Published in final edited form as:

Abdom Radiol (NY). 2020 November ; 45(11): 3386–3399. doi:10.1007/s00261-020-02783-1.

Liver Fat Quantification: Where Do We Stand?

Jitka Starekova², Scott B. Reeder^{1,2,3,4,5}

¹Department of Biomedical Engineering, University of Wisconsin, Madison, WI, USA

²Department of Radiology, University of Wisconsin, Madison, WI, USA

³Department of Medical Physics, University of Wisconsin, Madison, WI, USA

⁴Department of Medicine, University of Wisconsin, Madison, WI, USA

⁵Department of Emergency Medicine, University of Wisconsin, Madison, WI, USA

Abstract

Excessive intracellular accumulation of triglycerides in the liver, or hepatic steatosis, is a highly prevalent condition affecting approximately one billion people worldwide. In the absence of secondary cause, the term nonalcoholic fatty liver disease (NAFLD) is used. Hepatic steatosis may progress into nonalcoholic steatohepatitis, the more aggressive form of NAFLD, associated with hepatic complications such as fibrosis, liver failure and hepatocellular carcinoma. Hepatic steatosis is associated with metabolic syndrome, cardiovascular disease and represents an independent risk factor for type 2 diabetes, cardiovascular disease and malignancy.

Percutaneous liver biopsy is the current reference standard for NAFLD assessment; however, it is an invasive procedure associated with complications and suffers from high sampling variability, impractical for clinical routine and drug efficiency studies. Therefore, noninvasive imaging methods are increasingly used for the diagnosis and monitoring of NAFLD. Among the methods quantifying liver fat, chemical shift encoded MRI (CSE-MRI) based proton density fat-fraction (PDFF) has shown to the most promise. MRI-PDFF is increasingly accepted as quantitative imaging biomarker of liver fat that is transforming daily clinical practice and influencing the development of new treatments for NAFLD. Furthermore, CT is an important imaging method for detection of incidental steatosis, and the practical advantages of quantitative ultrasound hold great promise for the future. Understanding the disease burden of NAFLD and the role of imaging may initiate important interventions aimed at avoiding the hepatic and extrahepatic complications of

Corresponding author: Jitka Starekova, MD, Department of Radiology, UW Madison, WI, 600 Highland Ave, Madison, WI 53792, USA, *Telephone number:* (608) 504-4218, starekova@wisc.edu.

Author contributions:

All authors contributed to the study conception and design. Literature search and material collection were performed by Jitka Starekova and Scott Reeder. The first draft of the manuscript was written by Jitka Starekova and critically revised by Scott Reeder. All authors read and approved the final manuscript.

Publisher's Disclaimer: This Author Accepted Manuscript is a PDF file of an unedited peer-reviewed manuscript that has been accepted for publication but has not been copyedited or corrected. The official version of record that is published in the journal is kept up to date and so may therefore differ from this version.

Conflicts of interest:

No authors have any relevant conflicts. Unrelated to this work, Dr. Reeder consults with HeartVista and has ownership interests in Calimetrix, Reveal Pharmaceuticals, Collectar Biosciences, and Elucent Medical.

NAFLD. This article reviews clinical burden of NAFLD, and the role of non-invasive imaging techniques for quantification of liver fat.

Keywords

hepatic steatosis; nonalcoholic fatty liver; NAFLD; nonalcoholic steatohepatitis; NASH; non-invasive quantitative biomarker; magnetic resonance imaging; liver fat quantification

Introduction

Hepatic steatosis is a pathological condition, characterized by excessive intracellular accumulation of triglycerides in the liver. It is the hallmark histopathological feature of nonalcoholic fatty liver disease (NAFLD), the most common chronic liver disease afflicting an estimated 1 billion people worldwide [1–3]. Hepatic steatosis is common also in other conditions such as viral hepatitis, genetic lipodystrophies or as a consequence of alcohol abuse or treatment with steatogenic drugs [4–6]. Early identification of hepatic steatosis is important to prevent progression and adverse outcomes of the disease [4]. Prolonged, excessive fat accumulation increases the risk of hepatocyte injury, inflammation, fibrosis and eventually cirrhosis [1, 7, 8]. Complications of cirrhosis include liver failure and hepatocellular carcinoma [8, 9]. Further, NAFLD is an independent risk factor that predisposes patients to the subsequent development of diabetes mellitus type 2 and cardiovascular events [10–12]. Because of the limitations of percutaneous liver biopsy, noninvasive methods, particularly quantitative imaging biomarkers, are increasingly used for the diagnosis and monitoring of NAFLD, before isolated steatosis progresses to nonalcoholic steatohepatitis (NASH) [13]. This review will focus on the current status and recent progress of the radiological imaging methods for assessment of liver fat. After reading this article, readers should be familiar with the current understanding of NAFLD and the growing necessity for the use of non-invasive techniques in research and clinical fields.

Take home message:

Hepatic steatosis and nonalcoholic fatty liver disease (NAFLD) is a rapidly growing public health problem affecting over 1 billion people worldwide. The limited sampling and high variability of the current reference standard, percutaneous liver biopsy, has spurred the development of promising non-invasive imaging techniques used in diagnosis and monitoring of NAFLD. Currently, quantitative chemical-shift-encoded magnetic resonance imaging (CSE-MRI) is the most accurate, reliable non-invasive method to detect and quantify hepatic steatosis.

Complications of Liver Fat

The liver plays a central role in lipid metabolism. Lipid droplets in the liver serves as an energy reservoir, storing neutral lipids such as triglycerides and cholesterol esters during times of energy excess and releasing those during times of extended fasting [9]. Imbalances of lipid homeostasis present in metabolic diseases, obesity or other pathological conditions can lead to the excessive accumulation of lipids inside of hepatocytes [8]. In hepatic

steatosis, among the all lipids, triglyceride is the major lipid that accumulates in the liver [14]. If hepatic steatosis is present in the absence of any secondary causes (viral hepatitis, excess alcohol consumption, lipodystrophy, steatogenic drugs, etc.), a diagnosis of nonalcoholic fatty liver disease (NAFLD) can be made [15]. The presence of hepatocyte injury (ballooning degeneration), inflammation (presence of leukocytes) and/or scar (fibrosis), is termed nonalcoholic steatohepatitis (NASH), the more aggressive form of NAFLD [3, 16].

Why should we care about liver fat?

NAFLD is an increasing major public health concern worldwide [9]. Prevalent in up to 30% of adults in the Western countries and 18% in Asia, NAFLD is now the most common chronic liver disease worldwide, affecting as many as one billion people [3, 8]. In the United States alone, an estimated 80–100 million individuals are affected by NAFLD [3]. Particularly worrisome is the increasing trend of NAFLD diagnoses in younger individuals [17], with 3–10% of non-obese and approximately 50% of obese children reported to have NAFLD [7]. Compelling emerging evidence also suggests that metabolic abnormalities that originate as early as in utero may cause NAFLD in newborns [7]. Thus, childhood NAFLD is important to recognize because prolonged fat accumulation may lead to end-stage liver disease in early adulthood [7, 18].

The severity of NAFLD varies from isolated steatosis, i.e.: nonalcoholic fatty liver (NAFL) to more advanced forms such as NASH, fibrosis and NASH-related cirrhosis [3]. In addition to steatosis, NASH comprises necro-inflammatory changes of the hepatocytes, highlighting the progressive nature of the disease [3, 7–9, 19]. An estimated 33% of NAFL and NASH patients will progress to hepatocyte injury, inflammation and fibrosis, although an estimated 20% may exhibit disease regression to some extent [8]. Hepatic decompensation and cirrhosis have been shown to develop over a mean time space of 7.6 years in approximately 3% patients diagnosed with NAFLD [8]. Moreover, patients with decompensated NASH have a median survival of only 2 years [8].

Hepatic steatosis also is an independent risk factor for the development of hepatocellular carcinoma (HCC) [8]. Seven percent of patients with NAFLD related cirrhosis will develop HCC over a 10-year timeframe [20]. It is important to note that NASH is currently the second leading indication for liver transplantation in the United States and projected to be the most common one within the next 20 years [7, 8].

In addition to liver related morbidity and mortality, abnormal liver fat content has other important clinical implications. NAFLD has many related co-morbidities, including obesity, dyslipidemia, type 2 diabetes and metabolic syndrome [3], and is more prevalence in patients who are severely obese (90%) or who type 2 diabetes (76%) [21]. Furthermore, steatosis imparts an independent increased risk of kidney disease [22], cancer [1, 23] and cardiovascular disease [12, 17], thus also contributing to non-liver related morbidity and mortality. NAFLD patients have a 34–69% higher chance to die within the next 15 years, in comparison to the general population [8]. NAFLD is known to increase the risk of type 2 diabetes threefold and the risk of cardiovascular events eightfold [17]. Notably, NAFLD was

shown to be associated with presence of high-risk coronary plaques, independently from other traditional cardiovascular risk factors [12]. Thus, the consequences of elevated liver fat in association with cardiovascular diseases eclipses the mortality risk related to liver disease, i.e.: NAFLD/NASH.

In the liver, hepatic steatosis is an important consideration in diseases beyond NAFLD [9]. For example, its prevalence in patients infected with hepatitis C virus (HCV) is about 50%, and in genotype 3a HCV, as high as 75% [5]. Importantly, HCV patients with steatosis affecting more than 30% of hepatocytes are more likely to develop liver fibrosis [9]. Hepatic steatosis is also a common finding in human immunodeficiency virus (HIV) patients, with prevalence up to 36% and represents increased risk for fibrosis development in HIV without concomitant viral hepatitis infection [24]. Monitoring of liver fat content is therefore important for the prognosis and evaluation of disease progression in these patient populations. Hepatic steatosis is also a common side effect of alcohol consumption. While previously considered harmless, alcohol-induced steatosis enhances susceptibility of the liver to developing more advanced pathologies such as alcoholic steatohepatitis (ASH), cirrhosis and HCC [6].

In summary, hepatic steatosis is the histopathological hallmark of NAFLD but can occur in many other pathological conditions. Prolonged liver fat accumulation is not a benign condition. Patients with hepatic steatosis are at risk of developing serious complications, such as steatohepatitis, fibrosis, end-stage liver disease and hepatocellular carcinoma [6, 9, 19]. There are far too many patients with early stages of NAFLD to justify medical intervention in every patient. Rather, the prevalence and severity of later complications and consequences justifies the need for appropriate preventive measures and monitoring in a subset of patients with more advanced NAFLD.

Limitations of liver biopsies and need for imaging biomarkers

Percutaneous non-targeted liver biopsy is widely regarded as the reference standard for histopathological evaluation of fat content in the liver [25, 26]. Hepatic steatosis is typically graded on a 0–3 scale, based on the percentage of hepatocytes that contain intracellular lipid vacuoles [16]. Grade 0 (<5%) is considered normal, grade 1 (5–33%) is designated as mild, grade 2 (34–66% hepatocytes affected) is considered moderate, and grade 3 (>66% hepatocytes affected) is considered severe [7, 8, 16]. Non-targeted liver biopsy is usually performed to grade steatosis and to determine whether inflammation and significant fibrosis are present. Notably, liver biopsy is still required for the definitive diagnosis of NASH [27], and NAFLD-risk stratification requires distinguishing patients with inflammation and/or fibrosis, i.e.: distinguish patients with NASH from those patients with isolated steatosis i.e.:NAFL [8]. However, the disadvantages of this diagnostic approach are well known, and the emergence of non-invasive imaging biomarkers are leading to rethinking of the current diagnostic approach. Non-targeted liver biopsy is usually performed in specialized liver centers requiring sedation, periprocedural monitoring and involvement of an expert radiologist or hepatologist, and pathologist [25, 28]. Not only is biopsy expensive [29], but it also requires that both the patient and a care-giver give up a full day of work, an economic impact often not considered in cost-effective analyses of biopsy compared to imaging.

Although uncommon, biopsy can lead to complications such as bleeding and in rare cases, death [25]. It is worth highlighting that patients with liver disease may present with coagulopathy and thrombocytopenia which further increases the risk of biopsy complications [25]. Repeated liver biopsies are a traumatic experience for patients, leading to poor compliance and consequently unsatisfactory and inappropriate monitoring of the disease [20, 25]. Many providers and parents may be reticent to perform biopsies in children.

Importantly, the histopathologic features of NAFLD are patchy, at the spatial scale of a biopsy. As a result of this inhomogeneous distribution of fat in the liver [30], variability due to sampling error often occurs [26, 29]. Poynard et al. reported maximized accuracy on biopsy samples at least 3 cm long [31]. However, in practice up to 36% of biopsy samples do not reach half of this length [29]. The diagnostic performance for NASH based on a single biopsy specimen is limited, with a negative predictive value of 74% in patients with NAFLD [29]. There is also very high inter- and intra-observer variability in the assessment of liver biopsy samples [29, 32]. These limitations are an intrinsic limitation of biopsy. Indeed, sampling 1/50,000th of an organ with patchy disease features is inherently flawed. Volumetric imaging methods or other biomarkers that assess the entire organ, do not suffer from this fundamental limitation, although these methods may suffer from other limitations unrelated to volumetric assessment.

For these reasons, there is significant motivation for the development of non-invasive approaches for the management of NAFLD, including predictive models such as NAFLD fibrosis score, serum biomarkers (enhanced liver fibrosis test (ELFTM)) and imaging techniques that quantify hepatic steatosis, and liver stiffness (a biomarker of hepatic fibrosis) [27], and assess other potential quantitative imaging biomarkers.

Ultrasound

The accessibility and relatively low cost of conventional ultrasound (US) make it an attractive, non-invasive imaging technique. For hepatic steatosis, qualitative US assessment is based on evaluation of sonographic patterns, including increased liver echogenicity due to scattered by hepatic lipid droplets (Fig. 1) [33]. The US beam is also attenuated by liver fat, resulting in decreased beam penetration; therefore, structures in liver parenchyma, such as blood vessels become obscured [34]. Consequently, conventional US has been reported to have low accuracy (53–57%) for detecting low histologic grades of steatosis [35]. Further, conventional US has high inter-/intra-reader variability and is limited in patients with high body mass index (BMI) [30, 34]. To address these limitations, quantitative US-based methods are being developed based on assessment of two fundamental tissue parameters: attenuation coefficient (AC) and backscatter coefficient (BSC) [35].

AC reflects US beam energy loss in tissue [34]. Techniques based on spectral shift-, spectral difference- and hybrid methods have also been developed for the estimation of AC slope attenuation. Some of these methods have moderate diagnostic performance with area under the curve receiver operating characteristics (AUROC) values approaching 0.79 (95% CI 0.676–0.911) and have shown high correlation values with reference standard obtained via

MRI-based proton density fat fraction (MRI-PDFF) $\rho=0.69$ (0.53–0.81) and liver biopsy $\rho=0.55$ (0.34–0.71) [34].

BSC measures US energy ‘returned’ from tissue and is the quantitative analogue to echogenicity in conventional US [35]. Effective scatterer diameter and effective acoustic concentration are among techniques derived from BSC. High correlation values between BSC and MRI-PDFF $\rho=0.72$ (0.57–0.83) and liver biopsy $\rho=0.67$ (0.49–0.79) reference standards were obtained [34].

Among the quantitative US techniques, the controlled-attenuation parameter (CAP) method based on transient elastography implemented by FibroScan® (Echosens™, Paris, France; approved by FDA in 2013), is the most widely studied US-based approach. In a multimodality study in patients with biopsy-proven NAFLD, it was shown that using a threshold of 261 dB/m CAP had an AUROC of 0.85 (95% confidence interval of 0.75–0.96) for diagnosing the presence of steatosis (steatosis grades 1–3 versus 0) [36]. Other studies have compared the diagnostic accuracy of CAP and MRI-methods in steatosis assessment [36–38]. In a direct comparison of CAP and MRI-PDFF, MRI-PDFF performed better than CAP to predict histologic steatosis grade [36]. For diagnosis of any steatosis (grade 0 versus grades 1–3), MRI-PDFF had an AUROC of 0.99 (95% CI 0.98–1.00), CAP of 0.85 (95% CI 0.75–0.96) [36]. AUROC of MR-PDFF was 0.90 (95% CI 0.82–0.97) for differentiating grade 0–1 versus grade 2–3 and 0.92 (95% CI 0.84–0.99) for differentiating grade 0–2 versus grade 3, while the area under the curve value for CAP was 0.7 (95% CI 0.58–0.82) and 0.73 (95% CI 0.58–0.89) respectively [36].

A general methodical limitation of all US-based methods estimating liver fat content, including CAP, is that sonography exploits the attenuation of the propagated and reflected waves. While liver fat attenuates sound waves, many other liver pathologies such as hepatitis, hemochromatosis or fibrosis can also affect sound waves in the same manner [39, 40]. Thus, US-based methods are not specific to fat [39, 40]. Further, the performance of quantitative ultrasound is limited by machine and operator variability, as well as limitations related to obesity [30, 34]. For this reason, the cost advantages of US are greatly diminished by the fact that MRI-PDFF enables accurate estimation of fat content over the entire liver within ~20s [13].

Despite quantitative US being less accurate than MRI, dependent on machine and operator, and its lower sensitivity and specificity in obese patients [30, 34], it is less expensive and generally more accessible than MRI. Future improvements in the detection of lower grades of steatosis may improve its cost-effectiveness.

Computed Tomography

Computed tomography (CT) is a widely available imaging method capable of providing objective assessment of liver fat content [41, 42]. The X-ray attenuation of triglycerides is lower than normal liver parenchyma, leading to lower attenuation of the liver, expressed in Hounsfield units (HU), in the presence of hepatic steatosis [41, 43]. Using CT, the presence of hepatic steatosis can be predicted by absolute attenuation of liver parenchyma value (in

units of HU) or by the relative attenuation difference between liver parenchyma and spleen (Fig. 2) [43–45]. The attenuation value of normal liver should be 50–65 HU on unenhanced CT, 8–10 HU higher than the spleen [46].

For the diagnosis of hepatic steatosis (5% steatosis at histopathology), unenhanced CT has limited sensitivity of 50% and specificity of 77% [47]. Sensitivity and specificity increased for higher grade of steatosis (30% steatosis at histopathology) with 73% and 91% respectively [47]. For grade 2–3 (33% of hepatocytes with intracellular fat vacuoles) hepatic steatosis, the CT sensitivity rises to 93% (with positive predictive value of 76%) with a cutoff 58 HU, and to 73% sensitivity and 100% specificity using a cutoff value of 42 HU, below which a confident diagnosis of hepatic steatosis can be made [46, 48]. Quantification of hepatic steatosis is more accurate using unenhanced CT, compared to contrast-enhanced CT because liver attenuation increases in the presence of iodinated contrast. Further, CT enhancement is impacted by factors such as acquisition timing, contrast dose, contrast type, and altered organ perfusion [45, 46].

Importantly, single-energy CT has been shown to be more accurate than dual-energy CT [43]. Excellent correlation has been shown between single-energy CT and MRS and SECT ($r^2=0.86$), while dual-energy CT achieved only moderate correlation ($r^2=0.42$) [43]. The poor performance of dual-energy CT to detect and quantify hepatic steatosis is likely related to the fact that the energy-attenuation curves for fat are very similar to those for normal liver parenchyma, limiting the ability of dual-energy to differentiate the contribution of fat and liver parenchyma to the overall tissue attenuation.

CT provides a more objective assessment of hepatic steatosis than US [41], but a less objective assessment than MRI or MR spectroscopy (MRS). Excellent correlation has been reported between non-contrast CT and MR-spectroscopy ($r^2 = 0.86$, human study), and MRI-PDFF ($r^2 = 0.99$ phantom study; $r^2 = 0.83$ human study), respectively [41, 43]. However, the correlation between CT and MRS showed lower correlation in patients with PDFF 5.6% assessed by MRS ($r^2 = 0.07$), suggesting that CT may have limited sensitivity for mild hepatic steatosis [43].

An important drawback of CT is use of ionizing radiation, limiting the use of repeated examinations in adults and its use in children and other populations sensitive to ionizing radiation [49]. Further, CT voxel-based attenuation values are confounded by other factors, including the presence of glycogen, iron (Fig. 3), copper, and iodine [46]. Indeed, patients on long-standing amiodarone therapy are well known to have elevated liver attenuation observed by CT (Fig. 3) [50]. Further, the apparent CT attenuation of the liver may be influenced by beam hardening effects in patient with large body habitus and CT acquisition parameters are also known to impact the apparent CT attenuation, including kVp and vendor-specific filters [41, 43, 51]. Finally, differentiation between isolated steatosis and steatohepatitis is not currently possible with CT [52]. For these reasons, CT is not generally recommended for the primary evaluation of hepatic steatosis. However, as abdominal CT is more commonly used than MR for routine abdominal imaging (Fig. 4). CT-derived liver fat quantification is playing an increasingly important role as imaging biomarker of hepatic steatosis in the context of opportunistic screening [42, 53].

Magnetic Resonance

Magnetic resonance (MR) is an imaging modality with a rich array of contrast mechanisms capable detecting and quantifying liver fat content through the detection of the proton signals present in water and in fat [54]. Assessment of hepatic steatosis has evolved from conventional MRI methods that gives qualitative estimates of hepatic steatosis to fully quantitative MRS and MRI methods, which enable accurate and precise measurement of liver fat content [8, 13, 54, 55]. MRS and CSE-MRI, if performed properly can serve as confounder-corrected methods capable of quantifying proton density fat-fraction (PDFF) [56].

PDFF is defined as the ratio of unconfounded signal from the protons of mobile fat to the sum of the unconfounded signal from the protons of mobile fat and unconfounded signal from the protons of mobile water molecules [56]. MRI-based PDFF (MRI-PDFF) is expressed as a percentage (%) ranging from 0 to 100%, [55] correlating closely with histologic steatosis grades [57–60]. Furthermore, PDFF also correlates with the percentage of liver fat content as measured using semi-automatic histologic quantification performed on digitized biopsy specimens [61].

To standardize PDFF as a quantitative imaging biomarker (QIB) across different platforms and improve its value, standardization and practicality the Quantitative Imaging Biomarkers Alliance (QIBA) PDFF group has been recently established [62].

Conventional Magnetic Resonance Imaging

Conventional in-phase opposed-phase (IOP) imaging or fat suppression methods (T1 weighted or T2 weighted fast spin-echo) allows subjective evaluation of hepatic steatosis based on observable signal differences [20]. IOP exploits echo time (TE)-dependent phase interference effect between gradient echo signals of water and fat [20]. Because protons in water and fat molecules precess at different frequencies, they undergo phase interference at predictable intervals. Thus the signals of water and fat add at in-phase (IP) time and cancel at out-of-phase (OP) time [20].

Subjective evaluation of liver fat with IOP is possible within the limited dynamic range of 0–50% fat signal fraction [20]. This limit is generally considered acceptable since fat-fraction >50% in the liver is uncommon, although can occur. IOP allows for qualitative estimation of hepatic steatosis, however, is unsuitable for quantification of liver fat because of presence of confounding factors diminish its accuracy [20]. Furthermore, other conditions must be considered such as presence of concomitant hepatic steatosis and iron overload. The loss of signal intensity on in-phase images caused by iron depositions is a potential pitfall for accurate determination of hepatic steatosis using IOP [54].

Magnetic Resonance Spectroscopy

MRS is an MR-based technique that can directly measure the chemical compositions of the liver fat and quantify hepatic steatosis noninvasively [63, 64]. MRS is long established and widely accepted as the non-invasive reference standard for the liver fat quantification [55, 65, 66]. In a meta-analysis with histopathology as a reference standard, MRS showed mean

sensitivity of 73–89% and mean specificity of 92–96% across different cut-off values (0–33%) [66].

The most commonly used methods are point-resolved spectroscopy (PRESS) and stimulated-echo acquisition mode (STEAM) [64]. PRESS has higher signal to noise ratio (SNR) than STEAM, however STEAM is less affected by J-coupling and is generally preferred [64].

Although long accepted as the noninvasive reference standard for quantification of liver fat [55]. MRS-based PDFF can be challenging for several reasons. MRS is limited by spatial coverage, typically a single voxel (Fig. 5) [54]. Measurements of only small portion of liver can lead to potential bias in livers with heterogenous fat deposition, and negatively impact test-retest repeatability [55]. This source of variability is particularly important for longitudinal studies, because placement of voxel at identical locations in different exams may be difficult, if not impossible, to perform in many circumstances. Furthermore, MRS is limited by its complexity, operator- and device-dependency [13, 20]. Lack of widespread expertise to operate MRS acquisition and evaluation limits implementation in clinical routine and in drug development clinical trials”.

Chemical Shift-Encoded Magnetic Resonance Imaging

CSE-MRI separates MR signal into water and fat components by sampling spoiled gradient echoes at two or more echo times after signal excitation [20]. There are multiple confounders that are addressed by quantitative CSE-MRI methods in order to make accurate estimates of PDFF. These confounders include T1 relaxation [67], T2*decay [68], spectral complexity of fat [69, 70], and eddy currents [71, 72].

There are two main approaches to perform CSE-MRI. These include magnitude- and complexed-based approach [20]. Magnitude-based methods use only the magnitude of the complex MRI signal, are simpler to implement and are insensitive to phase errors, including B0 magnetic field inhomogeneities and eddy currents [72]. However, magnitude-based CSE-MRI has a limited dynamic range of 0–50% PDFF, lower signal to noise ratio (SNR) performance [20].

Complex-based methods utilizes both the magnitude and phase of the acquired MRI signal, thus allowing advantage of a full PDFF range from 0–100% (Fig. 6), higher SNR performance [72], and lower sensitivity to changes in temperature [73]. However, complex-based CSE-MRI suffers from occasional water-fat swaps in the presence of highly inhomogeneous B0 fields [74, 75]. Although PDFF estimation algorithms for complex CSE-MRI are generally more complicated than magnitude-based algorithms, the signal estimation process for both methods is invisible to the user, and both algorithms produce PDFF maps ready for region of interest (ROI) analysis. Unlike magnitude-based CSE-MRI methods, complex-based CSE-MRI methods are FDA approved on all major MRI vendors, for the purpose of quantifying tissue fat content. In comparison to the complicated postprocessing in MRS, CSE-MRI PDFF maps can be reconstructed automatically, and in real-time [20, 65].

CSE-MRI methods have been validated using phantom studies [76, 77], animal studies [78, 79], ex vivo human liver tissue studies [80], and in vivo adult and pediatric studies comparing CSE-MRI to liver biopsy [57, 81, 82] and to MRS [55, 83–86]. Compared to biopsy as a reference standard, MRI-PDFF showed high diagnostic accuracy for diagnosing presence of steatosis (histological grade 1–3) AUROC 0.99 (95% CI 0.98–1.00), and sensitivity of 96% and specificity 100% using a threshold of ~3.7% [36].

Finally, confounder-corrected $R2^*$ ($=1/T2^*$) maps from the same acquisition allows for simultaneous estimation of fat-corrected iron concentration estimates in the liver (Fig. 7), important for the diagnosis of hepatic iron overload in patients with coexisting fatty liver [20, 87].

Analysis of Parametric PDFF Maps

Analysis of the parametric PDFF maps is an area of active investigation and there is currently no consensus on the best approach to derive a summary quantitative PDFF estimates from volumetric PDFF maps acquired over the entire liver. Generally, it is recommended that multiple, large regions of interest (ROI) should be placed at multiple representative locations across the liver, avoiding large vessels, bile ducts and liver lesions. Based on a study conducted by Campo et al., the use of a 4-ROI (medial, lateral, anterior, posterior) or 9-ROI paradigm (Couinaud segments) are preferable for the lowest intra- and inter-observer variability [88]. Automated whole-liver segmentation may be a desirable approach and many groups are actively pursuing liver segmentation for automated PDFF analysis [89, 90]. Finally, we note emerging work that suggests that ROI and segmentation-based PDFF analysis should use the median, rather than the mean estimator to quantify PDFF. Asymmetric noise statistics in estimated PDFF maps can lead to SNR-dependent bias when the average PDFF value is measured from an ROI [91].

MRI-PDFF in Clinical Use

According to QIBA, the most important criteria to characterize a QIB are linearity, bias and precision [92]. In a recent meta-analysis MRI-PDFF demonstrated excellent linearity ($R^2=0.96$) with minimal bias (-0.13%) compared to MRS-PDFF as a reference standard, as well as high precision (reproducibility and repeatability coefficient of 4.1% and 2.9%) across scanner- platforms, manufacturer, imaging centers and field strength [55]. These are all important considerations for widespread dissemination of PDFF and standardization of MRI-based fat quantification [55]. Further, using quantitative biomarkers clinical diagnosis and decisions can be made using cutoff values to classify the presence, absence or severity of disease (eg hepatic steatosis) [54]. Due to the high accuracy and precision, easy post-processing, time effectiveness and wide availability, MRI-PDFF is increasingly recognized as the best method for the detection and accurate quantification of liver fat [55, 93–95] (Fig. 8, Table 1).

With increasing awareness of clinical burden of NAFLD, there is an urgent need for pharmacological therapy for NASH, the progressive form of NAFLD associated with increased risk of hepatic complications. However, despite ongoing investigation, there are currently no effective therapies approved for NASH [13]. The invasive nature of biopsy, its

sampling variability and subjective character of histopathological evaluation of biopsy samples are considered to be a major barriers for drug development for NASH [13].

MRI-PDFF enables quantitative assessment of steatosis, one of the components in NAFLD Activity Score (NAS), a tool developed for measurement of changes in NAFLD during clinical trials [96]. Several scenarios have been proposed for desirable and meaningful use of MRI-PDFF as a potential endpoint in NASH Trials. These include its use in situations when drugs or interventions have a high likelihood of pro- or antisteatotic effect [13]. Furthermore, early use PDFF in clinical trials could influence preliminary drug assessment, and in conjunction with clinical risk factors help identify patients suitable for inclusion in larger invasive biopsy NASH trails [13]. Indeed, MRI-PDFF proved to be useful as a surrogate to liver biopsy in assessing treatment response in a biopsy-proven trial designated to assess the efficiency of colesevelam versus placebo in NASH patients [97], and has been increasingly implemented in other clinical trials [98, 99].

Further acceptance of MRI-PDFF in treatment assessment of NAFLD and NASH will require long-term studies to determine the degree of liver fat reduction that is necessary to reduce the progression and risk of hepatic and extrahepatic complications [13]. In biopsy proven NASH, randomized study on ezetimibe, estimated reduction of ~30% liver fat PDFF was associated with a histologic response defined as 2-point improvement in NAFLD activity Score (NAS) [100]. Further studies are needed to validate this finding and to establish clinically meaningful cutoffs.

Among all imaging techniques, MRI-PDFF is preferred in clinical trials because its practicability, lower sampling variability, equivalence with MRS-PDFF and the ability to obtain PDFF values over the entire liver within a single breath-hold, or even in free breathing [77, 101, 102]. The use of CT in clinical trial is limited due to ionizing radiation. Furthermore, CT is limited by lower diagnostic performance in presence of lower fat content in the liver [43] as discussed above. Although quantitative US techniques do not reach the sensitivity and accuracy of PDFF [36, 43], future advances in the robustness, accuracy and precision of US-based methods are expected.

Conclusion

Hepatic steatosis is no longer considered to be “benign” or “simple”, but rather is recognized as an independent risk factor for progression into NASH with possible hepatic and extrahepatic complications. The increasing prevalence of NAFLD and recognition of the burden of disease, together with the limitations of liver biopsy has led to rapid development of non-invasive imaging methods capable of objective assessment of hepatic steatosis.

In particular CSE-MRI has been shown to be the most accurate and precise non-invasive method for the early identification, and quantification of liver fat content. MRI-PDFF is emerging as an increasingly accepted quantitative biomarker of liver fat, and will continue to transform clinical diagnostic algorithms and drug discovery trials for new treatments for NAFLD in the coming years.

Acknowledgments:

The authors wish to acknowledge support from the NIH (R01 DK088925, R01 DK100651). The authors also wish to acknowledge support from GE Healthcare and Bracco Diagnostics who provide research support to the University of Wisconsin. Dr. Reeder is a Romnes Faculty Fellow, and has received an award provided by the University of Wisconsin-Madison Office of the Vice Chancellor for Research and Graduate Education with funding from the Wisconsin Alumni Research Foundation.

REFERENCES

1. Byrne CD, Targher G (2015) NAFLD: A multisystem disease. *Journal of Hepatology* 62:S47–S64. 10.1016/j.jhep.2014.12.012 [PubMed: 25920090]
2. Estes C, Razavi H, Loomba R, et al. (2018) Modeling the epidemic of nonalcoholic fatty liver disease demonstrates an exponential increase in burden of disease: Estes et al. *Hepatology* 67:123–133. 10.1002/hep.29466 [PubMed: 28802062]
3. Perumpail BJ, Khan MA, Yoo ER, et al. (2017) Clinical epidemiology and disease burden of nonalcoholic fatty liver disease. *WJG* 23:8263–8276. 10.3748/wjg.v23.i47.8263 [PubMed: 29307986]
4. Chalasani N, Younossi Z, Lavine JE, et al. (2018) The Diagnosis and Management of Nonalcoholic Fatty Liver Disease: Practice Guidance From the American Association for the Study of Liver Diseases. 67:30
5. Adinolfi L (2001) Steatosis accelerates the progression of liver damage of chronic hepatitis C patients and correlates with specific HCV genotype and visceral obesity. *Hepatology* 33:1358–1364. 10.1053/jhep.2001.24432 [PubMed: 11391523]
6. Donohue TM Jr (2007) Alcohol-induced steatosis in liver cells. *World J Gastroenterol* 13:4974–4978 [PubMed: 17854140]
7. Nassir F, Rector RS, Hammoud GM, Ibdah JA (2015) Pathogenesis and Prevention of Hepatic Steatosis. *Gastroenterology & Hepatology* 11:167–175 [PubMed: 27099587]
8. Benedict M, Zhang X (2017) Non-alcoholic fatty liver disease: An expanded review. *WJH* 9:715 10.4254/wjh.v9.i16.715 [PubMed: 28652891]
9. Gluchowski NL, Becuwe M, Walther TC, Farese RV (2017) Lipid droplets and liver disease: from basic biology to clinical implications. *Nat Rev Gastroenterol Hepatol* 14:343–355. 10.1038/nrgastro.2017.32 [PubMed: 28428634]
10. Mantovani A, Byrne CD, Bonora E, Targher G (2018) Nonalcoholic Fatty Liver Disease and Risk of Incident Type 2 Diabetes: A Meta-analysis. *Dia Care* 41:372–382. 10.2337/dc17-1902
11. Adams LA, Anstee QM, Tilg H, Targher G (2017) Non-alcoholic fatty liver disease and its relationship with cardiovascular disease and other extrahepatic diseases. *Gut* 66:1138–1153. 10.1136/gutjnl-2017-313884 [PubMed: 28314735]
12. Puchner SB, Lu MT, Mayrhofer T, et al. (2015) High-Risk Coronary Plaque at Coronary CT Angiography Is Associated with Nonalcoholic Fatty Liver Disease, Independent of Coronary Plaque and Stenosis Burden: Results from the ROMICAT II Trial. *Radiology* 274:693–701. 10.1148/radiol.14140933 [PubMed: 25369449]
13. Caussy C, Reeder SB, Sirlin CB, Loomba R (2018) Noninvasive, Quantitative Assessment of Liver Fat by MRI-PDFF as an Endpoint in NASH Trials. *Hepatology* 68:763–772. 10.1002/hep.29797 [PubMed: 29356032]
14. Cairns SR, Peters TJ (1983) Biochemical Analysis of Hepatic Lipid in Alcoholic and Diabetic and Control Subjects. *Clinical Science* 65:645–652. 10.1042/cs0650645 [PubMed: 6627849]
15. Ludwig J, Viggiano T, McGill D, Oh B (1980) Nonalcoholic steatohepatitis: mayo Clinic experiences with a hitherto unnamed disease. *Mayo Clin Proc* 7:434–8
16. Brunt EM, Janney CG, Bisceglie AM, et al. (1999) Nonalcoholic steatohepatitis: a proposal for grading and staging the histological lesions. *Am J Gastroenterology* 94:2467–2474. 10.1111/j.1572-0241.1999.01377.x

17. Wesolowski SR, Kasmi KCE, Jonscher KR, Friedman JE (2017) Developmental origins of NAFLD: a womb with a clue. *Nat Rev Gastroenterol Hepatol* 14:81–96. 10.1038/nrgastro.2016.160 [PubMed: 27780972]
18. Nobili V, Alisi A, Valenti L, et al. (2019) NAFLD in children: new genes, new diagnostic modalities and new drugs. *Nat Rev Gastroenterol Hepatol* 16:517–530. 10.1038/s41575-019-0169-z [PubMed: 31278377]
19. Wilkins T, Tadkod A, Hepburn I, Schade RR (2013) Nonalcoholic Fatty Liver Disease: Diagnosis and Management. *Nonalcoholic Fatty Liver Disease* 88:8
20. Reeder SB, Sirlin CB (2010) Quantification of Liver Fat with Magnetic Resonance Imaging. *Magnetic Resonance Imaging Clinics of North America* 18:337–357. 10.1016/j.mric.2010.08.013 [PubMed: 21094444]
21. Ipsen DH, Lykkesfeldt J, Tveden-Nyborg P (2018) Molecular mechanisms of hepatic lipid accumulation in non-alcoholic fatty liver disease. *Cell Mol Life Sci* 75:3313–3327. 10.1007/s00018-018-2860-6 [PubMed: 29936596]
22. Targher G, Byrne CD (2017) Non-alcoholic fatty liver disease: an emerging driving force in chronic kidney disease. *Nature Reviews Nephrology* 5:297–310. 10.1038/nrneph.2017.16
23. Rinella ME (2015) Nonalcoholic Fatty Liver Disease: A Systematic Review. *JAMA* 313:2263–2273. 10.1001/jama.2015.5370 [PubMed: 26057287]
24. Pembroke T, Deschenes M, Lebouché B, et al. (2017) Hepatic steatosis progresses faster in HIV mono-infected than HIV/HCV co-infected patients and is associated with liver fibrosis. *Journal of Hepatology* 67:801–808. 10.1016/j.jhep.2017.05.011 [PubMed: 28527666]
25. Boyd A, Cain O, Chauhan A, Webb GJ (2020) Medical liver biopsy: background, indications, procedure and histopathology. *Frontline Gastroenterol* 11:40–47. 10.1136/flgastro-2018-101139 [PubMed: 31885839]
26. Jayakumar S, Middleton MS, Lawitz EJ, et al. (2019) Longitudinal correlations between MRE, MRI-PDFF, and liver histology in patients with non-alcoholic steatohepatitis: Analysis of data from a phase II trial of selonsertib. *Journal of Hepatology* 70:133–141. 10.1016/j.jhep.2018.09.024 [PubMed: 30291868]
27. Younossi ZM, Loomba R, Anstee QM, et al. (2018) Diagnostic modalities for nonalcoholic fatty liver disease, nonalcoholic steatohepatitis, and associated fibrosis. *Hepatology* 68:349–360. 10.1002/hep.29721 [PubMed: 29222917]
28. Bedogni G, Nobili V, Tiribelli C (2014) Epidemiology of fatty liver: An update. *World J Gastroenterol* 20:9050–9054. 10.3748/wjg.v20.i27.9050 [PubMed: 25083078]
29. Tapper EB, Lok AS-F (2017) Use of Liver Imaging and Biopsy in Clinical Practice. *N Engl J Med* 377:756–768. 10.1056/NEJMr1610570 [PubMed: 28834467]
30. Ozcan HN, Oguz B, Haliloglu M, et al. (2015) Imaging patterns of fatty liver in pediatric patients. *Diagn Interv Radiol* 21:355–360. 10.5152/dir.2015.14505 [PubMed: 26027765]
31. Poynard T, Lenaour G, Vaillant JC, et al. (2012) Liver Biopsy Analysis Has a Low Level of Performance for Diagnosis of Intermediate Stages of Fibrosis. *Clinical Gastroenterology and Hepatology* 10:657–663.e7. 10.1016/j.cgh.2012.01.023 [PubMed: 22343514]
32. Ratzliff V, Charlotte F, Heurtier A, et al. (2005) Sampling Variability of Liver Biopsy in Nonalcoholic Fatty Liver Disease. *Gastroenterology* 128:1898–1906. 10.1053/j.gastro.2005.03.084 [PubMed: 15940625]
33. Zhang YN, Fowler KJ, Hamilton G, et al. (2018) Liver fat imaging—a clinical overview of ultrasound, CT, and MR imaging. *BJR* 20170959. 10.1259/bjr.20170959
34. Ozturk A, Grajo JR, Gee MS, et al. (2018) Quantitative Hepatic Fat Quantification in Non-alcoholic Fatty Liver Disease Using Ultrasound-Based Techniques: A Review of Literature and Their Diagnostic Performance. *Ultrasound in Medicine & Biology* 44:2461–2475. 10.1016/j.ultrasmedbio.2018.07.019 [PubMed: 30232020]
35. Paige JS, Bernstein GS, Heba E, et al. (2017) A Pilot Comparative Study of Quantitative Ultrasound, Conventional Ultrasound, and MRI for Predicting Histology-Determined Steatosis Grade in Adult Nonalcoholic Fatty Liver Disease. *American Journal of Roentgenology* 208:W168–W177. 10.2214/AJR.16.16726 [PubMed: 28267360]

36. Park CC, Nguyen P, Hernandez C, et al. (2017) Magnetic Resonance Elastography vs Transient Elastography in Detection of Fibrosis and Noninvasive Measurement of Steatosis in Patients With Biopsy-Proven Nonalcoholic Fatty Liver Disease. *Gastroenterology* 152:598–607.e2. 10.1053/j.gastro.2016.10.026 [PubMed: 27911262]
37. Karlas T, Petroff D, Garnov N, et al. (2014) Non-Invasive Assessment of Hepatic Steatosis in Patients with NAFLD Using Controlled Attenuation Parameter and 1H-MR Spectroscopy. *PLoS ONE* 9:e91987 10.1371/journal.pone.0091987 [PubMed: 24637477]
38. Imajo K, Kessoku T, Honda Y, et al. (2016) Magnetic Resonance Imaging More Accurately Classifies Steatosis and Fibrosis in Patients With Nonalcoholic Fatty Liver Disease Than Transient Elastography. *Gastroenterology* 150:626–637.e7. 10.1053/j.gastro.2015.11.048 [PubMed: 26677985]
39. Guiu B (2013) Other Diffuse Liver Diseases: Steatosis, Hemochromatosis, et al. In: Hamm B, Ros PR (eds) *Abdominal Imaging*. Springer Berlin Heidelberg, Berlin, Heidelberg, pp 1027–1044
40. Hamer OW, Aguirre DA, Casola G, et al. (2006) Fatty Liver: Imaging Patterns and Pitfalls. *RadioGraphics* 26:1637–1653. 10.1148/rg.266065004 [PubMed: 17102041]
41. Pickhardt PJ, Graffy PM, Reeder SB, et al. (2018) Quantification of Liver Fat Content with Unenhanced MDCT: Phantom and Clinical Correlation with MRI Proton Density Fat Fraction. *American Journal of Roentgenology* 211:W151–W157. 10.2214/AJR.17.19391 [PubMed: 30016142]
42. Pickhardt PJ, Graffy PM, Zea R, et al. (2020) Automated CT biomarkers for opportunistic prediction of future cardiovascular events and mortality in an asymptomatic screening population: a retrospective cohort study. *The Lancet Digital Health* 2:e192–e200. 10.1016/S2589-7500(20)30025-X [PubMed: 32864598]
43. Kramer H, Pickhardt PJ, Kliever MA, et al. (2017) Accuracy of Liver Fat Quantification With Advanced CT, MRI, and Ultrasound Techniques: Prospective Comparison With MR Spectroscopy. *American Journal of Roentgenology* 208:92–100. 10.2214/AJR.16.16565 [PubMed: 27726414]
44. Hahn L, Reeder SB, del Rio AM, Pickhardt PJ (2015) Longitudinal Changes in Liver Fat Content in Asymptomatic Adults: Hepatic Attenuation on Unenhanced CT as an Imaging Biomarker for Steatosis. *American Journal of Roentgenology* 205:1167–1172. 10.2214/AJR.15.14724 [PubMed: 26587921]
45. Kodama Y, Ng CS, Wu TT, et al. (2007) Comparison of CT Methods for Determining the Fat Content of the Liver. *American Journal of Roentgenology* 188:1307–1312. 10.2214/AJR.06.0992 [PubMed: 17449775]
46. Li Q, Dhyani M, Grajo JR, et al. (2018) Current status of imaging in nonalcoholic fatty liver disease. *WJH* 10:530–542. 10.4254/wjh.v10.i8.530 [PubMed: 30190781]
47. Lee SS, Park SH, Kim HJ, et al. (2010) Non-invasive assessment of hepatic steatosis: Prospective comparison of the accuracy of imaging examinations. *Journal of Hepatology* 52:579–585. 10.1016/j.jhep.2010.01.008 [PubMed: 20185194]
48. Wells MM, Li Z, Addeman B, et al. (2016) Computed Tomography Measurement of Hepatic Steatosis: Prevalence of Hepatic Steatosis in a Canadian Population. *Canadian Journal of Gastroenterology and Hepatology* 2016:1–7. 10.1155/2016/4930987
49. Fazel R, Krumholz HM, Wang Y, et al. (2009) Exposure to Low-Dose Ionizing Radiation from Medical Imaging Procedures. *N Engl J Med* 361:849–857. 10.1056/NEJMoa0901249 [PubMed: 19710483]
50. Patrick D, White FE, Adams PC (1984) Long-term amiodarone therapy: a cause of increased hepatic attenuation on CT. *BJR* 57:573–576. 10.1259/0007-1285-57-679-573 [PubMed: 6733404]
51. Cheng X, Blake GM, Brown JK, et al. (2017) The measurement of liver fat from single-energy quantitative computed tomography scans. *Quant Imaging Med Surg* 7:281–291. 10.21037/qims.2017.05.06 [PubMed: 28811994]
52. Lee SS (2014) Radiologic evaluation of nonalcoholic fatty liver disease. *WJG* 20:7392 10.3748/wjg.v20.i23.7392 [PubMed: 24966609]
53. Chen X, Ma T, Yip R, et al. (2020) Elevated prevalence of moderate-to-severe hepatic steatosis in World Trade Center General Responder Cohort in a program of CT lung screening. *Clinical Imaging* 60:237–243. 10.1016/j.clinimag.2019.12.009 [PubMed: 31945662]

54. Reeder SB, Cruite I, Hamilton G, Sirlin CB (2011) Quantitative assessment of liver fat with magnetic resonance imaging and spectroscopy. *J Magn Reson Imaging* 34:729–749. 10.1002/jmri.22580 [PubMed: 22025886]
55. Yokoo T, Serai SD, Pirasteh A, et al. (2018) Linearity, Bias, and Precision of Hepatic Proton Density Fat Fraction Measurements by Using MR Imaging: A Meta-Analysis. *Radiology* 286:486–498. 10.1148/radiol.2017170550 [PubMed: 28892458]
56. Reeder SB, Hu HH, Sirlin CB (2012) Proton density fat-fraction: A standardized mr-based biomarker of tissue fat concentration. *J Magn Reson Imaging* 36:1011–1014. 10.1002/jmri.23741 [PubMed: 22777847]
57. Tang A, Tan J, Sun M, et al. (2013) Nonalcoholic Fatty Liver Disease: MR Imaging of Liver Proton Density Fat Fraction to Assess Hepatic Steatosis. *Radiology* 267:422–431. 10.1148/radiol.12120896 [PubMed: 23382291]
58. Tang A, Desai A, Hamilton G, et al. (2015) Accuracy of MR Imaging–estimated Proton Density Fat Fraction for Classification of Dichotomized Histologic Steatosis Grades in Nonalcoholic Fatty Liver Disease. *Radiology* 274:416–425. 10.1148/radiol.14140754 [PubMed: 25247408]
59. Middleton MS, Van Natta ML, Heba ER, et al. (2018) Diagnostic accuracy of magnetic resonance imaging hepatic proton density fat fraction in pediatric nonalcoholic fatty liver disease: Middleton et al. *Hepatology* 67:858–872. 10.1002/hep.29596 [PubMed: 29028128]
60. Middleton MS, Heba ER, Hooker CA, et al. (2017) Agreement Between Magnetic Resonance Imaging Proton Density Fat Fraction Measurements and Pathologist-Assigned Steatosis Grades of Liver Biopsies From Adults With Nonalcoholic Steatohepatitis. *Gastroenterology* 153:753–761. 10.1053/j.gastro.2017.06.005 [PubMed: 28624576]
61. Kukuk GM, Hittatiya K, Sprinkart AM, et al. (2015) Comparison between modified Dixon MRI techniques, MR spectroscopic relaxometry, and different histologic quantification methods in the assessment of hepatic steatosis. *Eur Radiol* 25:2869–2879. 10.1007/s00330-015-3703-6 [PubMed: 25903702]
62. QIBA PDFF Biomarker Comitee. In: PDFF Biomarker Comitee. http://qibawiki.rsna.org/index.php/PDFF_Biomarker_Ctte
63. Hamilton G, Yokoo T, Bydder M, et al. (2011) In vivo characterization of the liver fat 1 H MR spectrum: IN VIVO CHARACTERIZATION OF THE LIVER FAT 1 H MR SPECTRUM. *NMR Biomed* 24:784–790. 10.1002/nbm.1622 [PubMed: 21834002]
64. Hamilton G, Middleton MS, Bydder M, et al. (2009) Effect of PRESS and STEAM sequences on magnetic resonance spectroscopic liver fat quantification. *J Magn Reson Imaging* 30:145–152. 10.1002/jmri.21809 [PubMed: 19557733]
65. Kühn J-P, Hernando D, Mensel B, et al. (2014) Quantitative chemical shift-encoded MRI is an accurate method to quantify hepatic steatosis: Quantitative Chemical Shift-Encoded MRI. *J Magn Reson Imaging* 39:1494–1501. 10.1002/jmri.24289 [PubMed: 24123655]
66. Bohte AE, van Werven JR, Bipat S, Stoker J (2011) The diagnostic accuracy of US, CT, MRI and 1H-MRS for the evaluation of hepatic steatosis compared with liver biopsy: a meta-analysis. *Eur Radiol* 21:87–97. 10.1007/s00330-010-1905-5 [PubMed: 20680289]
67. Liu C-Y, McKenzie CA, Yu H, et al. (2007) Fat quantification with IDEAL gradient echo imaging: Correction of bias from T1 and noise. *Magn Reson Med* 58:354–364. 10.1002/mrm.21301 [PubMed: 17654578]
68. Yu H, McKenzie CA, Shimakawa A, et al. (2007) Multiecho reconstruction for simultaneous water-fat decomposition and T2* estimation. *J Magn Reson Imaging* 26:1153–1161. 10.1002/jmri.21090 [PubMed: 17896369]
69. Yu H, Shimakawa A, McKenzie CA, et al. (2008) Multiecho water-fat separation and simultaneous R 2* estimation with multifrequency fat spectrum modeling. *Magn Reson Med* 60:1122–1134. 10.1002/mrm.21737 [PubMed: 18956464]
70. Bydder M, Yokoo T, Hamilton G, et al. (2008) Relaxation effects in the quantification of fat using gradient echo imaging. *Magnetic Resonance Imaging* 26:347–359. 10.1016/j.mri.2007.08.012 [PubMed: 18093781]

71. Hernando D, Hines CDG, Yu H, Reeder SB (2012) Addressing phase errors in fat-water imaging using a mixed magnitude/complex fitting method. *Magn Reson Med* 67:638–644. 10.1002/mrm.23044 [PubMed: 21713978]
72. Yu H, Shimakawa A, Hines CDG, et al. (2011) Combination of complex-based and magnitude-based multiecho water-fat separation for accurate quantification of fat-fraction. *Magn Reson Med* 66:199–206. 10.1002/mrm.22840 [PubMed: 21695724]
73. Hernando D, Sharma SD, Kramer H, Reeder SB (2014) On the confounding effect of temperature on chemical shift-encoded fat quantification: Effect of Temperature on CSE Fat Quantification. *Magn Reson Med* 72:464–470. 10.1002/mrm.24951 [PubMed: 24123362]
74. Hernando D, Haldar JP, Sutton BP, et al. (2008) Joint estimation of water/fat images and field inhomogeneity map. *Magn Reson Med* 59:571–580. 10.1002/mrm.21522 [PubMed: 18306409]
75. Hernando D, Liang Z-P, Kellman P (2010) Chemical shift-based water/fat separation: A comparison of signal models. *Magn Reson Med* 64:811–822. 10.1002/mrm.22455 [PubMed: 20593375]
76. Hines CDG, Yu H, Shimakawa A, et al. (2009) T1 Independent, T2* Corrected MRI with Accurate Spectral Modeling for Quantification of Fat: Validation in a Fat-Water-SPIO Phantom. *J Magn Reson Imaging* 30:1215–1222. 10.1002/jmri.21957 [PubMed: 19856457]
77. Hernando D, Sharma SD, Aliyari Ghasabeh M, et al. (2017) Multisite, multivendor validation of the accuracy and reproducibility of proton-density fat-fraction quantification at 1.5T and 3T using a fat-water phantom: Proton-Density Fat-Fraction Quantification at 1.5T and 3T. *Magn Reson Med* 77:1516–1524. 10.1002/mrm.26228 [PubMed: 27080068]
78. Hines CDG, Agni R, Roen C, et al. (2012) Validation of MRI biomarkers of hepatic steatosis in the presence of iron overload in the ob/ob mouse. *J Magn Reson Imaging* 35:844–851. 10.1002/jmri.22890 [PubMed: 22127834]
79. Hines CDG, Yu H, Shimakawa A, et al. (2010) Quantification of Hepatic Steatosis with 3-T MR Imaging: Validation in ob/ob Mice. *Radiology* 254:119–128. 10.1148/radiol.09090131 [PubMed: 20032146]
80. Bannas P, Kramer H, Hernando D, et al. (2015) Quantitative magnetic resonance imaging of hepatic steatosis: Validation in ex vivo human livers: STEATOHEPATITIS/METABOLIC LIVER DISEASE. *Hepatology* 62:1444–1455. 10.1002/hep.28012 [PubMed: 26224591]
81. Idilman IS, Aniktar H, Idilman R, et al. (2013) Hepatic Steatosis: Quantification by Proton Density Fat Fraction with MR Imaging versus Liver Biopsy. *Radiology* 267:767–775. 10.1148/radiol.13121360 [PubMed: 23382293]
82. Permutt Z, Le T-A, Peterson MR, et al. (2012) Correlation between liver histology and novel magnetic resonance imaging in adult patients with non-alcoholic fatty liver disease - MRI accurately quantifies hepatic steatosis in NAFLD. *Aliment Pharmacol Ther* 36:22–29. 10.1111/j.1365-2036.2012.05121.x [PubMed: 22554256]
83. Idilman IS, Keskin O, Celik A, et al. (2016) A comparison of liver fat content as determined by magnetic resonance imaging-proton density fat fraction and MRS versus liver histology in non-alcoholic fatty liver disease. *Acta Radiol* 57:271–278. 10.1177/0284185115580488 [PubMed: 25855666]
84. Bashir MR, Zhong X, Nickel MD, et al. (2015) Quantification of Hepatic Steatosis With a Multistep Adaptive Fitting MRI Approach: Prospective Validation Against MR Spectroscopy. *American Journal of Roentgenology* 204:297–306. 10.2214/AJR.14.12457 [PubMed: 25615751]
85. Meisamy S, Hines CDG, Hamilton G, et al. (2011) Quantification of Hepatic Steatosis with T1-independent, T2*-corrected MR Imaging with Spectral Modeling of Fat: Blinded Comparison with MR Spectroscopy. *Radiology* 258:767–775. 10.1148/radiol.10100708 [PubMed: 21248233]
86. Pooler BD, Hernando D, Ruby JA, et al. (2018) Validation of a motion-robust 2D sequential technique for quantification of hepatic proton density fat fraction during free breathing: Hepatic PDFF During Free Breathing. *J Magn Reson Imaging* 48:1578–1585. 10.1002/jmri.26056 [PubMed: 29665193]
87. Hernando D, Levin YS, Sirlin CB, Reeder SB (2014) Quantification of liver iron with MRI: State of the art and remaining challenges: Liver Iron Quantification Using MRI. *J Magn Reson Imaging* 40:1003–1021. 10.1002/jmri.24584 [PubMed: 24585403]

88. Campo CA, Hernando D, Schubert T, et al. (2017) Standardized Approach for ROI-Based Measurements of Proton Density Fat Fraction and R2* in the Liver. *American Journal of Roentgenology* 209:592–603. 10.2214/AJR.17.17812 [PubMed: 28705058]
89. Stocker D, Bashir MR, Kannengiesser SAR, Reiner CS (2018) Accuracy of Automated Liver Contouring, Fat Fraction, and R2* Measurement on Gradient Multiecho Magnetic Resonance Images: *Journal of Computer Assisted Tomography* 42:697–706. 10.1097/RCT.0000000000000759
90. Wang K, Mamidipalli A, Retson T, et al. (2019) Automated CT and MRI Liver Segmentation and Biometry Using a Generalized Convolutional Neural Network. *Radiology: Artificial Intelligence* 1:180022 10.1148/ryai.2019180022 [PubMed: 32582883]
91. Roberts NT, Hernando D, Holmes JH, et al. (2018) Noise properties of proton density fat fraction estimated using chemical shift-encoded MRI: Noise Properties of PDFF. *Magn Reson Med* 80:685–695. 10.1002/mrm.27065 [PubMed: 29322549]
92. Raunig DL, McShane LM, Pennello G, et al. (2015) Quantitative imaging biomarkers: A review of statistical methods for technical performance assessment. *Stat Methods Med Res* 24:27–67. 10.1177/0962280214537344 [PubMed: 24919831]
93. Nouredin M, Lam J, Peterson MR, et al. (2013) Utility of magnetic resonance imaging versus histology for quantifying changes in liver fat in nonalcoholic fatty liver disease trials: *Hepatology*. *Hepatology* 58:1930–1940. 10.1002/hep.26455 [PubMed: 23696515]
94. Kim B, Kim SY, Kim KW, et al. (2018) MRI in donor candidates for living donor liver transplant: Technical and practical considerations: Living Donor Liver Transplant. *J Magn Reson Imaging* 48:1453–1467. 10.1002/jmri.26257 [PubMed: 30318644]
95. Parente DB, Rodrigues RS, Paiva FF, et al. (2014) Is MR Spectroscopy Really the Best MR-Based Method for the Evaluation of Fatty Liver in Diabetic Patients in Clinical Practice? *PLoS ONE* 9:e112574 10.1371/journal.pone.0112574 [PubMed: 25426708]
96. Kleiner DE, Brunt EM, Ferrell LD, et al. (2005) Design and Validation of a Histological Scoring System for Nonalcoholic Fatty Liver Disease. 41:9
97. Le T-A, Chen J, Changchien C, et al. (2012) Effect of colesvelam on liver fat quantified by magnetic resonance in nonalcoholic steatohepatitis: A randomized controlled trial. *Hepatology* 56:922–932. 10.1002/hep.25731 [PubMed: 22431131]
98. Cui J, Philo L, Nguyen P, et al. (2016) Sitagliptin vs. placebo for non-alcoholic fatty liver disease: A randomized controlled trial. *Journal of Hepatology* 65:369–376. 10.1016/j.jhep.2016.04.021 [PubMed: 27151177]
99. Loomba R, Sirlin CB, Ang B, et al. (2015) Ezetimibe for the treatment of nonalcoholic steatohepatitis: Assessment by novel magnetic resonance imaging and magnetic resonance elastography in a randomized trial (MOZART trial): Steatohepatitis/Metabolic Liver Disease. *Hepatology* 61:1239–1250. 10.1002/hep.27647 [PubMed: 25482832]
100. Patel J, Bettencourt R, Cui J, et al. (2016) Association of noninvasive quantitative decline in liver fat content on MRI with histologic response in nonalcoholic steatohepatitis. *Therap Adv Gastroenterol* 9:692–701. 10.1177/1756283X16656735
101. Luo H, Wiens C, Shimakawa A, et al. A novel fat and iron quantification technique with non-rigid motion-corrected averaging based on non-local means. 3
102. Pooler BD, Hernando D, Reeder SB (2019) Clinical Implementation of a Focused MRI Protocol for Hepatic Fat and Iron Quantification. *American Journal of Roentgenology* 213:90–95. 10.2214/AJR.18.20947

Fig. 1.

Conventional ultrasound (US) can detect liver fat droplets through increased scattering, which leads to increased liver parenchyma echogenicity, and through increased attenuation, which leads to blurring and poor visualization of deep structures. Shown are four examples of patients with liver fat ranging from normal (a) to increasingly severe hepatic steatosis (b-d). Arrows depict increased obscuration of the diaphragm with increasing severity of steatosis. Note decreased visualization of vessels and bile ducts with increasing liver fat content.

Fig. 2.

Triglycerides have lower X-ray attenuation than normal liver parenchyma, leading to decreased CT attenuation with increasing liver fat content. Shown are four examples of patients with varying degrees of liver fat content assessed by non-contrast CT. Using CT, the degree of hepatic steatosis can be quantified using the absolute CT attenuation of liver parenchyma (Hounsfield units) or by the relative attenuation difference between liver parenchyma and spleen. Examples include a normal liver (a) and patients with moderate (b), severe (c) and extreme (d) hepatic steatosis. HU, Hounsfield Unit. Decreased x-ray attenuation on non-contrast CT correlates closely and linearly with MRI-PDFF [41]. The MRI-PDFF equivalents to 65 HU, 35 HU, 12 HU and -8 HU are approximately 0.5%, 28%, 31% and 43%, respectively.

Fig. 3.

Factors that increase the X-ray attenuation of liver on CT will confound the ability of CT to quantify liver fat due to increased liver density, potentially masking the presence of fat which lowers tissue attenuation. As an example, CT images demonstrate effects of amiodarone (b) and severe iron overload (c) in two patients. In comparison to the patient with normal liver (a), amiodarone and iron increase liver attenuation (b, c) in similar way as hepatic steatosis. Thus, in the presence of amiodarone or iron a concomitant hepatic steatosis could be neither confirmed nor excluded using CT. Note, that due to an increased attenuation of the liver parenchyma liver vessels (depicted by arrows on image b and c) appear hypodense to the parenchyma.

Fig. 4. Longstanding hepatic steatosis can lead to NASH-related cirrhosis. As shown in this 55-year-old patient, severe steatosis (a) was noted 12 years earlier on a CT performed primarily to exclude pulmonary embolism. Follow-up CT showed progression into cirrhosis (b) with liver atrophy and surface nodularity depict by arrows).

Fig. 5.

MR spectroscopy (MRS) can provide confounder-corrected estimates of proton density fat-fraction (PDFF), typically within a single voxel [63]. Axial T2-weighted scout MR image (a) demonstrates the correct placement of the of the MRS voxel (white square), placed in the right lobe of the liver avoiding large vessels, lesions, bile ducts and the liver edge. Figure b shows an example (STEAM) MR spectrum in patient with mild steatosis, acquired at multiple echo times, demonstrating the presence of both water and fat peaks. Post-processing can be used to correct for the effects of T2 signal decay and the multi-peak spectral characteristics of fat.

Fig. 6.

Chemical shift encoded MRI (CSE-MRI) can quantify PDFF as a quantitative biomarker of liver fat content, across the entire liver within a single breath-hold. Shown are representative examples of complex-based CSE-MRI parametric PDFF maps of 3 children. The entire liver was evaluated within only a single 20s breath-hold. PDFF values of the first patient (age 11, male) showed minimal fat content in the liver (normal <6%). PDFF values of the second and third patient (age 11 and 12, male) revealed medium amount of liver fat, consistent with hepatic steatosis in young age.

Fig. 7.

CSE-MRI enables simultaneous estimation of liver fat- and iron concentration. Using confounder-corrected R2* ($1/T2^*$), the R2* maps are reconstructed from the same CSE-MRI acquisition data as PDFF maps. The R2* is quantitative biomarker of iron concentration, clinically relevant in patients who suffer from concomitant NAFLD and iron overload. Coexisting fat and iron deposition in the liver can be also present in viral hepatitis, HCC, hemosiderosis and hemochromatosis. As shown, Patient 1 had high fat but normal iron content in the liver, while patient 2 has both, severe hepatic steatosis and hepatic iron overload. Patient 3 had extreme high iron deposition in the liver due to hemochromatosis, however normal fat content.

Fig. 8.

Among the non-invasive modalities used for quantification of liver fat, MRI-PDFF is the most accurate technique for the evaluation of liver fat content. Shown is an example of conventional ultrasound image (a), CT image (b) CSE-MRI with PDF map (c) of 41-year-old male patient with severe hepatic steatosis. The US beam is attenuated by liver fat, resulting in decreased beam penetration; therefore, structures in liver parenchyma, such as blood vessels (star), and diaphragmatic outline (arrow) are obscured.

Table 1:

Comparison of ultrasound, CT and MRI-PDFF for clinical routine, clinical and drug discovery trials in hepatic steatosis

Modality	Mode of Imaging	Advantages	Disadvantages	Recommendations
Ultrasound	Sound waves	Relatively inexpensive Easy availability No side effects/Safe	Low accuracy for mild steatosis Interobserver variability Sampling variability Operator and vendor dependent Mostly qualitative Confounders: obesity, ascites, fibrosis, edema, hepatitis	Initial screening tool Not suitable for accurate grading and clinical trials
CT	Ionizing radiation	Relatively inexpensive Continuous availability Opportunistic screening Easy analysis Linear correlation CT/PDFF Short acquisition times Quantitative	Ionizing radiation Low accuracy for mild steatosis Acquisition parameters dependent Confounders: iron, amiodaron, glycogen, edema	Suitable for opportunistic screening Unenhanced CT preferable Not suitable for primary diagnostic and clinical trials
MRI-PDFF	Magnetic properties	Good price-performance ratio Direct, precise measurement High sensitivity and specificity Confounder correction possible Complex-base CSE easy analysis Quantitative Short acquisition times	Relative higher price Contraindications for MRI: implantable devices, claustrophobia Relatively limited availability	Imaging method of choice for primary diagnosis, disease monitoring and clinical trials