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Quantification of Liver Iron with MRI: State of the Art and Remaining Challenges

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Abstract

Liver iron overload is the histological hallmark of hereditary hemochromatosis and transfusional hemosiderosis, and can also occur in chronic hepatopathies. Iron overload can result in liver damage, with the eventual development of cirrhosis, liver failure and hepatocellular carcinoma. Assessment of liver iron levels is necessary for detection and quantitative staging of iron overload, and monitoring of iron-reducing treatments. This article discusses the need for non-invasive assessment of liver iron, and reviews qualitative and quantitative methods with a particular emphasis on MRI. Specific MRI methods for liver iron quantification include signal intensity ratio as well as R2 and R2* relaxometry techniques. Methods that are in clinical use, as well as their limitations, are described. Remaining challenges, unsolved problems, and emerging techniques to provide improved characterization of liver iron deposition are discussed.

Keywords

Liver iron; MR relaxometry; R2; R2*; susceptometry

INTRODUCTION

This article reviews current techniques for assessment of liver iron, with a particular emphasis on MRI. The article is divided into the following sections: (1) Pathophysiology of iron overload, (2) Mechanisms of iron overload, (3) Treatment of iron overload, (4) Current methods to assess liver iron that are not MR-based, and (5) MRI-based methods for quantification of liver iron, including remaining challenges and unsolved problems.

After reading this article, the reader should understand the scope of liver disease involving iron deposition, the methods currently used clinically to assess liver iron levels, and be familiar with emerging quantitative MRI methods for measuring liver iron.

PATHOPHYSIOLOGY

Iron is needed by all cells in the human body, particularly cells producing hemoglobin and myoglobin. However, excessive iron accumulation promotes the formation of toxic oxygen radicals that can lead to cell damage (1). Dietary absorption of iron is highly regulated by the body in response to physiologic needs (2). About 1–2 mg of dietary iron is absorbed daily. Iron absorption is performed by enterocytes in the proximal small intestine near the gastroduodenal junction (1,3–5). Under normal physiologic conditions, body iron concentration is maintained in a narrow homeostatic range, about 40 mg/kg in premenopausal women and 50 mg/kg in men (2,6). Approximately 80% of total body iron is functional, located in hemoglobin, myoglobin and iron-containing enzymes, and the remaining 20% is largely found in storage form inside the storage protein ferritin (7–9). Ferritin acts as a depot, storing excess iron (up to 4,500 atoms in a single ferritin polymer), and facilitates mobilization of iron as needed elsewhere in the body (1). Ferritin is found mainly in the liver, spleen and bone marrow (9). A smaller amount of iron is bound to transferrin, which plays an essential role in iron metabolism. Transferrin is involved in transporting iron between absorption, utilization, excretion, reclamation and storage areas (10).

Elimination of iron from the body occurs through sloughing of intestinal lining cells as well as skin cells, and also by menstruation in women (11). Similar to iron absorption, iron elimination also occurs at a rate of 1–2 mg/day (5), but the body does not regulate it. Thus, if excessive iron is delivered into the body, systemic iron overload will develop. Initially, ferritin as well as hemosiderin (a partially denatured form of ferritin) will safely store the excess iron. However, if the storage capacity of ferritin and hemosiderin is reached, free iron will accumulate in the cells of the affected organs (8). Free iron is toxic and can cause cellular and tissue injury (12). In iron overloaded states, the functional iron pool is generally unaffected (9).

Excess delivery of iron into the body can occur due to two primary mechanisms: 1) increased intestinal absorption, and 2) blood transfusions. Both mechanisms lead to systemic iron overload, including overload in the liver. Additionally, diminished mobilization of iron out of the liver can contribute to liver iron overload. The pattern of organ involvement differs depending on the mechanism of the overload, as described below. Further, in many clinical conditions more than one mechanism may be involved.

MECHANISMS OF IRON OVERLOAD

Increased intestinal absorption

Increased intestinal absorption can occur in hereditary hemochromatosis (HH), and conditions that stimulate physiologic gastrointestinal over-absorption (e.g., iron-loading anemias and dietary supplementation). Although there are other genetic mutations that can

lead to iron overload, HH is most often associated with mutations in the HFE gene, resulting in excessive iron absorption in the duodenum (2). The prevalence of homozygous HH is between 0.2% and 0.45% in the Caucasian population (7).

In HH, iron accumulation due to excessive intestinal absorption occurs mainly in organs rich in transferrin receptors, including the liver, heart, thyroid, gonads, pituitary, skin and pancreas (2). Extrahepatic reticuloendothelial system (RES) organs (spleen, marrow and lymph nodes) are relatively spared, except in very severe iron overloaded states when these organs are also affected.

The hallmark of HH is the deposition of hemosiderin in hepatocytes and biliary epithelium. Initially, iron deposition occurs in periportal hepatocytes (zone 1). As the disease progresses, iron deposition extends to midzonal (zone 2) and centrilobular (zone 3) hepatocytes, as well as to the biliary epithelium (11). Kupffer cells are typically spared until later stages in the progression of HH (13). In HH, liver iron concentration (LIC) may be up to 10 times the upper limit of normal, where the 95% upper limit of normal is 1.8 mg Fe/g dry tissue (14).

Conditions associated with increased intestinal absorption tend to have preferential liver manifestations due to earlier accumulation in and damage to hepatocytes. Manifestations include liver injury leading to progressive fibrosis and eventually cirrhosis, and sequelae such as portal hypertension, liver failure and hepatocellular carcinoma. Subsequent accumulation of iron in the pancreas can lead to type 1 diabetes and pancreatic exocrine dysfunction. Accumulation of iron in the heart can lead to cardiomyopathy and sudden death from cardiac arrhythmias. In untreated overload, iron can be visible in the skin, giving it a dark gray or bronze color. This uncommon feature of hemochromatosis has given rise to the name “bronze diabetes.”

Chronic blood transfusions

Patients who undergo chronic blood transfusions, as required for transfusion dependent anemias such as thalassemia and sickle cell disease (SCD), can develop iron overload. Multiple blood transfusions lead to preferential iron deposition in RE cells within the liver, spleen and bone marrow, where iron is initially sequestered as ferritin. Initially, the accumulation of iron in these organs has little clinical importance. However, after sufficient transfusions, the iron storage capacity of the RES (approximately 10g) is exceeded. Since each unit of transfused red blood cells contains about 250 mg of iron, the storage capacity of the RES will be reached after approximately 40 units of blood (7). Subsequent blood transfusions result in clinically relevant accumulation of iron in hepatocytes and in parenchymal cells of the pancreas, myocardium and endocrine glands (7). In iron overload associated with transfusion-dependent anemias, the LIC may exceed 20 times the upper limit of normal.

Clinical findings in transfusional iron overload are dominated by the damaging effects of iron on extrahepatic organs, including the heart and endocrine system. Despite high levels of iron in the liver, there is usually no or only mild hepatic impairment initially because the iron preferentially accumulates within Kupffer cells rather than hepatocytes. Hepatic fibrosis and cirrhosis may occur, but onset may be delayed compared to that observed in cases of

increased intestinal absorption of iron. Cardiac toxicity is the greatest clinical concern in transfusional overload, and death often occurs from cardiomyopathy or cardiac arrhythmias (15–17).

Chronic liver disease

A variety of chronic liver diseases (including chronic viral hepatitis, alcohol-induced liver disease, nonalcoholic fatty liver disease, and porphyria cutanea tarda) can also lead to hepatic iron overload (2,9,18). In chronic liver disease, iron deposition occurs in both ferritin and hemosiderin forms. Iron accumulation usually occurs initially in the RES, but accumulation in hepatocytes is also common (19–21). In chronic hepatopathies, the resulting liver iron accumulation is typically lower than in HH (2,18).

In cases of chronic hepatopathy, the underlying liver condition is the paramount abnormality. However, the presence of excess liver iron has been shown in some studies to contribute to disease progression, and it may portend a worse prognosis (20,22–26).

TREATMENT FOR IRON OVERLOAD

Treatment of iron overload is aimed at reducing body iron stores. The specific treatment will depend on the underlying cause of iron overload. Primary treatment for hemochromatosis is life-long therapeutic phlebotomy, which removes excess iron and prevents iron-mediated tissue damage (9,18,27).

Iron overload in patients with transfusion-dependent anemias cannot be treated with phlebotomy due to the patients' need for blood transfusions. Rather, these patients are treated with chelators, which bind to excess iron and facilitate its removal from the body. Currently, three iron chelators are available: deferasirox, deferiprone, and deferoxamine. Accurate treatment monitoring is needed for both phlebotomy and chelation therapy to maintain sufficiently low body iron levels while minimizing side effects (6,28–30).

QUANTIFICATION OF LIVER IRON

Need for liver iron quantification

Excess accumulation of iron in the liver is toxic, and can lead to liver damage, cirrhosis, liver failure, and hepatocellular carcinoma. Further, LIC (mg Fe/g dry tissue) correlates strongly with total body iron, therefore LIC can be used as a surrogate marker of body iron stores. Although the relation between liver iron stores and total body iron depends on the specific underlying disorder, the liver is the only organ where iron stores increase consistently with increasing total body iron (6).

In HH, liver iron quantification is used to identify individuals suitable for phlebotomy therapy and to monitor response to therapy. Quantitative measurement of liver iron content also provides prognostic information regarding the risk for developing complications such as hepatic fibrosis and cirrhosis (18,31). Further, iron quantification can exclude iron overload in individuals at risk for HH based on genetic studies. Finally, the ability to accurately perform repeated measurements of liver iron may be beneficial in the development of

guidelines for phlebotomy treatment. Although phlebotomy has been in use since 1950 to eliminate excess iron in HH patients, there are no evidence-based guidelines for its use (13).

In transfusion-dependent anemias, liver iron content serves as an indirect measure of total body iron (32) and can be used to guide, monitor and titrate therapy, of particular importance given the significant expense of chelation therapy using deferasirox (33). Further, liver iron also serves as a prognostic biomarker for endocrine and cardiovascular complications in patients with thalassemia. Therefore, measurement of liver iron can be used as a biomarker in research directed at detecting and monitoring HH or transfusional iron overload, as well as a surrogate biomarker in drug trials for drugs designed to treat these diseases. Additionally, in the management of chronic liver diseases, evaluation of liver iron content is considered relevant by many investigators due to the recognition that iron overload may be a co-factor in the progression of these diseases (20,22–26).

Ideally, a liver iron quantification technique should be sensitive and accurate over the entire clinically relevant range of LIC, eg: up to 40mgFe/g dry tissue (14,34). Additionally, it should be widely accessible, fast, inexpensive and noninvasive in order to enable widespread use, treatment monitoring in the clinic, and large studies for research and drug development.

Liver biopsy

The current gold standard to quantify iron overload is non-targeted percutaneous liver biopsy and subsequent measurement of LIC in biopsy specimens using atomic absorption spectrophotometry (11). Unfortunately, atomic absorption spectrophotometry for LIC quantification is available only at specialized centers and is a destructive technique that precludes the use of the specimen for histologic evaluation (9).

Due to the destructive nature of quantitative LIC measurements, iron deposition is more commonly evaluated on histologic sections using a semi-quantitative scale based on Perl's Prussian Blue staining of iron granules. The scoring system of Rowe and colleagues (35) is used frequently. The system is a 5-point scale, with a score assigned based on the magnification at which discrete iron granules can be resolved. A higher score corresponds to resolution of granules at lower magnification. Compared to quantitative LIC measurements, the advantage of semi-quantitative histological assessment is that it can simultaneously evaluate other key pathophysiologic features such as cell injury, inflammation, fibrosis, and parenchymal remodeling.

Regardless of whether biopsy specimens are analyzed quantitatively by spectrophotometry or semi-quantitatively by histology, liver biopsy suffers from two limitations: 1) its sampling variability, and 2) its invasiveness.

- For cirrhotic livers, the coefficient of variation of LIC measurements from two separate biopsies has been reported to be 41% (36); in nondiseased liver, average values of approximately 19% have been reported (37). The small size of the biopsy core and lack of information on spatial distribution may contribute to the relatively high variability of liver biopsy measurements. Therefore, biopsy-based iron

concentration measurements may not be representative of the entire liver, particularly in cases of heterogeneous iron deposition

- The invasiveness of biopsy is a limitation for all patients due to discomfort, anxiety, and rare complications including death (38,39). For these reasons, biopsy is generally not well suited for repeated measurements during treatment monitoring. Further, liver biopsy is contraindicated in some patients with hepatic iron overload, such as those with myelodysplastic syndrome, due to the risk of uncontrolled bleeding.

Due to these limitations of liver biopsy, there is a clinical need for noninvasive iron quantification techniques. These techniques are discussed next.

Non-invasive markers

Serum markers—Serum markers such as serum iron, transferrin and ferritin can be assessed through venipuncture, and provide the simplest and least expensive method to assess body iron stores. While they show some correlation with quantitative LIC (40), however, serum iron markers are often inaccurate and, because they are acute phase reactants, can be confounded by systemic conditions such as malignancy or inflammatory states (6,41–43). Although these limitations are widely recognized, serum markers remain the primary means of diagnosis, and are often used to initiate and modify chelation therapy (33).

Biomagnetic susceptometry—Magnetic susceptibility is a fundamental property of tissues, and is determined by the induced magnetic field generated by the tissue in response to an externally applied magnetic field. Ferritin clusters and hemosiderin iron aggregates are paramagnetic, producing an induced magnetic field in the same direction as the applied field, and with strength about four orders of magnitude smaller than the applied field. This paramagnetic response of iron aggregates is combined in the liver with the diamagnetic response (induced magnetic field in the opposite direction to the applied field) of non iron-loaded liver tissue (6,44–46). The paramagnetic change in susceptibility introduced by the presence of iron can be modeled as a linear function of the iron concentration (45). Thus, measurement of liver susceptibility can enable quantification of LIC.

Liver magnetic susceptibility can be measured non-invasively using a superconducting quantum interference device (SQUID) (6,44,46). A SQUID device is an extremely sensitive detector of magnetic fields. A SQUID-based biosusceptometer consists of a localized magnetic field source and a SQUID detector. To correct for contributions to the measured magnetic field from tissues other than the iron-overloaded liver, differential field measurements are typically made: one with the detector directly next to the abdomen over the liver, and one (baseline) with the detector a few centimeters away from the abdomen, with a container of water placed in between the abdomen and detector. The baseline field measurement arising mostly from water is meant to represent normal body tissue, and the difference between the two field measurements is dominated by the effect of liver iron.

The SQUID has been validated by experiments showing excellent correlation with biopsy-determined LIC (44). Some technical challenges may remain, eg: measuring liver

susceptibility in obese patients with a large volume of subcutaneous fat between the detector and the liver (47). However, the main limitation on the use of this device is its high cost and very limited availability, with only four devices currently available in the world (2,6). For these reasons, SQUID biosusceptometry is currently not widely used for liver iron quantification in clinical practice.

Ultrasound—Ultrasound is not able to detect liver iron overload (48). Although it is not used to directly assess iron overload, ultrasound can be used to assess the sequelae of liver injury related to iron overload, such as cirrhosis, HCC and portal hypertension (9).

Computed Tomography—The overall attenuation of liver in CT is increased in iron-overloaded states (see Figure 1). Iron overload of the liver parenchyma can be suspected on CT when liver density measures >75 HU provided that no IV contrast has been given. Although CT may be able to detect iron overload qualitatively (49,50), increased attenuation in the liver is not specific to iron overload (48). Several confounding factors, including steatosis, glycogen deposition, Wilson's disease, as well as certain drugs (eg: amiodone or gold) may alter hepatic attenuation (51–55). Recent studies have shown that dual-energy CT may be able to provide improved assessment of liver iron even in the presence of steatosis (51). However, due to exposure to ionizing radiation, CT is not optimal for repeated measurements during treatment monitoring.

Because of the limitations of these techniques, and due to the widespread availability of MRI scanners, significant efforts have been made to assess liver iron content using MR based methods.

MR-BASED METHODS TO ASSESS LIVER IRON

MRI is very sensitive to the presence of iron

The presence of iron in tissue affects the MRI signal in multiple ways: iron, typically in the form of ferritin and hemosiderin, shortens the relaxation times $T2^*$, $T2$ and $T1$ (ie: increases in the relaxation rates $R2^*$, $R2$ and $R1$). Additionally, because iron in the body is paramagnetic, it affects the susceptibility of tissue, resulting in macroscopic changes in the $B0$ field (the basis of SQUID biosusceptometry). The most prominent effect of iron is increasing $R2$ and $R2^*$ relaxation rates (56):

- $R2$ effects: $R2$ is well known to increase in the presence of tissue iron, although the exact mechanism is not well understood. Two explanations have been offered for the observed $R2$ increase with iron concentration. The first explanation is based on the fact that spins within an individual voxel are not stationary but undergo diffusion between excitation and echo generation. Because tissue iron introduces microscopic magnetic field inhomogeneities, each spin moves through an inhomogeneous field, and therefore its precession frequency changes between the excitation and the echo. Due to the random nature of diffusion, the phase accumulated by the spins prior to the refocusing pulse is not completely re-phased after the refocusing pulse (56,57). According to this explanation, the microscopic $B0$ inhomogeneities introduced by the presence of iron essentially result in a

diffusion weighting of spin-echo images, and this diffusion weighting increases with iron concentration. A second explanation is that relaxation occurs through proton chemical exchange between bulk water and exchangeable protons bound to iron-containing proteins. Iron electrons enhance the relaxation of the protein-bound water protons. Through chemical exchange, enhanced relaxation of bound water protons is then transferred to the bulk water protons, leading to R2 increase of bulk water (8,58,59). Regardless of the specific mechanism, it has been shown that R2 increases monotonically (although not linearly) with LIC (14,56).

- R2* effects: iron introduces microscopic B0 field inhomogeneities that result in rapid signal dephasing and increased rate of R2* relaxation in gradient-echo images. As has been shown both theoretically (56) and empirically (34), the relationship between R2* and liver iron concentration can be well modeled linearly ($LIC = aR2^* + b$).

Liver iron overload can also be assessed qualitatively by acquiring T2- or T2*-weighted MR images (see Figure 2):

- T2-weighted images, typically acquired using fast spin echo (FSE) sequences, can be used to detect iron overload. This appears as reduced signal intensity (compared to the non iron-overloaded case) in the liver and other affected organs (eg: the spleen), as a consequence of accelerated T2 decay. The optimum choice of echo time to visualize iron overload depends heavily on the expected LIC.
- T2* weighted images historically were obtained as single-echo gradient echo (GRE) sequences with long echo times (10–15 ms) to impart T2* weighting. In addition, opposed-phase and in-phase (OP and IP) GRE imaging can be used to detect liver iron overload. In OP and IP imaging, two GRE images are acquired with echo times where the water and main methylene fat signals are in opposed-phase and in-phase, respectively. Although OP and IP imaging was developed for liver fat detection, the second echo has a longer echo time than the first echo and therefore is more T2* weighted. The difference in T2* weighting between echoes can be exploited for iron detection. Most commercial 1.5T scanners implement the dual-echo sequence using an OP-IP sequence design, with the first echo obtained at around 2.3 ms (OP at 1.5T), and the second at around 4.6 ms (IP at 1.5T). On such sequences, appreciable signal loss between the first echo (OP) and the second echo (IP) indicates short T2* decay and suggests the presence of parenchymal iron. The normal liver has a long T2* relaxation time (greater than 20 ms) relative to the echo times acquired and loses only minimal signal intensity across closely spaced gradient echoes. Iron accumulation shortens the transverse relaxation and increases signal loss. Concomitant liver steatosis may confound the interpretation, however, because signal loss due to fat-water signal cancellation on the first echo (OP) may mask or even dominate R2* signal decay on the second echo (IP). Depending on the relative amounts of fat and iron in the liver, iron detection may fail completely (figure 3).

Although detection of liver iron using qualitative MR imaging is possible, qualitative methods do not reliably assess the degree of iron overload and cannot reliably guide

treatment initiation or therapy monitoring. These applications require quantitative methods that estimate LIC based on MR measurements. Ideally, such methods would estimate LIC accurately over its entire clinical range from 0.2 mg Fe/g dry weight to more than 40 mg Fe/g dry weight (14) in a manner that is robust to changes in protocol, platform-independent, and reproducible across most clinical MR imaging platforms.

Several quantitative MR-based strategies have been employed to measure LIC: signal intensity ratio (SIR) techniques based on T_2 -weighted or T_2^* -weighted imaging, quantitative relaxometry (largely R_2 - and R_2^* -based), and MR susceptometry. These techniques are reviewed next.

Signal intensity ratio techniques

In SIR methods, the signal intensity of the liver on spin-echo or GRE sequences is divided by the signal intensity of a reference tissue that does not accumulate iron, such as fat or skeletal muscle, or air outside the body (i.e., image noise) (60–62). Images are acquired using a body coil to avoid sensitivity variations arising from surface coils (48). Large regions-of-interest (ROIs) are placed in the reference object and the liver on the same slice, and the ratio of the mean signals is calculated.

The most widely used SIR method is the one described by Gandon and colleagues (48). In this method, five breath-hold GRE sequences are obtained in separate breath-holds, keeping the TR constant but using different flip angles (20° or 90°) to alter T_1 -weighting and different TEs (between 4–21 ms) to alter T_2^* -weighting. These five images result in different T_1 - and T_2^* weightings that can be used to measure liver iron. Specific protocols have been optimized at different field strengths (63). Liver and muscle signal intensity measurements are performed on three ROIs within the right lobe of the liver, and in two ROIs in the right and left paraspinal muscles, respectively. Thus, each of the five sequences results in a different liver/muscle signal intensity ratio. These five values are then combined to provide an LIC estimate using a specialized algorithm (<http://www.radio.univ-rennes1.fr/Sources/EN/HemoCalc15.html>). Calibrations are available at several field strengths up to 1.5T, but are not available at 3T.

The Gandon technique was evaluated in 149 patients, resulting in accurate LIC estimates (mean difference of 0.8 $\mu\text{mol/g}$, 95% confidence interval of [6.3,7.9]) over a range of LIC values from 3 to 375 $\mu\text{mol Fe/g dry weight}$ (0.17 to 20.9 mg Fe/g dry weight). This method saturates for LIC beyond 20.9 mg Fe/g dry weight, likely due to the relatively long TEs employed.

Although the Gandon method is used clinically, it has important limitations. The liver-to-muscle ratio depends on acquisition parameters. Therefore, standardization of parameters is necessary. Even with standardized parameters, the Gandon method results may depend on the scanner type (64). Further, it is unclear how factors such as liver steatosis affect SIR measurements. Although chemical shift based fat saturation can reduce the influence of fat on liver signals, this approach is complicated in subjects with severe iron overload or large body habitus. Finally, the Gandon method cannot quantify LIC values greater than 20.9 mg/g (375 $\mu\text{mol/g}$) and hence does not capture the entire relevant range of liver iron levels

(14), although refinements of this method have been developed to extend the dynamic range (65).

R2 relaxometry techniques

By measuring relaxation parameters that are potentially independent of scanning platform and coil configuration, MR relaxometry techniques may be able to overcome some of the challenges of SIR techniques. R2 relaxometry can be performed through acquisition of spin-echo data with increasing TEs. Specific acquisition and reconstruction techniques are described below.

Pulse sequence design—R2 can be measured with a series of single spin-echoes (SSE), each acquired after a separate excitation, or with a train of spin-echoes acquired after a single excitation using a Carr-Purcell-Meiboom-Gill (CPMG) sequence. R2 measurements made with a CPMG sequence will be lower than those made with a series of single spin-echoes (due to more frequent application of refocusing pulses) and will also depend strongly on the spacing between refocusing pulses (12,57). Spin-echo data can be acquired with or without fat suppression (66). Figure 4 illustrates the process of R2 relaxometry in subjects without and with iron overload.

Choice of TE—TEs should be selected such that the range of expected clinically relevant R2 values (eg: 17–400 s⁻¹ at 1.5T) can be reliably measured (67). Optimally, the first echo time should be as short as possible: 5 ms or less (67). The last echo time should be as long as reasonable without degradation by motion and other artifacts. Often, the longest acquired TE is 15–30 ms. Although the optimal number of echoes has not been determined, a reasonable approach is to acquire as many echoes as possible (within acquisition time limitations) between the first and last echoes, and to space the echoes uniformly or logarithmically.

R2 estimation signal models—R2 can be estimated from spin-echo signals acquired at multiple echo times by modeling the decay of the spin-echo signal magnitude with TE as a mono-exponential decay. Alternatively, more than one signal component can be included in the estimation. Bi-exponential modeling of the spin-echo signal is sometimes used to estimate two separate R2 components (fast and slow R2 decay components, respectively) (68,69). Bi-exponential signal modeling may be more accurate than mono-exponential modeling, although it also requires better SNR or more TEs in order to enable stable estimation.

More sophisticated models of T2 decay have been proposed for different applications, including estimating a distribution of R2 values (70). However, these models require acquisition of many TEs with very high SNR and their use has been limited mainly to brain imaging.

Existing MRI-based methods and calibrations—Early correlation studies showed a linear relationship (71–74) between R2 and LIC. However, these studies had moderate numbers of subjects, which may have complicated the detection of nonlinear relationships between R2 and LIC. A nonlinear relationship was calibrated by St Pierre et al. (14) in over 100 patients.

The St Pierre method uses 5 T2-weighted SSE free-breathing sequences with constant repetition time (TR) and increasing TE spaced at 3-ms intervals (TEs=6,9,12,15,18ms). Correction for instrumental gain drift (affecting image magnitude) between sequences acquired at different TEs is accomplished by placement of an external calibration phantom with very long T2 within the field of view (14). Postprocessing steps that include gain drift correction, respiratory motion correction, background noise subtraction, estimation of effective initial signal intensity at TE = 0, and bi-exponential modeling pixel by pixel (after smoothing voxel intensities over a 5 × 5 window kernel to reduce image noise) are required (12,69). For each pixel, the biexponential model estimates two R2 values corresponding to the short T2 and long T2 components, respectively. An average of the two R2 values, weighted by their respective population densities, is calculated as a composite R2 value at each voxel. A slice with large liver cross-section and without major motion artifacts is then selected and the mean composite R2 value in the liver is measured by placing a large ROI, excluding vessels and artifacts.

In a validation study of this technique, the investigators evaluated more than 100 patients with LIC values ranging from 0.3 to 42.7 mg Fe/g dry weight. Liver R2 demonstrated a curvilinear relationship with LIC over the entire LIC range with a correlation coefficient of 0.98 (14) and limits of agreement between -56% and 50%. St Pierre et al derived an empirical expression for R2 in terms of LIC that reflects this curvilinear relationship (figure 5). Wood et al (34) inverted this expression to enable prediction of iron concentration from a given R2 measurement:

$$[Fe] = (29.75 - \sqrt{900.7 - 2.283R2})^{1.424} \quad [1]$$

This empirical equation was derived at 1.5T for a specific set of imaging parameters and for the analysis method described by St. Pierre. This approach may not be generalizable to other sequences that measure R2. Notably, however, Wood et al (34) were able to reproduce the same calibration curve obtained by St. Pierre using different imaging parameters. Specifically, they used a 120° – 120° echo rather than conventional 90° – 180° spin echo, they fit the data with a monoexponential decay model with constant offset rather than a biexponential decay model, and they used a TE range of 3.5 to 30 ms rather than 6 to 18 ms.

An important advantage of the St. Pierre method is that it has received FDA approval, and is commercially available as FerriScan®, provided by ResonanceHealth®. The company provides fee-based LIC analysis of images acquired using their protocol. A practical limitation of this method is cost; in addition to the fee charged by ResonanceHealth, the method requires 10–20 minutes of imaging time.

The effects of TR on the R2 measurements provided by the St. Pierre method have been assessed by Pavitt et al. (75). In this study, the authors compared R2 measurements obtained with TR=1000 ms and TR=2500 ms. A systematic difference between R2 measurements obtained with the two protocols. It was concluded that either protocol can be used to quantify liver iron, but a conversion factor is needed to “translate” R2 measurements obtained with one protocol to the other. Recently, the reproducibility of the St. Pierre method has been validated in a multi-center study (76). In this study, the calibration of this

method (FerriScan) was validated in 233 iron-overloaded β -thalassemia patients. It was found that the calibration curve was independent of scanner platform, patient age, liver fibrosis stage, inflammation grade, and use of chelator therapy, although the limits of agreement between R2-based LIC and biopsy-based LIC were very broad (between 74% and -71%).

Spectroscopy-based—An alternative approach for measuring liver R2 in a single breath-hold consists of acquiring single-voxel MR spectroscopy (MRS) at increasing TEs. Wang et al (74) acquired single-voxel spectra at multiple echo times in human subjects with varying degrees of liver iron overload. The authors used the liver water peak to measure R2 relaxation values, and these values showed high correlation with corresponding LIC values measured from synchronous liver biopsies. More recently, several methods have been developed that permit measurement of liver fat and R2 (separately for water and fat signal components) in a single breath-hold, using stimulated echo acquisition mode (STEAM) spectroscopic sequences (77–80). These MRS methods have the advantage of enabling rapid acquisitions with relatively high SNR, but the disadvantage of not providing any anatomical information, which may be a limitation in cases of heterogeneous iron deposition (7). Figure 6 shows examples of liver STEAM spectroscopy datasets in patients with varying degrees of iron overload.

Field strength dependence of R2—Although most R2-LIC calibrations have been derived at 1.5T, some studies have analyzed the effect of field strength on R2 in the presence of liver iron. For instance, Bulte et al (68) studied R2 as a function of chemically-determined liver iron. In this work, the authors observed a biexponential R2 decay for LIC > 2mg Fe/g wet weight, and a nearly linear increase of the rapid R2 decay component with field strength.

Novel R2 relaxometry techniques for advanced liver iron characterization—

One of the main limitations of most MR-based liver iron quantification techniques is the inability to distinguish between different types of iron deposition, particularly ferritin and hemosiderin. It has long been recognized that hemosiderin iron deposits introduce non-monoexponential relaxation in CPMG echo trains. The CPMG signal can be described using a non-exponential decay model including contributions from hemosiderin as well as ferritin (57). This model explicitly includes the effects of CPMG echo spacing on observed signal decay, which vary in the presence of hemosiderin. This signal behavior can potentially be exploited to provide further characterization of liver iron deposition beyond LIC. A careful analysis of CPMG signals obtained with varying echo spacing has been proposed as a method to separately quantify ferritin and hemosiderin liver iron deposition (81,82). In Ref. (82), Tang et al used three different echo spacings (4ms, 8ms, 15ms) to enable separate estimation of ferritin and hemosiderin in 6 healthy volunteers and 20 subjects with transfusional iron overload.

Limitations—The nonlinear relationship between R2 and LIC introduces complexity into the calibration. Also, the slow increase in R2 with LIC at high LIC values may result in reduced precision of R2-based LIC measurements with severe iron overload. However, the

main limitation of R2-based liver iron quantification is the long acquisition times required for R2 mapping, particularly using SSE imaging sequences. In addition, these long acquisition times result in potentially severe motion artifacts in free-breathing acquisitions. These motion artifacts currently require careful manual delineation of ROIs in the St. Pierre method, complicating the workflow of iron quantification. Motion artifacts could be avoided by respiratory triggering, although at the cost of further acquisition time increases.

R2* relaxometry techniques

R2* relaxometry, based on gradient echo acquisitions, has the potential to overcome some of the limitations of R2-based techniques, due to its ability to provide full liver coverage without motion artifacts within a single breath-hold.

Pulse sequence—R2* relaxometry is typically based on 2D or 3D spoiled gradient echo (SPGR) multi-echo sequences. These sequences can acquire all echoes in each TR using an echo train (either with monopolar or bipolar readout), a single echo per TR, or a combination of the two (ie: multiple interleaved echo trains). Subsequently, R2* is measured from the rate of exponential signal decay of the gradient echo signal. Fitting of R2* can be performed on either a voxel-by-voxel basis or after averaging the measured signal within a region of interest (ROI). Figure 7 illustrates the process of R2* relaxometry in subjects without and with iron overload. Figure 8 shows R2* maps from a 21 year old patient undergoing chelation therapy.

Choice of echo times—TEs should be selected to maximize the ability of fitting algorithms to reliably measure the expected clinically relevant values of R2* (eg: 33–2000 s⁻¹ at 1.5T) (67). Optimally, the first echo should be as short as possible (1 ms or less) and the echo spacing should be short enough to capture the signal decay in cases of severe iron overload (eg: 1 ms). The total number of echoes and choice of last echo time depends on the precision requirements for low iron values, but typically 10–15 ms is sufficient (83). It may be necessary to utilize a small frequency matrix (192 or 224), fractional echo sampling, and high receive bandwidth (>100 kHz) to achieve the appropriate echo time combination.

R2* estimation—Estimation of R2* values from multi-echo SPGR data is generally performed by fitting an R2* signal model to the acquired data. There are a number of choices regarding the estimation process, including:

- **Signal model:** The multi-echo SPGR signal is usually modeled as an exponential decay with decay rate R2*. Exponential model fitting can be performed using either the complex signal, or the signal magnitude (83). An important disadvantage of magnitude-based methods is that magnitude data contains a “noise floor” with nonzero mean at low SNR due to the non-Gaussian noise distribution of magnitude MR signals. This noise floor can introduce systematic errors (underestimation) in magnitude-based R2* estimation, as shown in figure 9A-9B. The noise floor effect is particularly relevant for liver iron quantification because in the presence of severe iron overload R2* decay can be extremely rapid, with R2* of 1000 s⁻¹ or higher (ie: T2* of 1 ms or shorter).

In order to overcome noise floor effects, several approaches can be used: 1) baseline fitting, based on approximately modeling the noise floor as a constant offset (84); 2) signal truncation, (85,86), where echoes considered to be below the noise floor (at the end of the echo train) are discarded, or 3) complex fitting which obviates such errors as discussed below. Other methods, such as filtering-based techniques (87), have been introduced to reduce noise effects in magnitude fitting $R2^*$ relaxometry, and have been shown to work well in practice.

Complex fitting based techniques do not suffer from noise floor effects because the Gaussian distribution of the noisy signal is preserved (88). For this reason, complex $R2^*$ fitting provides accurate estimates of $R2^*$ without the need for complicated filtering algorithms (83).. A disadvantage of complex $R2^*$ fitting is that it requires the signal phase to be reliable, and it may be problematic in cases where eddy currents cause inconsistent phase shifts in different echoes (eg: bipolar acquisitions, multiple interleaved echo trains, or echo trains with unevenly spaced TEs). Recently, phase-corrected chemical shift encoded acquisitions have been developed that can address these phase shifts (89).

- Correction for fat: Hepatic fat accumulation is a common condition, affecting about 20–30% of the US population (90). In the presence of water and fat signal components, the signal acquired at a single voxel is not a simple monoexponential decay, but instead contains oscillations as water and fat signals become in and out of phase. These oscillations lead to errors in fat-uncorrected $R2^*$ estimation using an exponential signal model. Note that these errors depend heavily on the choice of TE combination (figure 9C-9D). Fat-corrected $R2^*$ measurements can be obtained by jointly performing fatwater separation and $R2^*$ estimation (83,91).

Similar to the fat-uncorrected case described above, fat-corrected $R2^*$ estimation can also be performed from complex or magnitude data. Noise-floor effects also affect magnitude-based fat-corrected $R2^*$ mapping. However, correction methods such as baseline fitting or truncation are not commonly used, likely because they introduce additional complexity and noise instability to the estimation problem.

It is possible to partially address the effects of fat in $R2^*$ mapping by acquiring images at TEs where the water and main methylene resonance of fat (-217 Hz from water peak at 1.5T) are “in-phase” (eg. 4.6ms, 9.2ms, etc, at 1.5T) (92,93). However, this approach does not account for the spectral complexity of the fat signal, and it has been shown recently that “in-phase” echoes do not permit accurate $R2^*$ estimation (ref Kuhn). In additional, this approach requires the acquisition of widely spaced echoes (4.6ms at 1.5T), which are not able to capture rapid signal decay in cases of severe iron overload (83,91).

Fat-suppressed $R2^*$ acquisitions are also possible, using either T1 based short-tau inversion recovery (STIR) (94), or chemical shift based techniques (95,96). However, these techniques may suffer from SNR loss or inaccurate fat suppression in the presence of T1 shifts, B0 inhomogeneities and peak broadening introduced by the presence of iron.

These limitations can be overcome by acquiring fat-unsuppressed SPGR images with short TEs, and performing simultaneous fat-water separation and $R2^*$ estimation (ie: fat-corrected

R2* estimation). As has been shown in recent studies, fat-corrected R2* mapping can provide robust (83,91), accurate (97) estimates of liver iron. Further, fat-corrected R2* estimation results in no SNR penalty in R2* mapping over a wide range of acquisition parameters and R2* values (83).

- Background B0 field variations: The presence of intra-voxel background field variations introduces additional signal decay in R2* mapping acquisitions. This additional signal decay can result in significant bias in R2* measurements, particularly near tissue-air interfaces such as near the dome of the liver. Techniques have been developed to address background field variation effects in R2* mapping by using complex data, estimating the field inhomogeneity and removing its effects (93,98–102). Additionally, it is likely that the effect of background field variations can be minimized if acquisitions are performed with high enough spatial resolution or if R2* measurements are obtained from “favorable” locations within the liver away from tissue-air interfaces (102).

Status of development and validation—Several studies have performed calibration of liver R2* (in s⁻¹) at 1.5T to biopsy-measured LIC (in mg Fe/g dry tissue). One such study by Wood et al calibrated R2* to LIC in 20 patients (34). To measure R2* with short TE_{init} and short TE, this study used single-echo gradient-echo acquisitions within a single breathhold, stepping TE from 0.8 to 4.8 ms in 0.25 ms intervals, for a total of 17 acquisitions. Other acquisition parameters were FOV of 48 × 24, flip angle 20, TR 25 ms, imaging matrix 64 × 64, and slice thickness 15 mm. Data were fitted to a mono-exponential decay curve with constant offset on a pixel-by-pixel basis, and R2* maps were obtained.

In this study, R2* estimates were compared to biopsy-determined LIC in 20 patients (with a total of 22 biopsies) with transfusion-dependent thalassemia or sickle cell disease. For those patients with LIC values ranging from 1.3 to 32.9 mg iron/g dry weight (19 of the 20 patients), a linear relationship between R2* and LIC was derived, with a correlation coefficient of 0.97 and limits of agreement of -46% to 44%. This relationship is described by the empirical equation

$$[Fe] = 0.202 + 0.0254R2^* \quad [2]$$

Equation 2 has been validated only for the particular field strength, imaging parameters, and reconstruction technique used by Wood; it cannot yet be generalized to other methods of R2* measurement. Of note, other authors who have attempted to measure R2* as a function of LIC have derived different calibration curves (103–106), likely due to differences in fat content, susceptibility, fitting methods, and/or differences in the biopsy-LIC measurements.

The main concern for R2*-based liver iron quantification has been the presence of confounding factors, which can lead to poor robustness (variability with different acquisition protocols), and poor reproducibility (variability with scanning platform). Several recent research projects have aimed to address this concern.

Inter-scanner and inter-site reproducibility of liver and cardiac T2* measurements have been assessed in several studies, demonstrating good reproducibility (107–110). Kirk et al (111)

assessed the inter-center reproducibility of liver and cardiac $T2^*$ in 49 patients. In this study, patients were scanned at different sites (in different countries) using breath-hold sequences on 1.5T scanners from different vendors. The study showed that, using consistent acquisition protocols (ie: similar acquisition parameters) and $T2^*$ quantification techniques, $T2^*$ measurements in heart and liver had good inter-center reproducibility (coefficient of variation $<6\%$ in both cases).

These reproducibility studies have largely focused on assessing reproducibility of $R2^*$ (or $T2^*$) measured with a similar acquisition and estimation technique at different sites. However, different studies (using different $R2^*$ quantification techniques) have produced different $R2^*$ -LIC calibrations. Recent results have shown that calibrations from different studies can be “translated”, overcoming some of the limitations of $R2^*$ mapping (112).

Further, liver $R2^*$ estimates corrected for all relevant confounding factors have been shown to be robust to changes in acquisition protocol at both 1.5T and 3T in normal volunteers as well as patients with iron overload (113). In addition, several multi-scanner and multi-center studies have shown good reproducibility of $R2^*$ mapping across sites (107–109,111). However, larger multi-center studies in iron overloaded populations are needed in order to establish the reproducibility of liver iron quantification using $R2^*$ -MRI across vendors and sites.

Field strength dependence of $R2^*$ —Most $R2^*$ calibration studies have been performed at 1.5T. Fortunately, the relationship between $R2^*$ measurements at 1.5T and 3T has been shown to be a factor of 2 (114). Therefore, liver iron quantification at 3T should also be possible (115,116). The key limitation of $R2^*$ -based iron quantification at 3T is the rapid decay of the signal at this field strength. This rapid decay may limit the dynamic range for accurate $R2^*$ -based iron quantification at 3T.

Limitations—Techniques that do not adequately correct for confounding factors will likely suffer from poor robustness (ie: $R2^*$ measurements will depend on the specific acquisition parameters). However, this is not a fundamental limitation of liver $R2^*$ mapping, as confounder-corrected $R2^*$ techniques have been developed where all relevant confounding factors are corrected. Nevertheless, cases of extremely rapid signal decay may impose a limitation for accurate liver iron quantification in the presence of massive iron overload (eg: LIC >30 mg Fe/g dry tissue), even at 1.5T. Conventional Cartesian acquisition techniques are inherently limited in the shortest TE_{init} that can be achieved. Ultra-short TE techniques based on radial acquisitions have been proposed to extend the dynamic range of $R2^*$ relaxometry techniques (104,117).

Performance of relaxometry-based MRI techniques

As described above, MRI relaxometry ($R2$ or $R2^*$) is based on acquiring images with increasing echo times using spin-echo or gradient-echo sequences, respectively. A relaxation signal model (as a function of echo time) is then fitted to the acquired images. Measurements in MRI relaxometry are often performed by fitting the signal model to the acquired signal at each voxel, followed by averaging the estimated voxel-wise relaxation parameter values over a ROI, ie: a “fit-first” approach. In order to address noise effects,

some techniques average the acquired signal over a manually-delineated ROI at each TE before fitting the corresponding signal model to the averaged signal (ie: “average-first”). It is unclear whether fit-first or average-first is superior in practice, in terms of bias and noise performance (67,118). One important advantage of fit-first relaxometry techniques is the ability to provide maps depicting iron deposition in different organs (figure 10) as well as the spatial distribution of iron within a given organ such as a liver (figure 11).

Among established relaxometry methods, the limits of agreement between R2 derived using the St Pierre method (–56% to 50%) or R2* derived using the Wood method (–46% to 44%,) and LIC were very broad. Both St. Pierre et al and Wood et al have speculated that broad confidence intervals may be attributable in part to spatial variability of iron concentration within the liver, as noted previously. However, a potentially contributing factor is that relaxometry does not directly measure iron content, but rather measures an MR relaxation rate (R2 or R2*). This relaxation rate depends not only on iron concentration but also on numerous other variables including iron particle size, shape, and distribution (119–122) as well as coexisting conditions such as steatosis, inflammation, and fibrosis. Hepatic fibrosis and inflammation are common in patients with iron overload (123,124). These conditions result in a decrease in R2 relaxation rate, which may partially offset the R2-lengthening effect of iron. Additionally, restricted diffusion of water molecules in liver parenchyma is associated with inflammation and fibrosis (125). This may reduce the sensitivity of R2 relaxation to the presence of iron because diffusion contributes to iron-mediated relaxation mechanisms (56,57).

Although there is not yet consensus regarding the optimal relaxation rate parameter (R2 or R2*) for LIC estimation, R2 and R2* values generated comparable noninvasive estimates of LIC in a study by Wood et al (34). If this is confirmed in additional studies, R2* techniques should be preferable given their rapid acquisition times.

Other MRI techniques for liver iron quantification

T1 relaxometry—The presence of iron overload also shortens T1 relaxation times. A correlation between T1 and iron concentration (or T2*) has been observed in previous studies, both in the liver (126), and in the heart (127,128). Thus, it may be possible to use liver T1 measurements to quantify liver or cardiac iron concentration. In liver T1 mapping, however, care must be taken to address the presence of liver fat, which has a short T1 and may confound T1-based assessment of liver iron overload.

MR susceptometry—Magnetic susceptibility is a fundamental property of tissue, and it is strongly correlated with iron concentration. Although dedicated SQUID-based liver susceptometry devices exist (as described above), these devices are not widely available and there is significant recent research interest in MR-based susceptometry techniques.

Liver iron quantification using MR susceptometry is based on the effect of iron on tissue magnetic susceptibility and the effect of susceptibility on the B0 field. The B0 field has a known mathematical relationship to the underlying magnetic susceptibility distribution. The B0 field can be measured in MRI using multi-echo gradient-echo sequences, and can be used to estimate the susceptibility distribution (129) using Quantitative Susceptibility

Mapping (QSM) techniques. From the liver susceptibility estimates, LIC can be assessed (45). However, QSM constitutes a challenging mathematical problem (129–131). Development of QSM techniques has focused largely on brain applications, eg: for assessment of brain iron deposition (132). Abdominal QSM is less developed than brain applications, likely due to several additional challenges, including the presence of motion, fat, and multiple tissue/air interfaces.

Holt et al (133) developed a phase-based method using a water bath as reference. Holt (134) performed a retrospective study by scanning 23 patients (19 with iron overload and 4 normal) who had previously undergone liver biopsy, as well as a prospective study on 29 patients with beta-thalassemia major who underwent liver biopsy within 1 week of their MRI studies. Correlation between MRI susceptometry and liver biopsy was $r = 0.889$ in the retrospective group and $r = 0.622$ in the prospective study.

Several MR-based liver susceptibility measurement techniques based on “boundary” methods have been developed. In these methods, the difference in B_0 field between two adjacent tissues enables localized measurement of the susceptibility difference between these tissues (135–137). Chu et al (135) measured the susceptibility difference between hepatic portal venous blood and the liver parenchyma as a marker of liver iron concentration. More recently, other techniques have performed liver iron quantification based on “boundary” B_0 field measurements between the liver and adjacent muscle (138) or between the liver and subcutaneous fat (139). These techniques have shown good correlation with $R2^*$ - and $R2$ -based measures of LIC, and demonstrate that the fundamental information for liver iron quantification can be captured by the measured B_0 field map (figure 12).

Although appealing, susceptibility estimation techniques present several limitations for iron quantification, including uncertainty in converting susceptibility measurements to liver iron concentration values. This conversion may depend on the relative concentration of ferritin and hemosiderin, which in turn may depend on the type of iron overload and give rise to inter-organ, inter-patient or inter-disease variability (139,140). Currently, the main drawback of susceptibility mapping techniques compared to relaxometry is likely the complication of the estimation problem. Although sophisticated QSM techniques have been proposed in recent years, significant development and validation are needed before QSM based liver iron quantification becomes clinically applicable.

CONCLUSION

Liver iron quantification is needed for the detection, quantitative staging and treatment monitoring of iron overload. Biopsy is considered the reference standard for assessment of liver iron, but is limited by high cost, sampling variability and invasiveness. Non-invasive techniques such as serum biomarkers, CT or SQUID-susceptometry are either inaccurate or have poor accessibility.

MRI is widely available and non-invasive, very sensitive to the presence of iron (both in hemochromatosis and transfusion-related iron overload), and has the potential to provide accurate liver iron quantification. T2 and T2* weighted imaging permit qualitative

assessment of liver iron. For quantitative assessment, calibration curves have been empirically derived that permit estimation of LIC from MR-based measurements of signal intensity ratios, as well as R2 or R2* relaxation rates. R2*-based methods are particularly promising due to their speed. R2* maps with whole liver 3D coverage can be acquired in a 20 second breath-hold. However, the confounders that affect R2* mapping, including noise, fat and background field variations, must be addressed in order to obtain accurate, robust and reproducible measurements of liver iron.

Finally, newer methods of quantitative susceptibility mapping (to date applied primarily in the brain) (129) to directly measure liver magnetic susceptibility, as well as advanced relaxometry methods for distinguishing different types of iron deposition may provide improved assessment of liver iron. Upon successful development and validation, these advanced methods may one day replace or complement current R2- or R2*-based methods for liver iron quantification.

In summary, signal intensity ratio and R2 mapping techniques are currently clinically available and provide validated liver iron concentration measurements. However, R2* relaxometry methods can provide rapid and accurate liver iron quantification in a single breath-hold. Additionally, the lack of motion artifacts (due to the rapid, breath-held acquisitions) and automatic reconstruction of R2* maps may simplify workflow for iron quantification. In our opinion, the lack of reproducibility of R2* methods has been due to technical development challenges rather than fundamental limitations of liver R2* mapping. Upon establishment of reproducibility, R2* relaxometry will likely be the preferred method for liver iron quantification.

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Figure 1.
Representative CT images in patients with liver iron overload. MRI-based R2* ($=1/T2^*$) maps also shown for comparison.

Figure 2.

T2- or T2*-weighted images enable qualitative assessment of liver iron. Images show representative T2- and T2*-weighted images in patients with different levels of iron overload. T2-weighted images were obtained using a 2D single spin echo sequence (TE=12ms). T2*-weighted images were obtained with a single breath-hold 3D SPGR sequence (TE=5.2ms).

Figure 3.

OP/IP imaging is confounded by the simultaneous presence of fat and iron. In this example, OP/IP images from a patient with severe steatosis and moderately elevated R2* only indicate mild steatosis. However, joint estimation of fat-fraction and R2* using a six-echo acquisition demonstrates a fat-fraction of 28% and R2* of 90 s⁻¹. The presence of fat and iron was confirmed by biopsy (bottom images).

Figure 4.

Outline of the R2 ($=1/T_2$) mapping process. In this technique, several spin-echo images are acquired with increasing echo times. Images are subsequently processed to estimate the R2 relaxation rate at each voxel. Examples show R2 maps from a normal volunteer without iron overload (top), and from a patient with iron overload (bottom).

Figure 5.

Relationship between relaxation rates R2 and R2*, and biopsy-determined liver iron concentration, as measured by St. Pierre et al, and by Wood et al, respectively.

Figure 6.

$R2$ can be measured from spectroscopy acquisitions in order to assess liver iron levels. Similar to imaging-based $R2$ mapping techniques, STEAM or PRESS spectra can be acquired at increasing echo times, and the rate of decay of the signal with echo time can be measured. Importantly, multi-echo spectra can be acquired quickly and with good SNR. However, they do not provide information on the spatial distribution of iron.

Figure 7.

Outline of the R2* ($=1/T2^*$) mapping process. In this technique, several gradient-echo images are acquired with increasing echo times. Images are subsequently processed to estimate the R2* relaxation rate at each voxel. Examples show R2* maps from a normal volunteer without iron overload (top), and from a patient with iron overload (bottom).

Figure 8.

R2* relaxometry can be used to monitor treatment for iron overload. Images show R2* maps from a 21 year old patient undergoing chelation therapy. Note the sharp decrease in R2* after one year of therapy.

Figure 9.

Effects of noise (A–B) and fat (C–D) on R_2^* estimation. (A–B) R_2^* fitting in the presence of noise, in high R_2^* case ($R_2^* = 500$ /s) (Adapted from Ref (83)). (A) Noise in complex MRI signals is zero-mean. Complex fitting provides unbiased R_2^* measurements over a wide range of R_2^* values. (B) Noisy magnitude signals results in a “noise floor” at low SNR, leading to underestimation of R_2^* when using magnitude fitting. (C) The presence of fat introduces additional oscillations in the MR signal, as fat and water become in and out of phase. (D) Measured R_2^* from 6-TE echo trains (fat-fraction=40%, $R_2^* = 30$ /s), using $TE_{init} = 1$ ms and varying dTE . Fat-water oscillations result in severe errors in fat-uncorrected R_2^* measurements. Note that errors occur even when the echo spacing is equal to 4.61 ms (ie: one cycle of the main fat peak around the water resonance), due to the spectral complexity of the fat signal.

Figure 10.

Different mechanisms for iron overload lead to distinct organ involvement, as depicted with R2* mapping. Note the similar liver R2* but different spleen R2* in a patient with hereditary hemochromatosis and a patient with transfusional hemosiderosis.

Figure 11.

Imaging-based techniques for liver iron assessment may be important in cases of heterogeneous iron deposition. Images show R2* maps from two patients with heterogeneous iron overload. In these patients, localized techniques such as biopsy or MR spectroscopy may not provide a representative measurement of liver iron.

Figure 12.

The presence of liver iron modifies the susceptibility of tissue, which in turn modifies the B₀ field map measured with MRI. Similar to the principle of SQUID biosusceptometry, MRI-based B₀ field mapping may be able to detect and quantify susceptibility changes caused by the presence of iron in the liver and other organs. Images show R2* maps and B₀ field maps in a normal volunteer without iron overload, and in a patient with iron overload.