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MR PHASE AND SUSCEPTIBILITY-WEIGHTED IMAGING
OF IRON DEPOSITION IN MULTIPLE SCLEROSIS
AND RADIATION-TREATED BRAIN TUMORS

by

Wei Bian

DISSERTATION

Submitted in partial satisfaction of the requirements for the degree of

DOCTOR OF PHILOSOPHY

in

Bioengineering

in the

GRADUATE DIVISION

of the

UNIVERSITY OF CALIFORNIA, SAN FRANCISCO

AND

UNIVERSITY OF CALIFORNIA, BERKELEY

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by

Wei Bian

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ABSTRACT

Magnetic Resonance Imaging is a non-invasive imaging technique that is widely used in medicine. Conventionally, MRI contrasts rely on differences in longitudinal or transverse relaxation times of different tissues and the resulting images display primarily anatomical information of the tissues. With progress in MRI techniques, image contrasts that provide functional or physiological information are now available. Tissue susceptibility is one of this kind and recently has been gaining interest in MRI, especially at high MR field strengths. To generate susceptibility contrast, MR phase and susceptibility-weighted imaging (SWI) are currently two imaging modalities that are used most often. In this work, we focused on using these imaging methods to study abnormal iron accumulation in multiple sclerosis (MS) and radiation-treated brain tumors. We first report in this dissertation a serial phase imaging study of chronic MS lesions in which the phase contrast, presumed due to iron deposition in the lesions, was investigated longitudinally. The observations from the study contribute to a better knowledge of the mechanism of the phase contrast in MS lesions and their evolution. Then we present a comparison study of SWI of iron-containing cerebral microbleeds (CMBs) between 3 Tesla and 7 Tesla MR scanners for patients who had brain tumors and received radiation therapy. This study was aimed at knowing how much sensitivity gain can be achieved when choosing 7T over 3T for detection of CMBs. Followed by the study, a gradient-echo sequence with multiple echoes is introduced, which is able to acquire MR angiography and susceptibility-weighted images simultaneously. This sequence provides a way to characterize CMBs together with veins and arteries in the brain. In addition, by integrating data from several echoes, the SWI can be made more flexible and its imaging quality of CMBs can be improved compared to sing-echo SWI. Finally, an automated CMB detection algorithm is developed, which is able to identify

CMBs on images from SWI with a high sensitivity. Its high accuracy and fast speed significantly reduces radiological time burden in identifying CMBs, which will speed up the exploring of the clinical relevance of CMBs.

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PUBLICISHED MATERIALS

Chapter 4 is based on the journal article titled “*A serial in vivo 7T magnetic resonance phase imaging study of white matter lesions in multiple sclerosis*”, published in *Multiple Sclerosis* in 2013 with authors: Wei Bian, Kristin Harter, Kathryn Hammond-Rosenbluth, Janine Lupo, Duan Xu, Douglas Kelley, Daniel Vigneron, Sarah Nelson, Daniel Pelletier.

Chapter 5 is based on the journal article titled “*Susceptibility-weighted MR imaging of radiation therapy-induced cerebral microbleeds in patients with glioma: a comparison between 3T and 7T*”, published in *Neuroradiology* 2014 with authors: Wei Bian, Christopher Hess, Susan Chang, Sarah Nelson, Janine Lupo.

Chapter 6 is based on the abstract titled “*Multi-Echo Acquisition of 3D TOF and SWI of Radiation-Induced Cerebral Microbleeds at 7T*”, presented 2013 in the 21st annual meeting of the international society of magnetic resonance in medicine (ISMRM) with authors: Wei Bian, Suchandrima Banerjee, Douglas Kelley, Susan Chang, Sarah Nelson, Janine Lupo.

Chapter 7 is based on the journal article titled “Computer-aided detection of radiation-induced cerebral microbleeds on susceptibility-weighted MR images”, published in *Neuroimage Clinical* in 2013 with authors: Wei Bian, Christopher Hess, Susan Chang, Sarah Nelson, Janine Lupo.

Wei Bian is the primary author of all above publications. His contributions included experiment design, data acquisition, data analysis, and manuscript composition.

Chapter 1 Introduction

1.1 Background

Magnetic resonance imaging (MRI) is an imaging technique that has been widely used in clinical radiology and preclinical studies. Its non-invasive nature, non-destructiveness (no ionizing radiation required), versatile soft tissue contrast, multi-dimensional imaging capability, and accurate presentation of functional as well as structural information have made it a powerful modality for the imaging the human body. Since its invention in 1970s (Lauterbur, 1973), MRI has advanced tremendously in imaging quality and flexibility, with major research efforts concentrated on improving image resolution, speeding up image acquisition, and exploring new sources of image contrast.

One novel image contrast mechanism that has recently emerged is provided by differences in tissue magnetic susceptibility. Unlike the contrast in conventional imaging, which is manifested directly on magnetic resonance (MR) magnitude signals such as proton density, T1 and T2, susceptibility weighting relies on information buried in the phase as well as the magnitude of the signal. Although the natural output of MRI comprises complex numbers that contains both phase and magnitude information, the phase images have previously been ignored because they are often contaminated by artifacts that are introduced by macroscopic susceptibility effect at air-tissue interfaces or inhomogeneity of the external magnetic field. It was not until late 1990s (Reichenbach et al., 1997) that researchers started to realize that macroscopic susceptibility artifacts can be largely eliminated by applying a high-pass filtering process, exposing local variations in susceptibility. These differences originate primarily from metal accumulation in tissues such as iron and calcium, but substances such as proteins and

lipids may also contribute (Haacke et al., 2005; Duyn et al., 2007; He et al., 2009). All of these substances and their structural arrangements will perturb the external magnetic field and make tissues feel slightly different field strengths, which will be reflected on the phase images from MRI. Processed phase images can be used either independently or further utilized to enhance the susceptibility effect on corresponding magnitude images and to provide an MR method referred to as susceptibility-weighted imaging (SWI).

With advances in MRI hardware, interest in improving the acquisition and post-processing of phase images and SWI has continued to grow. Firstly the homogeneity of the external magnetic field has been improved greatly in the past decades and reduced macroscopic susceptibility effect. Moreover the increase in the magnetic field strength, from 1.5 Tesla (T) to 3T, and more recently to ultra-high field of 7T has improved both the signal to noise ratio (SNR) and the susceptibility effect on MR images in general. The increased SNR obtained at high field can be used to trade off for a higher image resolution, while the increased susceptibility effect enhances local susceptibility contrast as long as the macroscopic susceptibility artifacts can be well suppressed at the same time. A critical issue is how SWI and phase imaging can benefit from these technical advances and exhibit their usefulness in clinical applications. Examples of these applications include phase imaging of multiple sclerosis (MS) lesions and SWI of cerebral microbleeds (CMBs) in brains of patients with neurological disorders.

Phase images from MS patients have displayed a subset of lesions that could not be seen on conventional MR images probably due to abnormal iron accumulation in these lesions (Hammond et al., 2008; Haacke et al., 2009). At the same time several morphological subtypes of these lesions have been identified, implying different distribution patterns for intracellular iron may exist (Hammond et al., 2008). Like some MS lesions, CMBs are also iron-rich lesions due

to previous microhemorrhage of small vessels in diseases such as hypertension, stroke, cerebral amyloid angiopathy (CAA), dementia, Alzheimer's disease, and radiation-treated brain tumors (Greenberg et al., 2009; Lupo et al., 2012). With high-resolution SWI at the high field strengths of 3T or 7T, the number of CMBs detected in abnormal human brains are much larger and their size limit for detection becomes much smaller (Nandigam et al., 2009; Conijn et al., 2011).

However, several aspects of these studies have not yet been investigated. In particular, it is not clear how the susceptibility contrast in MS lesions varies with time. In addition, it is also unclear whether the gain of SWI always scales with magnetic field strength or whether high field always gives a higher yield for SWI, particularly in CMB identification. Moreover radiological inspection for CMBs on SWI images can be challenging and time consuming, as lesion size is usually small and there may be plenty of them. As a start in overcoming this problem, several methods have been proposed for automated or semi-automated identification of CMBs on SWI images, however their detection sensitivity and computation speed are still low. On the other hand, although modifications of the basic SWI pulse sequence, such as adding several additional echoes, have been proposed, their implementation for clinical studies has lagged behind. The aim of this dissertation is to address these issues and provide tools that can aid in the more widespread clinical use of SWI and phase imaging.

1.2 Contributions

Our first contribution in this field was to perform a longitudinal phase imaging study of MS lesions using phase sensitive imaging. We found that the phase contrast of some of the MS lesions stayed unchanged over a long time, which challenged the previous speculation that the source of phase contrast of these lesions as abnormal iron accumulation in active macrophages. Our second contribution was to compare the sensitivity of SWI to CMBs in radiation-treated

brain tumors between 3T and 7T scanners. Unlike previous comparisons made between 1.5T and 3T or 7T, which consistently found that the number of CMBs detected increased with field strength, our comparison showed that the benefit of heightened susceptibility weighting at 7T could be offset by concomitantly increased susceptibility artifacts. The third contribution in this dissertation was our implementation of a multiple echo sequence at 7T that can simultaneously perform MR angiography (MRA) of arteries and SWI of veins and CMBs. We made the sequence different from other similar sequences by allowing it to acquire images at even higher resolution and using a different post-processing pipeline. More importantly, we evaluated the sequence performance on patients, which had not previously been attempted. Our final contribution was to develop a novel algorithm for automatic identification of CMBs on SWI images, which is the most sensitive and fastest one among those that have been described so far. This algorithm highlights potential CMBs for radiological inspection and confirmation, which can greatly alleviate the time burden on radiologists and promote the diagnostic value of SWI.

1.3 Organization of the dissertation

This dissertation is organized as follows: in chapter 2, basic principles of MRI are briefly reviewed. In chapter 3, MR susceptibility effect, susceptibility contrast on MR phase images, and technical aspects of SWI are described. In chapter 4, susceptibility contrast from MS lesions on MR phase images is presented, with an emphasis on its variation with time. In chapter 5, the SWI of CMBs in the brains of patients with brain tumors who were treated with radiation therapy is introduced, and the sensitivity of SWI to the CMBs is compared between 3T and 7T scanners. The results from the comparison highlight the compromises that need to be made in applying SWI to CMB detection at 7T. In chapter 6, a multi-echo MR imaging sequence that is able to simultaneously acquire MRA and perform SWI is proposed, and its post-processing pipeline

described. The clinical usefulness of the sequence is highlighted for its capability of characterizing radiation-induced CMBs in conjunction with microvessels including both small veins and arteries. In chapter 7, an algorithm that is able to automatically identify CMBs is presented and evaluated on patients with radiation-treated brain tumors. The sensitivity, computational speed, and false positive rate of the algorithm are compared to previously published approaches. Finally, Chapter 8 addresses conclusions from the study and makes suggestions for future work.

Chapter 2 Principles of MRI

In this chapter, we review the basic principles of MRI with a brief introduction of the physics of magnetic resonance, followed by an explanation of MRI signal detection. The conventional Fourier spatial encoding scheme, pulse sequence, and image formation process of MRI are then addressed. Finally the contrast mechanisms that are used in conventional MRI are described.

2.1 Nuclear Magnetism

2.1.1 Spin and magnetic moment

Elementary anatomic particles such as protons, and electrons have spin, which is a form of angular momentum. Although the analogy of these particles to a spinning top may help understand the concept, spin is an intrinsic property of the particles and does not correspond to rotation at the level of quantum mechanics (Levitt, 2001). Another essential property of the particles that is closely related to spin is magnetic moment, which is related to spin angular momentum through the following equation:

$$\boldsymbol{\mu} = \gamma \mathbf{S} \quad 2.1$$

where $\boldsymbol{\mu}$ and $\mathbf{S}$ are magnetic moment and spin angular momentum, respectively, and γ is gyromagnetic ratio. Both $\boldsymbol{\mu}$ and $\mathbf{S}$ are vectors (displayed as bold letters), with both their magnitude and direction quantized, and γ is a scalar that can have either positive or negative sign. Depending on the sign of γ , magnetic moment is either parallel or anti-parallel to spin angular momentum. As will be discussed, it is coexistence of spin and magnetic moment that makes MR experiment possible. While the experiment can be performed either on electron or on nucleon (proton and neutron), electron MR is far more difficult than nuclear MR for biological tissues, as

it requires much higher energy to be deposited to biological tissues (Levitt, 2001). Since this dissertation is concerned about nuclear magnetic resonance, only nuclear spin and nuclear magnetic moment will be discussed.

Inside a nucleus, protons and, separately, neutrons are usually paired up, which cancels their spins and magnetic moments as much as possible. Thus, nuclei with even numbers of protons and even numbers of neutrons such as ^{16}O and ^{12}C have no net nuclear spin and magnetic moment, while nuclei with odd numbers of protons and even numbers of neutrons (or vice versa) such as ^1H or odd numbers of both protons and neutrons such as ^{14}N have net nuclear spin and magnetic moments.

2.1.2 Precession of nuclear magnetic moment

In the presence of a static external magnetic field, nuclei with net spin will interact with the field as they possess net magnetic moment. However, unlike the interaction between a compass and the earth magnetic field, where the compass will align to the direction of the earth magnetic field and hence stay in a low magnetic energy state, the interaction between the nuclei and external magnetic field is a dynamic process. The difference lies on the fact that the nuclei possess spin angular momentum as well as magnetic moment. Because of the spin angular momentum, the external magnetic field will generate a torque on the magnetic moment, and create a rotational movement called precession, which can be described as

$$\frac{d\boldsymbol{\mu}}{dt} = \gamma \boldsymbol{\mu} \times \mathbf{B}_0 \quad (2.2)$$

where $\boldsymbol{\mu}$ is the net magnetic moment a single nucleus has and $\mathbf{B}_0$ is the external magnetic field. In the Cartesian coordinates, assuming the direction of external magnetic field $\mathbf{B}_0$ is along Z axis, the solution of this differential equation is:

$$\begin{aligned}
\mu_x(t) &= \mu_x(0) \cos \omega_0 t + \mu_y(0) \sin \omega_0 t \\
\mu_y(t) &= \mu_y(0) \cos \omega_0 t - \mu_x(0) \sin \omega_0 t \\
\mu_z(t) &= \mu_z(0)
\end{aligned}
\tag{2.3}$$

where

$$\omega_0 = \gamma B_0 \tag{2.4}$$

The solution implies: 1) the nuclear magnetic moment rotates around Z axis with an angular frequency of ω_0 ; 2) the magnitude of the magnetic moment remains unchanged in the process of rotation; 3) the angle between the magnetic moment and Z axis also remains the same. The rotational movement is called precession, which is illustrated in Figure 2.1, and ω_0 is precession frequency or Larmor frequency.

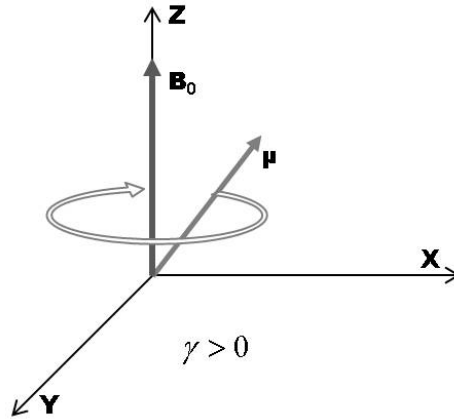


Figure 2.1 Clockwise precession of a nuclear magnetic moment with positive gyromagnetic ratio.

2.1.3 Net macroscopic magnetic moment

Now consider the ensemble behavior from all nuclei in a sample. In the absence of external magnetic field, the direction of any nuclear magnetic moment is uniformly distributed, pointing to all possible directions with equal probability. The total macroscopic magnetic moment of the sample is therefore close to zero. When an external magnetic field is turned on,

each individual nuclear magnetic moment will precess around the field. However, the angle between the magnetic moment and the external field is no longer a constant over time. On a microscopic scale, the molecular environment of each nucleus keeps changing due to the thermal motion of molecules. Since each molecule carries magnetic particles (e.g., electrons and nuclei), the variation of molecular environment cause each nucleus experiences a small local magnetic field that is fluctuating both in magnitude and in direction. Although the strength of this fluctuating field is very slight compared to the external field, it can oscillate at a frequency around the Larmor frequency, which is sufficient to drive the magnetic moment of each nuclear spin to wander around, eventually sampling all possible orientations in a long time (Levitt, 2001). However, the wandering motion is not isotropic, as it involves transition between different energy levels, created after the sample being placed in the external magnetic field, a phenomena called Zeeman splitting. It is more likely for a spin magnetic moment to align towards an orientation with lower magnetic energy than towards an orientation with higher energy. Since the orientation with low magnetic energy is parallel to the external field while the orientation with high magnetic energy is anti-parallel to the field, there will be more nuclear magnetic moment align towards the external field than against it, hence generating net macroscopic magnetic moment along the external field. The amount of the net magnetic moment (or macroscopic magnetization) at thermal equilibrium is proportional to the ratio of the difference in magnetic energy to thermal energy and governed by Boltzmann distribution, which is given by

$$\mathbf{M}_0 \propto \frac{\rho \gamma^2 h^2}{4kT} \mathbf{B}_0 \quad (2.5)$$

where ρ is the number of nucleus per unit volume, h is the Planck's constant, k is the Boltzmann constant, and T is the absolute temperature. At room temperature, the thermal energy is several

orders higher in magnitude than the difference in magnetic energy, the magnitude of $\mathbf{M}_0$ is therefore very small. To make $\mathbf{M}_0$ as large as possible, it is common to target a nucleus with the most abundance in the sample of interest. For the human body, the nucleus of this kind is hydrogen (the proton) in water molecules, which is of the largest amount in tissues. However, even for proton, the $\mathbf{M}_0$ is not large enough (only about 3.8 ppb (Schenck, 1996)) and hardly to be detected directly. To make it as a detectable signal, another non-static and oscillating magnetic field has to be applied.

2.2 Detection of MR signal

In MR experiments, the equilibrium macroscopic magnetic magnetization $\mathbf{M}_0$ can be tipped away from the external magnetic field direction by applying a radiofrequency magnetic field that is perpendicular to $\mathbf{B}_0$ but oscillating at the same frequency as the Larmor frequency. Since the field is applied only for a short time, it is usually referred as an rf pulse or $\mathbf{B}_1$ field. The role of the rf pulse can be understood by setting up a rotating frame, where its Z axis corresponds to the original $\mathbf{B}_0$ axis while both its X and Y axes are rotating around Z at the Larmor frequency ω_0 . In this rotating frame, precession with the frequency ω_0 is unobservable as if there were no external magnetic field. On the other hand, the rf pulse becomes a static field as it is rotating at the same speed as the rotating frame. Thus, in the rotating frame, the $\mathbf{M}_0$ begins to process around $\mathbf{B}_1$ at a frequency of $\omega_1 = \gamma B_1$ and move away from Z axis. The angle the $\mathbf{M}_0$ can rotates is called flip angle and given by $\theta = \omega_1 t$, where t is the duration of rf pulse. Figure 2.2 illustrates a 90 degree rotation of $\mathbf{M}_0$ in the rotating frame after a $\mathbf{B}_1$ is applied. The match of the frequency of an rf pulse to the Larmor frequency of an external magnetic field is so called *Magnetic Resonance*. The resonance allows a $\mathbf{B}_1$ that is much smaller than $\mathbf{B}_0$ to perform a

synchronized action on each individual processing nuclear magnetic moment, and eventually manifesting as a macroscopic rotation of $\mathbf{M}_0$.

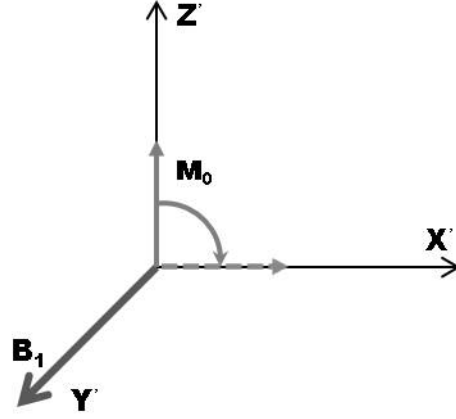


Figure 2.2 The rotation of $\mathbf{M}_0$ in the rotating frame after a $\mathbf{B}_1$ is applied

If the $\mathbf{B}_1$ is removed after the $\mathbf{M}_0$ is rotated away from $\mathbf{B}_0$ axis, $\mathbf{M}_0$ will precess around the $\mathbf{B}_0$ axis just like what has been described for a single nuclear magnetic moment in the section 2.1.2. For example, an rf pulse applied along the negative Y' axis in the rotating frame rotates the $\mathbf{M}_0$ of protons θ ($0 < \theta \leq 180^\circ$) degree away from $\mathbf{B}_0$ (or Z') axis and towards positive X' axis, and the resulting transverse magnetization will have the same magnitude as that of $\mathbf{M}_0$ and starts precessing clockwise around the $\mathbf{B}_0$ axis. According to the equation 2.3, the transverse magnetization is

$$\begin{aligned} M_x(t) &= M_0 \sin \theta \cos \omega_0 t \\ M_y(t) &= -M_0 \sin \theta \sin \omega_0 t \end{aligned} \quad (2.6)$$

or in complex form

$$\mathbf{M}_+(t) = M_x(t) + iM_y(t) = M_0 \sin \theta e^{-i\omega_0 t} \quad (2.7)$$

Note that $M_x(0) = M_0 \sin \theta$ and $M_y(0) = 0$ in this example.

The time-varying transverse magnetization $\mathbf{M}_+$ generates a time-varying magnetic field, which can in turn induce an oscillating electric field and current in a wire coil if it is put close to a sample. According to the reciprocity principle, the current flow or magnetic resonance signal detected by the wire coil is

$$s(t) = C\gamma B_0 M_+(t) \quad (2.8)$$

where C is a constant depending on the electronic gain during detection and the magnitude of $\mathbf{B}_1$ (assuming $\mathbf{B}_1$ is homogeneous over the sample). From the equation 2.5 and 2.7, 2.8 can be reformulated as

$$s(t) = C \frac{\rho \gamma^3 h^2 B_0^2}{4kT} \sin \theta e^{-i\omega_0 t} = C \frac{\rho \gamma h^2 \omega_0^2}{4kT} \sin \theta e^{-i\omega_0 t} \quad (2.9)$$

Compared to the $\mathbf{M}_0$ in equation 2.5, it is clear that the proportion of signal magnitude to γ and B_0 is increased by one order for both. It also implies that the signal from an MR experiment grows quadratically with the external magnetic field, which motivates the interests in high field MR experiments.

2.3 Spatial encoding

Usually the sample used in MR experiments is inhomogeneous, i.e., the proton density is a function of space. In the simplest case, suppose that proton density of a sample is a 1D function $\rho(x)$ along x axis, then the signal in equation 2.9 can be written as

$$s(x, t) = C \frac{\rho(x) \gamma h^2 \omega_0^2}{4kT} \sin \theta e^{-i\omega_0 t} \quad (2.10)$$

By absorbing all time and spatial independent constants into $\rho(x)$ and interpreting it as apparent proton density (Here including ω_0 into $\rho(x)$ is only for the sake of simplicity in mathematics and

caution should be taken when comparing signal from different field strengths), 2.10 can be reduced to a simpler form

$$s(x, t) = \rho(x)e^{-i\omega_0 t} \quad (2.11)$$

The total signal from the sample is therefore an integral over x

$$S(t) = \int \rho(x)e^{-i\omega_0 t} dx \quad (2.12)$$

The goal of spatial encoding is to find a way to recover $\rho(x)$ at every x from measured signal $s(t)$, which contains contributions from the magnetization everywhere along x . A great method to achieve this goal is to apply a small perturbation of the magnitude of the external magnetic field and make the Larmor frequency is dependant on x . Physically this can be done by superposing $\mathbf{B}_0$ with a gradient magnetic field, which is a small field pointing to the same direction as $\mathbf{B}_0$ but with its magnitude vary linearly along x . Assuming the slope of the variation is G_x , then the Larmor frequency as a function of x is given by

$$\omega_x(x) = \gamma(B_0 + G_x x) = \omega_0 + \gamma G_x x \quad (2.13)$$

Now the equation 2.12 becomes

$$S(t) = e^{-i\omega_0 t} \int \rho(x)e^{-i\gamma G_x x t} dx \quad (2.14)$$

The term $e^{-i\omega_0 t}$ in the equation 2.15 can be looked as the carrier of the signal that contributes to final image formation. Since in a MR experiment we already know ω_0 , the carrier can be removed by demodulation, leaving the signal as

$$S(t) = \int \rho(x)e^{-i\gamma G_x x t} dx \quad (2.15)$$

If we define $k_x = \gamma G_x t$, then the equation 2.15 can be rewritten as

$$S(k_x) = \int \rho(x)e^{-ik_x x} dx \quad (2.16)$$

Mathematically, the integral in the above equation stands exactly for the Fourier transform of $\rho(x)$, and $S(k)$ and $\rho(x)$ are just Fourier transform pairs. Knowing $S(k_x)$, one can easily obtain $\rho(x)$ by doing a inverse Fourier transform, which is

$$\rho(x) = \int S(k_x) e^{ik_x x} dk \quad (2.17)$$

The idea of 1D spatial encoding can be generalized to 2D or 3D cases by applying linear gradient fields to multiple directions. For example, when both x and y gradient fields are used, the 2D form of equations 2.15 to 2.17 are as follows

$$S(t) = \int \rho(x, y) e^{-i\gamma(G_x x + G_y y)t} dx \quad (2.18)$$

$$S(k_x, k_y) = \int \rho(x, y) e^{-i(k_x x + k_y y)} dx dy \quad (2.19)$$

$$\rho(x, y) = \int S(k_x, k_y) e^{i(k_x x + k_y y)} dk_x dk_y \quad (2.20)$$

where the last two equations are 2D Fourier and inverse Fourier transforms, respectively.

From the perspective of signal processing, $S(k_x, k_y)$ is referred to signal in spatial frequency domain, but in the community of MRI the domain is usually called as ‘k-space’. The larger the k , the higher the spatial frequency it represents. Since objects in MR experiments have limited dimensions, some finite samples of the signal $S(k_x, k_y)$ from a k-space are sufficient to reconstruct the proton density $\rho(x)$ in its discrete form through discrete Fourier transform, as long as the sampling rate follows the Shannon’s sampling theorem.

2.4 Pulse sequence and image formation

To form the described signal in a k-space and have it sampled as required, the timing of rf pulse, spatial encoding gradients, and sampling devices has to be carefully designed in MRI. The

sequence of these components is called a pulse sequence. A typical 2D MR pulse sequence is shown in Figure 2.3(A).

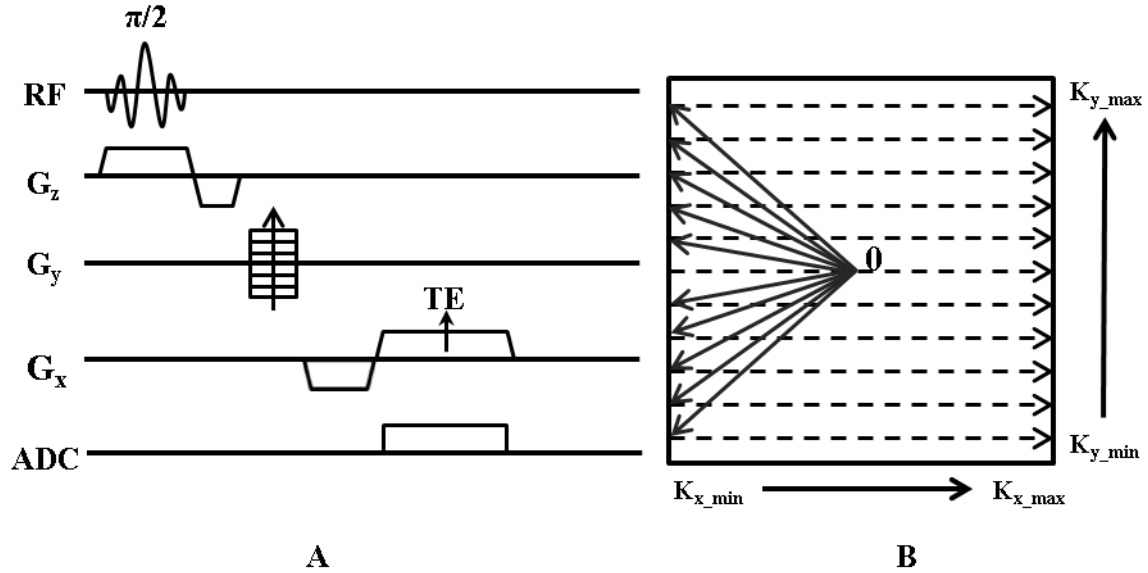


Figure 2.3 A 2D gradient-echo pulse sequence (A) and its k-space trajectory (B)

The sequence starts with a 90 degree rf pulse, which excites the protons and rotates macroscopic magnetization into the transverse plane. In practice rf pulses are not designed to have a single value of the Larmor frequency of $\mathbf{B}_0$ but a spectrum of frequencies (the width of the frequency spectrum is the bandwidth of the rf pulse). This is not only because it is hard to make a rf pulse oscillating at a single frequency, but also because it is often desirable in MRI to excite protons in a thin slice (in 2D) or a slab (in 3D). The slice/slab selection is made possible by applying an rf pulse with a limited bandwidth, simultaneously, with a selection gradient field that allows $\mathbf{B}_0$ (hence the Larmor frequency) varies with the location just like spatial encoding gradient field. The gradient could be along any direction, depending on the plane (axial, coronal, or saggittal) of interested. The thickness TH of the slice spin is determined by the bandwidth BW of the rf pulse and the gradient G_s of the selection field, which can be expressed as

$$TH = \frac{BW}{\gamma G} \quad (2.21)$$

After the rf pulse is turned off, the slice selection gradient field will be kept on but with negative slope for an additional time that is equal to the half of rf pulse duration. The negative part of the slice selection field refocuses the procession coherence of protons in the slice, which has been disrupted by the positive part of the field. Because of the refocusing, the slice selection gradient does not introduce location dependent magnetization, which distinguishes itself from spatial encoding gradients.

Followed by the rf pulse and slice selection gradient, the y gradient field is turned on for a time of τ_y to do the spatial encoding along y such that $k_y = \gamma G_y \tau_y$. Since signal sampling has not started yet at this moment, the current k_y will be kept same for all following samples. After the y gradient is turned off, all protons will again process at the Larmor frequency but with different phases along y , which was accumulated during y gradient was on, and this why the y gradient is usually called *phase-encoding* gradient. Once y gradient is off, x gradient is on but first with a negative slope for a time of τ_x , which simply makes the sampling of k_x begin with a negative value of $-\gamma G_x \tau_x$. Once the sign of the x becomes positive, the analog to digital converter (ADC) is turned on (marking the start of signal sampling in k -space) simultaneously for a time of $2\tau_x$, as long as the duration of positive x gradient. Since protons process at different frequencies along x when signal is being sampled, the sampled signal contains a spectrum of frequencies, and this gives x gradient a common name as *frequency-encoding* gradient. If the sampling interval is Δt , then $k_{x_n} = -\gamma G_x (\tau_x - (n-1)\Delta t)$, where n is from 1 to N (the number of total samples) and $\Delta t = 2\tau_x/(N-1)$. At the end of sampling, each of these N samples will have different k_x but with same k_y , implying that only a line of a 2D k -space has been sampled. To reconstruct the image

corresponding to the k-space, more lines need to be sampled. Therefore the pulse sequence has to be cycled to sample k-space lines with different k_y , which is achieved by making G_y stepwise, with its value stepping from negative to positive to cover both negative and positive parts of k-space along k_y . The duration of one complete pulse sequence cycle is called time of repetition (TR) and the time interval from the center of the rf pulse to the center of the positive lobe of x gradient is called time of echo (TE). The term ‘echo’ comes from the fact that at the time of TE, the effect, separately, from the negative and positive x gradient on protons perfectly cancel each other, leaving $k_x = 0$ and bringing once incoherent procession of magnetic moments along x due to the application of negative gradient back to coherence. The sequence itself is therefore named as *gradient-echo* sequence in MRI. After a sufficient number of repetitions, 2D sampling of k-space is completed, and the original image can be reconstructed by performing the 2D inverse Fourier transformation on the sampled data. Since the sampling occurs in spatial frequency domain, according to the Shannon’s sampling theorem, the width is the reconstructed image (Field of views: FOV) is

$$\begin{aligned} FOV_x &= \frac{1}{\Delta k_x} = \frac{1}{\gamma G_x \Delta t} \\ FOV_y &= \frac{1}{\Delta k_y} = \frac{1}{\gamma G_y \tau} \end{aligned} \quad (2.22)$$

and the image resolution is

$$\begin{aligned} \Delta x &= \frac{1}{2k_{x_max}} = \frac{1}{2\gamma G_x \tau_x} \\ \Delta y &= \frac{1}{2k_{y_max}} = \frac{1}{2\gamma G_y \tau_y} \end{aligned} \quad (2.23)$$

2.5 Image contrast

In the above discussions, the strength of signal depends on proton density in each pixel, the more densely the protons pack in a pixel, the higher signal from that pixel. However, the sources of image contrast in MRI is not limited to proton density, there are several other contrasts that reflect different physical properties of protons other than their density. Two of these properties that have been omitted so far are longitudinal and transverse relaxation times.

The longitudinal relaxation, which is characterized by a constant T_1 , is the time interval that a macroscopic magnetization reestablishes its equilibrium state along the direction of the external magnetic field after it has been rotated into the transverse plane. The mechanism behind the longitudinal relaxation is the interaction between individual spin and the oscillating magnetic field as the result of molecular motion surrounding the spin, which has already been discussed in section 2.1.3. The longitudinal relaxation can only occur when the frequency of the oscillating magnetic field matches the Larmor frequency of the external field. During the relaxation, energy is exchanged between the spin and its molecular environment. The transverse relaxation, which is characterized by a constant T_2 , is the time interval that processing nuclear magnetic moments gradually lose their phase coherence and the overall net macroscopic magnetic moment is reduced. The loss of phase coherence of precession is due to the slight change in the local magnetic field each proton experiences, which makes the Larmor frequency to be different among protons. The local field change is also caused by the interaction between individual spin and its surrounding oscillating magnetic field. However, the source of the oscillating magnetic field can be either from thermal motion of molecules or from spins themselves. In addition, for transverse relaxation to occur, the frequency of the oscillating magnetic field does not have to match the Larmor frequency, hence no energy exchange is required.

After including both longitudinal and transverse relaxations, the procession of macroscopic magnetic moment in an external magnetic field can be described by the well known Bloch equation:

$$\frac{d\mathbf{M}(t)}{dt} = \gamma \mathbf{M}(t) \times \mathbf{B}_0 + \frac{1}{T_1} (\mathbf{M}_0 - \mathbf{M}_z(t)) - \frac{1}{T_2} \mathbf{M}_+(t) \quad (2.24)$$

which is an extension of the equation 2.2. If a MR sequence uses a 90 degree rf pulse with a repetition time of TR and data is assumed to be sampled instantly at an echo time of TE, the solution of the Bloch equation for the transverse magnetization at TE is

$$M_+ = M_0 (1 - e^{-\frac{TR}{T_1}}) e^{-\frac{TE}{T_2}} \quad (2.25)$$

where M_0 is the strength of the initial longitudinal magnetization. The equation shows that as the result of the transverse relaxation, the signal strength from MRI decays exponentially with T2. In addition, the equation implies how to weight an image towards a certain contrast while suppress other contrasts by manipulating TR and TE. For example, an image is weighted towards proton density contrast when TR is long and TE is short; and it is weighted towards T1 contrast and away from T2 contrast when both TR and TE are short; whereas it is T2 weighting dominated when both TR and TE are long. Since proton density can not be changed, its contrast is always present to certain degree even in the cases where T1 or T2 weighting is dominated. Figure 2.4 shows MR images of a human brain with these contrasts.

In practice, ideal T2-weighted contrast is hard to obtain using the gradient echo sequence, because two extra dephasing effects exist. First, the external magnetic field cannot be made purely homogeneous due to technical challenges. Second, objects show susceptibility effect after being placed into external magnetic field, which will be described in Chapter 3. Both external

field inhomogeneity and susceptibility effect change the field strength protons can experience, which gives more variation of their procession frequency in addition to that caused by T_2 relaxation. Therefore, protons move out of phase much faster, so do the rate of signal decay.

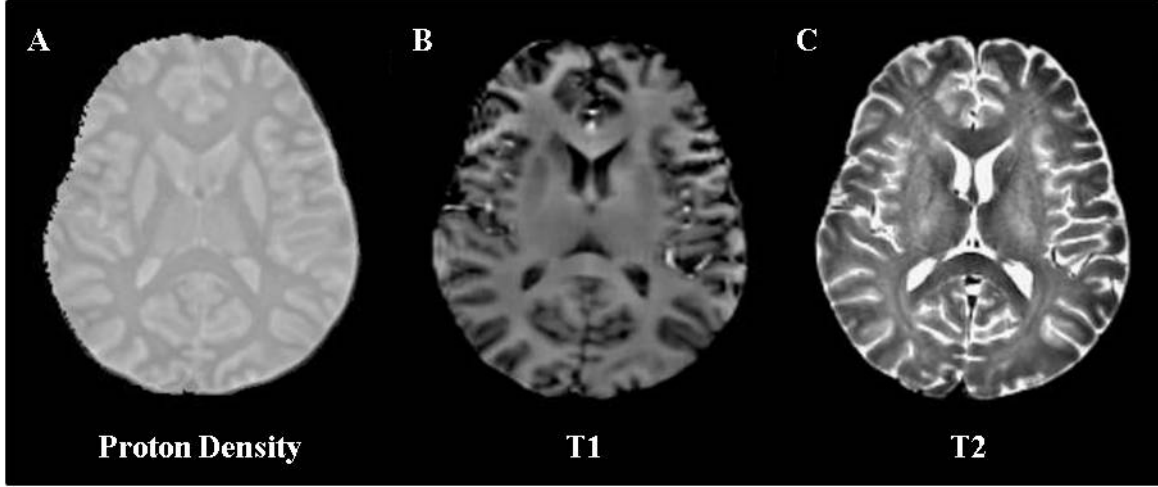


Figure 2.4 MR images with different conventional contrasts

(A) Proton-density-weighted, (B) T1-weighted, (C) T2-weighted images from a normal volunteer. All images are acquired at a 7T scanner with TEs/TRs of 0.25ms/2000ms, 5.6ms/556ms, and 85ms/2000ms, respectively for the proton-density-, T1- and T2-weighted images. Gray matter and cerebral spinal fluid (CSF) are brighter than white matter on the proton-density weighted image, as they contain more protons than white matter. The contrast of these tissues on T1- and T2-weighted images is roughly complementary: white matter has the highest signal intensity on T1-weighted images followed by gray matter and CSF, whereas the order is opposite on the T2-weighted image, where the highest signal is from CSF followed by gray and white matter. The resulting contrast is due to the differences in T1 and T2 relaxation times in these tissues, of which $T1/T2_{\text{white matter}} < T1/T2_{\text{gray matter}} < T1/T2_{\text{CSF}}$.

The relaxation due to external field inhomogeneity and susceptibility effect can be represented by a time constant T_2' , and the relaxation that includes contributions from both T_2 and T_2' relaxation is referred as T_2^* relaxation. Their relation is as follows

$$\frac{1}{T_2^*} = \frac{1}{T_2} + \frac{1}{T_2'} \quad (2.26)$$

The T_2 relaxation is irreversible as it is caused by random interactions at atomic or molecular level, whereas the relaxation due to external field inhomogeneity and susceptibility effect, which

are somewhat deterministic, is sometimes reversible, for example, by using a spin-echo sequence diagrammed in Figure 2.5. Compared to the gradient-echo sequence in Figure 2.3, the spin-echo sequence in Figure 2.5 has an additional 180 degree rf pulse added at the time of $TE/2$, which inverts the sign of extra phase accumulated due to external field inhomogeneity and susceptibility effect. Then protons continue to collect extra phase at the same rate that will cancel the previously inverted phase at the time of TE , hence form an echo finally. A comparison between T2- and T2*-weighted images is shown in Figure 2.6.

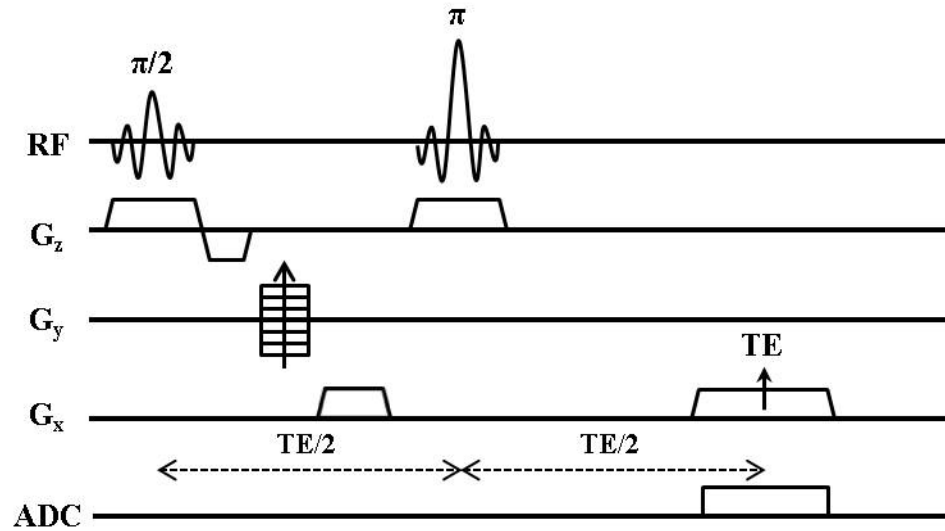


Figure 2.5 A 2D spin-echo pulse sequence.

Although T2* weighted images from gradient-echo sequence may lead to a reduction in signal and contain unwanted information or artifacts due to the imperfection of external magnetic field, it also contains useful information that reflects local susceptibility difference. Moreover, the difference can be further enhanced, making final MR images susceptibility-weighted and display susceptibility contrast. The MRI method towards this goal is called susceptibility-weighted imaging, which will be introduced in the following chapter.

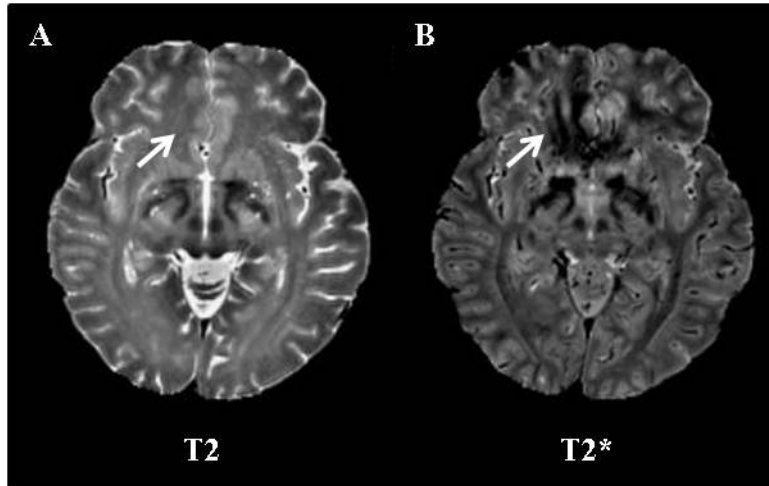


Figure 2.6 Spin-echo T2 and gradient-echo T2* images

Both images were acquired at a 7T scanner with TE=12ms and TR=250ms for the gradient-echo images and TE = 85ms (effective) and TR=2000ms for the spin-echo image using a fast spin-echo sequence. Compared to the T2-weighted image (A), there is a significant signal drop in the region (arrows) anterior to the sinus on the T2*-weighted image, which is due to the susceptibility effect from the air in the sinus.

Chapter 3 MR phase and susceptibility-weighted imaging

In this chapter, the concept of magnetic susceptibility is introduced and its effect on MR phase images is described. Methods for processing phase images are then presented, which include phase unwrapping and multi-coil phase combination. Lastly, the sequence, image acquisition and post-processing methods that are applied to SWI are detailed.

3.1 Magnetic susceptibility

Magnetic susceptibility is an intrinsic tissue property, which measures how likely it is a substance got magnetized after placed in an external magnetic field. The relation between magnetic susceptibility and induced magnetization in an isotropic material can be written as

$$M = \chi H \quad (3.1)$$

Since

$$H = \frac{B_0}{\mu_0 - M}, \quad (3.2)$$

the equation 3.1 becomes

$$M = \frac{\chi B_0}{\mu_0(1 + \chi)} \quad (3.3)$$

where $\mu_0 = 4\pi \times 10^{-7}$ Henry/Meter is the permeability in empty space, B_0 is the strength of applied external magnetic field, and χ is the magnetic susceptibility and a dimensionless constant. For materials with χ much smaller than one, we have

$$M = \frac{\chi B_0}{\mu_0} \quad (3.4)$$

Thus, the induced magnetization is proportional to the main magnetic field and the magnetic susceptibility. The sign of χ can be either positive or negative. Materials with positive χ are paramagnetic while those with negative χ are diamagnetic.

Because of the induced magnetization, the actual field strength inside a sample is no longer equal to the external magnetic field strength, and the variation of the field inside the sample is

$$\Delta B_s(x, y) = g\Delta\chi B_0 \quad (3.5)$$

where g is a factor depending on the shape of the sample and its geometric relation to the field (Haacke et al., 2011), and $\Delta\chi$ is the susceptibility difference between the inside and outside of the sample. The equation implies that the field strength variation is proportional to the external field strength and spatially varies due to its dependence on the geometry factor g .

As with proton density, T1 and T2, susceptibility can be utilized as a source of MRI contrast. This can be generated either by high-pass filtering MR phase images or on images acquired from a pulse sequence that provides susceptibility-weighted imaging (SWI).

3.2 Susceptibility effect on MR phase images

In section 2.3, it has been shown that a 2D image reconstructed from MRI is the Fourier transform of the signal sampled in k-space, i.e.,

$$\rho(x, y) = \int S(k_x, k_y) e^{i(k_x x + k_y y)} dk_x dk_y \quad (3.6)$$

Since the objects that are subjected to MRI are real valued, the reconstructed image $\rho(x, y)$ should also be a real number. However, in practice this is not true and, when $\rho(x, y)$ becomes complex it can be described as

$$\rho(x, y) = |\rho(x, y)| e^{i\phi(x, y)} \quad (3.7)$$

where $|\rho(x, y)|$ is the magnitude of the image and $\Phi(x, y)$ is the phase of the image.

The phase term is introduced by spatially varying $\mathbf{B}_0$. Three major sources of $\mathbf{B}_0$ variation is the non-uniformity of $\mathbf{B}_0$ itself, susceptibility effect that has already been mentioned, and chemical shift, which characterizes the ability of electrons surrounding protons to shift local field strength. After a sample is placed into an external magnetic field, the field will induce electron current, which shields protons a bit from the external field and thus changes the field strength that the protons feel. Like susceptibility, chemical shift is a dimensionless constant and depends on the local molecular environment of protons, which also varies as a function of space. The variation of local field due to chemical shift is proportional to the strength of the external field, and their relation is

$$\Delta B_{cs}(x, y) = \delta(x, y)B_0 \quad (3.8)$$

When all these aspects are taken into consideration, the equation 2.13 in its 2D form becomes

$$\omega_x(x, y) = \gamma(B_0 + \Delta B_0(x, y) + \Delta B_{cs}(x, y) + \Delta B_s(x, y) + G_x x + G_y y) \quad (3.9)$$

After demodulation and following the same derivation as in section 2.3, the equation 2.20 becomes

$$\rho(x, y) = e^{-\gamma(\Delta B_0(x, y) + \Delta B_{cs}(x, y) + \Delta B_s(x, y))TE} \int S(k_x, k_y) e^{i(k_x x + k_y y)} dk_x dk_y \quad (3.10)$$

where, for simplicity, it is assumed that all ΔB is time independent and the data sampling happens instantly at TE. Since ΔB is only restricted to three components that have been described, the inverse Fourier transform in equation 3.10 will be a real image that has no imaginary part, i.e.,

$$|\rho(x, y)| = \int S(k_x, k_y) e^{i(k_x x + k_y y)} dk_x dk_y \quad (3.11)$$

Now equation 3.10 becomes

$$\rho(x, y) = |\rho(x, y)| e^{-\gamma(\Delta B_0(x, y) + \Delta B_{cs}(x, y) + \Delta B_s(x, y))TE} \quad (3.12)$$

And the phase term is

$$\phi(x, y) = -\gamma(\Delta B_0 + \Delta B_{cs} + \Delta B_s)TE \quad (3.13)$$

The susceptibility effect that causes field variation can be both global and local. The former represents susceptibility difference in large spatial scale such as that introduced by the geometry of the object, while the later in small spatial scale such as that introduced by local iron deposition. Then the equation 3.13 can be reformulated as

$$\phi(x, y) = -\gamma(\Delta B_0 + \Delta B_{cs} + \Delta B_{sg} + \Delta B_{sl})TE \quad (3.14)$$

where ΔB_{sg} and ΔB_{sl} are field variations induced by global and local susceptibility effects, respectively.

For a long time, only the magnitude image has typically been used in clinical radiology, with the phase information ignored due to the contaminations from the external magnetic field inhomogeneity and global susceptibility effect. More recently, however, phase information has begun to play a role in diagnosis due to techniques that are able to remove these artifacts having been developed.

3.3 Phase image processing

3.3.1 Phase Wrapping

To make phase information from MRI reflect only local susceptibility contrast, the original phase image has to be further processed. This is not only for removing unwanted contaminations but also for the purpose of correcting measured phase to its actual values.

In mathematics, the phase angle Φ of a complex number

$$c = x + iy \quad (3.15)$$

is calculated as

$$\phi = \tan^{-1}\left(\frac{y}{x}\right) \quad (3.16)$$

i.e., the inverse tangent of the ratio of the number's imaginary part to its real part. Since the range of the inverse tangent is $[-\pi, \pi)$, any measured phase is limited within this range. This means that any true phase exceeding the range will be forced to have a value in the range, causing phase aliasing or so called phase wrapping. The true and measured phases have a following relationship

$$\phi_{true} = \phi_{measured} + 2n\pi \quad (3.17)$$

where n is an integer. Because of phase wrapping, $\phi_{measured}$ will have discontinuities even though it is continuous in general. Figure 3.1A shows a phase image derived from the equation 3.16.

3.3.2 Phase unwrapping methods

The goal of phase unwrapping is to recover the true phase value for every pixel on a phase image, making it continuous and free of wraps. The basic idea is to either add or subtract multiple 2π to or from the measured phase for every pixel. This seems to be simple in theory, but in practice phase unwrapping could be a complicated and ambiguous process, which becomes even worse when noise is presented or performed in higher dimension. One of the most popular phase unwrapping algorithms is PRELUDE (Jenkinson, 2003), which is freely available, fully automated in 3D, and specifically designed for unwrapping phase in MRI. The algorithm solves the unwrapping problem by minimizing a cost function that penalizes phase differences across boundaries.

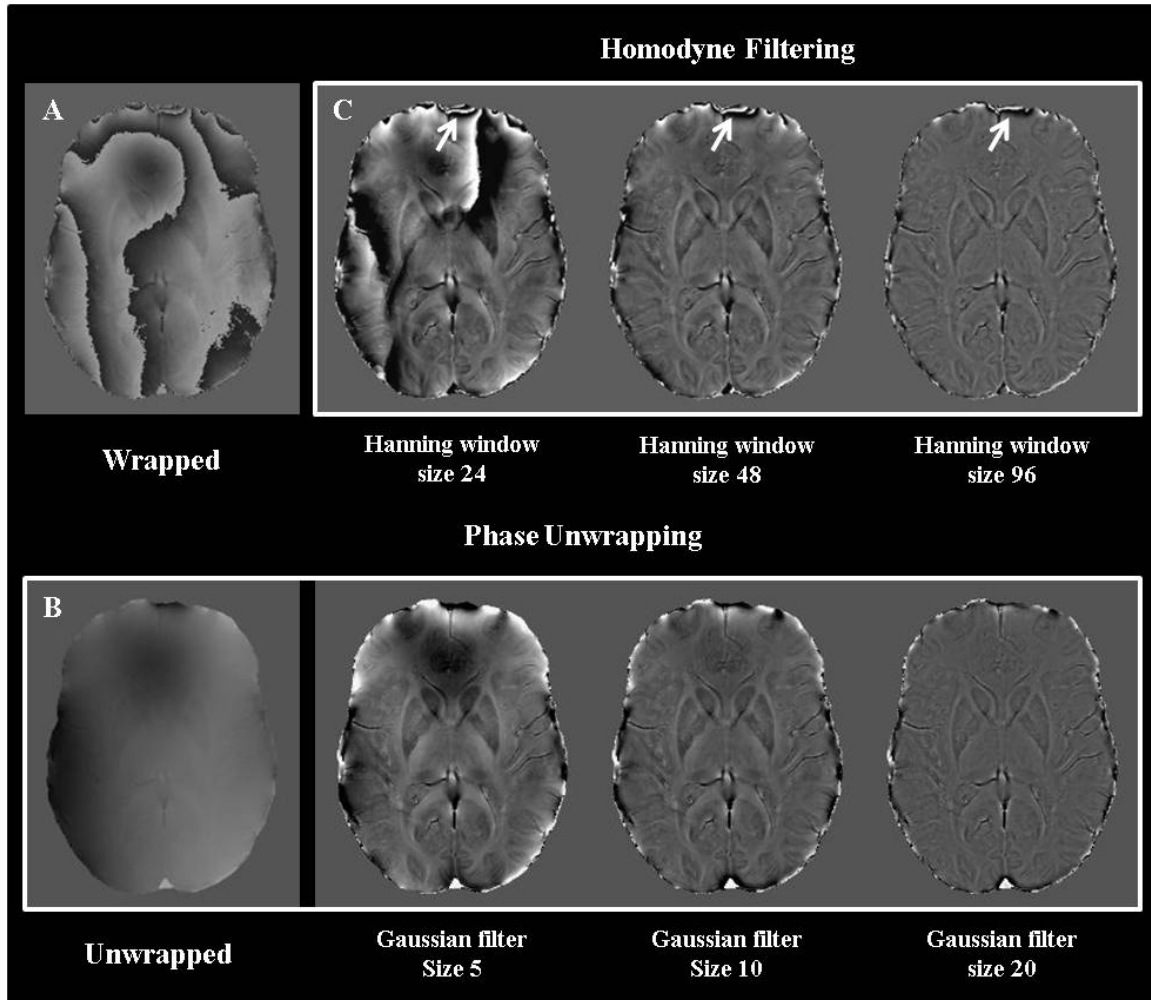


Figure 3.1 Phase unwrapping and Homodyne filtering of a wrapped phase image

Phase discontinuities can be clearly seen on the wrapped phase image (A). The unwrapped phase image after phase unwrapping in (B) is dominated by low frequency components with little contrast from tissues. After applying the Gaussian filter, the low frequency components are removed and tissue structures start manifesting. On the other hand, all phase images are almost free of residual wraps no matter what filter size was used. However, as filter size goes larger, the image loses its contrast gradually. This similar observation can be found in the Homodyne filtering in (C), but some residual wraps (*arrows*) are always presented, especially at regions close to air-sinus interface, although the amount of wraps can be reduced by increasing the filter size.

Since phase unwrapping aims only to correct aliasing, the resulting image still contains contributions from all of the field variation components described in equation 3.14. To remove

unwanted components, a filtering process can be applied to the unwrapped phase image. In general, the sources that give rise to the external magnetic field variation and global susceptibility-induced field variation are high spatial-frequency objects, whereas sources that produce chemical shift and local susceptibility-induced field variation are low spatial-frequency objects. Therefore, it is possible to remove the low spatial frequency phase components by high-pass filtering the unwrapped phase image. The high-pass filtering is equivalent to subtracting a low-pass filtered image from the original image and can be implemented by convolving the original image with a Gaussian function. An example of phase unwrapping and high-pass filtering is shown in Figure 3.1B.

The above process can be made even faster and simpler by skipping phase unwrapping and directly performing a high-pass filtering on the original image data in k-space, a process that is called homodyne filtering (Noll et al., 1991). The method first applies a low-pass window function (e.g., Hamming or Hanning) on the original k-space data and then inverse Fourier transforms the windowed k-space data back into image space. The resulting complex image is a low-pass filtered version of the original image

$$\rho_L(x, y) = |\rho_L(x, y)| e^{-\gamma(\Delta B_0(x, y) + \Delta B_{sg}(x, y))TE} \quad (3.18)$$

which contains phase information that only has contributions from the external magnetic field inhomogeneity and global susceptibility effect. After dividing this complex image by the original complex image, i.e.,

$$\rho(x, y) = |\rho(x, y)| e^{-\gamma(\Delta B_0(x, y) + \Delta B_{cs}(x, y) + \Delta B_{sg}(x, y) + \Delta B_{sl}(x, y))TE} \quad (3.19)$$

a high-pass filtered image is created

$$\rho_H(x, y) = \frac{\rho(x, y)}{\rho_L(x, y)} = |\rho_H(x, y)| e^{-\gamma(\Delta B_{cs}(x, y) + \Delta B_{sl}(x, y))TE} \quad (3.20)$$

The phase image from the high-pass filtered image is

$$\phi_H(x, y) = -\gamma(\Delta B_{cs} + \Delta B_{sl})TE \quad (3.21)$$

which contains phase information contributed only from chemical shift and local susceptibility effect. In tissues, the most significant chemical shift comes from protons in fat. However, in normal brain tissues, fat concentration is small and has a shorter T2/T2* than water (when a GRE sequence is used), which generates very little signal on a heavy T2*-weighted image. Thus, the chemical shift effect from fat is usually ignorable in T2*-weighted MR neuroimaging. To make the phase image independent to TE and represent only a frequency shift map, one can normalize it by the TE value.

The drawback of Homodyne filtering is that incomplete unwrapping may occur in regions with sharp phase transition (Figure 3.1C). Whereas phase unwrapping preserves all of the spatial frequency of phase information, the image tends to have less residual phase wraps after subsequent high-pass filtering.

One critical step in the above processing is the selection of the appropriate window size of the low-pass filter. A narrow window could result in large number of residual low frequency phase components or wraps, whereas a wide window could reduce contrast between large structures. Figure 3.1B&C shows the effect of low-pass filter size on the final high-pass filtered phase images. Usually, an optimal window size is determined empirically after trials with a few different values.

3.3.3 Multi-coil phase image combination

The use of multiple phased array coils has become more and more popular due to the higher SNR that can be achieved with surface coil elements that are smaller and closer to objects than conventional volume coils. More importantly, the signal redundancy arising from multi-coil receivers can be utilized to accelerate image acquisition by reducing the number of data sampling, e.g., the number of phase-encoding lines in traditional Cartesian k-space sampling.

One issue arising from using multi-coil receivers is that images from each individual coil have to be combined together to generate a single image. Ideally this should be done by weighting each coil value of a pixel by the complex sensitivity of the corresponding coil in that pixel and then adding up coil values from all elements. Although the coil sensitivity can be estimated from a separate calibration scan, it is hard to eliminate calibration errors due to, for example, misregistration between calibration and real image scans. For simplicity, the magnitude image from each coil can be thought as a reasonable approximation to the sensitivity of that coil, and this leads to an efficient way to combine the magnitude image, which is called a sum of squares reconstruction (Rommer, 1990), i.e.,

$$\rho_{comb}(x, y) = \sqrt{\sum_{coil=1}^n |\rho_{coil}(x, y)|^2} \quad (3.22)$$

where ρ_{comb} is the combined magnitude image, ρ_{coil} is the complex image from each coil, and n is the number of coil. Clearly all phase information is eliminated from the combined image.

One intuitive way to combine multi-coil phase values is to calculate the phase angle after complex summation across all coils

$$\phi_{comb}(x, y) = \angle \left(\sum_{coil=1}^n |\rho_{coil}(x, y)| e^{i\phi_{coil}(x, y)} \right) \quad (3.23)$$

where Φ_{comb} and Φ_{coil} are the combined phase and the phase from a single coil, respectively. Although the method is straightforward, it often causes a destructive rather than a constructive addition of phase from different coils, resulting in the loss of SNR on the combined phase image. The reason behind of this is that there is a spatially varying constant phase offset Φ_o superimposed on the actual phase. Since Φ_o depends on the geometry relation between each pixel and each coil, the offset varies between coils and between pixels from the same coil. A constant offset has to be removed from Φ_{coil} before 3.23 can be used. Φ_o can be estimated on a pixel by pixel basis or on a global sense if the variation in phase offset between pixels in a same coil can be ignored. In the former case, it can be derived from the phase difference from echoes at different TEs (Robinson et al., 2011), and in the latter case a region of interest is first defined (e.g., a small center part of image) and then the average phase from the region is calculated and treated as the phase offset for each pixel in the relevant coil (Hammond et al., 2007). Once Φ_o is known, equation 3.23 can be write as

$$\phi_{comb}(x, y) = \angle \left(\sum_{coil=1}^n |\rho_{coil}(x, y)| e^{i(\phi_{coil}(x, y) - \phi_{0_coil}(x, y))} \right) \quad (3.24)$$

The phase images combined in this way is subject to subsequent phase unwrapping or homodyne filtering.

Alternatively, phase combination can be done after phase unwrapping or homodyne filtering has been applied to each coil individually. In this way, there is no need to estimate Φ_o because, as a low frequency component, it will be filtered out after the high-pass filtering. The unwrapped phase from each coil can be combined in a weighted average manner

$$\phi_{comb}(x, y) = \angle \left(\sum_{coil=1}^n W_{coil} e^{i\phi_{coil}(x, y)} \right) \quad (3.25)$$

where W_{coil} could be

$$W_{coil} = \frac{|\rho_{coil}(x, y)|}{\sqrt{\sum_{coil=1}^n |\rho_{coil}(x, y)|^2}} \quad (3.26)$$

or

$$W_{coil} = \frac{|\rho_{coil}(x, y)|^2}{\sum_{coil=1}^n |\rho_{coil}(x, y)|^2} \quad (3.27)$$

The difference between 3.26 and 3.27 is that the latter weighs more heavily on coils with higher SNR than the former. One drawback of this method is that phase unwrapping or homodyne filtering on individual surface coil may fail in the regions with low SNR as each coil is only sensitive to a small local region of a loaded object. Thus, masking low SNR regions may be required before doing phase unwrapping and homodyne filtering.

Unwrapped and combined phase images contain information that display local susceptibility contrast of tissues, such as veins, iron deposition in deep gray matter in normal brain, or abnormal iron accumulation in neurological diseases.

3.4 Susceptibility weighted imaging

Although phase images themselves are very useful, they can be further utilized to enhance T_2^* contrast on magnitude images. This can be done by creating a phase mask from the phase image and then applying the phase mask to the magnitude image.

3.4.1 Phase mask for SWI

The generation of phase mask is arbitrary and flexible depending on specific phase values of interest (Haacke et al., 2011). For example, for veins or iron-rich tissues, which are paramagnetic and have negative phase values, the mask can be defined as

$$\phi_{mask}(x, y) = \begin{cases} \frac{\pi + \phi(x, y)}{\pi}, & \text{if } -\pi < \phi < 0 \\ 1, & \text{else} \end{cases} \quad (3.28)$$

i.e., negative phase values are mapped to a value between 0 and 1 while all other phase values are set to 1. The phase mask defined like this is so called negative phase mask. In contrast, for diamagnetic tissues, a positive phase mask can be defined

$$\phi_{mask}(x, y) = \begin{cases} \frac{\pi - \phi(x, y)}{\pi}, & \text{if } 0 \leq \phi(x, y) < \pi \\ 1, & \text{if } -\pi < \phi(x, y) < 0 \end{cases} \quad (3.29)$$

where negative phase values are set to 1 while 0 and positive phase values are mapped to a value between 0 and 1. More complicated phase masks can be defined depending on particular applications, such as masks defined by a few segmented or nonlinear functions. An example of a negative phase mask and its original phase image are shown in Figure 3.2(A) and (B), respectively.

To create a susceptibility-weighted magnitude image $\rho_s(x, y)$, a phase mask is multiplied to the original magnitude image $\rho(x, y)$ for a few times.

$$\rho_s(x, y) = \rho(x, y) \cdot \phi_{mask}^m(x, y) \quad (3.30)$$

The number of multiplications m is empirically chosen to achieve an optimal contrast-to-noise ratio. A value of 4 is usually set for m (Haacke et al., 2011). To have a better visualization of

veins or other susceptibility-enhanced structures, $\rho_s(x,y)$ can be minimum-intensity projected through several slices. A comparison between an original magnitude image, its SWI image and minimum intensity projection (MIP) of SWI image are shown in Figure 3.3C~E.

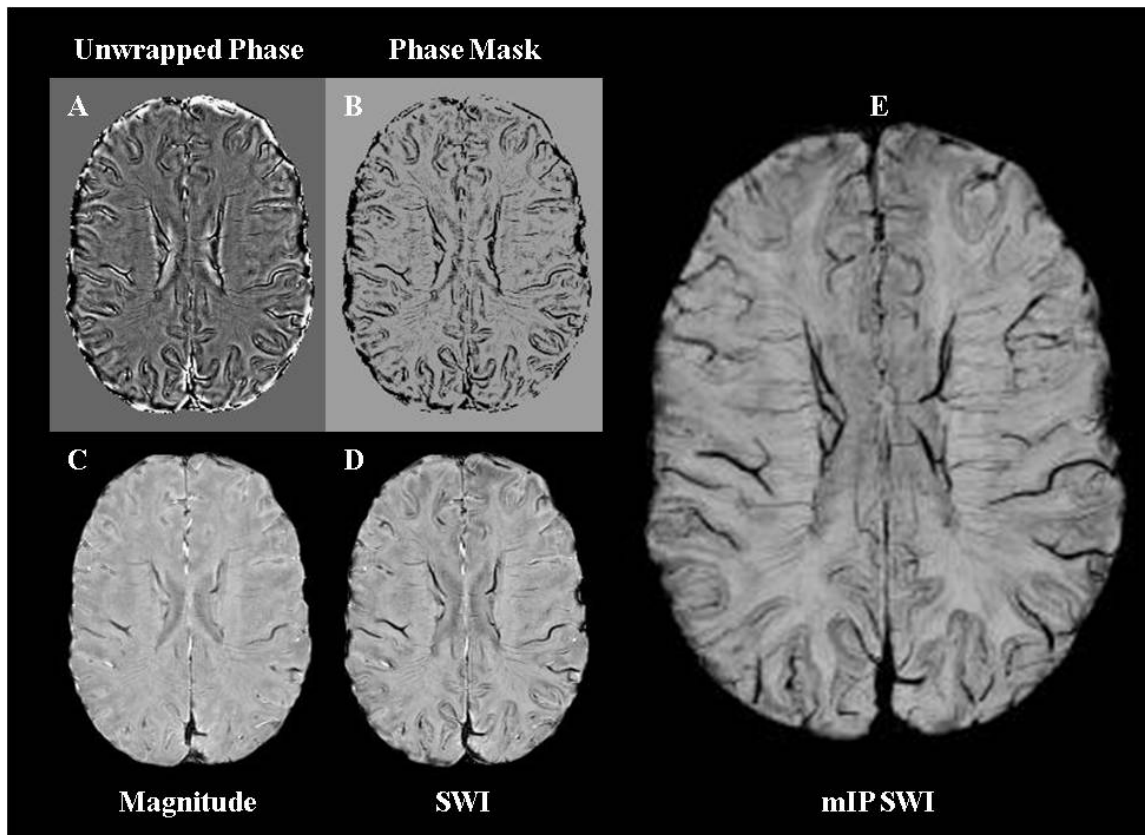


Figure 3.2 Images that are produced through the pipeline of SWI processing.

In the processing of SWI, the phase image (A) is first unwrapped to create a negative phase mask. Then the magnitude image (C) is multiplied by the phase mask for 4 times to produce the SWI image (D), which is further processed through minimum intensity projection over 8 mm to generate the SWI image (E). Compared to the original magnitude image, the contrast of veins is significantly enhanced on both SWI and MIP SWI.

3.4.2 SWI sequence

The gradient-echo sequence is the workhorse for SWI, because it manifests local susceptibility effect as $T2^*$ contrast on magnitude images and phase contrast on phase images.

Although it is possible to use a 2D gradient-echo sequence for SWI, the 3D GRE sequence is usually preferred, as the latter usually has higher SNR and/or higher resolution than the former. Figure 3.3 shows a 3D gradient-echo sequence, which has one more encoding gradient added in the slice selection direction (along z) compared to the 2D GRE sequence in Figure 2.3. The flip angle θ is usually small for SWI in order to create a flat contrast between white matter, gray matter and cerebral spinal fluid.

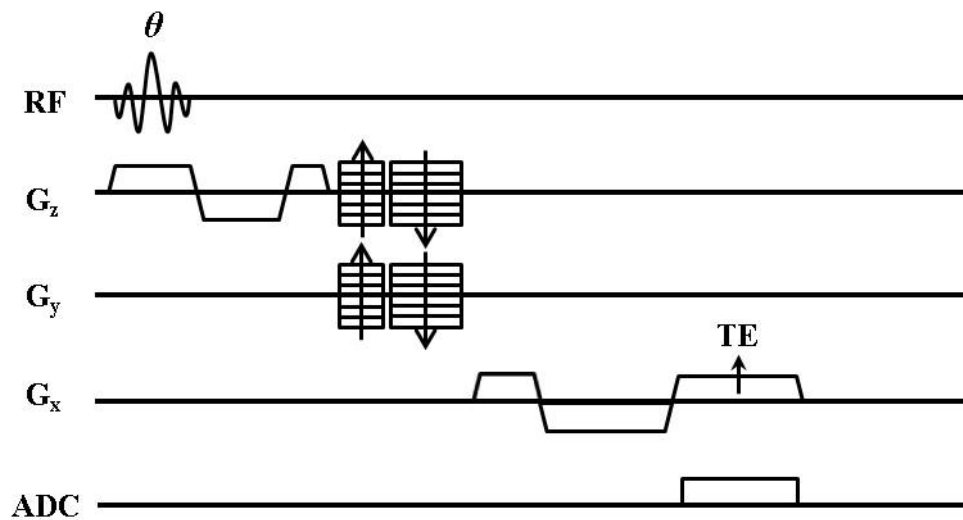


Figure 3.3 A typical 3D gradient-echo sequence used for SWI

In addition to the encoding gradient, flow compensation gradients are also added in all three spatial directions in the 3D gradient-echo sequence. When encoding gradients are turned on, rapid flow in blood will force protons in the blood to accumulate additional phase, which is proportional to the velocity of the protons and cannot be refocused by encoding gradients themselves. Since SWI should be only sensitive to phase changes introduced by local susceptibility effect, the phase accumulated from the rapid flow must be compensated. If the flow is first order, i.e., with a constant velocity, the additional flow compensation gradients can compensate or eliminate the phase accumulated by the flow at TE.

3.4.3 Data acquisition of SWI

When using the 3D GRE sequence for SWI, high imaging resolution is usually preferred, because when pixel size become smaller, the degree of global field inhomogeneity across a pixel is reduced, which suppresses the global $T2^*$ effect on the magnitude image. On the other hand, partial volume effect on the phase image is reduced by using a smaller pixel size, which helps preserve the contrast of local phase offset. Although high resolution may cause the loss of local $T2^*$ contrast on the magnitude image, the contrast can be recovered and enhanced after applying the phase mask to the magnitude image.

Because of the high resolution imaging, the acquisition time of SWI could be long. To make it clinically tolerable, its acquisition time can be made shorter by using parallel imaging techniques such as SENSE (Pruessmann et al., 2009), GRAPPA (Griswold et al., 2002) or ARC (Brau et al., 2008). These techniques allow k-space to be undersampled along the phase encoding or slice selection direction or both, hence reducing the total number of sequence repetitions. However, both high resolution and parallel imaging lead to SNR loss on images, and this is one of reasons why SWI at higher magnetic fields is preferred, as the SNR is higher at the higher field strengths or the higher SNR can be used to trade off for an even higher resolution and/or faster parallel imaging acceleration than those with lower fields. More importantly, the susceptibility effect also scales with field strength, which is the major factor that drives SWI toward higher field strength.

Since the susceptibility difference between veins or tissues with iron deposition and surrounding parenchyma tissues is small (usually around a few to tens ppm), SWI of these objects has to use long TEs to allow them to accumulate sufficient phase offset in the images.

While a long echo time is favorable for displaying susceptibility effect, there are several problems that arise when acquiring images at long TEs. Firstly, the SNR is relatively low. Secondly, objects of interest may be masked by overlapping structures that have high magnetic susceptibility. For example, veins in basal ganglia may not be visible as the latter is an iron-rich tissue, which may look as dark as veins at long TEs. Thirdly, and most important of all, phase wrapping is inevitable at long TEs. In regions with severe phase wrapping, it is difficult to eliminate all of the wraps, resulting in artifacts in the phase images. To alleviate these problems, multi-echo SWI has been proposed (Du et al., 2009; Denk et al., 2010), which uses GRE sequence with multiple echoes at different TEs. Both SNR and CNR can be improved by averaging images from several echoes. By looking at individual images local susceptibility effect can be detected in images from shorter TEs. Also improved phase unwrapping algorithms can be developed by using complementary information from several echoes (Feng et al., 2013) and the quality of multi-coil phase image combination is enhanced by more efficiently eliminating phase variations that depend upon coil geometry (Robinson et al., 2011).

Chapter 4 Longitudinal MR Phase imaging of Multiple Sclerosis Lesions

In this chapter, the presence of susceptibility effect on MR phase images from patients with Multiple Sclerosis is introduced. The potential sources of the susceptibility contrast in white matter lesions are proposed based on the observations from a longitudinal *in vivo* MR imaging study at 7T, which tracked the evolution of phase contrast in the lesions for up to 2.5 years.

4.1 Introduction

Multiple sclerosis (MS) is an inflammatory and demyelinating neurodegenerative disease of the central nervous system. Previous magnetic resonance imaging (MRI) studies have shown that MS lesions show high contrast in phase and susceptibility-weighted images (SWI) (Hammond et al., 2007; Hammond et al., 2008; Haacke et al., 2009; Eissa et al., 2009; Mainero et al., 2009; Pitt et al., 2010; Grabner et al., 2011; Bagneto et al., 2011). However, the source of this contrast remains unclear. Accumulating evidence has suggested that the underlying contrast mechanism for MS lesions seen on phase images is at least in part due to abnormal iron deposition, content or presence (Pitt et al., 2010; Bagneto et al., 2011; Duyn et al., 2007; Yao et al., 2009; He et al., 2009). The purpose of this study was to evaluate the clinical utility of serial phase imaging for MS and to have a better understanding of the time course of phase contrast and its implication for iron accumulation in MS lesions

MRI is sensitive to iron accumulation in MS (Haacke et al., 2005; Stankiewicz et al., 2007) and is associated with hypointensity on conventional T2 or T2*-weighted MR images (Drayer et al., 1987; Bakshi et al., 2002), which can be used to make quantitative measurements of iron concentration by mapping relaxation time, R2 or R2* (Haacke et al., 2005; Stankiewicz et al., 2007; Khalil et al., 2009). However, these methods may be confounded by changes in water diffusion and relaxation times (Haacke et al., 2005; Stankiewicz et al., 2007;), which can lead to

inaccurate measurement of iron, especially for MS white matter plaques, in which iron deposition and free water components in edema, inflammation and gliosis are usually co-localized. Recently, with the combination of T2*-weighted gradient echo (GRE) sequence and advanced image post-processing methods, MR phase and SWI images that are sensitive to paramagnetic susceptibility effects of iron have been applied for MS (Hammond et al., 2007; Hammond et al., 2008; Haacke et al., 2009; Eissa et al., 2009; Mainero et al., 2009; Pitt et al., 2010; Grabner et al., 2011; Bagneto et al., 2011). Phase accumulation as a result of local perturbation of the main magnetic field due to iron deposition can be revealed on high-pass-filtered phase images, or further processed and combined with magnitude images to create SWI images (Haacke et al., 2009). Since susceptibility effects are independent of water relaxation and scale with field strength, phase images of MS subjects at high field provide contrast specific to field perturbations and show excellent contrast of local iron in white matter plaques. Furthermore, phase images have demonstrated potential clinical utility in identifying a subset of MS lesions that are not detected on magnitude images (Hammond et al., 2007; Hammond et al., 2008; Haacke et al., 2009; Eissa et al., 2009; Mainero et al., 2009; Pitt et al., 2010; Grabner et al., 2011; Bagneto et al., 2011).

Imaging iron in MS is of interest because abnormal elevation of iron concentrations has been proposed to have a pathogenic role in MS by promoting oxidative damage (Stankiewicz et al., 2007; Levine et al., 2004), although it is more likely to represent an epiphenomenon rather than a causal event. The increased iron in MS may come from several different sources, including damaged oligodendrocytes and myelin (Levine et al., 2004), blood brain barrier (BBB) compromise (Craelius et al., 1982), macrophage infiltration (Levine et al., 2004; Craelius et al., 1982; Adams, 1988) and astrocytes (Mehindate et al., 2001). Oligodendrocytes and myelin have

high concentrations of iron in order to catalyze their high-level lipid production (Levine et al., 2004). This iron can be released in the extracellular compartment when these cells, the primary targets of MS, are destroyed. Impairment of the BBB may increase iron concentration by allowing blood iron to leak into perivascular regions (Craelius et al., 1982; Adams, 1988). Iron from these sources can be sequestered in macrophages and microglia, making them iron-rich as well (Levine et al., 2004; Craelius et al., 1982; Adams, 1988).

Because the cellular activity of MS lesions changes over time, the presence and spatial distribution of iron in MS lesions is expected to vary as lesions form and evolve and this variability should be detectable by serial clinical MR phase imaging. The purpose of this pilot study was to test this hypothesis by following the evolution of MS lesions in serial images acquired at a field strength of 7T. Such high field strength permitted acquiring high-resolution images with heightened phase contrast in clinically acceptable scan times.

4.2 Materials and Methods

4.2.1 Patients

Five clinically stable (no clinical attacks documented during the observation period) relapsing-remitting MS subjects were serially scanned at 3T and 7T. The demographics were 3 females, 2 males, mean age of 51 years, mean disease duration of 17 years, mean EDSS of 3.1. All subjects were on the same injectable FDA-approved therapy for the duration of the study and all gave their written informed consent to participate in the longitudinal study approved by the UCSF Committee of Human Research.

4.2.2 Image Acquisition

The subjects were serially scanned using a whole-body GE 7T scanner (GE Healthcare, Waukesha, WI) equipped with an 8-channel receive phased array coil (Nova Medical, Wilmington, MA). Axial oblique 2D T2*-weighted GRE images were acquired at a spatial resolution of $195 \times 260 \mu\text{m}$ or $350 \times 350 \mu\text{m}$ with TE/TR of 12 to 15/250 milliseconds, flip angle of 20° , slice thickness of 2mm, matrix/field of view $1024 \times 768/20\text{cm}$ or $512 \times 512/18\text{cm}$, 3 NEX (number of excitations), and scan time of approximately 9 or 6.5 minutes. Nine consecutive slices were prescribed parallel to the inferior callosal line from a mid-sagittal scout image to assure coverage of the corpus callosum, the location where MS lesions are frequently seen. If more coverage was needed, another nine slices superior to the previous block were also acquired using the same sequence. Immediately after the 7T scan, the subjects also received a post contrast Gadolinium-enhanced (Gd) clinical scan with a GE 3T scanner to determine whether any contrast-enhancing lesions were present. The mean follow-up time for the five subjects was 21.8 months (ranging from 16 to 31 months) with an average of 3.2 scans/subject (16 scans total). The interval between scans varied from 1 month to 31 months.

4.2.3 Image Processing

The magnitude and phase images were reconstructed using the method previously described (Hammond et al., 2008). In brief, the magnitude signal from each channel in the complex image volume was combined using a root-sum-of squares algorithm to obtain a magnitude image; individual phase images from each channel were then unwrapped on a slice by slice basis using the PRELUDE algorithm (Jenkinson, 2003) to generate a full range of phase images; each unwrapped image was high-pass filtered by complex division of the unwrapped image by a low-pass filtered unwrapped image; the filtered phase images from each coil were

finally combined by weighting by the magnitude image and summing across channels. The follow-up magnitude images were first co-registered (3D rigid body registration) to magnitude images from the baseline using software based on the VTK CISG registration toolkit (Studholme et al., 1999), and the transformation matrix obtained from magnitude image co-registration was then applied to the corresponding phase image at each time point.

4.3 Results

A total of 75 phase lesions were found in the 5 subjects at baseline. None of the lesions showed Gadolinium-enhancement (Gd-). As the source of iron in an MS lesion may differ based on the appearance or morphology of the phase contrast, lesions were divided into two categories according to their phase contrast patterns: (1) nodular and uniform hypointense lesions (n=70) and (2) ring lesions with a hypointense rim at the edge of a lesion (n=5). Fifty-nine of the 70 nodular lesions were observed both on magnitude and phase images, while the remaining 11 were only visualized on phase images. The ring of all 5 ring phase lesions (from 4 different patients) was either not detected on magnitude images or better contrasted on the phase images.

Figure 4.1 shows the 5 ring phase lesions followed from baseline to month 1, month 4 and month 16 (lesions 1-3), month 28 (lesion 4), and month 31 (lesion 5). None of the phase ring lesions disappeared over time, even when followed for up to 31 months. Similarly, none of the nodular phase lesions disappeared over time (Figure 4.2). As can be seen, once detected, all of the lesions maintained the same contrast, size, and morphological appearance over time on both phase and magnitude images.

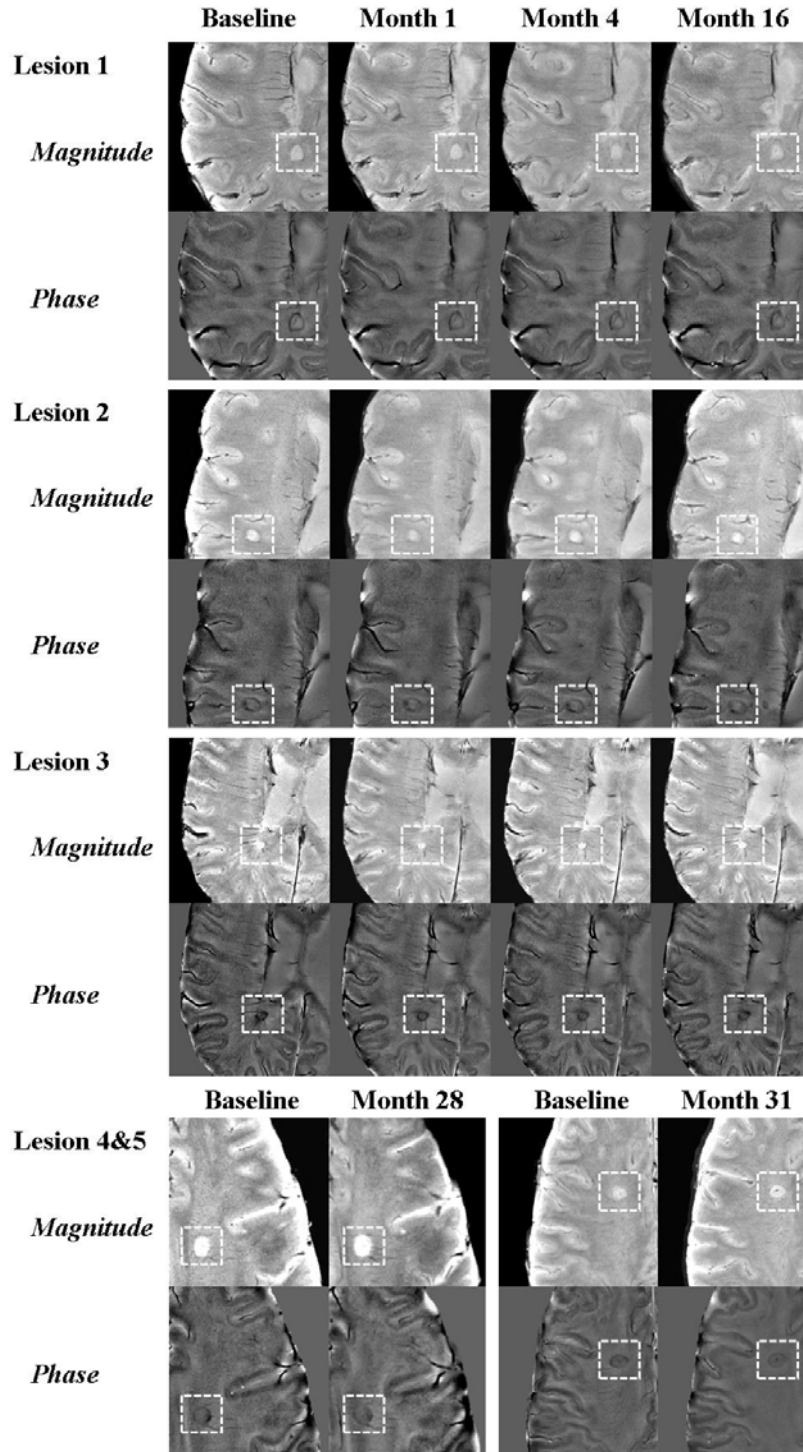


Figure 4.1 Evolution of all five ring lesions observed in the study

Evolution of all five ring lesions observed in the study. Lesion 1 and 2 were from a same patient, and the other 3 were from 3 different patients. Lesion 1-3 had follow-ups at 1, 4 and 16 month after their baseline scans, lesion 4 had one follow-up at 21 month after the baseline scan, and lesion 5 had one follow-up at 31 month after the baseline scan. Note that none of the lesions underwent obvious change in terms of image intensity and morphology.

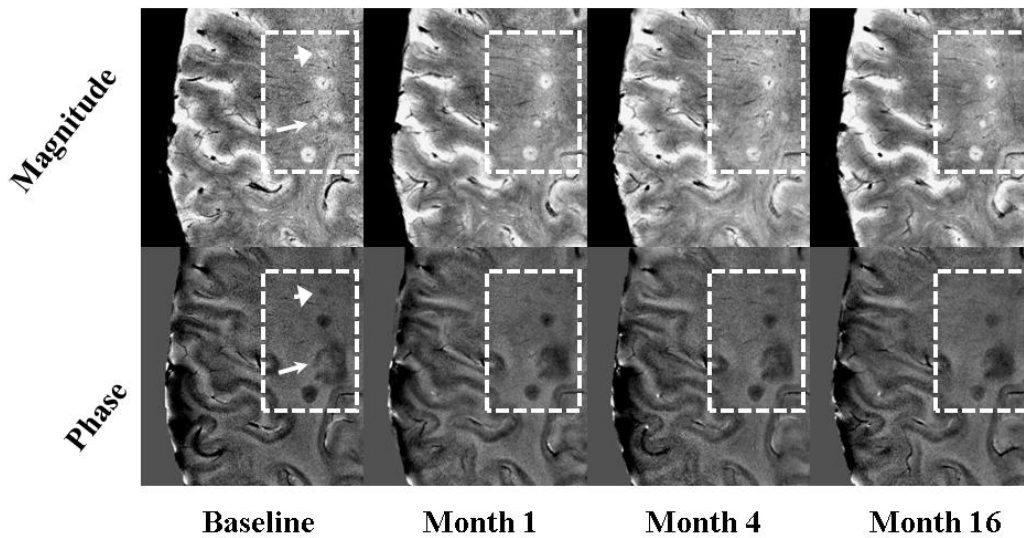
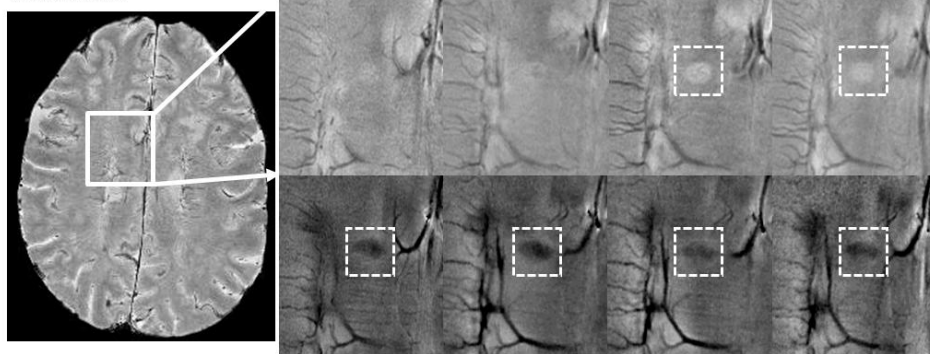


Figure 4.2 Evolution of representative nodular-like lesions observed in a patient

Evolution of representative nodular lesions observed in a patient. The lesions were followed at month 1, 4 and 16 after baseline. Similar to ring lesions, the overall appearance and image intensity showed no change over time. There was a lesion (arrow head) shown on the magnitude image but not well-defined on the phase image. While the other lesions were seen on the both magnitude and phase images, one conglomeration of magnitude lesions (arrow) had different morphology on the phase images

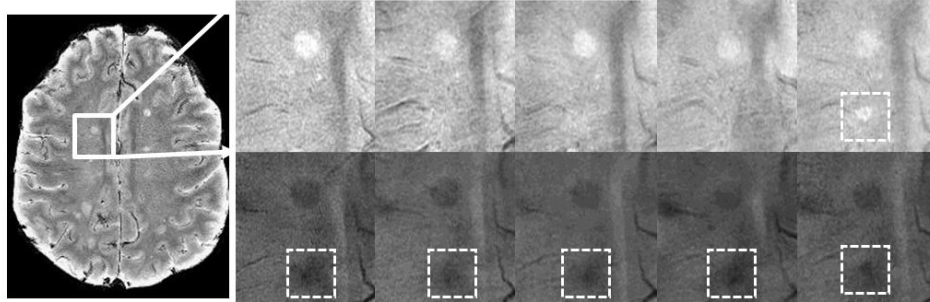
Lastly, we made the following observation during follow-ups. Three magnitude lesions (lesions 6-8 in Figure 4.3), not prospectively identified as abnormal signals initially, were either better appreciated using phase contrast imaging (lesion 6 & 8) or preceded (lesion 7) by phase changes. Lesion 6 showed minimal magnitude contrast at baseline, which was not obvious and read as abnormal until month 4, whereas it had already shown substantial and stable phase contrast since baseline. Lesion 7 (from a different patient) developed an abnormal magnitude contrast signal at month 16 but had showed apparent phase contrast on all scans since baseline. Lesion 8 (from the same patient as lesion 7) appeared at month 16, but the abnormality was more clearly seen on the phase rather than magnitude images (again the corresponding magnitude abnormality was subtle). Of note, all 3 lesions were Gd- on the corresponding 3T post-Gadolinium images.

Lesion 6



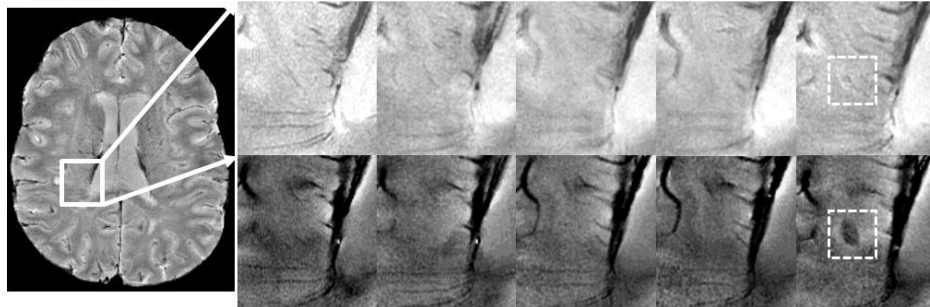
Baseline Month 1 Month 4 Month 16

Lesion 7



Baseline Month 1 Month 4 Month 13 Month 16

Lesion 8



Baseline Month 1 Month 4 Month 13 Month 16

Figure 4.3 Three magnitude lesions that were initially not prospectively identified.

Lesion 6 showed minimal magnitude contrast (top row) at baseline but not obvious until month 4, whereas its corresponding phase lesion had been seen on the phase image (bottom row) since baseline scan. Lesion 7 (from a different patient) was first seen on the magnitude image 16 months after the baseline scan, but its phase lesion already arose at the baseline scan. Lesion 8 (from the same patient as lesion 7) arose on the phase image at month 16. At the same time, the signal abnormality on the magnitude image was so subtle such that it was only slightly observable on retrospective review.

4.4 Discussion

This pilot study followed several white matter lesions from patients with clinically stable relapsing-remitting MS for up to 2.5 years using a T2*-weighted GRE imaging sequence at the high field strength of 7T and using conventional contrast-enhancing scans acquired at 3T. Two major observations were made. Firstly, phase lesions did not disappear over time and, secondly, phase lesions preceded the appearance of hyperintense lesions on magnitude images.

Two distinct types of MS lesion appearance were observed in the phase images, (1) ring lesions showing a dark rim at the lesion edge and; (2) nodular lesions showing roughly uniform hypointensity. These have both been reported in several previous studies (Hammond et al., 2007; Hammond et al., 2008; Haacke et al., 2009; Eissa et al., 2009; Mainero et al., 2009; Pitt et al., 2010; Grabner et al., 2011; Bagnato et al., 2011). None of the lesions observed in the MRI phase images at baseline showed obvious changes in radiological features such as intensity, size, and morphology through the course of the follow-up, suggesting that all baseline phase lesions were chronic lesions. This is further supported by the findings that lesions never showed enhancement on post-contrast T1-weighted images, which is generally a hallmark of active lesions (Kermode et al., 1990).

The ring phase lesions were associated with central penetrating veins, surrounded by a classic abnormal ovoid signal on the corresponding magnitude image. Although this pattern could be consistent with the presence of infiltrating iron-rich activated macrophages surrounding a demyelinating area, as reported in previous studies (Pitt et al., 2010; Bagnato et al., 2011), the persistence of the signal on serial images (over 2.5 years in some cases) speaks against the acute active stage of the macrophages, because only few activated macrophages can be found in chronic lesions (Adams et al., 1990; Brück et al., 1995). However, since the phenotype of

macrophages may undergo changes with chronicity, as shown in histopathological studies ((Lucchinetti et al., 2000; Barnett et al., 2006), activated pro-inflammatory macrophages that contain myelin debris (and hence iron) may later become anti-inflammatory, promote tissue repair, and stay for an extended period of time (Lassmann et al., 2011). It is unclear why the macrophages could stay there for such a long time, but similar observations have been found in MR studies of intracerebral hemorrhage, where perivascular collections of iron in hemosiderin within macrophages may be visible indefinitely on MRI (Dimigen et al., 2004).

Phase contrast in chronic nodular lesions (Figure 4.2) may be from astrogliosis, the scar-like tissue that usually appears within a few months of the appearance of a fresh lesion and is a pathological hallmark of chronic lesions. Astrocytes have been reported to accumulate iron in the absence of oligodendrocytes and myelin (Connor et al., 1990). A possible mechanism for astrocytes to become iron-rich under neurological affliction could be the overexpression of heme oxynenase-1, an enzyme that promotes mitochondria iron sequestration in oxidative stressed astrocytes (Mehindate et al., 2001; Schipper et al., 2004) The sustained presence of nodular lesions may be explained by the prolonged survival of iron-laden astrocytes due to their robust ability to antioxidant defenses, cytoprotection, and anaerobic metabolism compared with other neural cell types (Schipper et al., 2004).

This study also demonstrated that longitudinal changes in lesion contrast could, in some cases, be better appreciated on phase compared with magnitude images. In one case (lesion 7 in Figure 4.3), the presence of a phase lesion even preceded the appearance of a hyperintense signal on the corresponding magnitude images. Although larger controlled experiments are needed to validate this observation, the importance of this finding and its potential impact on our understanding of the pathological course of MS were worth reporting in this pilot study. As

previously described, abnormal signal changes from magnetization transfer imaging (Goodkin et al., 1998; Pike et al., 2000) and diffusion-weighted imaging studies (Werring et al., 2000) preceded MS lesions detected by conventional MR images. Such phase contrast changes could also reflect an early process of myelin breakdown or abrupt loss of white matter tissue integrity as pointed in an experimental study (He et al., 2009). Alternatively, the discrepancy between phase and magnitude images may also be explained by there being opposing contrast mechanisms on magnitude and phase images due to different pathological processes. The T2* prolongation induced by inflammation and edema may be offset by T2* shortening due to iron deposition, leading to poor visibility of hyperintense lesions on T2* magnitude images.

There are several limitations in this pilot study. All patients investigated had a diagnosis of stable relapsing-remitting MS. To obtain a comprehensive understanding of the role of phase imaging in studying the evolution of MS, future studies would need to include patients with different types of MS (e.g., early RRMS, secondary and primary progressive MS) and include contrast-enhancing lesions. Such studies should also address the sensitivity of phase over conventional post-contrast T1-weighted imaging. The qualitative findings of these pilot results call for a longer and larger sample size study with rigorous standardized frequent imaging protocol, including the use of quantitative susceptibility mapping (Schweser et al., 2011) to generate intrinsic magnetic susceptibility map from phase images. Such quantitative mapping may offer a more robust tool to quantify the susceptibility contrast from MS lesions, especially in longitudinal monitoring of lesion progression. Lastly, one cannot emphasize enough the need for additional histopathological-MRI studies specifically investigating the source of iron content in different central nervous system regions, such as chronic lesions, active lesions, and normal appearing white and gray matter MS tissue.

4.5 Conclusions

Phase lesions from 5 relapsing-remitting patients were serially followed using a 7T high field MR scanner. All 5 ring phase lesions observed at baseline remained unchanged for up to 31 months, challenging the notion that such lesions reveal the presence of acute active iron-rich macrophages. It suggests that either different phenotypes of iron-rich macrophages persist longer than previously expected or other mechanisms of tissue injury contribute to the contrast. Also the appearance of some lesions may be better appreciated on phase rather than on magnitude images, suggesting that phase imaging could potentially be used to further study early indicators of emerging lesions.

Chapter 5 Susceptibility-weighted Imaging of CMBs at 3T and 7T

In this chapter, the usefulness of susceptibility-weighted imaging (SWI) of cerebral microbleeds (CMB) is demonstrated when compared to traditional magnitude images. The quality of SWI images and their ability in characterizing CMB are compared between 3T and 7T scanner for patients with radiation treated brain tumors. Benefits and pitfalls of SWI at ultra-high field strength of 7T for CMBs are also discussed.

5.1 Introduction

Gliomas are the most common type of primary brain tumors with heterogeneous and diffuse histopathology. Radiation therapy is a mainstay their treatment with the goal of removing as much residual tumor as possible following maximal safe surgical resection. However, even with modern technology and treatment planning strategies, radiation therapy can cause injury to normal brain tissue (Valk et al.; 1991). One of the principal effects of radiation injury is cerebral hemorrhage, which over time results in the formation of cerebral microbleeds (CMBs) that comprise focal perivascular collections of hemosiderin and persist for years after receiving treatment (Shobha et al., 2009; Lupo et al., 2012).

Since hemosiderin is a paramagnetic ferric-containing protein and its susceptibility effect results in local dephasing and subsequent loss of signal, CMBs are manifested as small, round, hypointense lesions on T2*-weighted images obtained using gradient echo sequences. The clinical relevance of detecting CMBs in cerebrovascular disorders such as cerebral amyloid angiopathy and hypertensive encephalopathy has been widely discussed, but their role as a potential diagnostic and prognostic marker is still under debate (Cordonnier et al., 2007; Greenberg et al., 2009; Charidimou et al., 2011). While relatively few studies have addressed

radiation therapy-induced CMBs in gliomas, we have recently found that these lesions are distinct from calcification, are not related to patient age, increase in number over time since irradiation, and correlate with the dose and the target volume defined for radiation therapy (Lupo et al., 2012). This indicates that their burden may be a useful measurement of parenchymal radiation injury and therefore provide information for treatment evaluation.

Although the susceptibility effect scales with field strength and MR magnitude images that were acquired from scanners with field strength of 3T or 7T have been reported to detect more CMBs compared to those from 1.5 T (Theysohn et al., 2011; Conijn et al., 2011), it is not clear how much sensitivity is gained with 7T over 3T for CMB detection, which is often confounded by macroscopic susceptibility artifacts and/or iron-rich tissues. Susceptibility-weighted imaging (SWI) combines information from both magnitude and phase images from a T2*-weighted gradient echo sequence, further enhancing the susceptibility effect on the final images and thus improving the detection sensitivity of CMBs (Nandigam et al., 2009; Ayaz et al., 2010; Theysohn et al., 2011); yet, there has been recent debate as to whether SWI is necessary at 7T where there is already heightened susceptibility in magnitude images. The purpose of this study was to compare 3T and 7T SWI and magnitude images for the detection of radiation therapy-induced CMBs in patients with treated gliomas and evaluate how the presence of susceptibility-induced artifacts and altered contrast affect detection sensitivity at 7T.

5.2 Materials and Methods

5.2.1 Patients

Ten patients with gliomas who received radiation therapy between 2 and 15 years prior to the date of imaging were recruited for this study. All patients were scanned at both 3T and 7T field strengths on the same day. This varied population allowed for a wide range of CMB

locations, contrasts, and sizes, thereby creating a broad spectrum of detection sensitivities from which to evaluate the different acquisition strategies. The study was approved by our Committee of Human Research, and written informed consent was obtained from all subjects.

5.2.2 MR imaging and image processing

High-resolution T2*-weighted imaging with a 3D flow-compensated spoiled gradient echo sequence was performed using whole-body 3T and 7T scanners (GE Healthcare Technologies, Milwaukee, WI, USA) with eight-channel phased array coils. The TE/TR was 28/46 ms at 3T and 16/50 ms at 7T, which were optimized respectively for CMB detection at individual scanner. A GRAPPA-based parallel imaging acquisition was implemented with either a twofold (3T) or threefold (7T) acceleration factor and 16 autocalibrating lines to keep the total acquisition time within 7 min. A flip angle of 20°, 24 cm FOV, in-plane resolution of 0.5×0.5 mm, and 2 mm slice thickness were used for both field strengths (Lupo et al., 2009).

The raw complex k-space data from all channels were transferred off-line to a SunBlade 2000 Workstation (Sun Microsystems, Santa Clara, CA, USA), and post-processing was performed using in-house programs developed with Matlab 7.1 software (MathWorks Inc., Natick, MA, USA) on a Linux cluster running Sun's N1 Grid Engine. A GRAPPA-based reconstruction was utilized to restore the full complex k-space data of each individual coil before employing standard SWI processing (Lupo et al., 2009). Phase masks were constructed from the full complex k-space data of each individual coil element through complex division by a low-pass filtered image and scaling the resulting negative phase values between 0 and 1 (Haacke et al., 2004). To generate a susceptibility-weighted image, the phase mask was then multiplied with the corresponding magnitude image from each channel four times, and the resulting images from each channel were combined by the square root of sum of squares method. A 72×72 Hanning

low-pass filter was used for phase mask generation at 3T, while a larger filter size of 96×96 was used at 7T due to the higher frequency phase wraps present at 7T compared to 3T. A low pass filter with edge completion was applied to the combined images to minimize any residual intensity variation across the image. Finally, minimum intensity projection (MIP) images were generated through 8-mm-thick slabs of overlapping volumes between 3T and 7T images for both magnitude images and SWI in order to provide uniform coverage for analysis.

5.2.3 Microbleed counting

Microbleeds were identified as small hypointense foci that did not correspond to vessels on consecutive slices, and counted in normal-appearing tissue outside the tumor region. Veins that traversed the axial image plane mimicking CMBs were excluded if they were of linear structure or skewed from the axis, as illustrated in Figure 5.1.

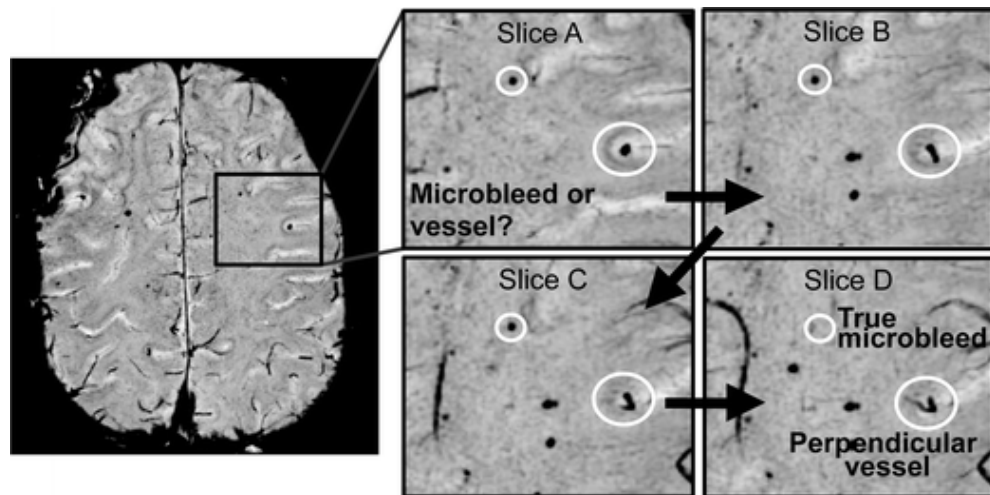


Figure 5.1 Illustration of how CMBs were identified.

CMBs are round hypointense foci that excluded vessels, tumor, or surgical borders on consecutive slices.

CMBs were counted independently by two trained raters with 6 and 11 years experience in brain tumor imaging, taking into account that the same lesion could be conspicuous on adjacent slices. In ambiguous cases, both reviewers reached a consensus after discussion with a

subspecialty certified neuroradiologist. To reduce recall bias, different image sets from a same patient were examined at least 1 week apart and in random order.

5.2.4 Statistical analysis

A two-sample paired Wilcoxon signed-rank test was performed to test whether there was a significant difference in the number of CMBs between image sets. The significance level was set to an alpha of 0.05.

5.3 Results

A total of 208 CMBs (mean, 20.8; range, 10–42) were detected using 3T SWI and 159 (mean, 15.9; range, 9–31) on the corresponding magnitude images. The 7T SWI and magnitude images detected 236 (mean, 23.6; range, 13–43) and 153 (mean, 15.3; range, 7–30) CMBs, respectively. Table 5.1 shows the CMB count and tumor location for each patient, and the results of statistical comparisons between different image sets are listed in Table 5.2.

5.3.1 Image processing: SWI vs. magnitude

There was a significant difference at both field strengths (7T: $p=0.002$; 3T: $p=0.004$) in the number of CMBs seen on magnitude and SWI images, with 54.2 % more CMBs detected at 7T and 30.8 % more CMBs detected at 3T. The contrast of CMBs to surrounding brain tissue was also greatly improved on 7T SWI compared to magnitude images, as shown in Figure 5.2a. When gain in the number of CMBs for SWI versus magnitude images was compared between 3T and 7T on a patient-by-patient basis, the gain was larger at the higher field strength ($p=0.037$).

Table 5.1 CMB Counts from 10 Patients on 3T and 7T images

Patients	3T			7T			Tumor Location
	SWI	Magnitude	Gain with SWI (%)	SWI	Magnitude	Gain with SWI (%)	
1	12	10	20.0	16	11	45.5	Frontal Lobe
2	10	9	11.1	13	11	18.2	Frontal Lobe
3	18	11	63.6	28	15	86.7	Frontal Lobe
4	13	11	18.2	18	11	63.6	Frontal Lobe
5	18	13	29.0	30	18	43.3	Parietal Lobe
6	40	31	38.5	43	30	66.7	Cerebellum/Brain stem
7	23	21	9.5	31	23	34.8	Cerebellum/Brain stem
8	18	18	0.0	15	7	114.3	Frontal Lobe/Basal Ganglia
9	14	13	7.7	13	8	62.5	Frontal Lobe/ Basal Ganglia
10	42	22	90.9	29	19	52.6	Temporal Lobe
Total	208	159	30.8	236	153	54.2	

Table 5.2 Significant Differences in CMB Detection between Different Image Sets

	Comparison Groups	<i>p</i> Value (N=10)	<i>p</i> Value (N=7)*
Field Strength	SWI: 3T vs. 7T	0.193	0.016
	SWI: 3T only vs. 7T only	0.193	0.016
	Magnitude: 3T vs. 7T	0.984	0.094
	3T SWI vs. 7T Magnitude	0.023	--
	% Gain with SWI: 3T vs. 7T	0.037	--
Image Processing	3T: SWI vs. Magnitude	0.004	--
	7T: SWI vs. Magnitude	0.002	--

Asterisk (*) indicates excluding the last 3 patients in Table 5.1, who had tumors located in deep brain tissue.
P-values in *bold* are significant

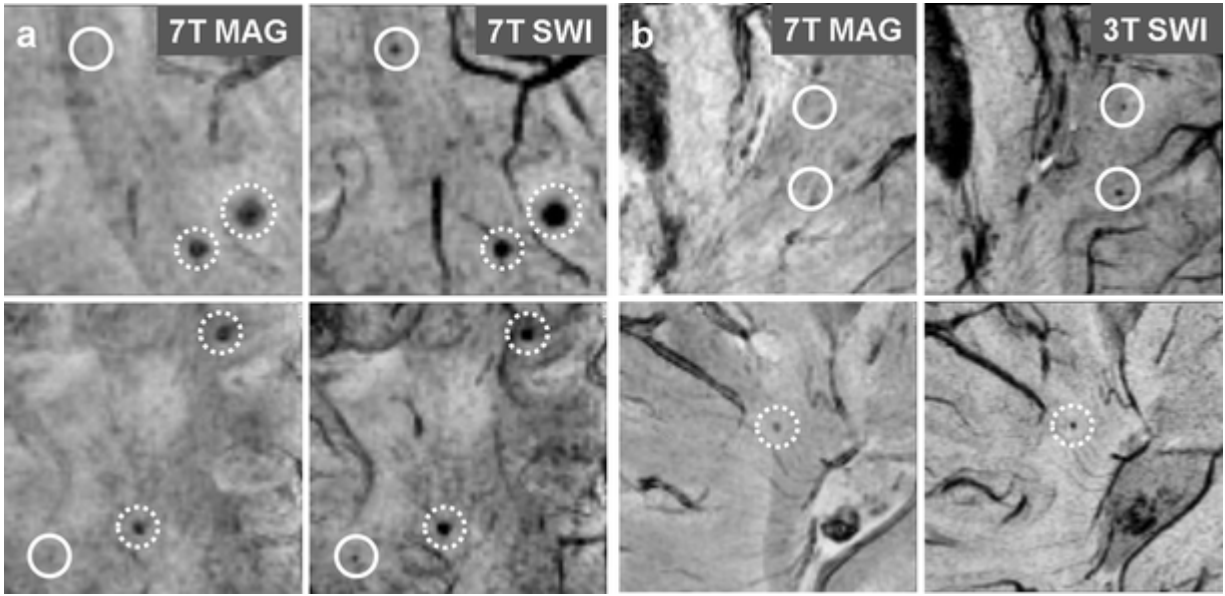


Figure 5.2 Comparison of CMB contrast between SWI and magnitude images

(a) seven tesla magnitude and SWI images from patients 7 (top row) and 10 (bottom row). Solid circle: CMBs seen on 7T SWI only; dashed circle: CMBs better contrasted on 7T SWI compared to 7T magnitude images. (b) Three tesla SWI and 7T magnitude images from patients 3 (top row) and 2 (bottom row). Solid circle: CMBs seen on 3T SWI only; dashed circle: CMBs better contrasted on 3T SWI compared to 7T magnitude images

5.3.2 Field strength: 7T vs. 3T

Seven (patients 1–7 in Table 5.1) of ten patients had more CMBs identified on 7T SWI. The other three patients (patients 8–10 in Table 5.1), who had more CMBs detected at 3T, had tumors in the temporal lobe or basal ganglia. When all ten patients were considered, there was no significant difference in the number of CMBs detected at the two field strengths. However, when the three patients with tumors in the temporal lobe or basal ganglia were excluded, the difference was significant ($p=0.016$). A representative example is shown in Figure. 5.3a&b, where panel a shows heightened CMB contrast at 7T SWI compared to 3T SWI and panel b shows CMBs masked by susceptibility artifacts at 7T SWI. Of the CMBs detected by SWI, 112 were seen at 7T but not identified at 3T, while 84 were seen at 3T but not identified at 7T. There was no

difference in CMB detection between 7T and 3T magnitude images, even after removing the latter three patients. In addition, 3T SWI detected significantly more CMBs than 7T magnitude images ($p=0.023$), as shown in Figure 5.3b.

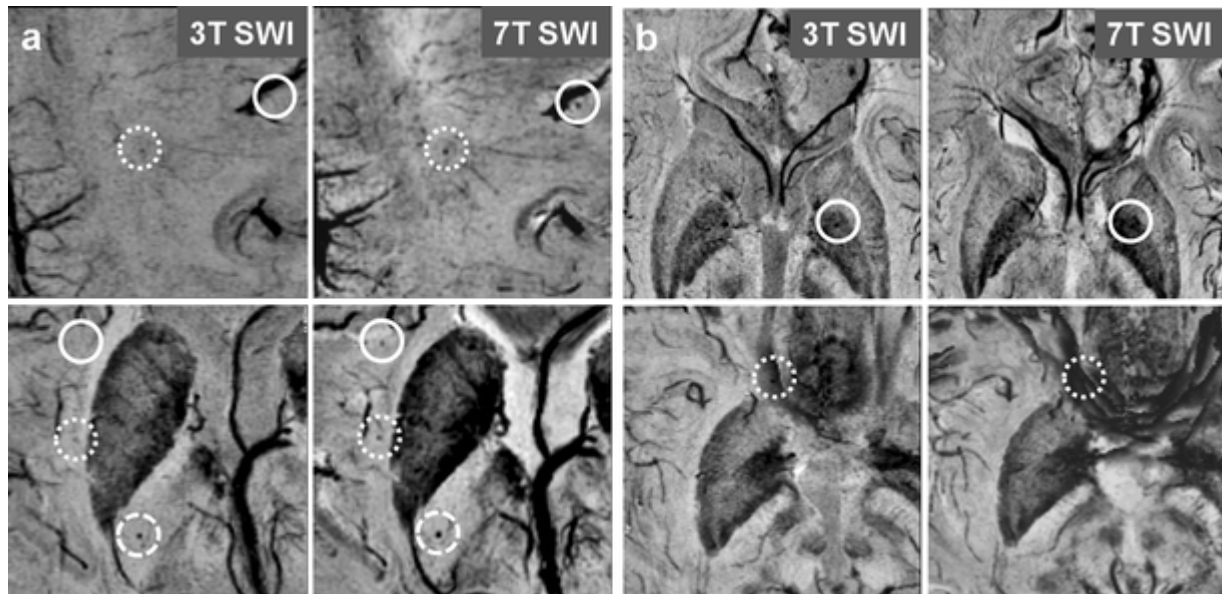


Figure 5.3. Comparison of CMB contrast between 3T and 7T SWI images

(a) Three tesla (left column) and 7T (right column) SWI images from patients 2 (top row) and 3 (bottom row). Solid circle: CMBs seen on 7T SWI only; densely dashed circle: CMBs better contrasted on 7T SWI compared to 3T SWI; loosely dashed circle: CMBs seen on both 7T and 3T SWI. (b) Three and 7T SWI images from patients 8 (top row) and 3 (bottom row). Solid circle: CMBs seen on 3T SWI but masked by the enhanced susceptibility effect of iron within the globus pallidus on 7T SWI; dashed circle: CMBs seen on 3T SWI but masked by the phase wrap artifacts on 7T SWI

5.4 Discussion

The results of this study demonstrate that detection of radiation therapy-induced CMBs benefits from applying SWI at 7T in two ways. First, 7T SWI images detected more CMBs compared to magnitude images from the same field strength. This is consistent with observations from previous studies (Nandigam et al., 2009; Theysohn et al., 2011). Second, 7T magnitude images were less sensitive in detecting CMB than 3T SWI images. Note that the incremental

gain in sensitivity achieved by using SWI was even larger at 7T compared to 3T, probably because of the increased SNR and susceptibility contrast on phase images from the former.

Although 3T and 7T SWI showed no significant difference in CMB detection for the entire patient group, there were more CMBs detected at 7T for the seven patients (patients 1–7 in Table 5.1) who had tumors located remotely from deep brain structures. The improved sensitivity of 7T SWI to CMBs in this subgroup of patients may help early diagnosis of radiation-induced microvascular injury (Shobha et al., 2009; Lupo et al., 2012). Initial studies have shown that the number of CMBs increases over time since receiving radiation therapy and often extend well beyond the initial high-dose volume and into the contralateral hemisphere as time progresses (Lupo et al., 2012). Although the exact role of CMBs remains to be seen, they have been implicated as prognostic markers of neurocognitive impairment in other diseases such as TBI, CAA, stroke, mild cognitive impairment, and Alzheimer’s disease (Scheid et al., 2003; Goos et al., 2009; Ayaz et al., 2010; Werring et al., 2010; van Norden et al., 2011; Charidimou et al., 2013). Thus, earlier detection of them using ultra-high field scanners (7T and above) may be beneficial in the clinic for patient management and prevention of further cognitive decline. This may be especially relevant for designing future treatment strategies for patients with low-grade gliomas, who have longer progression-free survival compared to more aggressive high-grade tumors, where there is debate as to whether the benefits of radiotherapy outweigh the potential negative effects (Shaw, 1990).

The decreased CMB detection sensitivity at 7T for the remaining three patients (patients 8–10 in Table 5.1) was mostly due to macroscopic susceptibility artifacts often located near air-tissue interfaces that result in residual phase wrapping that is difficult to eliminate with standard SWI filtering methods (see the bottom row in Figure 5.3b), as the filter size must be selected to

balance heightened phase contrast with removal of residual high-frequency phase wraps. Enhanced susceptibility of iron-rich tissues such as the basal ganglia (see the top row in Figure 5.3b) also can obscure CMBs at 7T. Although in this study we used standard SWI processing that would be routinely available in the clinic for comparison purposes, more advanced post-processing and local field correction algorithms (Jin et al., 2008; Liu et al., 2012) should be applied to mitigate these artifacts and recover the missed CMBs at 7T.

Previous studies are consistent with higher field strength and SWI providing improvements in detecting CMBs (Nandigam et al., 2009; Theysohn et al., 2011). Nandigam et al. (Nandigam et al., 2009) compared CMB detection between 3T and 1.5T and found that SWI images detected significantly more CMBs at 3T. The population considered in that study was patients with cerebral amyloid angiopathy, for whom the characteristic superior location of CMBs made them less likely to be obscured by susceptibility artifacts. Another comparison study between 7T and 1.5T SWI performed by Theysohn et al. (Theysohn et al., 2011) found an improved sensitivity of CMB detection at 7T for patients with vascular dementia, where all CMBs that were visible at 1.5T were also detected at 7T. This result was not surprising given that these microbleeds were relatively large and of high contrast in order to be detected at 1.5T. In our comparison between 3T and 7T, 40.4 % of CMBs that were observed on 3T SWI were not visible on 7T SWI. Our results suggest that the increase in susceptibility contrast may be offset by there being more artifacts at 7T relative to 3T, and is further supported by the observation that there was no difference in CMB detection for magnitude images between 3T and 7T.

Knowing that macroscopic susceptibility artifacts can degrade the sensitivity of detecting CMBs is important for selecting methods for imaging glioma patients who have received radiation therapy. This may also be true in hypertensive arteriopathy, where CMBs are located

primarily in deep brain (Charidimou et al., 2011). Seven-tesla SWI would be preferred over 3T SWI as long as the tumor location is away from deep brain, because the heightened contrast may be especially critical in visualizing small CMBs or following them as they evolve over time. A better understanding of the evolution of CMBs would help in the exploration of their relationship to microvasculature damage and prognostic values.

The limitations of this study are similar to those that have been reported in previous studies (Nandigam et al., 2009; Theysohn et al., 2011; Conijn et al., 2011). Since sequence timing parameters must be altered between field strengths, the selection of these parameters can also affect CMB detection. However, since we individually optimized both imaging acquisitions and post-processing steps for CMB detection at each field strength with similar scan times, the degree of bias should be minimized. Another limitation was that the angle of obliquity of the image acquisition was not necessarily identical between 3T and 7T, even though care was taken during prescription to cover the same region. Although performing a minimum intensity projection through 8 mm of tissue helps mitigate any discrepancy due to this variation in head position, we were also careful to search adjacent slices for microbleeds that were not visible on all sets of images. Overall, differences in coverage between field strengths were minimal, and our analysis was restricted to the joint FOV of both acquisitions.

5.5 Conclusions

In conclusion, our study suggests that 7T SWI may be more sensitive to radiation therapy-induced CMBs than SWI at 3T, as long as the location of CMBs is not in areas with heightened susceptibility artifacts. Tumor location should be considered in conjunction with field strength when designing protocols for detecting radiation therapy-induced CMBs in patients with glioma. The gain in CMB detection sensitivity due to SWI processing is significant for detecting

CMBs at both 3T and 7T. However, studies with larger patient sample size must be done to confirm what we concluded here.

Chapter 6 Multi-echo SWI of CMBs, veins, and arteries

In this chapter, a 4-echo gradient echo sequence at 7T is presented. The sequence is able to simultaneously perform 3D time-of-flight (TOF) MR angiography (MRA) and susceptibility-weighted imaging (SWI) of radiation-induced cerebral microbleeds (CMBs), intracranial arteries, and veins. The sequence was evaluated clinically and its imaging quality was compared to single-echo sequences for 3D TOF MRA and SWI individually. The advantages of the multi-echo sequence over the single-echo ones are discussed.

6.1 Introduction

Radiation therapy (RT) is a widely utilized treatment in the management of patients with high-grade brain tumors and in lower grade tumors that progress. RT is often used either in conjunction with chemotherapy after surgical resection to reduce residual tumor burden or alone in surgically inaccessible tumors. Despite its effectiveness and recent modern advances to constrain radiation dose distribution more effectively to the tumor, collateral injury to normal brain tissue is always present and includes coagulative necrosis, white matter demyelization, cortical atrophy, and endothelial proliferation (Valk et al., 1991). RT is also correlated with the development of vascular abnormalities including cavernous malformations (Jain et al., 2005), moyamoya-like progressive steno-occlusive disease (Ullrich et al., 2007), accelerated atherosclerosis, and other forms of large vessel arteriopathy (Omura et al., 1997). At the microvascular level, histopathological analyses reveal a spectrum of radiation injury that includes endothelial disruption, fibrinoid necrosis, luminal narrowing, and occlusion, which leads to the formation of cerebral microbleeds (CMBs) in otherwise normal-appearing brain tissue (Fazekas et al., 1999; Fajardo et al., 2005). These hemosiderin-containing deposits begin

to appear approximately two years post-RT and continue to increase in number over time (Zeng et al., 2011; Lupo et al., 2012; Tanino et al., 2013). More recent studies have demonstrated a correlation between the number of CMBs and the dose and the target volume defined for radiation therapy (Lupo et al., 2012; Tanino et al., 2013), pointing to the potential use of these lesions as a surrogate quantitative marker of radiation injury.

While the origin of CMBs has not been completely defined, ionizing radiation is known to have a greater effect on smaller caliber vasculature and is more likely to damage arteries than veins (Fajardo et al., 1999). A strategy to non-invasively image arteries and veins simultaneously, and to assess the spatial distribution of CMBs relative to these structures would help to establish a relationship between CMB formation and underlying vascular pathology, and aid in clinical and basic science studies of CMB that arise in cerebral amyloid angiopathy (CAA) (Greenberg et al., 1999), stroke (Werring et al., 2005), Alzheimer's disease (Goos et al., 2009), mild cognitive impairment (Werring et al., 2010), and dementia (Ayaz et al., 2010).

CMBs can be observed noninvasively on MR images using T2*-weighted gradient echo sequences as small hypointense lesions, often with spherical shape (Greenberg et al., 2009). These imaging features are enhanced by susceptibility-weighted imaging (SWI), an MR imaging technique that is more sensitive to CMBs than conventional T2*-weighted imaging (Akter et al., 2007; Nandigam et al., 2009; Zeng et al., 2011; Lupo et al., 2012; Tanino et al., 2013). SWI also permits accurate visualization of intracranial veins as hypointense due to the presence of iron-containing deoxyhemoglobin (Reichenbach et al., 1997; Haacke et al., 2004). In contradistinction, arterial contrast in 3D time-of-flight (TOF) MR angiography (MRA) is determined by flow-related enhancement and background suppression (Nishimura et al., 1990). 7T 3D TOF-MRA improves visualization of intracranial arteries compared to 3T, with upwards

of an 80% increase in contrast-to-noise ratio (CNR) (von Morze et al., 2007). Using GRAPPA-based acquisition and reconstruction, its resolution can be also increased within the same scan time (von Morze et al., 2008). Much of the signal-to-noise ratio (SNR) loss due to the parallel imaging acquisition was regained with the higher resolution, since the effects of partial voluming and/or intravoxel dephasing were reduced. While the utility of SWI for characterizing both CMBs (Akter et al., 2007; Nandigam et al., 2009; Zeng et al., 2011; Lupo et al., 2012; Tanino et al., 2013) and intracranial veins (Reichenbach et al., 1997; Haacke et al., 2004) and 3D TOF-MRA for visualizing intracranial arteries (Nishimura et al., 1990; Parker et al., 1991; von Morze et al., 2007; von Morze et al., 2008) has been demonstrated throughout the literature, accounting for prescan, the combined time spent on the two separate sequences can be about 15~20 minutes. Moreover, accurate registration is nearly impossible to attain due to the lack of both anatomical contrast and structural similarity between the TOF-MRA and SWI, in addition to the blurring of sub-millimeter CMBs and microvasculature after the interpolation step of the registration. Thus, a combined MRA-SWI sequence using multiple gradient echoes can not only reduce scan time but also obviate the need for image coregistration, which might benefit our understanding of CMB formation by providing metrics that reflect the interaction among vascular structures that are not confounded by inaccuracies in the coregistration.

The ability to obtain a simultaneous acquisition of 3D TOF-MRA and SWI in a single imaging sequence with multiple echoes has been recently demonstrated in a normal volunteer at 3T (Du et al., 2009). Implementation of the sequence at 7T is likely to improve image quality due to elevated SNR, improved background suppression for TOF-MRA, and heightened susceptibility contrast for SWI, resulting in improved delineation of both CMBs and microvasculature (Lupo et al., 2012; von Morze et al., 2007). While previous studies have

demonstrated the capability of implementing a dual- or multi-echo acquisition on normal volunteers (Du et al., 2008; Du et al., 2009; Deistung et al., 2009; Bae et al., 2010), there have been no efforts at simultaneously optimizing contrast of CMBs and microvasculature, creating combined SWI images from multiple echoes, reconstruction and processing improvements, or clinical evaluation. The goal of this study was to design a 7T multi-echo sequence and SWI reconstruction pipeline using data from multiple echoes that would result in: 1) comparable image quality to that of separate TOF-MRA and SWI acquisitions, and 2) high-resolution vascular images for the simultaneous depiction of arteries, veins, and CMBs in patients with brain tumors treated with prior radiation therapy.

6.2 Method and Materials

6.2.1 Sequence Design

A multi-echo sequence was created by adding 3 additional echoes to a single-echo, multi-slab 3D spoiled gradient recalled (SPGR) echo sequence with TOF capabilities on a GE 7T system (GE Healthcare, Waukesha, WI) equipped with a 32-channel phased array receive coil insert situated within a volume transmit coil (Nova Medical, Wilmington, MA). The first echo was used to create TOF-MRA images, while the remaining 3 echoes were combined to generate a composite SWI image. The sequence diagram of the final empirically optimized acquisition scheme is shown in Figure 6.1. Because adequate background suppression and heightened contrast of arteries on the TOF-MRA images requires minimizing the TE of the first echo and overall TR, flow compensation was performed only in the readout direction and all echoes were partially acquired with a 65% sampling coverage. This resulted in a TE1/TE2/TE3/TE4 of 2.7/10.5/13.2/20.9ms and a TR of 40ms when using a bandwidth of 41.67kHz, in-plane matrix of 512×384, and FOV of 24 cm. A small flip angle of 25° was used for excitation and a multiple

overlapping thin-slab acquisition (MOTSA) (24), an approach that is widely used for TOF-MRA, was applied to all 4 echoes. Three slabs with 36 1mm-thick slices and 12 slices of overlap were used to minimize signal saturation for the TOF-MRA images from the first echo while maintaining a large enough 3D volume to achieve adequate SNR for SWI image obtained from the later echoes. The acquisition was accelerated in the phase encoding direction with an autocalibrating partially parallel imaging strategy that utilized an acceleration factor of 3 and 16 auto-calibrating lines, resulting in a total acquisition time of 10.6 minutes.

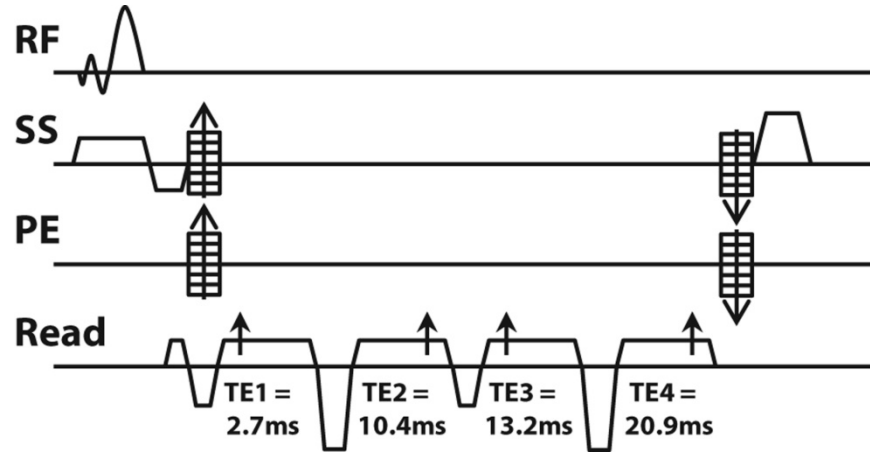


Figure 6.1 The diagram of the proposed 3D multi-echo spoiled gradient echo sequence.

The sequence contains 4 partially acquired (65%) echoes at the TEs (arrows) of 2.7, 10.4, 13.2 and 20.9ms. The data from the first echo are used to generate TOF-MRA, while the data from the remaining 3 echoes are combined to generate SWI images. Flow compensation was performed only in the readout direction using a pre-wind bipolar gradient for the first echo and flyback gradients for the other 3 echoes. (RF: rf pulse; SS: Slice selection gradient; PE: Phase encoding gradient).

6.2.2 Image Reconstruction

The raw complex k-space data from all 32 coils were transferred off-line to a Linux workstation, where post-processing was performed using in-house programs developed with MATLAB 7.0 software (MathWorks, Natick, MA). Our processing pipeline for reconstruction and combination of multi-echo, multi-channel data is illustrated in Figure 6.2. For each

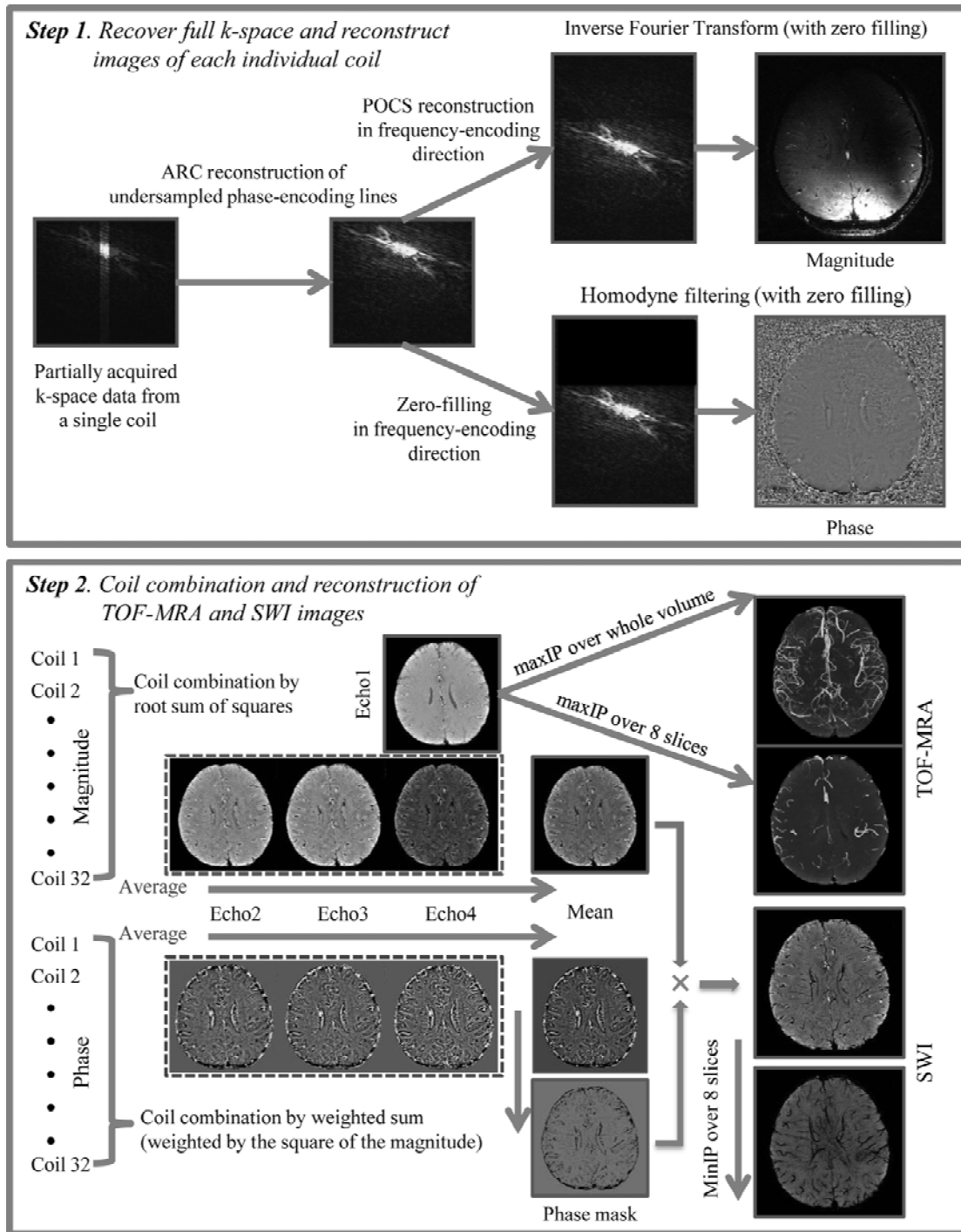


Figure 6.2 Image reconstruction and post-processing pipelines for the multi-echo sequence.

individual coil, missing phase-encoding lines were recovered using an autocalibrating parallel imaging reconstruction method: auto-calibrating reconstruction for Cartesian sampling (ARC)

(Brau et al., 2008). For reconstruction of magnitude images at each echo, all partially-acquired k-space echoes were recovered to their full extent by projection onto convex sets (POCS) (Haacke et al., 1991). The full FOV 512×384 k-space data from all echoes were then zero-padded out in the phase-encoding direction to create a 512×512 matrix before taking the inverse Fourier transform. Magnitude images from each coil were combined using the root sum of squares (Roemer et al., 1990) and skull stripped using FMRIB Software Library's (FSL) Brain Extraction Tool (BET) (Smith, 2002). The final magnitude images from the first echo are used for TOF-MRA while those from the final 3 echoes are used for subsequent SWI processing described as follows.

During SWI processing, the data from the final 3 echoes were firstly processed individually before a composite SWI image is created (Figure 6.2). To generate high-pass filtered phase images for each coil, the complex k-space data after ARC reconstruction from echoes 2-4 were zero-filled in the frequency-encoding direction, followed by homodyne filtering with Hanning filter sizes of 72, 88, and 104 for the 2nd, 3rd, and 4th echoes respectively. These filter sizes were empirically determined for each TE to preserve local contrast while removing macroscopic background phase wraps. The high-pass filtered phase images from each coil were combined by weighted sum (weighted by the square of corresponding magnitude intensity). The phase images from echoes 2~4 was then averaged to produce a mean phase image, from which a negative phase mask was created by linearly scaling negative phase values between zero and one and setting positive phase values to one. The mean magnitude image from the 3 echoes was also produced and multiplied with the phase mask 4 times to generate the final composite SWI image.

6.2.3 Clinical evaluation

The proposed sequence was evaluated on 8 patients (4 males and 4 females with a mean age of 45.2 years and a range from 29.8 to 67.1 years) who had multiple confirmed CMBs due to prior radiation therapy of a resected glioma. The time between the 7T imaging examination and start of radiation ranged from 3 to 15 years. The study was approved by our institutional Committee for Human Research and written informed consent was obtained from all patients. Conventional single-echo TOF-MRA and SWI sequences with the same acceleration, FOV, image matrix, flip angle, and coverage were scanned in addition to the combined multi-echo sequence on all patients. The other parameters utilized for each sequence are listed in Table 6.1.

Table 6.1 Acquisition parameters for single- and multi-echo sequences

	3D SWI	3D TOF-MRA	3D Multi-echo MRA/SWI
Slice thickness	2 mm	1 mm	1 mm
Slab thickness	1 slab	3 slabs	3 slabs
	36 slice	30 slices/slab	36 slices /slab
		6 overlapping slices	12 overlapping slices
Bandwidth	15.25 kHz	41.76 kHz	41.76 kHz
K-Space coverage	Full	Partial (65%)	Partial (65%)
TR	50 ms	30 ms	40 ms
TE(s)	16 ms	2.7 ms	2.7, 10.5, 13.2, 20.9 ms
Acquisition time	4 mins/50 secs	6 mins/40 secs	10 mins/36 secs

Parameters that are same to all 3 sequences: FOV 24 cm, flip Angle 25°, acquisition matrix 512×384, in-plane resolution 0.46×0.63 mm², and acceleration factor 3.

6.2.4 Data Analysis

In order to evaluate the quality of the depiction of arteries, veins, and CMBs in images generated from the combined multi-echo sequence, maximum and minimum intensity

projections (maxIP and minIP) through 8mm-thick slices were created for all TOF-MRA and SWI images, respectively. These projected images were used to both count the number of CMBs detected on SWI images and quantify the contrast-to-noise ratio (CNR) of *small*- (diameter $\leq$ 1mm) and *intermediate*- (1mm<diameter $\leq$ 2mm) sized vessels. CMBs are defined as round and hypointense foci with their diameters smaller than 5 mm. The detailed description of the definition of CMBs can be found in the section 2 of chapter 5. CMBs were firstly identified on minIP SWI using an automated detection algorithm, which will be described in chapter 7. The algorithm is highly sensitive to radiation-induced CMBs on minIP SWI images was employed to identify CMBs in approximately 1 minute. The algorithm uses geometric features such as shape, area, and circularity to distinguish potential CMBs from other sources of hypointense signal, including linear vessels and susceptibility artifacts. Two raters independently inspected the resulting masks in random order to remove any remaining false positives, include any CMBs that were missed by the automated detection, and count the identified CMBs. The mean phase images were also examined to verify that all identified CMBs were not due to the presence of calcification, which would appear hyperintense on phase images since calcium is diamagnetic (Lupo et al., 2012). The inter-rater agreement of the CMB counting was measured by calculating an intraclass correlation coefficient (ICC) based on absolute agreement. The final CMB counts that were obtained from each rater were averaged to quantify the number of true CMBs for each patient. CNR was calculated from 10 regions of interest (ROIs) for each size vessel on both the TOF-MRA and SWI images. Each vessel segment selected was carefully matched in size and length between the single and multi-echo sequences to minimize any effects of motion or partial voluming between scans and spaced to span the entire supratentorial brain coverage to minimize any bias from spatial variations in contrast due to the parallel

reconstruction. This resulted in 40 total vessel segments for each patient. ROIs of background brain parenchyma were generated by first dilating each vessel ROI (with a kernel size of .5 mm and 1 mm for small and intermediate vessels, respectively) and then subtracting the vessel ROI from the dilated ROI. Examples of vessel and background ROIs are illustrated in Figure 6.3.

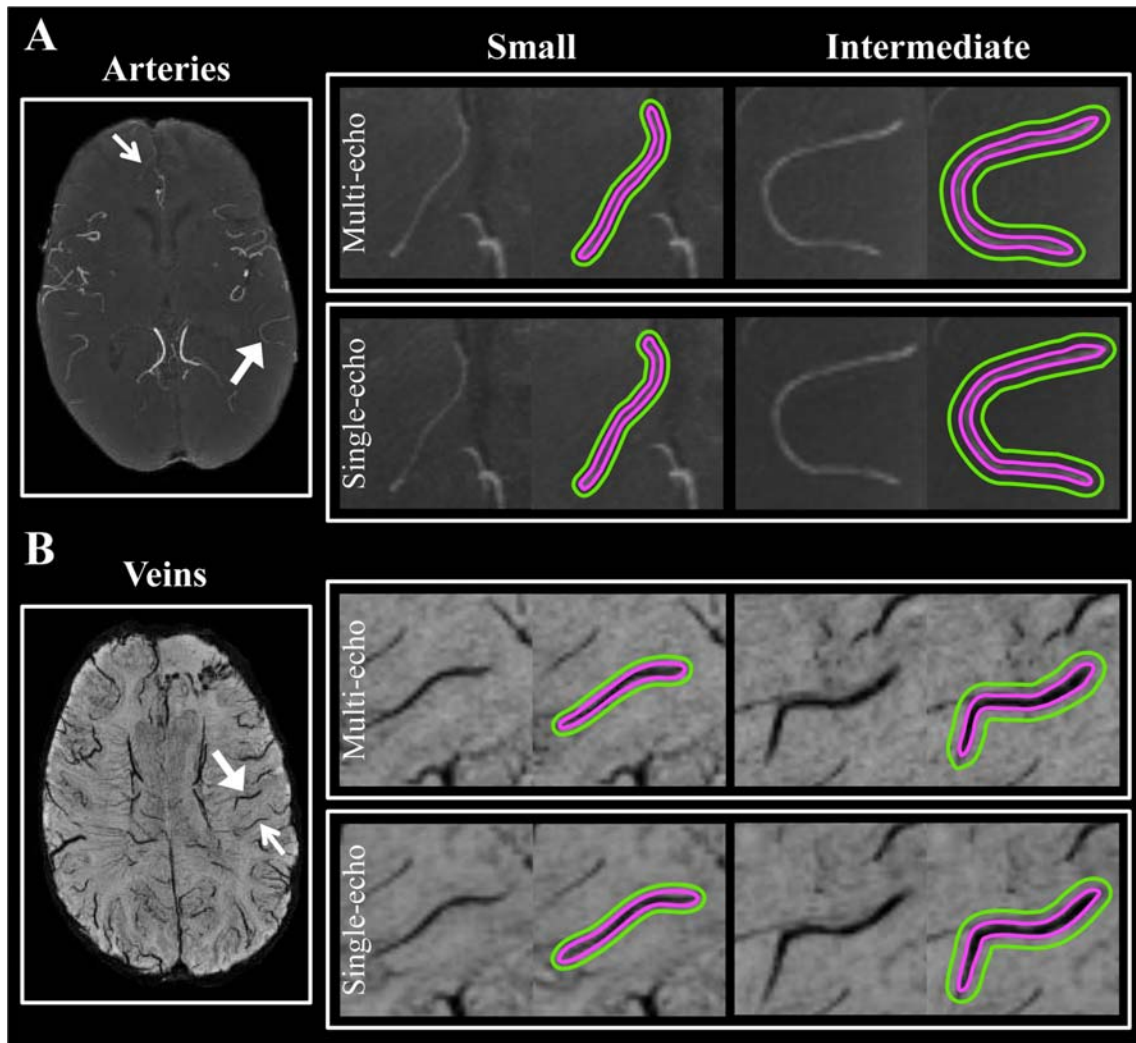


Figure 6.3 ROIs of veins and arteries were defined on (A) TOF-MRA and (B) SWI images.

The TOF-MRA and SWI images were maximally and minimally projected over 8mm thickness, respectively. ROIs were paired between the multi-echo images (top rows in A & B) and corresponding single-echo images (bottom rows in A & B). Each vessel ROI (pink) was dilated (green) to create a corresponding background tissue ROI after subtracting out the original vessel ROI. The ROIs were defined for intermediate diameter (between 1 and 2mm, large arrows) and small diameter (less than 1mm, small arrows) arteries and veins.

Noise was estimated from the standard deviation of a homogeneous region of corpus callosum for both the TOF-MRA and SWI according to Denk et al (Denk et al., 2010). CNR was calculated as the difference in the median values between the vessel and background ROIs divided by the noise. The 10 CNR values from each vessel type were then averaged for each patient. A Wilcoxon signed-rank test was used to test for significant differences in the averaged CNR and the number of CMBs between image acquisitions.

6.3 Results

The benefit of combining data from echoes 2-4 in the creation of a composite SWI image compared to using data from a single echo time is illustrated in Figure 6.4A. SNR is improved for the combined composite SWI image compared to the SWI image generated from echo 3 (whose TE was similar to that of single-echo SWI sequence), as evident by a more homogeneous appearance of the ventricles on the composite image due to the reduction of noise. Plots of average CNR for both small and intermediate veins on minIP SWI images from echo 3 and all echoes combined for all 8 patients are shown in Figure 6.4(B). As expected, combining the 2nd-4th echoes of the multi-echo sequence resulted in an average CNR that was 52.2% and 45.0% higher than those measured on the minIP SWI images from the third echo only for *small* and *intermediate* veins, respectively (both $p < 0.008$, Wilcoxon signed rank test).

The overall image quality was comparable between the single and multi-echo sequences, with a slight degradation of background suppression on the TOF-MRA and higher noise level on the SWI images generated from the multi-echo sequence. Locally, both *small* and *intermediate* veins were similarly depicted on the single- and multi-echo sequences. In addition, single-echo SWI could better delineate large draining veins because of their higher CNR, while the multi-echo SWI was better at detecting smaller veins and CMBs, primarily due to its higher slice

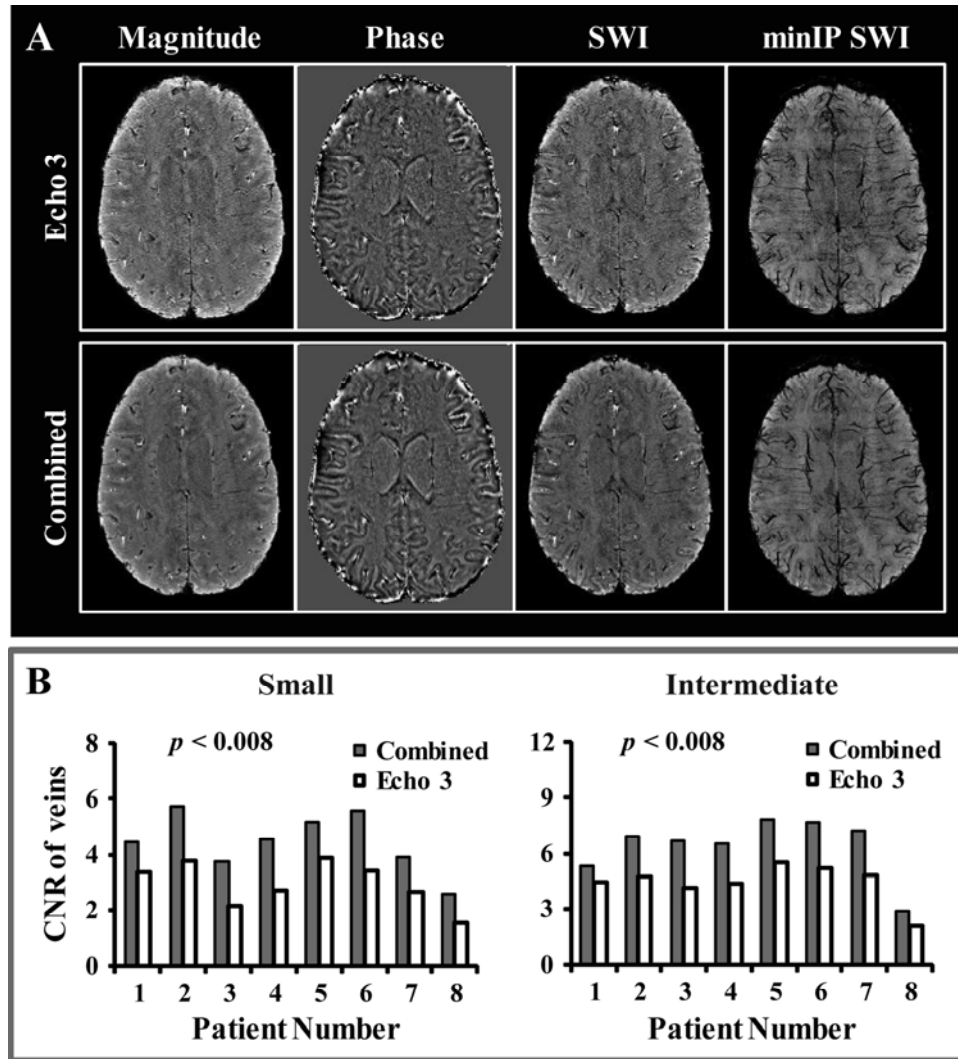


Figure 6.4 Comparison of quality and CNR of images from echo 3 and combined processing.

(A) Magnitude, phase, SWI, and minIP images from echo 3 (top row) and combined processing (bottom row), where both magnitude and phase images from echoes 2 to 4 were first created individually and then averaged to generate mean magnitude and phase images before generating the final SWI images. The SNR was greatly improved for the combined composite SWI image compared to that from the echo 3 (whose TE was similar to that of single-echo SWI sequence), which is evident by a more homogeneous display of the ventricles on the former. (B) Plots of average CNR for both small and intermediate veins from minIP SWI images of echo 3 and the composite SWI image for all 8 patients. The composite SWI image had significantly higher CNR for both vein sizes in all patients.

resolution. A typical example of this is shown in Figure 6.5A. On the other hand, all arteries had similar contrast on the multi-echo TOF-MRA compared to the single-echo one, despite the

slightly worse background suppression of the multi-echo acquisition, due to the longer TR (Figure 6.5B).

Figure 6.5C shows plots of average CNR for each type of vessel calculated from the multi- and single-echo sequences from all 8 patients. On average, CNRs of *small* and *intermediate* veins were not significantly different between the single- and multi-echo SWI images (3% difference with $p = 0.95$ for *small* veins and 6.7% difference with $p = 0.46$ for MRA increased 7.6% and 9.5%, respectively, for the multi-echo sequence compared to values obtained from the single-echo sequence. Although this difference was considered significant for *small* arteries ($p < 0.03$), it was not for *intermediate* ones ($p > 0.05$).

Figure 6.6A displays an overlay of a thresholded slice of TOF-MRA (maxIP over 8mm) on the minIP SWI image from the multi-echo sequence. From this overlay, it is apparent that some CMBs are clearly arising from venous vessels (as denoted by the pink dashed-box) while others appear to originate from arterioles (green dashed-box). No registration is necessary to visualize the vascular origin of newly-forming CMBs and accurately quantify parameters that describe their relationship to surrounding microvascular networks. Table 6.2 lists the number of CMBs detected from both sequences for each patient. Both raters consistently detected more CMBs on the multi-echo minIP SWI images than on the single-echo minIP images ($p < 0.008$ for the rater 1 and $p < 0.024$ for the rater 2). The ICCs between the two raters on CMB counting from all 8 patients were 0.995 for both the single- and multi-echo images. Since the ICC calculation is heavily influenced by the number of CMBs and can be artificially elevated when the variability in CMB counts among patients is large, we repeated the calculation excluding the two patients who had more than 200 CMBS. This more reliable estimation of inter-rater agreement resulted in ICCs of 0.920 and 0.872 for the single- and multi-echo images,

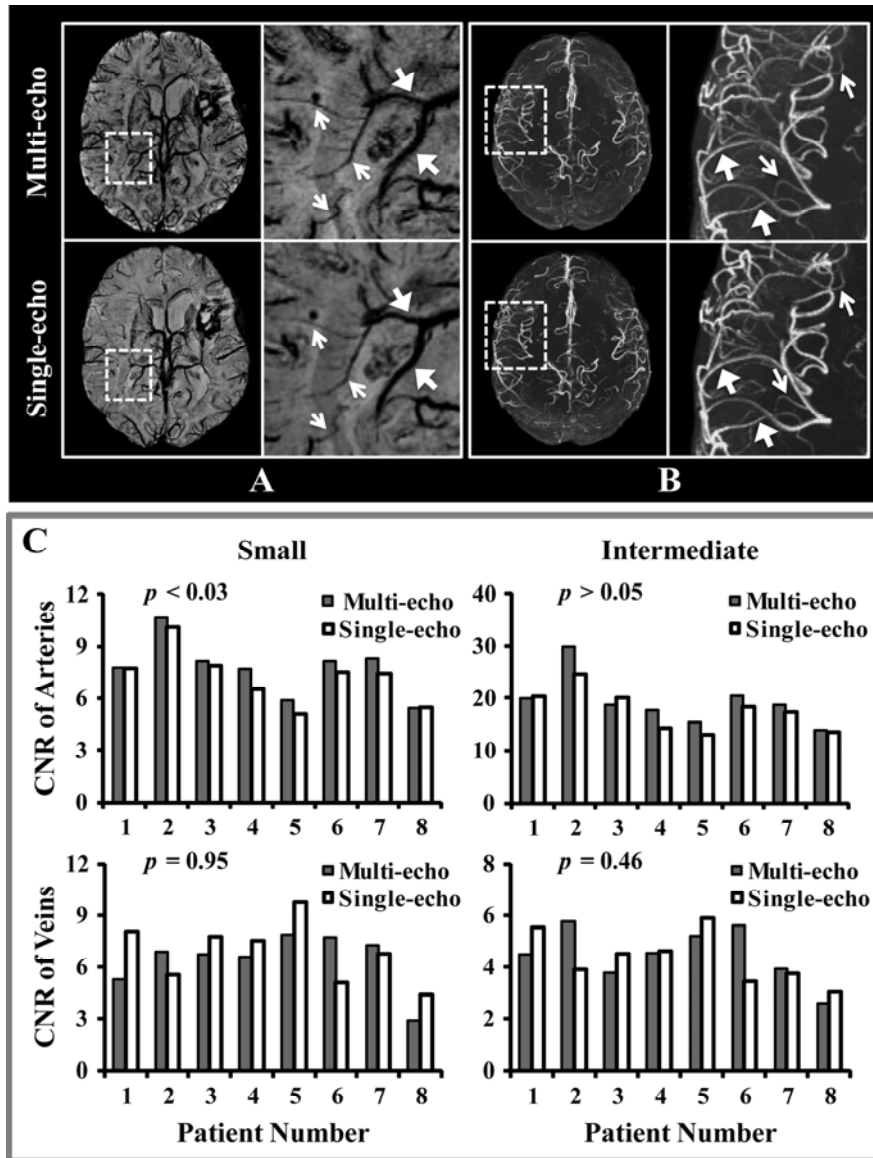


Figure 6.5 Comparison of quality and CNR of images from multi- and single-echo sequences.

(A) minIP SWI and (B) maxIP TOF-MRA for both multi-echo and individual single-echo sequences. For a better illustration purpose, a local region (dashed-box) from each is zoomed in and shown on the left column. On (A), single-echo SWI better delineates larger veins (large arrows) because of its high CNR, while multi-echo SWI better illustrates smaller veins and CMs primarily due to its higher slice resolution. On (B), all arteries including both larger (large arrows) and smaller (small arrows) arteries have similar contrast on both multi- and single-echo TOF-MRA, despite the slightly worse background suppression of the multi-echo acquisition due to a longer TR. (C) Plots of average CNR for each type of vessel from all 8 patients' images acquired from both multi- and single-echo sequences. Except for small arteries, which had a slightly significant higher CNR on TOF-MRA from the multi-echo sequence, all other vessels had comparable CNRs between images from the multi- and single-echo sequences.

respectively. When the CMB counts from both raters were averaged, the multi-echo sequence detected 18.3% more CMBs than the single echo minIP SWI image ($p < 0.008$), with 945.5 and 798.5 total CMBs identified, respectively. Each individual patient also had more CMBs identified on the multi-echo images. These additional CMBs were typically smaller in size as shown in Figure 6.6 (B).

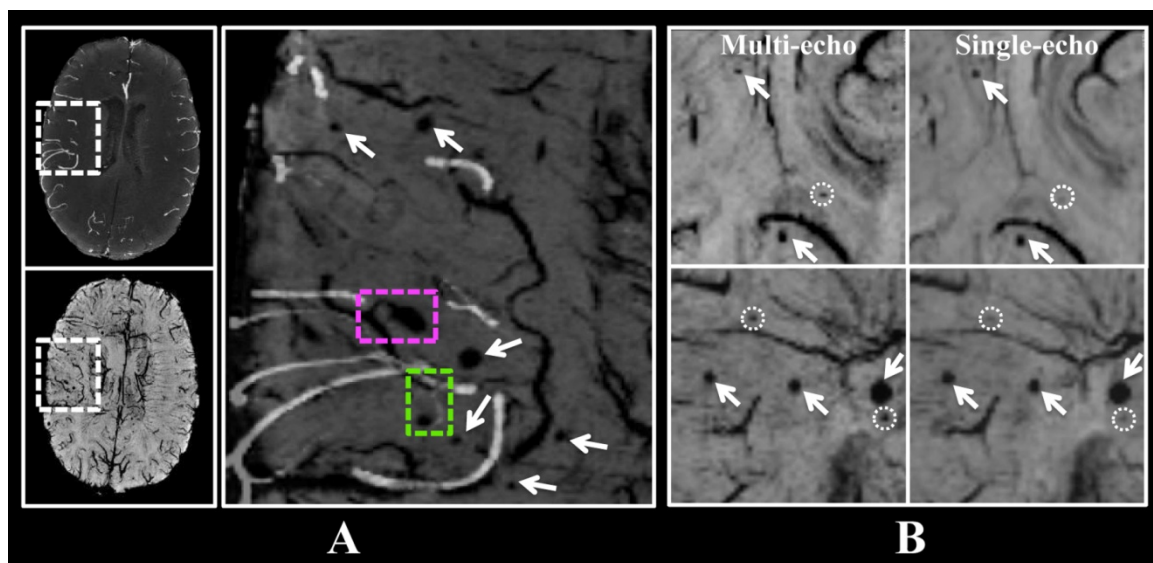


Figure 6.6. Benefits from using the multi-echo sequence.

(A) The multi-echo sequence allows arteries from TOF-MRA images (maxIP over 8 mm) to be overlaid on CMBs and veins from minIP SWI images without the need for registration. From the overlaid image (zoomed in to a local region specified by the dashed-box on the original images) on the left column, it can be clearly seen that one CMB is arising from venous vessels (pink dashed-box) while another one appears to originate from arterioles (green dashed-box). (B) minIP SWI Images from two different patients (top and bottom rows) showing CMBs (arrows) that are visualized on both single- and multi-echo images, and three additional CMBs that are only detected on multi-echo SWI (dashed-circles).

Table 6.2 CMB Number on minIP SWI images from single- and multi-echo sequences

Patients	Single-echo			Multi-echo		
	Rater1	Rater2	Average	Rater1	Rater2	Average
1	330	317	323.5	372	364	368
2	36	26	31	44	31	37.5
3	28	11	19.5	42	29	35.5
4	35	28	31.5	53	48	50.5
5	22	10	16	30	12	21
6	83	72	77.5	99	78	88.5
7	228	224	226	271	269	270
8	70	77	73.5	75	74	74.5
Total	832	765	798.5	986	902	945.5

6.4 Discussion

The heightened susceptibility contrast recently available with higher field strength scanners has greatly motivated the investigation of the clinical relevance of CMBs in neurological disorders such as cerebral amyloid angiopathy (CAA) (Greenberg et al., 1999), stroke (Werring et al., 2005), Alzheimer’s disease (Goos et al., 2009), and radiation injury (Zeng et al., 2011; Lupo et al., 2012; Tanino et al., 2013). The majority of prior MR imaging studies of CMBs have focused primarily on characterizing CMBs properties such as number, size, and location, how the selection of imaging parameters affects these properties, and their relationship to neurocognitive decline (Akter et al., 2007; Nandigam et al., 2009; Conijn et al., 2010; Theysohn et al., 2011; Zeng et al., 2011; Lupo et al., 2012; de Bresser et al., 2012; Tanino et al., 2013). Despite its implication in radiation injury and CMB formation, the spatial relationship between CMBs and surrounding microvasculature has received only limited attention. The ability to simultaneously visualize CMBs, arteries, and veins in direct spatial correspondence on one image will facilitate the quantification of metrics that reflect the interaction among these

structures, greatly aiding in the characterization of the mechanism of radiation-induced vascular injury as well as understanding the role of CMB presence in other diseases.

Motivated by these desires, we implemented a multi-echo, multi-slab gradient echo sequence with parallel imaging capability at 7T in this study. The sequence was able to acquire 3D TOF-MRA and SWI simultaneously and provided comparable image quality to each individual single-echo sequence for the depiction of intracranial arteries, veins, and radiation-induced CMBs without the need for image registration. The additional flexibility in SWI processing with multiple echoes also allowed for thinner slices, which not only improved CMB detection even further, but also enhanced the contrast of smaller veins. With the same coverage as the both single-echo sequences and twice the slice resolution of the single-echo SWI, the multi-echo sequence had an acquisition time that was less than the total acquisition time of two individual single-echo acquisitions and was comparable to that of previous studies (Du et al., 2008; Du et al., 2009; Deistung et al., 2009; Bae et al., 2010). If the multi-echo sequence were implemented at 3T and used to acquired images with the same coverage and resolution, the total acquisition time would become longer, as the TEs of all echoes except the first one has to be made longer to achieve sufficient susceptibility weighting for images from these echoes, which in turn increases the minimum TR can be used. This is another benefit in addition to CNR increase one could get by implementing the multi-echo sequence at 7T rather than 3T.

Successful implementation of a multi-echo sequence for 3D TOF-MRA and SWI requires the careful selection of imaging parameters to achieve adequate background suppression on TOF-MRA and reasonable SNR preservation on SWI. The required longer TR and thicker slabs for the SWI results in reduced background suppression and lower CNR for the TOF-MRA images, while the multi-slab, partial-echo acquisition with thinner slices necessary for TOF-

MRA degrades the quality of SWI images. Since our aim was to image small intracranial vessels and CMBs, our first constraint was to maintain a minimum slice and slab thickness of 1mm and 36mm respectively, which created a challenge in further optimization of other sequence parameters. Compared to previous studies that utilized comparable in-plane resolution (Du et al., 2008; Du et al., 2009; Deistung et al., 2009; Bae et al., 2010), our slice and slab thickness were at least 17% and 30% thinner, respectively. To achieve this desired resolution, a composite SWI image was created from three echoes in order to improve the degraded CNR of SWI images due to the thin slice/slab thickness. Although the TR of the TOF-MRA mainly limited the number of additional echoes that could be incorporated, the addition of echoes at TEs over 22ms would not have provided any further benefit to the composite SWI image because of the concomitant susceptibility distortions that occur at long TEs at 7T. Despite these tradeoffs, the combination of parameters chosen and modified SWI processing of the later echoes preserved the quality of both TOF-MRA and SWI images with good contrast.

Both *small* and *intermediate* arteries had a higher CNR on TOF-MRA from the multi-echo sequence. This can be explained by the thicker slab thickness and longer TR of the multi-echo sequence that increase the signal from both arteries and background tissue. Although the latter leads to less efficient background suppression, the longer TR enhances the in-flow effect of slowly flowing blood spins in small arteries, which further increases the signal from arteries. At the same time, since all other TOF-MRA acquisition parameters (i.e. resolution, bandwidth, TE, and sampling coverage) were kept constant between the multi- and single-echo sequences, their noise level should be similar. Therefore, it is not surprising that a longer acquisition time for the multi-echo TOF-MRA resulted in a slightly higher CNR.

To explain the observed difference in CNR for veins, the image reconstruction/post-processing strategy must also be taken into consideration in addition to the acquisition parameters of the two sequences. Compared to the single-echo SWI sequence, all individual echoes of the multi-echo sequence had half the slice thickness, 2.7 times higher bandwidth, 6.3 times shorter TR, and 35% less sampling coverage, resulting in the reduced CNR and SNR as shown on images from echo 3 in Figure 6.4A. However, averaging magnitude and phase images from echoes 2~4, greatly improved CNR of veins in the composite minIP SWI image, by 45.0% for *intermediate* veins and 52.2% for *small* veins compared to minIP SWI images generated from the 3rd echo only. This result was similar to findings of previous studies that generated SWI images from data acquired at multiple echoes (Brainovich et al., 2009; Denk et al., 2010; Gilbert et al., 2012; Feng et al., 2013). Because of this processing pipeline, the CNR of both *intermediate* and *small* veins from the composite multi-echo SWI images were not significantly different from values obtained from the conventional single-echo images. The trend towards improved CNR observed in extremely small vessels with the multi-echo sequence was likely due to the thinner slice thickness of the multi-echo sequence, which reduced partial voluming effects for these vessels (Figure 6.5A). Likewise, the thinner slices also allowed for improved detection of small CMBs, as reflected by the 18.3% more CMBs identified using the multi-echo sequence (Figure 6.6B). The reliability of this result was confirmed by the excellent inter-rater agreement for CMB counting as indicated by the high ICC metrics. This result was not surprising due to the initial automated detection step that was employed and stresses the importance of automated detection algorithms for obtaining reliable, consistent CMB counts, especially when there is a large number of CMBs present. The ability to detect these smaller, lower contrast CMBs is critical for accurately characterizing their initial onset and evolution over time, a distinct

advantage of using a multi- rather than dual-echo sequence (Du et al., 2008; Deistung et al., 2009; Bae et al., 2010) for this application. In addition, the multi-echo design can achieve heavier susceptibility weighting by incorporating information from images acquired at long TEs, which may not be used in a dual-echo design due to the afraid of image contamination from susceptibility artifacts arising when echo time is too long (Du et al., 2009). Also our interest in implementing a multi- instead of dual-echo sequence lies on recent reports (Feng et al., 2013) that the quality of phase unwrapping and multi-channel phase combination, which are critical steps in processing SWI, may be improved by utilizing information from multiple echoes.

Despite its many advantages, there are several limitations in the acquisition and analysis of our current implementation. To keep the TE of the first echo and total TR as short as possible, flow compensation was only performed in the readout direction in our multi-echo sequence. When flow is not compensated in the phase-encoding directions, signal dropout and displacement of vessels often result, especially for large arteries with fast flow (Deistung et al., 2009). However, we did not observe these artifacts and were more interested in the appearance of smaller vessels that are most sensitive to ionizing radiation and exhibit slow flow. This results in, if any, only minor signal dropout and displacement. In addition, the higher bandwidth of the multi-echo sequence further minimized these artifacts. To minimize the saturation effect of a large slab thickness, extended slab overlap was used in our sequence, which increased total scan time. However, it provided an additional advantage of mitigating some residual wrapping of the scalp in the superior slab. Our data analysis was limited by our definition of CNR, as more traditional methods to measure CNR were no longer valid after implementation of parallel imaging, phase mask filtering, minimum or maximum intensity projection, and averaging. However, our calculation of CNR, where each ROI was defined on the same region for both

multi- and single-echo images, followed the definition utilized by several prior studies that performed similar analyses of CNR on SWI images (Denk et al., 2010; Feng et al., 2013). In addition, to account for spatial variations due to parallel imaging, several sets of vessels from each patient were uniformly selected throughout various brain locations; median instead of mean values were calculated to account for possible intensity inhomogeneity within vessel ROIs. All of above considerations should make our approach to measure CNR a feasible one in practice. Finally, although the current resolution of the proposed multi-echo sequence may still limit the detection of all microvessels and capillaries that directly lead to the formation of an observed CMB, identifying the effects of RT on slightly larger vascular beds surrounding CMBs is still helpful in understanding the mechanism of vascular injury.

6.5 Conclusions

In conclusion, a 3D gradient echo sequence with 4 echoes that is able to perform TOF-MRA and SWI simultaneously was successfully implemented at 7T. The resulting image quality obtained from the multi-echo sequence was comparable to the relevant single-echo sequences for the depiction of both arteries and veins and improved CMB detection. The merging of these objects on a registration-free image will ultimately facilitate the exploring of the mechanisms of radiation-induced vascular injury and the similar injuries and associated CMBs found in other neurologic disorders such as CAA, stroke, TBI, and Alzheimer's disease.

Chapter 7 Automated Detection of Cerebral Microbleeds

In the previous two chapters, we have shown the superiority of SWI in visualizing cerebral microbleeds (CMBs). Visual inspection of CMBs on SWI images, however, is a lengthy, arduous task that is highly prone to human error because of their small size and wide distribution throughout the brain. To explore the clinical relevance of CMBs, a fast and accurate method to detect them is required. In this chapter, an automated algorithm for identifying cerebral microbleeds (CMBs) on minimum intensity projected susceptibility-weighted MR images is introduced. Its performance on patients with brain tumors who had developed CMBs after having radiation therapy is presented and compared to those from previously published methods. Finally, its advantages and limitations are discussed.

7.1 Introduction

Cerebral microbleeds (CMBs) are small, frequently perivascular collections of brain parenchymal hemosiderins induced by prior hemorrhage. On MR T2*-weighted gradient echo (GRE) magnitude images, CMBs appear as small, rounded, hypointense lesions of variable size due to susceptibility-related signal loss within iron-containing hemosiderins that accumulate paramagnetic ferric atoms (Charidimou, 2011; Cordonnier et al., 2007; Greenberg et al., 2009). Since the susceptibility effect scales linearly with magnetic field strength (assuming a fixed TE time), the contrast of CMBs is greatly enhanced by higher field strengths (e.g., at 3T or 7T) and susceptibility-weighted imaging (SWI) (Ayaz et al., 2010; Conijn et al., 2011; Nandigam et al., 2009). Because this heightened contrast has facilitated the detection of CMBs, there is a growing interest in exploring their diagnostic and prognostic values in diseases such as cerebral amyloid angiopathy (CAA) (Greenberg et al., 1999), stroke (Cordonnier et al., 2007; Fiehler, 2006;

Werring et al., 2005) , neurodegenerative disorders (Cordonnier and van der Flier, 2011), traumatic brain injury (TBI) (Scheid et al., 2003), and radiation therapy-induced injury in patients with gliomas (the most common brain tumors) (Lupo et al., 2012). Although their putative role in neurocognitive function and implications for clinical management are still being evaluated (Charidimou, 2011; Cordonnier et al., 2007; Greenberg et al., 2009), there is accumulating evidence that CMBs reflect the severity of microvascular damage in brain due to microangiopathy (Vernooij et al., 2008), TBI (Scheid et al., 2003), or radiation therapy (Lupo et al., 2012).

Visual inspection of CMBs on MR images is especially difficult due to their small size (with radii often < 2 mm for radiation-induced CMBs) and wide distribution throughout the brain. In CAA and following cranial radiation, the sheer large number of CMBs can render manual lesion counting impractical or impossible. Detection is further confounded by the presence of normal anatomical structures with heightened magnetic susceptibility that mimic the appearance of CMBs on T2*-sensitive sequences, such as deoxyhemoglobin-containing intracranial veins. These characteristics make the identification of CMBs a lengthy and arduous task that is prone to human error and substantial intra-rater and inter-rater variability (Cordonnier et al., 2009; Gregoire et al., 2009). An automatic, computer-aided CMB detection method that can both minimize the burden of visual inspection and improve the accuracy of detection of CMBs is therefore highly desirable.

Several methods have been proposed for CMB detection (Barnes et al., 2011; Kuijf et al., 2012; Seghier et al., 2011). Seghier et al. (2011) implemented an intensity-based statistical classification algorithm in which T2*-weighted magnitude images are first registered to a standard template that maps voxelwise probabilities of individual brain structures such as gray

matter, white matter, cerebral spinal fluid, and CMBs. Gaussian mixture modeling is then applied to distinguish CMBs from other brain structures. They identified patients with CMBs at a sensitivity of 77% and a detection rate of 50% for total true CMBs without giving the number of false positives. Barnes et al. (2011) proposed a technique for intensity-based local statistical thresholding that assumes the Gaussian distribution of background tissue in a small region and then identifies hypointense CMBs as outliers. False positives were reduced by constructing a support vector machine that incorporates shape, size and intensity as features for each hypointense region to distinguish CMBs from mimics. They achieved a detection sensitivity of 81.7% with 107.5 false positives per patient. Kuijf et al. (2012) developed an algorithm for CMB detection based on the fast radial symmetry transform (FRST), which enhances local objects with spherical or near-spherical geometry (Loy and Zelinsky, 2003). By using the transform, the algorithm achieved a detection rate of 71.2% with 17.2 false positives per patient.

Despite the reported success of these computer-aided methods for detecting CMBs, there remains a need to improve diagnostic accuracy with simpler processing, less computation time, and greater robustness in the presence of anatomic distortion such as brain tumors, resections, and infarcts. In addition, all of the above methods are designed to detect CMBs on T2*-weighted magnitude or SWI images without minimum intensity projection (MIP) processing, which helps distinguish CMBs from hypointense veins and is used in clinical practice for visual inspection of CMBs (Lupo et al., 2012), even by the groups who have developed these automated methods (Ayaz et al., 2010; de Bresser et al., 2012). Direct adaptation of their methods for CMB detection on images may be nontrivial as the original geometric coordinates and characteristics of CMB vary after the process, rendering some of the prerequisites associated with these features invalid for these methods.

In this study, we propose a new approach for CMB detection with higher sensitivity and faster computation than has been previously reported, even in the presence of anatomic disease. This approach aims to detect CMBs on SWI images, and the detection process is divided into two main steps: 1) initial putative CMB detection using the 2D FRST, 2) subsequent false positive reduction by characterizing geometric features of putative CMBs through region growing. Although the FRST has already been used to detect CMBs on T2*-weighted magnitude images (Kuijf et al., 2012), the transform was performed in 3D, which requires isotropic image acquisition, and even a perfectly spherical paramagnetic object under isotropic acquisition becomes elongated along the direction of the main magnetic field on T2*-weighted images because the contour of the external field perturbation caused by the object is not spherical (Schenck, 1996). Also, the transform has been modified and utilized in new ways in our implementation. To illustrate the effectiveness of the proposed approach, we applied the method to a series of patients with CMBs induced by radiation treatment for resected gliomas.

7.2 Methods and Materials

7.2.1 CMB Detection Algorithm

The proposed algorithm can be divided into two primary steps: 1) identification of putative CMBs using 2D FRST; and 2) false positive reduction of putative CMBs identified in the first step using 3D region growing followed by geometric feature examination. A flowchart depicting the steps for this algorithm is given in Figure 7.1. Details of the implementation will be described in the following sections.

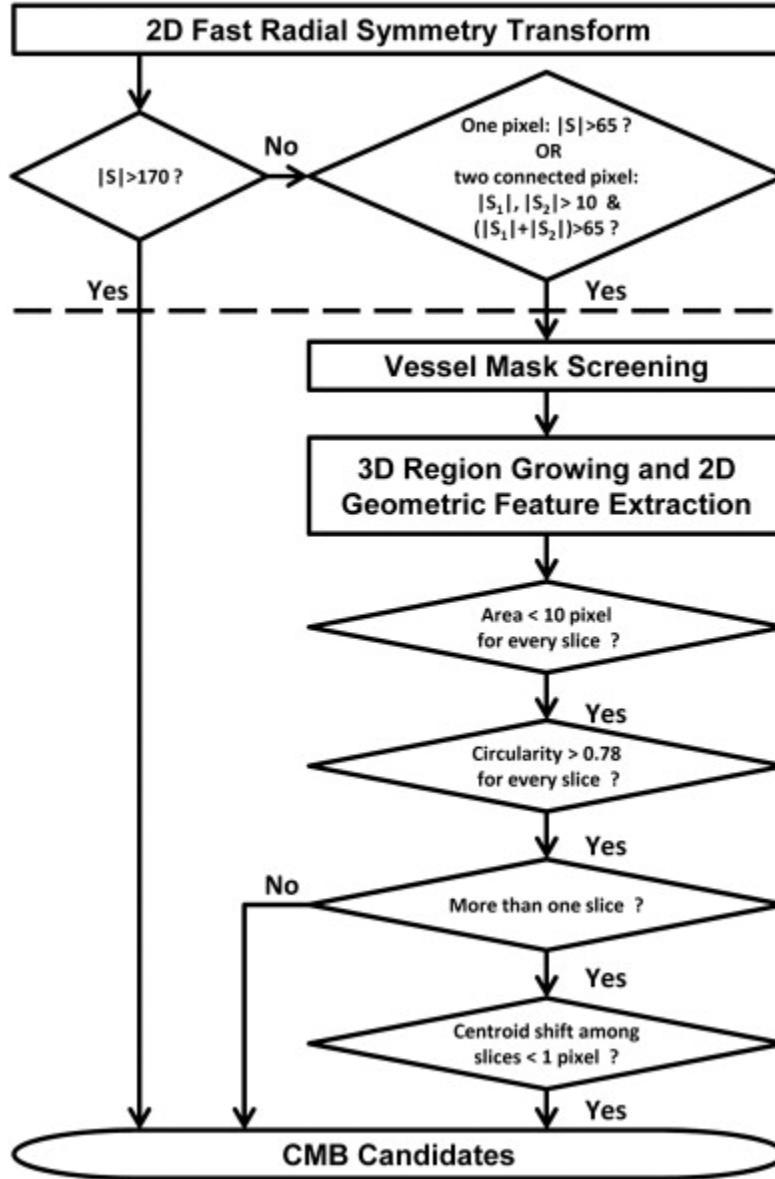


Figure 7.1 Schematic diagram for the proposed CMB detection algorithm

The processing above the dashed line belongs to the step of initial putative CMB detection, while the below belongs to the step of false positive reduction. The values of optimized parameters are given, where S refers to the intensity of FRST map.

7.2.1.1 Detection of Putative CMBs Using 2D FRST

Step1. FRST

The inherently circular morphology of CMBs on SWI images makes lesion geometry an ideal feature for automated detection. For this purpose, a modified version of the FRST that was developed by Loy et al. (2003) was adopted in our algorithm. Our goal in this initial step was to select a set of parameters that would identify the greatest possible number of true microbleeds, regardless of the number of false positives.

FRST is a gradient-based transform that begins with a computation of the gradient of each pixel using the 3×3 Sobel operator. If a pixel $\mathbf{p}$ lies on the edge of a circular disk, then the direction of its gradient $\mathbf{g}(\mathbf{p})$ is orthogonal to the edge, pointing to (if the circular disk is hyperintense) or away from (if the disk is hypointense) the center of the circle. The pixel that is at a distance n pixel away from $\mathbf{p}$ along the direction of $\mathbf{g}(\mathbf{p})$ is defined as a *positively-affected pixel*, whereas the pixel that is at a distance n pixel away from $\mathbf{p}$ along the direction opposite to that of $\mathbf{g}(\mathbf{p})$ is defined as a *negatively-affected pixel*. Since CMBs are hypointense objects on MR images, we only need to consider negatively-affected pixels, whose coordinates are given by

$$p_{-ve}(\mathbf{p}) = p - \text{round}\left(\frac{\mathbf{g}(\mathbf{p})}{g(\mathbf{p})}\right)n \quad (7.1)$$

where “round” rounds each vector element to the nearest integer and n is the radius of the circular features to be detected. Since there is no prior knowledge about the radius of the circular object to be detected, the transform is performed at a set of radii $n \in N$ $n \in N$. At each radius, an orientation projection image O_n and a magnitude projection image M_n are generated as the followings:

$$O_n(\mathbf{p}_{-ve}(\mathbf{p})) = O_{n_prev}(\mathbf{p}_{-ve}(\mathbf{p})) - 1 \quad (7.2)$$

$$M_n(\mathbf{p}_{-ve}(\mathbf{p})) = M_{n_prev}(\mathbf{p}_{-ve}(\mathbf{p})) - \|g(\mathbf{p})\| \quad (7.3)$$

The initial value of each pixel in O_n and M_n is set to zero. For each negatively affected pixel, the value of that pixel $\mathbf{p}_{-ve}$ in the O_n and M_n is decreased by 1 and $\|g(\mathbf{p})\|$ from O_{n_prev} (previous O_n) and M_{n_prev} (previous M_n), respectively. All edge pixels on the boundary of a hypointense circle with a radius of n will have their negatively-affected pixels located at the center of the circle, highlighting the central pixel in O_n and M_n . To prevent corruption by noise, gradient magnitudes smaller than the 95th percentile (background pixels on our SWI images have gradient of zero) were ignored in our experiments when computing O_n and M_n . This threshold was found to adequately suppress background noise while maintaining reasonable sensitivity to CMBs with low contrast.

Once the O_n and M_n are calculated, the radial symmetry contribution at radius n can be defined as:

$$F_n(\mathbf{p}) = \frac{M_n(\mathbf{p})}{k_n} \left(\frac{|\tilde{O}_n(\mathbf{p})|}{k_n} \right)^\alpha \quad (7.4)$$

where

$$\tilde{O}_n(\mathbf{p}) = \begin{cases} O_n(\mathbf{p}) & \text{if } O_n(\mathbf{p}) < k_n \\ k_n & \text{otherwise} \end{cases} \quad (7.5)$$

k_n is a scaling factor used to normalize O_n and M_n across different radii such that the symmetry map of objects with different size can be represented on a similar scale. The α parameter is used to characterize radial strictness, with higher values enhancing features with radial symmetry (i.e.

dots) and attenuating features without radial symmetry (i.e. lines). The full transform map is computed by summing the symmetry contribution over all the radii: $S = \sum_{n \in N} F_n$

In the original transform proposed by Loy et al. (2003), F_n has to be convolved with a Gaussian kernel in order to spread the influence of the p-ve as a function of the radius n . Because we are only interested in knowing the center location of each potential CMB during the initial detection step, this convolution is skipped in our algorithm. In addition, instead of initializing $On_initial$ with zero, a small negative value was used as the initial value for each pixel in $On_initial$. This modification increases the weight given to small radii over the other radii and thus improves the sensitivity of the transform to small circular objects (Riccardi, 2006). An example of a slice of SWI image and its 2D FRST map are shown in Figure 7.2a&b, respectively.

Step 2. Intensity screening of FRST map

Pixels that have transform value S with absolute value larger than an empirical threshold t_1 on the FRST map are directly identified as CMB candidates without 3D region growing. This upper bound defines hypointense regions with relatively large radius and high contrast, consisting primarily of true CMBs and very few false positives. To recover CMBs with smaller radii and lower contrast, pixels with $|S|$ falling between t_1 and a lower threshold t_2 (also determined empirically) are also considered as candidate CMBs. While a smaller absolute value for the lower threshold t_2 boosts sensitivity to true CMBs that are intended to be recovered, this choice for the parameter also results in a larger number of false positive lesions. The majority of these false positive CMBs are subsequently removed by performing the steps illustrated in Figure 7.1. To overcome the uncertainty introduced by partial volume averaging of sub-millimeter CMBs, we also consider regions with $|S| < t_2$ but impose two additional constraints: 1)

the region must contain two connected pixels with both of their absolute values of S larger than t_3 (with $t_3 < t_2$); and 2) the absolute value of the sum of the two connected pixels is larger than t_2 . This step prevents the elimination of true CMBs with less circular morphology where the peak of the FRST is blurred into neighboring pixels. The rationale for how these threshold values were determined can be found in the Parameters Selection section.

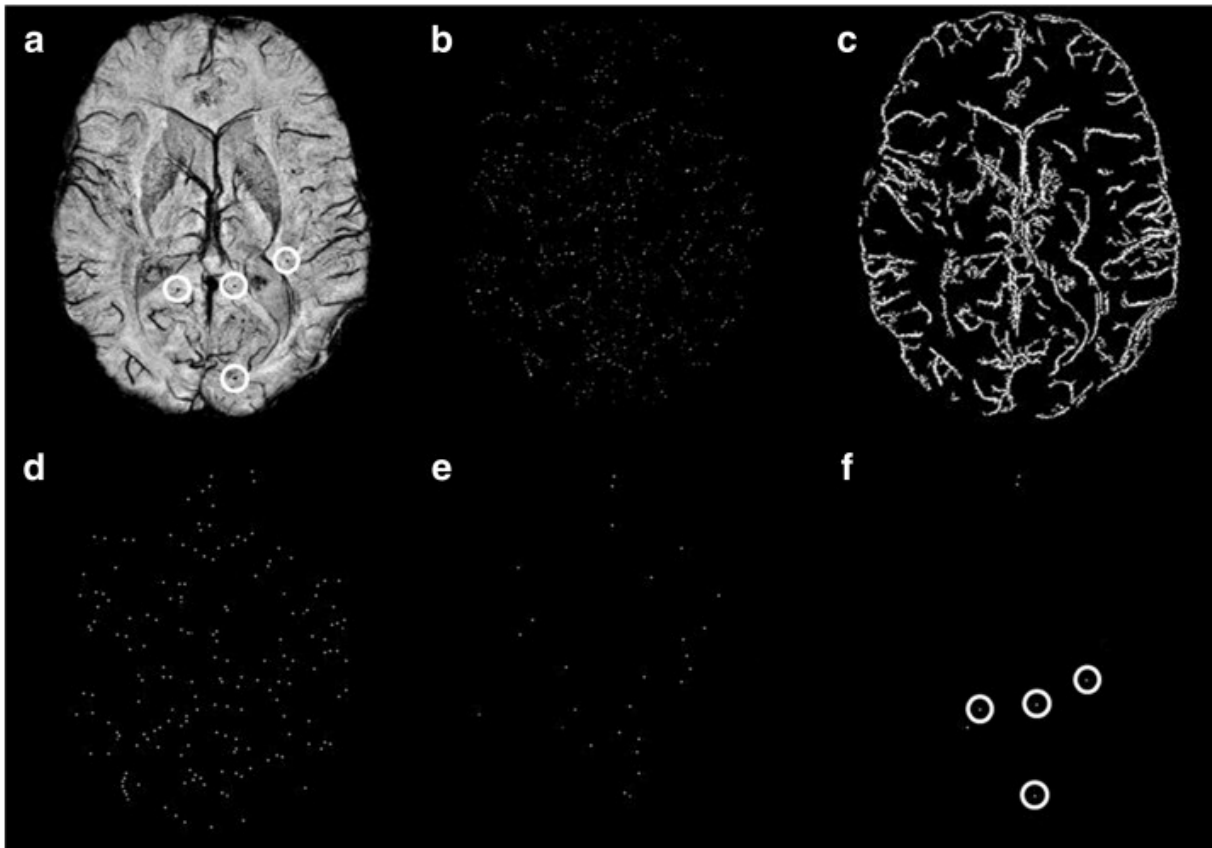


Figure 7.2 Example outputs from 2D FRST and 3D region growing.

(a) A representative SWI slice from a glioma patient with 4 CMBs (circles) on the displayed slice. (b) FRST map before thresholding. (c) Vessel mask, highlighting vessels and the edge of the brain. (d) FRST map after thresholding but before applying the vessel mask in (c). (e) FRST map after thresholding and masking (c). The bright foci are the centers of potential CMBs. Compared to the map in (d), the number of putative CMB candidates are greatly reduced by applying the mask. (f) The final output of detected CMB candidates after false positive reduction. All 4 CMBs (circles) were detected with 3 false positives left.

7.2.1.2 False Positive Reduction

The number of false positives generated during the initial detection step of our algorithm is large due to the low thresholds t_2 and t_3 used for the FRST map to intentionally maximize detection sensitivity in the first step. Stringent false positive reduction via the employment of a vessel mask, 3D region growing, and geometric feature examination is performed to eliminate the majority of these misidentified CMBs before final visual evaluation.

Step 1. Vessel mask screening using FRST outputs

Beyond its capability in the enhancement of the center of circular objects, we also found empirically that FRST helped to reduce false positives on original maps. Specifically, the orientation projection map O_l (O_n computed at $n=1$) emphasizes vessels and the edges of brain, regions where CMB mimics are typically found. The projection map O_l can therefore be used to create a binary mask in which a pixel value is 1 if its O_l decreased at least by one from its initial value (denoting vessels and the edges of brain), or 0 otherwise. The final mask is generated after removing binary regions with an area smaller than 25 pixels (a value that was comparable to the observed maximal CMB area). Only smaller CMBs undergo this vessel mask screening to prevent true large CMBs from being eliminated by the mask. Application of this mask to the FRST maps will reduce the number of initial false positives as shown in Figure 7.2 (c-e).

Step 2. 3D Region Growing

Because all thresholds used in the initial detection are applied to the 2D FRST map slice by slice, it is likely that a putative CMB will be detected multiple times on different slices. The central seed point from which to begin region growing is first determined by finding the minimum intensity of a 3D locally connected region (26-connectivity) that contains all detected

pixels of a putative CMB on SWI images. 3D region growing is subsequently performed from this center to neighboring pixels based on 26-connectivity if $|I_n - I_s| < MID$, where I_n , I_s and MID are the intensity of the neighboring pixels, seed point intensity, and the *maximum intensity difference* between pixels, respectively. The growing stops when either $|I_n - I_s| > MID$, or the distance from the seed point exceeds a maximum grown radius both in-plane (MP) and in the slice direction (MS).

Step 3. 2D Geometric feature examinations

After 3D region growing, 2D geometric features were extracted on every slice of the grown region. We chose to quantify 2D measures of area and circularity rather than 3D measures of volume and sphericity because the latter are distorted after processing. Area is measured as the total number of pixels that comprise a putative CMB and is utilized to remove false positives corresponding to large objects. Circularity (C) is defined as the ratio of the area of the CMB shape to the area of a circle having the same perimeter ($C = (4\pi \times \text{area}) / \text{perimeter}^2$). Circularity measures range from 0 to 1 depending on the length of the line, with circles having a value of 1 and lines a value approaching 0. This parameter removes objects that are irregular or elongated such as vessels. For each of the remaining CMBs, a centroid is then calculated at each slice location in order to remove false positives with slices that are shifted away from the central axis, such as transverse vessels that have a circular cross-section on 2D slices.

7.2.1.3 Parameter Selection

I. FRST

Three parameters of the FRST, k_n , n , and α , were empirically selected to achieve the highest possible sensitivity to CMBs. In our experiments, the normalization factor k_n was set to 5 for $n = 1$ and 8 for $n > 1$. These values are less than the values suggested by Loy et al. (2003), but

improve the sensitivity of the transform to smaller CMBs. As suggested by Kuijf et al. (2012), α was set to 3, and the set of radii n was allowed to vary from 1~3 pixels in order to encompass the majority of our CMBs.

II. Thresholding of FRST map

In order to first select easily identifiable true CMBs with $|S| \geq t_1$, which have relatively large radius and high contrast, a sufficiently high threshold should be used for t_1 . We therefore reviewed FRST outputs from multiple representative CMBs of intermediate size, and based on this review, empirically chose the threshold to be 170. In contrast, a sufficiently low threshold should be used for t_2 and t_3 to detect CMBs that have a smaller radius and lower contrast such that pixels with $|S| \geq t_2$ but $\leq t_1$ are also considered as potential CMB candidates. To determine the values for t_2 and t_3 , the FRST outputs from representative CMBs with extremely low contrast or small radii were investigated, and values of 65/10 were chosen for t_2/t_3 to ensure that nearly all true CMBs were included after thresholding. For any two connected pixels in the FRST map, a potential CMB is selected only if their combined $|S|$ is larger than t_2 and each individual $|S|$ is greater than t_3 . Because the t_1 value of 170 was set solely to achieve heightened detection specificity while a t_2/t_3 value of 65/10 was determined to maximize detection sensitivity, the empirical estimation of these threshold parameters from representative CMBs is feasible without the risk of over-training of these parameters.

III. Volume Control in 3D Region Growing

The main requirement for our region growing algorithm was the ability to sufficiently grow the extent of the initial region so that enough elongation is achieved to distinguish small vessels from CMBs, while preventing the generation of false negatives adjacent to neighboring hypointense regions. The parameters that control the extent of the grown region MP and MS,

were determined based on prior knowledge of CMB size (whose typical maximal diameter is approximately 6 pixels or 3 mm in diameter) and the fact that only smaller CMB candidates will undergo region growing. The values we selected for MS and MP allowed a region to maximally traverse 3 image slices (6mm) with a maximum diameter of 10 pixels (5 mm), respectively, both of which have exceeded the typical maximal size of CMBs we investigated. While an acceptable threshold for MP and MS can be easily determined, receiver operating characteristic (ROC) curve analysis with patients from a training set was necessary to establish a threshold for the MID between a grown pixel and the seed point, due to its inherently greater variability because the standard deviation of pixel intensity within CMBs and vessels can vary with image contrast

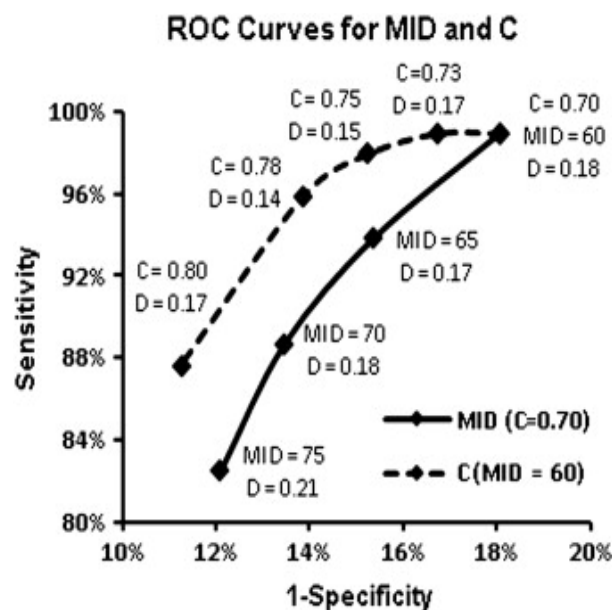


Figure 7.3 ROC curves for maximal intensity difference and circularity.

ROC curves constructed using the data from a training set that contains 5 patients and 116 true CMBS. The curves were used to determine cutoff thresholds for maximal intensity difference (MID) between the seed point and the final grown point after region growing (*solid line*; assuming $C = 0.70$), and circularity in geometric feature examination (*dashed line*; assuming $MID = 60$). When points on both curves are considered all together, $C = 0.78$ and $MID = 60$ are the optimal cutoffs in terms of their shortest distance ($D = 0.14$) to the upper left corner (Note the scales of horizontal and vertical axes are not equal). The distance was also calculated for $MID = 65$ with $C = 0.75$ and $MID = 65$ with $C = 0.78$, but neither combination gave a distance that was less than 0.14. Thus, the cutoff values for MID and C were set to 60 and 0.78, respectively.

and anatomical location. Based on the ROC curve analysis illustrated in Figure 7.3, a MID of 60 (jointly considered with C) resulted in the best overall detection performance based on minimizing distance to the upper left corner and was thus utilized in the final parameter set when validating our algorithm.

IV. Determination of thresholds for Geometric Feature Extraction

Similarly to region-growing parameters MP and MS, the maximum area on any 2D slice for a given CMB was also determined by its physical size. A conservative value of 10 pixels, which corresponds to a diameter of 3.6 pixels or 1.8 mm, was selected. A centroid shift of 1 pixel (0.5mm) was allowed in either direction to account for partial-voluming with discretization. CMB candidates with parameter values larger than these thresholds are removed. Because true CMBs that are less than 1 mm in diameter can appear more rectangular than circular in shape as limited by our pixel resolution, the range of circularity values was quite large. As a result, the threshold for circularity was determined by constructing and analyzing ROC curves jointly for MID and C as shown in Figure 7.3. When all points on both curves were considered together, C=0.78 and MID=60 were optimal cutoffs.

7.2.2. Validation of CMB Detection Algorithm in Patients

7.2.2.1. Patient Population

Fifteen patients with gliomas, who had undergone T2*-weighted MR imaging at our research center, were retrospectively selected to train parameters and evaluate the performance of our algorithm. To be eligible for this study, patients had to have received fractionated external beam radiation therapy, which damages microvasculature in normal brain parenchyma and results in the formation of CMBs. Because CMBs are not typically observed in the initial 1~2

years following cranial irradiation (Lupo et al., 2012), patients were only included if radiation was completed at least 2 years prior to MR imaging. Finally, patients were only included if they had at least 10 potential CMBs on initial screening, as these cases are where automated methods are most desired. The patients were randomly divided into two sets: a training set that included 5 patients and a test set that included 10 patients. The training set was used to construct ROC curves for C and MID to determine their optimal values, while the test set was used to evaluate the performance of the algorithm.

7.2.2.2. MR Imaging

MR images were acquired on a GE 3T whole-body system (GE Healthcare, Waukesha, WI) with an 8-channel phased array receive coil (Nova Medical, Wilmington, MA). High resolution T2*-weighted imaging using a 3D flow-compensated spoiled gradient echo sequence was performed using TE/TR=28/56ms, flip angle 20°, 24cm FOV, in-plane resolution of 0.5 x 0.5mm, 2mm slice thickness and a total slice number of 40 targeted to the area of glioma resection. A GRAPPA-based parallel imaging acquisition was implemented with a 2-fold acceleration factor in order to keep the total acquisition time under 7 minutes.

7.2.2.3. Image Reconstruction and Preprocessing

Standard SWI post-processing techniques were applied to the reconstructed k-space data for each coil, and then combined and intensity corrected (Haacke et al., 2004; Lupo et al., 2009). The skull and background were removed from reconstructed images by applying a brain mask created from the combined magnitude image with FSL's brain extraction tool software (Smith, 2002). Images were then normalized to an intensity range of 0~255 using 0 and the 98th percentile intensity of original images as the original minimum and maximum intensity, respectively. Finally, minimum intensity projection images through 8 mm-thick slabs (4 slices),

with a 6 mm-thick (3 slices) overlap between each consecutive projection, were generated from the intensity-normalized images and used for CMB identification.

7.2.2.4. *Visual Assessment of True CMB Burden*

CMBs were counted by two raters, one subspecialty-certified neuroradiologist and one trained reader. CMBs were counted independently by each reader, and discrepancies were resolved by consensus review. Raters initially counted CMBs using the proposed algorithm with parameters set for high sensitivity but low specificity. Both raters distinguished true CMBs from false-positive CMBs and additionally searched for true CMBs missed by the algorithm. The algorithm developer (WB) was blinded to the true lesion counts as determined by the interpreters, while both raters were blinded to the parameter selection. Our way of counting CMBs is driven by the finding, observed both by Kuijf et al. (2012) and by our initial experience in developing the algorithm, that an automated technique may be able to detect extra true CMBs apart from those identified by visual inspection alone. A gold standard of true CMBs, therefore, would be better constructed by comprising CMBs identified not only by visual inspection but also by automated detection.

CMBs in our gold standard were further divided into two groups by the interpreters: *definite* and *possible*. *Definite* CMBs were defined as those lesions with sufficient circular shape and hypointensity to be considered unambiguously as CMBs and not mimics on visual analysis. *Possible* CMBs were characterized by one or more of following deviations from *definite* CMBs: 1) less circular shape; 2) less tissue contrast; 3) a location or appearance that made it difficult to distinguish with confidence as representing a mimic such as a small “end-on” cortical vessel. Our criteria to scoring CMBs into the two categories are similar to those used in the literature (Conijn et al., 2011; de Bresser et al., 2012; Gregoire et al., 2009). In addition, regions where

CMBs are unlikely to occur, such as the ventricles and tumor cavity, as well as areas within a 5mm margin of the tumor cavity (which were frequently lined by confounding post-operative blood products) were excluded from the assessment.

7.3 Results

A total of 420 true CMBs were detected from 15 patients, of which 116 were from 5 patients in the training set and 304 were from 10 patients in the test set. Among the CMBs from the test set, 153 were classified as “*Definite*” and 151 as “*Possible*”. The *definite* CMBs had a larger average size (1.16 mm in diameter) and lower average minimum intensity (35.7) than *possible* CMBs (0.92 mm and 68.4). The minimum diameter detected was 0.57mm (1.13 pixel) and the largest 2.46mm (4.92 pixels). The number, diameter, and minimum intensity of these CMBs from the test set are listed in Table 7.1.

Table 7.1 Characteristics Of True CMBs Identified From 10 Patients in the Test Set

	Number			Diameter	Minimum Intensity	
CMBs	total	mean	range	Mean (mm/pixel)	Mean	Max
Definite	153	15.3	2~47	1.16/2.31	35.7	118
Possible	151	15.1	5~36	0.92/1.84	68.4	147
All	304	30.4	8~83	1.04/2.08	52.0	147

Table 7.2 summarizes the performance of the algorithm on the test set, which was able to correctly identify 263 of 304 total true CMBs, resulting in a sensitivity of 86.5%. Of these correctly identified CMBs, 16.7% (all definite) were directly identified after the FRST and did not undergo region growing and geometric feature examinations. Separating CMBs into two categories improved the sensitivity of definite CMBs to 95.4%, while, as expected, our algorithm

was less sensitive (77.5%) to possible CMBs. Figure 7.4 shows several examples of true CMBs detected by various steps of the algorithm. Of the 41 CMBs that were missed, 12 had low contrast with mean minimum intensity of 116.8 and a mean $|S|$ of 56, which was out of the selection range on the FRST maps. After applying the vessel mask to the FRST maps, 6 more true CMBs were lost because of their close proximity to vessels or tissue boundaries. The remaining 23 missed CMBs were removed in region growing and geometric feature examinations. A representative example of false negatives created at each of these steps is shown in Figure 7.5.

Table 7.2 CMB Detection Algorithm Performance Evaluated on the Test Set

CMB	True	Detected	Sensitivity	False Negative			False Positive
				FRST	Application of FP Mask	Region Growing	
Definite	153	146	95.4%	0	1	6	-
Possible	151	117	77.5%	12	5	17	-
Total	304	263	86.5%	12	6	23	449

The initial detection using the FRST identified 3162 potential CMB candidates, 90.8% of which were false positives. Only 1.0% of these false positives were produced after initial thresholding of the FRST maps with $|S| > 170$ and therefore did not undergo false positive reduction. The remaining false positives were generated after thresholding the FRST maps with lower thresholds. If the vessel mask had not been used, the number of false positives would have been 6 times as large. After region growing, 84.4% of these false positives were eliminated with a final average of 44.9 false CMBs identified per patient (range 23~65). The majority of these stemmed from tortuous, terminating, or transverse vessels and 17.4% (7.8/patient) were found in

ventricles, tumor cavities and areas of susceptibility artifacts at air-tissue interfaces as demonstrated in Figure 7.6.

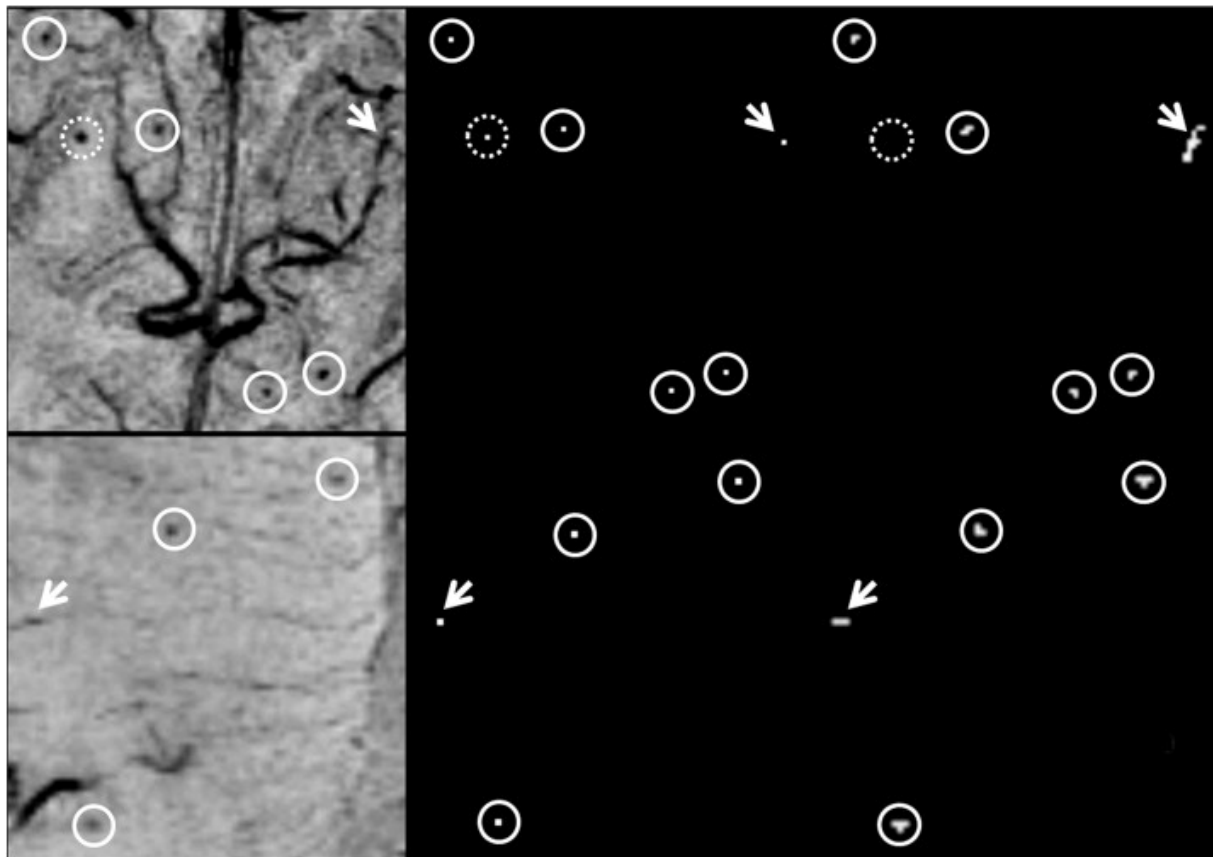


Figure 7.4 Representative examples of true CMBs detected by our algorithm.

Top row: The center of 5 Definite CMBs (circles) and 1 false positive (arrow) from a vessel on SWI image (left) are highlighted on the FRST map (middle). After 3D region growing and geometric feature examination, all true CMBs were identified and the false positive was eliminated because of the linear shape of its grown region (right). One true CMB (dashed circle) was directly identified because its $|S| > 170$, and thus did not undergo subsequent analysis.

Bottom row: three possible CMBs of low image contrast were correctly identified and one false positive was eliminated after region growing and geometric feature examination.

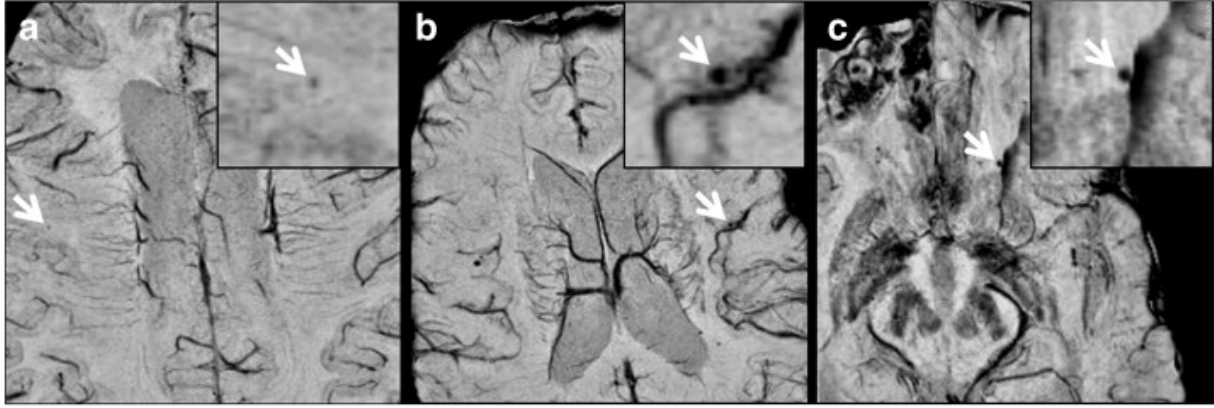


Figure 7.5 Typical false negatives found in our images.

(a) A true *possible* CMB that was missed due to low contrast compared to surrounding tissue. (b) A missed true *definite* CMB that was removed by the vessel mask because of its proximity to a neighboring vessel. (c) A true *definite* CMB that was eliminated after geometrical feature examination due to region growing into susceptibility artifacts introduced by unsuccessful phase unwrapping during SWI processing.



Figure 7.6 Typical false positives found in our images.

(a) Four false positives (arrows) seen at the ending, cross section, or turning point of vessels. (b) Two false positives found in the tumor cavity. (c) A false positive presented in the susceptibility artifacts in the air-tissue interface.

The efficiency and performance of our algorithm compared to previously published methods is presented in Table 7.3. The computation time of the algorithm was approximately 1 minute per patient using one core of a Linux workstation with Intel core 2 quad processors at 3.0GHz and 8 GB of RAM. Our algorithm achieved the highest sensitivity with the lowest

percentage of false positives per CMB and the fastest computation. Our results were also validated on a larger sample size both in terms of the total number of CMBs and the average number of CMBs per patient.

Table 7.3 Comparison of Efficiency Among CMB Detection Algorithms

Algorithm	Number of Patients	True CMBs		Sensitivity	False Positives			Computation Time
		Total	Mean		Total	Per Patient	Per CMB	
Seghier et al.	30	114	3.8	50%	NA ^a	NA	NA	3 Minutes (PC with 64-bit 3.2 GHz CPU, 12 GB RAM)
Barnes et al.	6	120	20	81.7%	645	107.5	5.4	1 Minute ^b
Kuijf et al.	18	66	3.7	71.2%	309	17.2	4.7	1 Hour ^c (One core of a standard workstation)
Bian et al.	10	304	30.4	86.5%	449	44.9	1.5	1 Minute (One core of a Linux workstation with quad processors at 3.0 GHz, 8 GB RAM)

^a The information was not provided in the publication.

^b The information of computation power was not specified in the publication.

^c The time used for image segmentation and registration was not included.

7.4. Discussion

Development of computer-aided detection methods for CMBs is challenging because of their small size, wide distribution, and the presence of other structures that mimic their appearance. The need to identify radiation-induced CMBs with low contrast is a further factor that complicates their detection. Despite several previous attempts to address this problem, it remains a topic of active research. In this study, we have presented a semi-automated CMB detection algorithm that has achieved superior performance with a sensitivity of 86.5% and computation time of 1 minute.

Achieving high sensitivity was the top priority in designing our algorithm because it has more impact than specificity on the performance that can be achieved. From a practical standpoint, visual inspection to remove false positive lesions is far easier than identifying CMBs

missed by the detection algorithm, as the eye can more rapidly dismiss lesions falsely labeled as CMBs than it can search the entire imaging volume for true CMBs. The overall performance of our algorithm depended on both the size and contrast of CMBs, which is supported by the heightened sensitivity that was observed for *definite* CMBs compared to *possible* ones (95.4% vs. 77.5%). The newly formed radiation-induced CMBs that were evaluated in this study are typically harder to detect than CMBs caused by trauma or other mechanisms of disease, as they are often smaller in size and have varied contrast. Thus, we anticipate that our algorithm would be even more robust when applied to other patient populations.

The high sensitivity of our algorithm may be explained to the strength of our false positive reduction strategy. Screening of the FRST map by applying a vessel mask allowed the use of a much lower threshold for the FRST map and maximized sensitivity to smaller and low contrast CMBs during the initial detection step. This method facilitated the detection of 96.1% of all true CMBs, which included 100% of *definite* CMBs and missed a relatively small number of *possible* low-contrast CMBs. If this vessel mask had not been applied, it would have been necessary to use a higher threshold, which would have sacrificed the detection sensitivity. Although true CMBs can be masked if they are adjacent to the structures that are included, the number of these CMBs was small (2.0%; 1 *definite* and 5 *possible*) for our experiments.

Another significant contributor to our high sensitivity in initial detection was the 3D region growing process. Although a potential pitfall that is inherent for region growing is the occasional inclusion of neighboring pixels containing nearby vessels or susceptibility artifacts (Figure 7.5c), we only applied region growing to a subset of CMBs with lower contrast and smaller size. Bypassing region growing for large CMB candidates afforded reduced thresholds for geometric features, which lead to more efficient false positive reduction. This separate

processing pipeline for large and small CMBs resulted in the removal of only 7.6% (6 *definite* and 17 *possible*) of true CMBs during subsequent region growing.

Our method achieved a higher sensitivity, faster computation speed, and a reduced number of false positives compared with other related approaches. Also the performance of our algorithm was validated on a larger sample size in terms of total number of CMBs and incidence per patient (Table 3), demonstrating the robustness of our method. In particular there was heightened sensitivity (86.5% vs. 71.2%) compared to the other method based on FRST (Kuijf et al., 2012). Moreover, it should be noted that many of our CMBs spanned a smaller number of pixels than Kuijf et al (2012) (average diameter: 2.08 vs. 2.28 pixels) and hence posed a greater challenge. This improved sensitivity applied even to the *possible* CMBs that had an average diameter of 1.84 pixels and after normalization had an average minimum intensity up to 20.4% of the maximum.

Unlike previously published studies, our elevated sensitivity is achieved without compromising computation speed. The fast computation of our algorithm originates from its simple design. Skull extraction and intensity normalization are the only preprocessing steps required, whereas other methods utilize image registration (Kuijf et al., 2012; Seghier et al., 2011) and/or segmentation (Kuijf et al., 2012) routines prior to applying their detection algorithms. In the initial detection step, gradient-based FRST can quickly locate local hypointense regions as potential CMBs. The computation time for FRST is further reduced in our algorithm by eliminating the convolution step originally proposed by Loy and Zelinsky (2003). In addition, the transform is repeatedly computed only at 3 radii in our experiments, while it was computed by Kuijf et al. at 18 radii. Finally, 3D region growing is performed within only a small local region for each candidate CMB, and only 3 geometrical features (area,

circularity and centroid) are quantified to sequentially remove falsely identified CMBs, whereas the method of Barnes et al. (2011) utilizes 14 features associated with the shape, intensity, and size of CMBs in a support vector machine to perform the classification, which takes up to multiple days for parameter training.

3D region growing was implemented in our algorithm in order to reduce the number of false positives that were present after the FRST step. This allowed geometric features of a potential CMB to be extracted and was used in our study to eliminate 86.1% of false positives. The remaining false positives (1.5/CMB and 44.9/patient) originated mostly from vessels, susceptibility artifacts, or the surgical cavity. Overall, our algorithm produced a smaller percentage of false positives per true CMB than previous ones. Since radiation-induced CMBs are often smaller and of lower contrast, there is a trade-off between producing a small number of false positives and maintaining the high detection sensitivity achieved in our study. Kuijf et al. (2012) used gray/white matter masks to exclude brain structures such as ventricles and sulci, where false positives are often observed. The disadvantage of this approach is that it requires the acquisition of a T1-weighted image as well as the application of registration and segmentation algorithms that prolong the total processing time. In addition, all of our patients received intracranial tumor resection, which produces structures that mimic CMB (see Figure 7.6b). These structures can be removed quickly during the final visual inspection because of their obvious anatomical location. A strict performance comparison of automated detection algorithms is difficult at this point, as the size and contrast of CMBs may vary with other experimental factors such as field strength and resolution (Nandigam et al., 2009). The manual review process and patient inclusion criteria are also all different among these studies. It is desirable in the future to construct a standard CMB database, in which CMBs are categorized by their disease type, MR

imaging field strength, distribution or other related factors. This will not only help objectively evaluate automated CMB detection algorithms but also facilitate the training process for these algorithms.

The success of our algorithm demonstrates the advantage of using a vessel mask to remove false positives in achieving a high sensitivity while maintaining a reasonable specificity in CMB detection. While methods that are able to create a high quality vessel mask in 3D using SWI have been proposed in literature (Koopmans et al., 2008), we used the vessel mask from the FRST transform in our approach because of its robustness and simplicity in integrating with our algorithm's pipeline. The better delineation and continuity of veins on SWI compared to non-projected images facilitated both the generation of a more reliable vessel mask by FRST and a greater extent of region growing on these structures, both of which aided in reducing false positives. The improved false positive reduction can in turn be used to enhance detection sensitivity, e.g., set up a low threshold to screen the FRST map and make the initial detection highly sensitive. Limitations associated with the usage of SWI images include susceptibility artifacts at air-tissue interfaces and magnified background noise that is introduced during reconstruction of SWI images that can be a potential source of false positives (Figure 7.6c). Also the original location of CMBs cannot be determined on these images because of the projection processing, but it can easily be recovered on the original non-projected images. The projection processing may also accidentally project some CMBs in or close to certain dark structures that do not surround them in actual anatomy, leading to decreased FRST response or leakage in region growing, which both increase the number of false negatives. Despite these limitations, using FRST on SWI images is especially advantageous for the detection of radiation-induced CMBs, whose essentially low contrast on conventional T2*-weighted magnitude images as the

result of its small size is greatly improved on SWI (Lupo et al. 2012). Furthermore, processing MR image using has been suggested as a required step for visual CMB detection, especially when high resolution images acquired at high field strengths (e.g., 3T or 7T) are used (de Bresser et al., 2012). As visual inspection (e.g., further false positive removal) of the output from automated CMB detection is still required, it is desirable to design an algorithm that operates on the same images that are used for subjective interpretation.

Finally, there are several limitations associated with our algorithm itself. First, while 3D region growing is a simple and fast method to segment putative CMBs, its capability to discriminate desired objects from close but dissimilar background is limited, making the segmentation leakage being more likely to happen for CMBs with low image contrast. To improve the accuracy of the segmentation, more advanced and finer techniques such as active contours and level sets may be used (Osher and Sethian, 1988), but at the expense of computation time. Second, at the very first and last image slices, the efficiency of geometric examination becomes lower due to limited space for region growing along slice selection direction. This may result in there being a higher number of false positives on these slices than on inner slices. Like previous published methods, training is an inevitable step that is required to apply our algorithm to detect CMBs from different types of diseases, different MR field strengths, and even different scan parameters such as TE, spatial resolution, and slice thickness. Size and contrast variation are the principal reasons that training is necessary, as most of parameters selected in our algorithm require that the range of CMB size and intensity be considered. However, the time used for training our algorithm is small as most of the parameters used in the algorithm can be empirically determined by studying a few representative CMBs as long as prior knowledge about the size of CMBs is available. Optimal values for other parameters such as MID and C can be

determined empirically or formally by constructing ROC curves on a small training dataset such as we did in this study.

7.5. Conclusions

This study presented a method for semi-automated CMBs detection that uses SWI, 2D FRST and 3D region growing. The FRST is used for initial lesion detection, and false positives are removed from putative CMBs identified in the first step using a region growing process with geometric feature examination. Our method achieved higher sensitivity with an acceptable number of false positives and faster computation time when compared to previously developed methods. Although it was evaluated for CMBs arising in the setting of prior radiation therapy for gliomas, its superior performance is likely to be of interest for detecting CMBs associated with other neurologic disorders, including CAA, hypertension, TBI and stroke.

Chapter 8 Summary and future work

In this dissertation, we investigated several aspects of MR phase and susceptibility-weighted imaging, including their sequence design, post-processing, and clinical applications for characterizing abnormal iron deposition in MS lesions and CMBs in patients with brain tumors who were treated with radiation therapy. In this chapter, the results from our research are summarized and future directions related to our work are proposed.

8.1 Summary of results

8.1.1 MR phase imaging of MS lesions

To investigate the clinical applications of phase images, we performed a longitudinal phase imaging study of chronic MS lesions and found no obvious change in phase contrast of the lesions, suggesting that the iron accumulated in the lesions may not be contained in active macrophages as previously hypothesized. We also noticed that some lesions appeared on phase images before they were present on magnitude images, indicating that phase images may be more sensitive to certain subtypes of MS lesions. The observations from this and future studies that use this technology may contribute to a better understanding of the mechanism of phase contrast, the dynamic progress, and the pathology of MS lesions.

8.1.2 Data acquisition for SWI of CMBs

We implemented a Gradient-echo sequence with 4 echoes at 7T that is able to simultaneously acquire MR angiography and SWI images, which was a primary goal when this multi-echo acquisition approach was firstly proposed (Du et al., 2008). The quality of both MRA and SWI images from our multi-echo sequence are comparable to that of images from individual single-echo sequences. Beyond this achievement, we made the sequence more sensitive to small

CMBs by using a higher slice resolution that was not achievable for a single-echo SWI sequence. The higher slice resolution was made possible in two ways, by creating a mean SWI image from several echoes, and by implementing the sequence at 7T. Because of the higher resolution, more CMBs were detected and most of them are small in size, which may be helpful in studying the formation and evolution of CMBs. Overall our multi-echo sequence opens a window towards the characterization of both CMBs and its surrounding microvasculature, which is the ultimate source of CMBs.

Although SWI is generally preferred at higher field strengths, this is not always true for imaging CMBs. In comparing SWI of CMBs between 3T and 7T, we showed that the sensitivity of images to CMBs was affected by the distribution of CMBs as well as the field strength. When CMBs are located in the regions close to air-tissue interfaces in the brain, attempting to image them at a high field strength such as 7T may not be ideal as susceptibility artifacts could dominate under these circumstances.

8.1.3 Automated detection of CMBs on SWI images

Although SWI has been widely recognized as a sensitive tool to visualize CMBs, efforts in exploring the clinical relevance of CMBs are hindered by the time taken to identify them, which is impacted by their small size, wide distribution in the brain, and presence of many mimics. In this dissertation, we have presented an automated method to identify CMBs on SWI images. Our method is sensitive and fast with an acceptable number of false positive output, which can be quickly be removed by manual inspection. The performance of our method is superior in terms of detection sensitivity and computational speed compared to several other automated CMB detection methods. The efficiency of the quantification of the number, size, and location of CMBs can be greatly improved using our algorithm, which will advance the

investigation of the role of CMBs as a diagnostic and/or prognostic marker for neurological disorders.

8.2 Future work

8.2.1 Phase contrast mechanisms in MS lesions

When inspecting MS lesions on phase images in our study, we attributed the main source of phase contrast for MS lesions to iron deposition. However, this speculation may not reflect the whole story and has been challenged by recent studies (Yablonskiy DA et al., 2012 ; Yao B et al., 2012), which suggested that myelin/axonal integrity could also contribute to changes in phase shifts. To validate this hypothesis, a promising direction is to utilize information from diffusion-weighted MR imaging and MR spectroscopic imaging, albeit at far lower spatial resolution. The former technique can generate white matter tractography while the latter can measure levels of N-acetyl aspartate (NAA) in white matter, which may be considered as measures of myelin/axonal integrity.

8.2.2 Clinical relevance of CMB

In this dissertation, we focused on finding efficient technical solutions for visualizing and identifying CMBs that are induced by radiation therapy based upon the design of MR imaging sequence and an automated detection algorithm. We anticipate that future research will be able to use these methods to investigate the clinical relevance of CMBs. For example, we noticed in our study that the number of radiation induced CMB numbers could vary dramatically, from a few to hundreds, even when radiation dose and delivery were similar. It is currently unclear what factors could contribute to the variation, and the answer to this observation may help to identify a

subgroup of patients with brain tumors who are vulnerable to radiation therapy, which will be a significant contribution to designing treatment plans for these patients.

8.2.3 Multi-echo SWI

We achieved a higher resolution SWI in this work by averaging images acquired from multiple echoes. However, the benefits of using multiple echoes are not limited to improved image resolution or SNR. The recent trend of integrating information from multiple echoes to improve multi-channel phase image combination (Robinson et al., 2012), phase unwrapping (Feng W et al., 2013; Robinson et al., 2013), susceptibility artifact suppression (Oh SS et al., 2013) and quantitative susceptibility mapping (Wu et al., 2012; Xu et al., 2013) has extended the potential of multi-echo gradient-echo imaging. It is likely that multi-echo SWI will continue to be an intensive research area with interests focusing on developing a variable of novel post-processing techniques.

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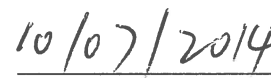
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