687, 0.818, and 0.818, respectively. As for R2*, hepatic elasticity correlated with mean, skewness, and 10th, 50th, 90th, and 99th percentiles (Fig. 5). The correlation coefficients were 0.619, -0.589, 0.566, 0.632, 0.647, and 0.637, respectively. There were no significant correlations between the other parameters.

Fig. 1 QSM (above) and R2* (below) images of patients with F0 stage fibrosis (median of elasticity: 5.9 kPa). The mean values of susceptibility and R2* of each ROI are as follows: S1 (−8.516 ppb and 30.552 Hz, respectively), S2 (−0.281 ppb and 31.715 Hz, respectively), S3 (−28.683 ppb and 33.127 Hz, respectively), S4 (−26.204 ppb and 34.878 Hz, respectively), S5 (−5.43 ppb and 29.837 Hz, respectively), S6 (−7.475 ppb and 30.199 Hz, respectively), S7 (−1.787 ppb and 27.493 Hz, respectively), and S8 (−34.063 ppb and 32.471 Hz, respectively). The corresponding averages of all ROIs are −14.055 ppb and 31.284 Hz, respectively. kPa, kilopascals; QSM, quantitative susceptibility mapping.

Fig. 2 QSM (above) and R2* (below) images of patients with fibrosis of F4 stage (median of elasticity: 11.6 kPa). The mean values of susceptibility and R2* of each ROI are as follows: S1 (10.471 ppb and 44.398 Hz, respectively), S2 (−35.837 ppb and 44.253 Hz, respectively), S3 (114.814 ppb and 48.765 Hz, respectively), S4 (1.109 ppb and 45.959 Hz, respectively), S5 (24.982 ppb and 42.688 Hz, respectively), S6 (76.480 ppb and 45.674 Hz, respectively), S7 (−18.986 ppb and 47.778 Hz, respectively), and S8 (−2.072 ppb and 43.482 Hz, respectively). The corresponding averages of all ROIs are 21.370 ppb and 45.419 Hz, respectively. kPa, kilopascals; QSM, quantitative susceptibility mapping.

Table 1 summarizes the results of the nonparametric Spearman's rank correlation test with regard to the relationship of elasticity measured using transient elastography and histogram parameters of QSM and R2* with the fibrosis stages. Four parameters of susceptibility were significantly ($P < 0.05$) correlated with fibrosis stages of patients (mean: $\rho = 0.637$; 50th percentile: $\rho = 0.671$; 90th percentile: $\rho = 0.706$; and 99th percentile: $\rho = 0.723$). Two parameters of susceptibility were moderately correlated with fibrosis stages (variance: $\rho = 0.486$, $P = 0.0780$ and kurtosis: $\rho = 0.526$, $P = 0.0529$), but these correlations were not significant. There

was no significant correlation of R2* histogram parameters with the fibrosis stage.

The mean and 50th, 90th, and 99th percentiles of susceptibility in the F2–4 group were significantly higher than those in the F0–1 subgroup. The average values of each of these parameters were 8.043 ppb (standard error [SE], 4.7942), 7.884 ppb (SE 5.0005), 69.2567 ppb (SE 11.096), and 125.804 ppb (SE 17.649) in the F2–4 group, and −14.084 ppb (SE 5.5358), −13.674 ppb (SE 5.7741), 25.2500 ppb (SE 12.813), and 59.806 ppb (SE 20.379) in the F0–1 group. The corresponding P values were 0.0106, 0.0154, 0.0234, and

Fig. 3 Coronal planar reconstructed images of patients with fibrosis of F0 and F4 stages. No serious artifacts are visible near the heart (arrows) and intestinal tract (arrowheads).

0.0307 (Fig. 6). Moreover, the mean and 50th percentile of susceptibility in the F3–4 group were significantly higher than those in the F0–2 subgroup. The average values of each of these parameters were 12.566 ppb (SE 6.2708) and 11.475 ppb (SE 6.7250) in the F3–4 group and -9.221 ppb (SE 4.6740) and -8.483 ppb (SE 5.0125) in the F0–2 group. The corresponding *P* values were 0.0165 and 0.0348 (Fig. 7). In contrast, there were no significant differences in all the parameters of R2* between these subgroups (Figs. 8 and 9).

Table 2 summarizes the areas under the curve (AUCs), optimal cutoff values, sensitivities, specificities, and accuracies of elasticity determined using UTE and histogram parameters of QSM and R2* for differentiating significant from nonsignificant hepatic fibrosis. The data on the same items for differentiating between mild and severe fibrosis are summarized in Table 3. The QSM parameters that showed AUCs of > 0.8 in the F0–1 versus F2–4 subgroups were mean, variance, and 50th, 90th, and 99th percentiles. In a comparison of F0–2 versus F3–4 subgroups, mean and 10th, 50th, 90th, and 99th percentiles of QSM showed AUCs of > 0.8 . In contrast, only the 1st percentile of R2* showed an AUC of > 0.8 for differentiating nonsignificant from significant fibrosis.

The intra- and interobserver ICC of QSM histogram parameters are summarized in Table 4. Interobserver reliabilities among three radiologists were almost perfect

for four parameters (variance: 0.977; 1st percentile: 0.860; 90th percentile: 0.930; and 99th percentile: 0.909), substantial for two parameters (mean: 0.703 and 10th percentile: 0.768), and moderate for one parameter (50th percentile: 0.582), which demonstrates a good reproducibility of the ROI measurements. The intraobserver reliabilities were almost perfect for seven parameters (mean: 0.921; variance: 0.989; 1st percentile: 0.957; 10th percentile: 0.929; 50th percentile: 0.924; 90th percentile: 0.981; and 99th percentile: 0.980) and moderate for two parameters (skewness: 0.449 and kurtosis: 0.565), which demonstrates good repeatability.

Discussion

In this study, several histogram parameters of intrahepatic susceptibility and R2* were significantly correlated with US-based elasticity. Among them, positive correlations were noted for mean and 50th, 90th, and 99th percentiles. Moreover, the mean and 50th, 90th, and 99th percentiles of hepatic susceptibility were significantly correlated with pathological fibrosis stages, but no parameters of R2* were significantly correlated. In addition, these parameters of QSM differentiated significant from nonsignificant hepatic fibrosis and severe from mild fibrosis with high AUC, whereas those of R2* did not. Therefore, hepatic

Fig. 4 Scatter plots between elasticity and histogram parameters of QSM. Hepatic elasticity measured by UTE significantly correlates with mean, variance, and 1st, 50th, 90th, and 99th percentiles of intrahepatic susceptibility. kPa, kilopascals; QSM, quantitative susceptibility mapping; UTE, ultrasonography-based transient elastography.

susceptibility by QSM can be a noninvasive biomarker of liver fibrosis.

Since the liver fibrosis generally does not appear homogeneous,⁵ analysis of a specific segment of the liver, even if it is near the spot of biopsy, might not reflect the progression of fibrosis precisely. In addition, the measurement spot of US and MRI cannot be exactly same as that of biopsy. Therefore, eight ROIs/hepatic segment were placed in the liver parenchyma, and values were averaged across all ROIs to evaluate diffuse changes among all the segments of the organ.

The results of our *in vivo* study were different from a previous *ex vivo* study. Jafari et al. showed that $R2^*$ in human fibrotic liver explant samples significantly increased compared with that in nonfibrotic samples, while the difference in susceptibility was nonsignificant.²⁷ In contrast, our study showed significant increase in susceptibility. Our study used WF separation method for QSM reconstruction, while the *ex vivo* study used simultaneous phase unwrapping and

removal of chemical shift (SPURS).³² We performed histogram analyses of images, while only median values were investigated in the *ex vivo* study. Moreover, the etiology of samples in the *ex vivo* study was not mentioned. These factors might make the difference in results between our study and the *ex vivo* study.

Mean and 50th, 90th, and 99th percentiles of susceptibility were significantly correlated with pathological fibrosis stage determined using the New Inuyama classification system. We speculated that an increase in the value of these parameters observed in this study might be caused by iron deposits. For example, Nelson et al. observed that iron deposition in (NAFLD liver biopsies was associated with more advanced fibrosis.³³ In addition, prior research using a rat model of radiation-induced liver fibrosis showed that iron deposition, confirmed using Prussian blue staining, increased gradually from stage F0 to stage F2.³⁴ In viral hepatitis, however, no association was found between the amount of iron

Fig. 5 Scatter plots between elasticity and histogram parameters of R2*. Hepatic elasticity measured by UTE significantly correlates with mean, skewness, and 10th, 50th, 90th, and 99th percentiles of intrahepatic R2*. kPa, kilopascals; UTE, ultrasonography-based transient elastography.

deposited and fibrosis in previous studies.^{35,36} However, Son et al. showed that liver volume tended to decrease gradually with the progression of liver fibrosis in patients with viral hepatitis,³⁷ which might decrease the density of parenchymal tissue or ROI and relatively increase susceptibility caused by intrahepatic iron deposits. In summary, iron deposition or shrinkage of liver parenchyma might increase the mean and larger percentile of intrahepatic susceptibility.

In the comparison between susceptibility and R2*, the mean and 50th, 90th, and 99th percentiles had a similar positive correlation with US-based elasticity. However, the 1st and 5th percentiles showed a negative correlation with susceptibility and a positive correlation with R2*. In addition, the variance of susceptibility significantly correlated

with elasticity. These results suggested that the distribution of susceptibility became wider with the increase in elasticity, but that of R2* did not. Existing reports suggest that intrahepatic fibrosis contributes nonlinearly to R2*, and it is weakly diamagnetic and contributes inversely and linearly to susceptibility.³⁸ Therefore, the mixture of fibrosis and iron deposition with the advancement of liver fibrosis—a mix of magnetically opposing materials—causes a wider distribution of magnetic susceptibility.

Fibrosis interferes with R2* and depends on the microenvironment of the water–fibrosis interaction,³⁹ which is very complex and difficult to quantify. Accordingly, we assumed that intrahepatic susceptibility parameters have a stronger relationship with elasticity than with R2* because of fibrosis. In our study, the mean and percentile (50th, 90th, and 99th) of

Table 1 Correlation of all measurements with liver fibrosis stages

	ρ	<i>P</i> value
Elasticity	0.680	0.0074
QSM		
Mean	0.637	0.0142
Variance	0.486	0.0780
Skewness	0.244	0.4002
Kurtosis	0.526	0.0529
1st Percentile	0.074	0.7999
10th Percentile	0.368	0.1948
50th Percentile	0.671	0.0086
90th Percentile	0.706	0.0048
99th Percentile	0.723	0.0035
R2*		
Mean	0.327	0.2525
Variance	0.219	0.4513
Skewness	0.377	0.1832
Kurtosis	-0.104	0.7235
1st Percentile	0.431	0.1238
10th Percentile	0.418	0.1362
50th Percentile	0.316	0.2697
90th Percentile	0.345	0.2268
99th Percentile	0.409	0.1458

QSM, quantitative susceptibility mapping.

susceptibility strongly correlated with fibrosis stage, and they were significantly higher in the group with significant fibrosis (F2–4) than those in the F0–1 (minimal fibrosis) subgroup. In addition, these parameters of QSM showed excellent performance for differentiating not only significant from nonsignificant fibrosis but also severe from mild fibrosis in ROC analyses. Conversely, correlation of R2* parameters with fibrosis stage was moderate at best, and there were no differences in all parameters of R2* in subgroup comparison. Almost all R2* parameters showed lower diagnostic performance for differentiating liver fibrosis. These results of R2* might be due to the nonlinear interference of fibrosis.³⁸ A similar superiority of susceptibility over R2* has been reported in the diagnosis of Parkinson's disease (PD). The discrimination between PD patients and normal controls is better in susceptibility than in R2*.⁴⁰

Several parts of the results do not agree among correlation analysis, ROC analysis, and subgroup comparison. First, variance of susceptibility was strongly correlated with intrahepatic elasticity, moderately correlated with pathological fibrosis stages, and showed an AUC of > 0.8 for differentiating significant from nonsignificant liver fibrosis, but there was no significant difference in variance of susceptibility between subgroups. Second, 90th and 99th percentiles of susceptibility showed an AUC of > 0.8 for differentiating severe from mild fibrosis; however, there were no significant difference in these parameters between mild and severe fibrosis subgroups. These mismatches might result from the characteristics of F4 stage fibrosis. In our study, all four patients with F4 were histopathologically diagnosed 1 year 9 months to 15 years 7 months (median of 11 years 6 months) before the MRI, and all were cases of viral hepatitis. The interval between histopathological staging and MRI was much longer than that in the F0–F3 group (less than 1 year, median of 23 days). Previous studies suggested that treatment with antiviral drugs improved fibrosis in the cirrhotic liver.^{41,42} Therefore, these long intervals of F4 diagnosis might reflect the improvement of fibrosis, which might result in mismatches among correlation analysis, ROC analysis, and subgroup comparison.

Our data suggest that intrahepatic heterogeneous magnetic susceptibility extracted by histograms may serve as good imaging aids for the objective assessment of hepatic fibrosis. In the future, it is interesting to conduct the prospective study and compare QSM and MRE in patients who are candidates of liver biopsy.

This study has several limitations. First, 5 of the 14 patients had liver biopsy, which was a very small size of sample: accordingly, the pathological staging of fibrosis may be biased. Second, the ROI of images was manually placed without reference to US-based elastography and pathological findings; thus, the value of the parameters may be biased. Third, each sample had data that were averaged across all segments, and we did not analyze the comparisons per segment. Fourth, the good intra- and interobserver reproducibility in our study could be attributed to the experience of the present investigators in performing liver QSM; hence, the reproducibility of this study may decline in cases of less experienced readers. Fifth, liver diseases of different etiologies may have various histological alterations; hence, studies focusing on patients with a single etiology are needed. Sixth, in our study, as mentioned above, all four cases of F4 were viral hepatitis, and all cases had much longer intervals between histopathological staging and MRI than the cases of F0–F3, which might reflect the improvement of fibrosis in F4 cases; however, in most cases of previous studies, F4 only improved to F2 or F3 by histological classification. It was rare that F4 improved to F0 or F1.^{41,42} Therefore, we classified those patients in the

Fig. 6 Box-Whisker plots of QSM parameters between F0-1 and F2-4 subgroups. The mean and 50th, 90th, and 99th percentiles of susceptibility in the F2-4 group are significantly higher than those in the F0-1 group ($P < 0.05$). There are no significant differences in the other parameters. QSM, quantitative susceptibility mapping.

Fig. 7 Box-Whisker plots of QSM parameters between F0-2 and F3-4 subgroups. The mean and 50th percentile of susceptibility in the F3-4 group are significantly higher than those in the F0-2 group ($P < 0.05$). There are no significant differences in the other parameters. QSM, quantitative susceptibility mapping.

Fig. 8 Box-Whisker plots of $R2^*$ parameters between F0-1 and F2-4 subgroups. There are no significant differences in all parameters.

Fig. 9 Box-Whisker plots of $R2^*$ parameters between F0-2 and F3-4 subgroups. There are no significant differences in all parameters.

Table 2 AUCs derived using ROC analysis for differentiating significant from nonsignificant fibrosis

F0–1 vs. F2–4	AUC (95% CI)	Optimal cutoff	Sensitivity	Specificity	Accuracy
Elasticity	1.000 (1.000–1.000)	8.7	1.000	1.000	1.000
QSM					
Mean	0.875 (0.680–1.069)	−6.0269	0.875	0.833	0.857
Variance	0.812 (0.577–1.047)	1228.651	0.875	0.666	0.785
Skewness	0.708 (0.412–1.004)	0.159719	0.625	0.833	0.714
Kurtosis	0.770 (0.440–1.101)	0.33479	1.000	0.666	0.857
1st Percentile	0.500 (0.157–0.842)	−152.250	0.250	1.000	0.571
10th Percentile	0.625 (0.308–0.941)	−43.167	0.500	0.666	0.642
50th Percentile	0.875 (0.681–1.068)	1.1250	0.750	1.000	0.857
90th Percentile	0.937 (0.803–1.071)	42.000	0.875	1.000	0.928
99th Percentile	0.937 (0.803–1.071)	82.1250	0.875	1.000	0.928
R2*					
Mean	0.750 (0.466–1.033)	33.24212	0.750	0.833	0.785
Variance	0.500 (0.157–0.842)	5.49029	0.250	1.000	0.571
Skewness	0.687 (0.343–1.031)	−0.158959	0.750	0.833	0.785
Kurtosis	0.645 (0.314–0.977)	0.56550	0.875	0.500	0.714
1st Percentile	0.822 (0.584–1.061)	27.75000	0.750	0.833	0.785
10th Percentile	0.791 (0.526–1.057)	31.75000	0.625	1.000	0.785
50th Percentile	0.729 (0.430–1.028)	33.25000	0.750	0.833	0.785
90th Percentile	0.729 (0.430–1.028)	36.37500	0.750	0.833	0.785
99th Percentile	0.750 (0.453–1.044)	39.75000	0.625	1.000	0.785

AUC, area under the curve; CI, confidence interval; QSM, quantitative susceptibility mapping; ROC, receiver operating characteristic.

study into two subgroups: F0–F1 versus F2–F4, and F0–2 versus F3–4. Finally, although the multipeak fat correction was performed in our study, intrahepatic fat might increase susceptibility to a certain degree. The mechanism of increase may be different for susceptibility and R2*, and the rate of increase in susceptibility may be lower than that in R2*,^{43–46} but further research is needed to improve our understanding in this regard.

Conclusion

Intrahepatic histogram analysis showed that susceptibility and R2* significantly correlated with elasticity measured by UTE. Susceptibility significantly correlated with pathological fibrosis stage, but R2* did not. The comparison and ROC analysis between histogram parameters and pathological fibrosis stage showed that QSM-based histogram analysis could distinguish not only significant liver fibrosis (F2–4) from nonsignificant fibrosis (F0–1) but also severe fibrosis

(F3–4) from mild fibrosis (F0–2). QSM imaging of liver can reflect the mixture of fibrosis and iron deposition with the advancement of fibrosis stage more precisely than R2* can. QSM is expected to be a useful surrogate biomarker for detecting liver fibrosis.

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Conflicts of Interest

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Table 3 AUCs derived using ROC analysis for differentiating severe from mild fibrosis

F0–2 vs. F3–4	AUC (95% CI)	Optimal cutoff	Sensitivity	Specificity	Accuracy
Elasticity	0.733 (0.4502–1.0164)	8.7	1.000	0.666	0.785
QSM					
Mean	0.866 (0.645–1.087)	2.4674	1.000	0.777	0.857
Variance	0.755 (0.480–1.031)	1228.651	1.000	0.555	0.714
Skewness	0.688 (0.339–1.038)	0.159719	0.800	0.777	0.785
Kurtosis	0.711 (0.424–0.997)	0.40102	1.000	0.555	0.714
1st Percentile	0.622 (0.277–0.967)	−88.833	0.800	0.555	0.642
10th Percentile	0.822 (0.549–1.095)	−43.167	0.800	0.888	0.857
50th Percentile	0.888 (0.671–1.106)	1.1250	1.000	0.888	0.928
90th Percentile	0.866 (0.645–1.087)	42.0000	1.000	0.777	0.857
99th Percentile	0.866 (0.645–1.087)	82.1250	1.000	0.777	0.857
R2*					
Mean	0.600 (0.231–0.968)	33.24212	0.800	0.666	0.714
Variance	0.622 (0.301–0.943)	9.95394	1.000	0.333	0.571
Skewness	0.711 (0.424–0.997)	−0.158959	0.800	0.666	0.714
Kurtosis	0.511 (0.178–0.843)	0.56550	1.000	0.444	0.642
1st Percentile	0.644 (0.298–0.990)	27.75000	0.800	0.666	0.714
10th Percentile	0.644 (0.270–1.018)	30.50000	0.800	0.666	0.714
50th Percentile	0.600 (0.231–0.968)	33.25000	0.800	0.666	0.714
90th Percentile	0.600 (0.231–0.968)	36.37500	0.800	0.666	0.714
99th Percentile	0.644 (0.450–1.016)	38.37500	0.800	0.666	0.714

AUC, area under the curve; CI, confidence interval; QSM, quantitative susceptibility mapping; ROC, receiver operating characteristic.

Table 4 Intra- and interobserver agreements on QSM histogram parameters

	Interobserver ICC	Intraobserver ICC
Mean	0.703 (0.435–0.881)	0.921 (0.764–0.974)
Variance	0.977 (0.945–0.997)	0.989 (0.967–0.996)
Skewness	0.060 (−0.206–0.447)	0.449 (−0.657–0.821)
Kurtosis	0.023 (−0.230–0.409)	0.565 (−0.296–0.860)
1st Percentile	0.860 (0.698–0.948)	0.957 (0.871–0.986)
10th Percentile	0.768 (0.536–0.910)	0.929 (0.787–0.977)
50th Percentile	0.582 (0.271–0.822)	0.924 (0.771–0.975)
90th Percentile	0.930 (0.840–0.975)	0.981 (0.944–0.994)
99th Percentile	0.909 (0.796–0.967)	0.980 (0.939–0.993)

ICC, intraclass correlation coefficient; QSM, quantitative susceptibility mapping.

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