Advanced MR Technique Development for Improved Characterization of Multiple Sclerosis

by

Meredith Auman Metcalf

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
Copyright 2008

by

Meredith Auman Metcalf
Acknowledgements

The writing of this dissertation was an individual effort, yet it was obviously not possible for me to complete this experience without the practical and personal support of many people. Thus, I am extremely grateful to all my superiors and colleagues at UCSF and UC Berkeley, my family, and my friends for their support and patience over the last six years. I would especially like to recognize the following people:

First and foremost, I would like to thank Dan Vigneron, my primary thesis advisor, for his guidance and encouragement throughout my graduate experience. I would not have finished my PhD without his encouragement, nor could I have completed this work without his input and constant reassurance that he was available to help if I needed it. I would also like to recognize Roland Henry for his supervision and assistance during the first half of my dissertation work. I am extremely grateful for his brilliant ideas, bubbly personality, and his ability to relate to me as I worked my way through the process of scientific- and self-discovery.

I was fortunate to work with many brilliant doctors and scientists while at UCSF who I would like to recognize for their efforts. I would especially like to acknowledge the work of Dr. Daniel Pelletier for serving on my qualifying exam and dissertation committees, as well as for his general enthusiasm for research and directing a large portion of my multiple sclerosis-related projects. Thanks to
Sharmila Majumdar for steering me through this process as my graduate advisor and also for serving as a member of my qualifying exam and dissertation committees. The help and encouragement I received from Sarah Nelson, John Kurhanewicz, and Steve Conolly as members of my qualifying exam committee were also very important, and I appreciate it very much.

A significant amount of advice, knowledge, distractions, and entertainment came from my interactions with peers and members of collaborating research groups at UCSF and Stanford. I would especially like to point out the scientific help given to me by Doug Kelly, Darin Okuda, Pratik Mukherjee, Radhika Srinivasan, Lucas Carvajal, John Pauly, Taeksoo Kim, Jeff Berman, Sugwon Chung, and Kathleen Garrison. Also, I could not have completed this experience without the advice and group outings provided by Duan Xu, Albert Chen, Cornelius VonMorze, Sri Veeraraghavan, and Marisa Thorne.

Most importantly, I would like to recognize the love and support I received from my parents Glenn and Susan Metcalf, my sister Amy Metcalf-Sams, and my boyfriend Andrew Burke, without whom I cannot imagine completing this experience.
Advanced MR Technique Development for Improved Characterization of Multiple Sclerosis

by

Meredith Auman Metcalf

Magnetic resonance imaging (MRI) provides many ways to study human brain anatomy as well as advance our understanding of brain structure, function, and processes that occur with disease. This dissertation project investigated the clinical utility of new and existing MRI techniques to the study of changes that occur in the brains of patients with multiple sclerosis (MS). The new MRI techniques were developed and tested with the goal of attaining more sensitive methods for disease detection than those that are currently available to clinician scientists. These new bioengineering developments included both novel MR acquisition and data analysis techniques for improved non-invasive, quantitative assessments of multiple sclerosis disease processes.

There are many quantitative MRI methods that can be used to detect tissue differences in patient populations when compared to controls. This dissertation research probed the utility of volumetric measurements, T1-relaxation times, and diffusion tensor imaging (DTI) metrics to detection of tissue damage and disease in early stage MS. New DTI methods were developed and new ultra high-field MR hardware and techniques were investigated to examine their potential in
identifying previously undetectable brain changes in patients. This bioengineering project included acquiring patient and healthy control data, MRI technique development and optimization, developing and implementing appropriate image-processing methods for parameter measurement, cross-sectional and longitudinal investigations, and determining appropriate statistical approaches for comparative studies.

It was determined that T1-relaxation and DTI measures of mean diffusivity ($D_{av}$), fractional anisotropy (FA), and transverse diffusion (EvT) were sensitive enough to detect brain tissue changes at the earliest presentation of disease. Although volume measurements were not different as a group at presentation, atrophy was detectable via volumetry at one-year post baseline. Additionally, DTI measures were able to predict this change, an indication of a causal relationship between microscopic damage and overall tissue loss. The diffusion technique development and high field MRI methods demonstrated the feasibility to collect images with advantages in image quality, signal to noise (SNR), resolution, and ability to detect disease over what was previously available. These developments provide the platform for future work in MRI and its application in MS to better characterize disease.
# Table of Contents

Chapter 1: Introduction 1

1.1 Motivation 1

1.2 Dissertation Objectives 2

1.3 References 4

Chapter 2: Background 6

2.1 Overview 6

2.2 Fundamentals of Magnetic Resonance Imaging 6

  2.2.1 Origin of the MR Signal 7
  2.2.2 Relaxation 10
    2.2.2.1 Longitudinal and Transverse Relaxation 10
    2.2.2.2 Image Weighting 12
  2.2.3 Image Acquisition 12
    2.2.3.1 Spatial Localization 12
    2.2.3.2 Pulse Sequence Basics 14

2.3 Techniques Used in this Dissertation 16

  2.3.1 Image Segmentation and Volumetry 16
  2.3.2 T1-Relaxation Measurements 18
  2.3.3 Diffusion Tensor Imaging 19
    2.3.3.1 Basic Principles of Diffusion 19
    2.3.3.2 Diffusion Tensor Imaging 20
    2.3.3.3 Steady State Free Precession Diffusion Imaging 22
    2.3.3.4 Fiber Tracking 22
  2.3.4 High Magnetic Field Strength Imaging 23

2.4 Multiple Sclerosis 25

  2.4.1 Brief Overview of Disease 25
  2.4.2 Application of MRI to MS 26
    2.4.2.1 Conventional MRI 27
    2.4.2.2 Advanced MRI Techniques 29

2.5 References 33
Chapter 3: Early MRI Detectable Brain Changes in Patients with Clinically Isolated Syndromes (CIS) Suggestive of MS 41

3.1 Introduction 41
   3.1.1 Application of MR Techniques to Multiple Sclerosis 41
   3.1.2 Project Goal in Patients with Clinically Isolated Syndromes 42

3.2 Materials and Methods 43
   3.2.1 Patients 43
   3.2.2 Image Acquisition 44
   3.2.3 Image Processing 44
      3.2.3.1 Calculation of Maps 44
      3.2.3.2 Lesion Identification 47
      3.2.3.3 Tissue Segmentation and Volume Calculations 47
      3.2.3.4 DTI Segmentation 48
   3.2.4 Histogram Analysis 49
   3.2.5 Statistical Analyses 49

3.3 Results 50
   3.3.1 Volume Measurements 50
   3.3.2 T1-Relaxation Time Estimates 51
   3.3.3 DTI 52
   3.3.4 Correlations 55

3.4 Discussion 57
   3.4.1 Earliest Detectable Measurement of Atrophy in CIS 57
   3.4.2 T1 as a Marker of Disease Processes 58
   3.4.3 DTI as a Marker of Disease Processes 58
   3.4.4 Interpretation of Parameter Correlations 59

3.5 Conclusion 60

3.6 References 61

Chapter 4: Longitudinal Brain Changes in Patients with Clinically Isolated Syndromes (CIS) Suggestive of MS 64

4.1 Introduction 64
   4.1.1 MR Measures of Disease in Clinically Isolated Syndromes 64
   4.1.2 Goal of Longitudinal Study 64

4.2 Materials and Methods 65
   4.2.1 Research Participants 65
   4.2.2 Image Acquisition 66
   4.2.3 Image Post-Processing 66
4.2.3.1 Lesion Identification 66
4.2.3.2 MRI-Derived Brain Tissue Parameter Calculation 67
4.2.3.3 Fiber Tracking 68
4.2.4 Statistical Methods 69

4.3 Results 69
4.3.1 Clinical Data 69
4.3.2 MRI Metrics Measured Over One Year 70
  4.3.2.1 Brain Volume Measurements 70
  4.3.2.2 T1-Relaxation Estimates 71
  4.3.2.3 DTI Parameters 72
4.3.3 Baseline Measures as a Predictor of Volume Change 73
4.3.4 Fiber Track Specific Changes 75

4.4 Discussion 78
4.4.1 Interpretation of Longitudinal Metrics 78
4.4.2 Predictability of Volume Change 80

4.5 Conclusion 80

4.6 References 81

Chapter 5: Steady-State Free Precession Diffusion Imaging at High Field Strength (7 Tesla) 84

5.1 Introduction 84
  5.1.1 Diffusion Tensor Imaging 84
  5.1.2 Limitations of Current DTI Tools 84
  5.1.3 3D DW-SSFP Pulse Sequence 85
  5.1.4 High Field Strength MRI 86

5.2 Materials and Methods 87
  5.2.1 Calculation of Sequence Parameters 87
  5.2.2 Image Acquisition 88
  5.2.3 Image Post-Processing 89
    5.2.3.1 Data Processing and Image Reconstruction 89
    5.2.3.2 Motion Correction 89

5.3 Results 90
  5.3.1 3D DW-SSFP Pulse Sequence Parameters 90
  5.3.2 Preliminary 3D DW-SSFP Imaging Results 91

5.4 Discussion 93
List of Tables

Table 3-1: T1, DTI, and volumetry correlations 56
Table 3-2: T1 and DTI correlations 57

Table 4-1: CIS patient and control population clinical information 70
Table 4-2: Correlation coefficients for baseline T1 and DTI vs volume changes measured over one year. 74

Table 6-1: T2*-weighted GRE Imaging Parameters 102
Table 6-2: TE optimization for 7T T2*-weighted GRE 105
List of Figures

Figure 2-1: $^1$H nuclei orientation in the absence and presence of $B_0$ 8
Figure 2-2: Precession and excitation of the NMV 9
Figure 2-3: $T_1$ and $T_2$ relaxation curves 12
Figure 2-4: Basic 2D MRI pulse sequence 15
Figure 2-5: Lesion appearance on T2 and T1-weighted MRI 28

Figure 3-1: T1-weighted image and derivatives from a CIS patient 45
Figure 3-2: Calculated DTI maps. (a) $\lambda_1$, (b) $\lambda_{23}$, (c) $D_{av}$, and (d) FA 46
Figure 3-3: Anisotropy segmentation overlaid on FA map 49
Figure 3-4: z-score plots of the nWM, nGM, and nBV for each subject 51
Figure 3-5: T1-estimate analysis normalized histograms z-score plots 52
Figure 3-6: Histogram analysis for NAWM and HA-NAWM $\lambda_1$ and $\lambda_{23}$ 54

Figure 4-1: CIS nWM, nGM, and nBV z-scores at baseline and 1-year 71
Figure 4-2: CIS NAWM T1 z-scores at baseline and 1-year 72
Figure 4-3: CIS HA-NAWM DTI z-scores at baseline and 1-year 73
Figure 4-4: Baseline EvT correlation to nGM volume change over 1-year 74
Figure 4-5: Correlation of baseline ADC with nBV change over 1-year 75
Figure 4-6: T2 and ev2 difference maps for a patient and control 76
Figure 4-7: Fiber track specific FA changes through time 77

Figure 5-1: Phase navigated 3D SSFP diffusion weighted sequence 86
Figure 5-2: Magnetic susceptibility effects on 3T EPI and SSFP images 87
Figure 5-3: 7T $b=0$, $b=1000\text{s/mm}^2$, and $D_{av}$ diffusion weighted imaging 92
Figure 5-4: Phase error motion correction using efficient refocusing 93

Figure 6-1: 7T Image Intensity Before and After Filtering 104
Figure 6-2: 7T 2DGRE for Optimization of TE 106
Figure 6-3: 7T T2*-Weighted Images from a Health Control 107
Figure 6-4: 7T T2*-Weighted Images from a MS Patient 108
Figure 6-5: Juxtacortical and Subcortical Lesions Images at 7T 109
Figure 6-6: Cortical Grey Matter Lesion at 7T 109
Figure 6-7: 7T Sagittal Comparison of Control vs MS 111
Figure 6-8: 7T DTI Scan from Control Subject 112
Figure 6-9: 7T DTI Scan from an MS Patient 113
Chapter 1: Introduction

1.1 Motivation

Multiple sclerosis (MS) is a chronic disease of the central nervous system (CNS) in which the body’s immune system attacks its own myelin.\(^1\) In these attacks myelin is lost in multiple areas, and is replaced with scar tissue known as sclerosis. In addition to the damage incurred on the protective sheath, the nerve fiber itself may be injured in these attacks. Over time, the damage from multiple episodes combines to slowly degrade neurological function.\(^2\) Well over 2,500,000 people around the world are currently living with some form of MS, however, the prevalence varies around the globe from 2 – 150 cases per 100,000 people, depending on the country or specific population.\(^3\) The etiology and complex pathophysiology of MS are still not well understood despite the pervasiveness of the disease and the fact that it has been studied intensely in the more than 100 years since it was first described in 1868 by Jean-Martin Charcot.\(^4\)

Over the past two decades, MRI has emerged as a powerful noninvasive tool to assist in the diagnosis and monitoring of MS.\(^5\) Conventional MR imaging techniques such as T1, T2, and proton density (PD) weighted images provide excellent tissue contrast between regions of MS-induced inflammation and sclerosis, and healthy brain tissue. These morphologic images in turn allow physicians to visualize lesions and evaluate spatial dissemination in time, making the diagnosis of MS more robust and clinically feasible at an earlier stage of disease. Due to the excellent ability of MRI to detect and monitor lesions, MRI
has emerged as a key supportive therapeutic outcome measure in MS-related clinical trials. Visualizing lesions on MR images has revolutionized the way MS is diagnosed and treated, however, these anatomic findings do not elucidate the underlying mechanisms for disease.

In MRI, adjacent tissues develop contrast when there are differences in MR-sensitive parameters. It is now well known that these parameter variations can be used to visualize lesions in MS, but what causes these changes is also of value. Understanding the underlying disease processes will increase the potential for developing more effective therapies, which in turn, will improve the quality of life for patients living with this disease. In recent years, MRI research in MS has focused more on finding new ways to measure, interpret, and understand the tissue abnormalities that occur not only in the lesions, but also in the tissues of the CNS that may appear healthy in conventional MR images.

1.2 Dissertation Objectives

The goal of this project was to develop and apply both image post-processing methods and novel acquisition techniques to improve the understanding of the underlying pathophysiology in multiple sclerosis through enhanced detection of small lesions and finer monitoring of MRI changes in the brain parenchyma as well as investigating their variations through time. This work included rigorous image analysis in cross-sectional and longitudinal studies of early stage MS. The findings in these early studies supported the idea to implement new strategies for
image collection and analysis on state-of-the-art high-field scanners that were determined to be feasible to use on MS patients. This dissertation is organized into the following chapters, each of which focuses on a specific one of the multiple bioengineering studies that together comprise the overall project designed to attain the project goals. Specifically:

**Chapter 2** provides a brief overview of background information relevant to the work completed for this dissertation. This chapter begins with basic MR physics and the image acquisition techniques. From there, the chapter transitions to applicable imaging methods and post-processing techniques including MR-derived metrics such as T1 measures, brain volumetry, and diffusion tensor imaging (DTI). This chapter concludes with a short introduction to Multiple Sclerosis (MS) and the use of MRI in this disease.

**Chapter 3** utilizes the post-processing techniques and derived MR metrics from Chapter 2 to study the brains of patients with a clinically isolated syndrome (CIS) suggestive of MS, the earliest possible stage of disease. The findings are compared with control subjects and interpretations are presented as to the underlying mechanisms for the apparent brain changes.

**Chapter 4** expands on the patient cohort investigated in Chapter 3, and looks at how the MR parameters change with time, and well as how those parameters
correlate with physical changes in the patient. Hypotheses are posed for the disease processes that occur to bring about and sustain these changes.

Chapter 5 presents steady-state free precession diffusion weighted imaging (SSFP-DWI), and investigates using this technique to achieve diffusion weighting at higher field strengths. This method provides high signal-to—noise ratios (SNR) diffusion images without the distortions apparent in standard echo-planar imaging (EPI). An optimized protocol was developed for using this technique to image at high field strengths and the calculation of the diffusion tensor using this method is discussed.

Chapter 6 describes the initial exploration of imaging MS using ultra high-resolution phased array MRI at 7 Tesla. The specialized coils and sequence optimizations are discussed as well as challenges and future promises of high-field imaging in MS patients.

Chapter 7 summarizes the major results in the previous chapters, discusses the clinical relevance of the work, and offers possible directions for future studies.

1.3 References


Chapter 2: Background

2.1 Overview

This chapter provides a brief overview of magnetic resonance physics and a biological foundation for the research projects completed as part of this dissertation. Basic MR physics is presented beginning with the origin of the MR signal through the general principles of image acquisition. Advanced imaging techniques and applicable post-processing methods that were used in this work are then explained based on the basic principles that were laid out in the first portion of the chapter. Advanced methodologies covered include brain volumetry, T1-relaxation parameter measurement, diffusion weighted imaging, including diffusion tensor imaging (DTI) and fiber tracking, and high magnetic field strength MRI. The background information concludes with a short introduction to Multiple Sclerosis (MS) and the many applications of MRI in this disease.

2.2 Fundamentals of Magnetic Resonance Imaging

Magnetic resonance imaging (MRI) is an imaging technique used in medical settings to produce high quality images of the inside of the human body. MRI is based on the principles of nuclear magnetic resonance (NMR), a spectroscopic technique discovered independently in 1946 by both Felix Bloch and Edward Purcell.\(^1\) In the period between 1950 and 1970, NMR was developed and used for chemical and physical molecular analysis. In 1971 Raymond Damadian showed that the nuclear magnetic relaxation times of tissues and tumors differed,
thus motivating scientists to consider magnetic resonance for the detection of disease.\textsuperscript{3} The following decades included significant research and development to achieve the current situation, where MRI one of the most widely applied medical imaging techniques in clinical practice. This section presents an introduction of the basic principles of MRI.

2.2.1 Origin of the MR Signal
The principles of MRI rely on the spinning motion of specific nuclei known as MR active nuclei. Nuclei that have a net charge and have spin acquire a magnetic moment and are able to align with an external magnetic field. Important examples of MR active nuclei include $^1\text{H}$, $^{13}\text{C}$, $^{15}\text{N}$, $^{17}\text{O}$, $^{19}\text{F}$, $^{23}\text{Na}$, and $^{31}\text{P}$; however, this dissertation and the majority of clinical MRI focus on imaging $^1\text{H}$ which is very abundant in the human body and has a relatively large magnetic moment, thus results in a detectable signal.\textsuperscript{4}

In the absence of an external magnetic field, the magnetic moments of the hydrogen nuclei in the body are randomly oriented. When a patient is placed in a strong static magnetic field such as in the bore of an MRI machine, the magnetic moments of the $^1\text{H}$ nuclei, primarily in water and lipid molecules, align with the magnetic field. Some of the $^1\text{H}$ align parallel, whereas a smaller number of the nuclei align anti-parallel to the main magnetic field. Quantum physics describes the properties of electromagnetic radiation in terms of discrete quantities of energy; however, the physics and mathematics of quantum mechanics are
beyond the scope of this bioengineering dissertation. The resulting magnetization can also be described in a classical sense as the net magnetization vector (NMV), which is the resulting difference in the parallel and anti-parallel nuclei, and it is this net magnetization that is detected in MRI.

![Figure 2-1. A) In absence of an external magnetic field, $^1$H are randomly oriented. B) In the presence of $B_0$, nuclei align and result in a net magnetization, which becomes the MRI signal.](image)

Each hydrogen nucleus that makes up the net magnetization is spinning on its axis, and the influence of the main magnetic field ($B_0$) produces an additional spin called precession. The frequency at which the nuclei precess about the main magnetic field is governed by the Larmor equation, which states that:

$$\omega_0 = \gamma \times B_0 \quad \text{Equation 2-1}$$

where $\omega_0$ is the resulting precessional frequency, $\gamma$ is the gyro-magnetic ratio, a constant for each specific nuclei (for $^1$H, $\gamma = 42.57$ MHz/T), and $B_0$ is the external magnetic field strength. Resonance is a phenomenon that occurs when an
object is exposed to an oscillating perturbation that has a frequency close to its own natural frequency of oscillation. When the $^1\text{H}$ nuclei are exposed to energy at the precessional frequency, they can absorb energy from that source. Keeping with the classical mechanical description, the result of this energy input from a radiofrequency (rf) pulse is called excitation, and can be thought of as the tipping of the net magnetization of the spins away from $B_0$ and into the transverse plane.

![Figure 2-2. A) Without external perturbation, the NMV precesses about $B_0$. B) Following a short burst of rf energy at the resonant (Larmor) frequency, the NMV tips away from $B_0$, and precesses in the transverse plane.](image)

As a result of resonance and energy put into the system by a short burst of rf, the net magnetization precesses in the transverse plane. Due to Faraday's laws of induction, the moving magnetic field caused by the body's protons rotating in the transverse plane induces a voltage in the receiver coil. This resultant voltage is
the MR signal, the frequency of which is the same as the Larmor frequency, and the magnitude depends on the amount of magnetization in the transverse plane.

The Bloch equation (Equation 2-2) can be used to describe the behavior of a magnetization vector, \( \mathbf{M} \), under any conditions. The three terms on the right side of the equation describe precession of the magnetization vector, as well as how the longitudinal \( (M_z) \) and transverse \( (M_{xy}) \) magnetizations return to equilibrium.

\[
\frac{\partial M(t)}{\partial t} = \gamma M(t) \times B + \frac{1}{T_1} (M_0 - M_z(t)) - \frac{1}{T_2} M_{xy}(t) \quad \text{Equation 2-2}
\]

2.2.2 Relaxation

During rf excitation, protons absorb energy via resonance, but when the rf is turned off, the absorbed energy begins to be given off by the protons and dissipated to their surroundings. Sticking with the classical mechanical description of the NMV, this corresponds to the magnetization returning from the transverse plane to realign with \( B_0 \). The processes by which the \(^1\)H nuclei return to their equilibrium states are accounted for by the \( T_1 \) and \( T_2 \) terms in the Bloch equation (Equation 2-2, above), and are known as \( T_1 \) and \( T_2 \) relaxation.

2.2.2.1 Longitudinal and Transverse Relaxation

\( T_1 \) relaxation is caused by the nuclei giving up their energy to the surrounding environment or lattice, and can be referred to by many names, including spin lattice relaxation, longitudinal relaxation and \( T_1 \) relaxation. Energy released to
the surrounding lattice causes nuclei to recover their longitudinal magnetization. The rate of recovery is an exponential process, with a recovery time constant called $T_1$. This is the time it takes 63% of the longitudinal magnetization to recover in the tissue.

$T_2$ decay is caused by nuclei exchanging energy with neighboring nuclei. The energy exchange is caused by the magnetic fields of each nucleus interacting with its neighbor. This form of relaxation is often termed spin-spin relaxation, transverse relaxation, or simply $T_2$ relaxation, and results in a decay or loss of transverse magnetization. This rate of decay is also an exponential process, so that the $T_2$ relaxation time of a tissue is its time constant of signal loss. It is defined as the time it takes 63% of the transverse magnetization to be lost. Inhomogeneities in the main magnetic field can cause additional dephasing of the transverse magnetization. This dephasing, combined with $T_2$ relaxation is defined as $T_2^*$ relaxation.$^5$
2.2.2.2 Image Weighting

One of the main advantages of MRI compared with other imaging modalities is the excellent soft tissue discrimination in the images. The contrast characteristics of different tissues in images depend on many variables including tissue composition and acquisition timing parameters. In MRI, the most basic forms of contrast, proton density, $T_1$, and $T_2$-weighting, are achieved because different tissue types have varying concentrations of $^1$H, and also different relaxation times resulting in different MR signals.$^3$

2.2.3 Image Acquisition

2.2.3.1 Spatial Localization

As previously mentioned, a short burst of RF energy at the precessional frequency tips the NMV so that it is flipped into the transverse plane. The resulting coherent transverse magnetization precesses at the Larmor frequency,
inducing a current in the receiver coil, and thus constitutes the MR signal. After the system has detected the signal, it must be able to resolve the signal spatially in three dimensions so that it can position each component at the correct point to create an image. Three basic steps that are used to accomplish this task are slice selection, frequency encoding, and phase encoding.

Slice selection in MRI is the excitation of spins in a plane through the object. In order to do this, a gradient coil is switched on during the period that the RF pulse is applied. When the gradient is on, the magnetic field and thus the precessional frequency of $^1$H will vary in a linear fashion in the direction of the gradient according to Equation 2-1. A slice can therefore be selectively excited by transmitting RF with a bandwidth of frequencies coinciding with the Larmor frequencies of spins in a particular slice as defined by the slice select gradient.

Once a slice has been selected, the signal coming from it must be located along both axes of the image. The first step towards the in-plane localization is phase encoding. When the phase encoding gradient is switched on, the magnetic field strength and therefore the precessional frequency of nuclei along the axis of the gradient is altered. As the speed of precession of the nuclei changes, so does the accumulated phase of the magnetic moments along their precessional path. There is now a phase difference or shift between nuclei positioned at different locations along the axis of the gradient. When the phase encoding gradient is switched off, the magnetic field strength experienced by the nuclei returns to $B_0,$
and therefore the precessional frequency of all the nuclei returns to the Larmor frequency; however, the spatially dependent phase difference between the nuclei remains.

The final step in spatial localization occurs as the signal is received, and is called frequency encoding. When the frequency encoding gradient is applied along the axis remaining to be encoded, the frequency will vary at each point in that direction. Because this process is being carried out as the computer is reading the signal, the resulting waveform data is composed of all these different frequencies, each of which corresponds to a particular location in space.

The Fourier transform is used to reconstruct the data into an image of a particular slice using the phase and frequency information contained in the signal.

2.2.3.2 Pulse Sequence Basics

Many different schemes have been devised for combining the localization gradients and rf excitation to create an image. Although there are many ways to accomplish the task, almost all MR image acquisitions will include some variation of the three steps of localization mentioned in the previous section. A basic representation of a pulse sequence is shown in Figure 2-4 below, and is in a similar format to those acquisition methods that are described in the remainder of this dissertation.
Figure 2-4. Basic 2D MRI pulse sequence showing the steps required for spatial localization.

Many of the sequence parameters used for acquisition are set directly by the operator, but some, such as gradient strength, are set by the MR system software based on the user’s input and hardware constraints. The type of MR acquisition that is performed, along with the parameters that are used, affect the resulting image characteristics, including image weighting and quality. The echo time (TE), the repetition time (TR), and flip angle of the rf pulse (α) factor in image weighting, or whether the acquisition results in an image that is PD, T₁, or T₂-weighted. Other parameters including field-of-view, number of frequency and phase encoding steps, and slice thickness, among many others, affect image
qualities such as spatial resolution, signal-to-noise (SNR), imaging time, and image quality in general. Imaging parameters have to be optimized for each unique acquisition method. This process is dependent on the anatomy that is being imaged, and is also a function of the information desired to be obtained in the imaging procedure.

2.3 Techniques Used in this Dissertation

2.3.1 Image Segmentation and Volumetry

Image segmentation is one of the most important steps leading to the analysis of processed image data. Its main goal is to divide an image into parts that have a strong correlation with objects of the real world contained in the image. In terms of MRI, segmentation is often used to separate anatomy from background noise, a particular tissue type from surrounding anatomy, or diseased from normal tissue. In the brain, segmentation is often used to distinguish brain parenchyma from surrounding skull, dura mater, skin, and fat tissue, as well as to break down the images into regions corresponding to grey matter (GM), white matter (WM), cerebral spinal fluid (CSF), and disease, when present.

Brain segmentation was used in many steps in the work that comprises this dissertation. In almost every project, segmentation was used in to separate the brain from background noise and surrounding tissue as the first step to image post-processing. More advanced image segmentation was a particularly important task in the early work that dealt with clinically isolated syndrome (CIS)
MS patients. Patients' brains were segmented into GM, WM, and CSF using fully automated programs; however, a higher-level of input had to be given in order to identify lesions in many cases and classify the lesion regions correctly. Also, CIS patient DTI scans were segmented based on anisotropy, so that regions of highly structured tissue (high anisotropy) could be studied without confounding information from less structured regions.

Structural Image Evaluation, using Normalization, of Atrophy adapted for cross-sectional measurement, or SIENAX, is a fully automated brain segmentation tool developed by the FMRIB Analysis Group at The University of Oxford. SIENAX performs segmentation of brain from non-brain tissue in the head and estimates the outer skull surface, with data from a single time-point. The brain and skull images are then registered to a standard space brain and skull image pair, which normalizes for skull size. Next a probabilistic brain mask derived in standard space is used to make sure that certain structures such as eyes/optic nerve have not been included in the brain segmentation. Finally tissue-type segmentation is carried out, including partial volume estimation, and a normalized brain volume estimate is calculated based on the skull registration. It was necessary to modify this program to account for lesions in this work dealing with MS patients. Due to lesion appearance in T1-weighted images, the input to SIENAX, this tissue is usually misclassified as GM. A modification step was added to the volume calculation process that took pre-identified lesions, kept them separate from both the GM and normal-appearing WM, and accounted for the volume appropriately.
in the final step.

The anisotropy segmentation used in the CIS patients was based on previous work in which the normal-appearing brain was segmented into three regions based on the diffusion anisotropy properties. Voxels corresponding to the 75th to 100th percentile of the anisotropy distribution were classified as high-anisotropy regions, those with a 50th to 75th percentile were classified as mid-anisotropy regions, and those voxels with a 0 to 50th percentile were classified as low-anisotropy regions. This step was modified in these projects to be specifically sensitive to changes in the WM, so the NAWM was isolated first, then broken down into the aforementioned three anisotropy-based regions.

2.3.2 $T_1$-Relaxation Measurements

Saturation recovery and inversion recovery pulse sequences that use multiple points in the relaxation curve to compute $T_1$ are the standard MR methods used to determine $T_1$-relaxation values. Although these techniques provide ideal measurements of $T_1$, they are not viable in most clinical studies that try to cover a large region of interest, yet need to do so in a reasonable imaging time.

Because the standard method is unpractical in many situations, considerable work has been focused on finding more clinically feasible methods to make $T_1$ measurements. Some of the strategies produced have shown that 3D images
collected at two flip angles and/or TRs can be used to make estimates of $T_1$.\textsuperscript{9-11} The signal intensity from a SPGR with $TE \ll T2^*$ and a flip angle $\alpha$ is given by

$$S = \frac{S_0(1 - f)\sin(\alpha)}{1 - \cos(\alpha) \cdot f} \quad \text{Equation 2-3}$$

where $f = \exp(-TR/T_1)$ and $S_0$ is a constant describing the imager gain and proton attenuation. Although this method gives an estimate of the relaxation constant and not the exact value, it has been shown to be robust and reproducible, thus valuable in researching clinical changes as long as it is the difference and not the exact value that is measured.\textsuperscript{12}

In this work, $T_1$ maps were calculated by substituting the ratio of signal intensities from SPGR's with two flip angles ($8^\circ$ and $40^\circ$), and solving Equation 2-3 for $T_1$. Histograms of the $T_1$ estimates were calculated in the WM of CIS patients and resulting parameters were compared to normal controls to investigate the sensitivity of $T_1$ to early stage disease activity.

2.3.3 Diffusion Tensor Imaging

2.3.3.1 Basic Principles of Diffusion

Molecular diffusion refers to the random motion of molecules, also called Brownian motion, which results from the thermal energy of the molecules. Diffusion-weighted imaging has been around since the 1980's when NMR imaging principles were combined with the principles introduced earlier to encode molecular diffusion effects in the NMR signal using bipolar gradient pulses.\textsuperscript{13} Diffusion MRI is a powerful tool because during their random walks, molecules
probe tissue structure at a microscopic scale far beyond the resolution achievable by typical MRI.\textsuperscript{14} The overall result is that a diffusion-weighted MR image reflects the displacement distribution of the water molecules which thus provides information on the structure and organization of the corresponding tissues.

2.3.3.2 Diffusion Tensor Imaging

Diffusion is a three-dimensional process, and since the diffusion of molecules is affected by interaction with tissue structures, the molecular mobility may be different in all directions. In the brain, the diffusivity of water is affected by the presence of semi-permeable membranes such as those of axons, by the particular arrangement of neurons in a given region, and by how densely packed the tissue is.\textsuperscript{15} In basic diffusion MRI, diffusion is described using a scalar parameter known as the diffusion coefficient (D), where the effect on the imaging signal is attenuation is given by

\[ A = e^{-bD} \quad \text{Equation 2-4} \]

where \( b \) is a constant that accounts for gradient characteristics such as timing, amplitude and shape.\textsuperscript{14} In the case of an anisotropic medium such as brain tissue, diffusion can no longer be characterized by a single coefficient, but requires a tensor (\( D \)) given as\textsuperscript{16}

\[ D = \begin{bmatrix} D_{xx} & D_{xy} & D_{xz} \\ D_{yx} & D_{yy} & D_{yz} \\ D_{zx} & D_{zy} & D_{zz} \end{bmatrix} \quad \text{Equation 2-5} \]

In order to solve for the elements of the diffusion tensor, images must be
collected along several gradient directions, using diffusion-sensitized pulse sequences such as echo-planar imaging (EPI). In addition to at least one non-diffusion weighted image, directional sampling is performed in at least six different directions; scanning additional directions will boost SNR, and provide additional information about the underlying tissue organization. The diffusion tensor can be determined from the set of equations given by Equation 2-6 using the set of diffusion-weighted images collected.\textsuperscript{14}

\[ A = \exp(-\sum_i \sum_j b_{ij}D_{ij}) \]  \hspace{1cm} \text{Equation 2-6}

After solving for the diffusion tensor, $D$, the eigenvalues of the diagonalized tensor may be determined and the mean diffusivity ($<D>$) and fractional anisotropy (FA) can be calculated as by Equation 2-7 and 2-8, respectively.

\[ <D> = \langle \lambda \rangle = \frac{\lambda_1 + \lambda_2 + \lambda_3}{3} \]  \hspace{1cm} \text{Equation 2-7}

\[ FA = \sqrt[3]{\frac{\left[ (\lambda_1 - \langle \lambda \rangle)^2 + (\lambda_2 - \langle \lambda \rangle)^2 + (\lambda_3 - \langle \lambda \rangle)^2 \right]}{2(\lambda_1^2 + \lambda_2^2 + \lambda_3^2)}} \]  \hspace{1cm} \text{Equation 2-8}

The mean diffusivity and fractional anisotropy, along with the calculated eigenvalues, can be used as surrogate markers for tissue microstructure, and have been shown to be useful in investigating an array of pathologies. In the projects that make up this work, DTI parameters are used to detect changes in the normal-appearing WM of patients with a clinically-isolated syndrome suggestive of MS.
2.3.3.3 Steady State Free Precession Diffusion Imaging

The effect of spin diffusion using spin-echo based sequences such as EPI has been studied extensively, and the exponential attenuation given by Equation 2-4 is widely accepted for diffusion imaging. Although these spin-echo sequences are the most widely used for diffusion-weighted MRI, steady state free precession (SSFP) sequences may also be made highly sensitive to diffusion by addition of a large unpaired gradient into the sequence. In the steady state, the measured signal is made up of magnetization tipped into the transverse plane by the most recent RF pulse, as well as a weighted combination of spin echo and multiple stimulated echo pathways from previous RF excitations. Although the diffusion weighting of a SSFP-based sequence is not as straightforward as its spin-echo counterpart, DW-SSFP has many advantages including excellent SNR, CNR, as well reduced gradient switching, RF power, and magnetic susceptibility.

In this dissertation, a 3D DW-SSFP sequence was implemented, optimized and tested for diffusion imaging at high field strengths. The initial results obtained on the 3.0 and 7.0 Tesla systems highlighted the strengths and some of the possible pitfalls of using this method of acquisition.

2.3.3.4 Fiber Tracking

The DTI parameters calculated from a set of diffusion images can also be used to determine connectivity in the brain. In each voxel, the axonal tracts can be
assumed to run in the direction of the primary eigenvector. Fiber tracking algorithms use each voxel's diffusion tensor to follow the path of neuronal fibers from voxel to voxel in the brain. There are several methods by which tracking can be done; this work used an algorithm based on FACT (Fiber Assessment by Continuous Tracking). This method starts tracking from user-defined voxels and following the direction of the primary eigenvector produces the tracts. When the fiber trajectory reaches the edge of the voxel, the direction is changed to match the next voxel's primary eigenvector.

The output of the fiber tracking software is anatomical connectivity, but the application for tracking in this project was to look at the DTI parameter values along the tracts that were created. An MS lesion was given as the starting point for tracking, in order to isolate tracts with MS-related injury. Diffusion parameters along these tracts were investigated to determine if they were indicative of Wallerian degeneration.

2.3.4 High Magnetic Field Strength Imaging

Magnetic resonance machines with a static magnetic field ($B_0$) of 1.5T have been available and served as the standard field strength in MRI since the mid 1980's. Although imagers with higher static magnetic field strengths were developed and explored uniquely for research purposes in the 1980's and 1990's, it was not until the beginning of this century that these high field systems have been applied for clinical imaging studies. Because of new developments
and pushing the limits of field strength, the term high field has undergone a transition in the past decade; in the 1990's high field typically meant 1.5T, but it now commonly refers to imaging at 3 Tesla and beyond. In this dissertation, high field strength is used to refer to work being carried out at 3 and 7 Tesla.

The most obvious advantage of moving to higher field strength is the increase in spin polarization, or in other words, greater signal available. Because the signal-to-noise ratio is proportional to voxel size, one use for the increase in signal is to obtain higher spatial resolution.\(^5\) Alternately, the increased SNR can be used to decrease the scan time;\(^5\) however, the practical implementation of this shortening depends on other factors such as RF power deposition, which is increased at high fields.

Higher magnetic field also affects many tissue and imaging parameters. For example, the T1 relaxation time elongates with an increase in field strength, but the T2 stays relatively constant and the T2* relaxation times are expected to shorten with increasing fields due to a rise in susceptibility-induced gradients.\(^{23-25}\) Other factors that have to be accounted for when implementing pulse sequences at higher fields include an increase in chemical shift and magnetic susceptibility effects, a rise in rf power deposition, and challenges in achieving B\(_1\) homogeneity.\(^{26-28}\)

The aforementioned differences and challenges will certainly be addressed with
improved MR methods specialized for high field MRI so as not to limit the technology; however, a practical limit on field strength especially at 7 Tesla and higher will likely be set by physiological effects in humans that result at high magnetic field. Several effects have been noted including dizziness, nausea, magnetophosphenes, headache, metallic taste, and ECG disturbances.\textsuperscript{30,31} Although these effects are transient and not known to have any lasting effects, there is a limit to subject willingness, and this will be a large factor in the transition of 7 T and higher MR systems from research to clinical applications.

High field imaging at 3.0 and 7.0T was used in this dissertation project as the catalyst for multiple technique developments and optimizations. The increase in signal was used to attain ultra-high-resolution anatomical images on volunteers and MS patients. Also, diffusion sequences were optimized and evaluated at high field, and applied for volunteer and MS patient studies.

\section{2.4 Multiple Sclerosis}

\subsection{2.4.1 Brief Overview of Disease}

Multiple sclerosis is a chronic inflammatory disease of the central nervous system characterized by inflammation, demyelination, axonal loss, and gliosis. The pathology of MS presents both as very specific regions of neuronal injury, and also as diffuse tissue abnormalities separate from obvious disease activity. The focal component of the disease results from acute attacks in which an inflammatory lesion develops, followed by destruction of myelin, and in severe
cases, transection of axons. When the lesion is no longer active, inflammation subsides and tissue repair begins; however, scar tissue seen as MS plaques remain. The diffuse portion of the disease occurs outside the lesions, and although the pathophysiology is not entirely known, it is suspected to have multiple causes including microscopic lesions and Wallerian degeneration initiated by inflammatory lesions. Over time, repetitive attacks followed by periods of recovery lead to a slow degradation of neurological function and an overall loss of brain parenchyma.

2.4.2 Application of MRI to MS

Magnetic resonance imaging techniques are used extensively in MS for clinical management and scientific research, providing valuable information about tissue structure and function. MRI is now the most widely used paraclinical diagnostic aid and longitudinal monitoring technology. MRI metrics provide primary outcome measures for phase 1 and 2 trials and supportive outcome measures for phase 3 trials of MS therapies. Conventional imaging techniques including T2-weighted, T1-weighted, and gadolinium-enhanced imaging are used routinely to identify lesions in the brain and spinal cord; however, these methods are not equally sensitive to lesions located in the white and grey matter, and mounting evidence suggests that these measures lack the specificity required for analyzing the underlying pathology and fail to capture clinically-relevant diffuse disease. Quantitative techniques such as MR spectroscopy, magnetization transfer, diffusion imaging, atrophy measurements, and T1 and T2 relaxometry have been
applied to identify abnormalities of both the white and grey matter in MS that go unseen in conventional imaging. In order to identify lesions and properly study both the white matter and grey matter, as well as differentiate between focal and diffuse disease, both traditional and advanced imaging methods need to be included in the imaging protocol.

2.4.2.1 Conventional MRI
MS plaques on conventional proton-density and T2-weighted brain images appear as bright areas, as shown on the left image in Figure 2-1, and are often referred to as T2-hyperintense lesions. These lesions in patients with MS are nonspecific for the underlying pathology, which may include inflammation, demyelination, gliosis, edema, Wallerian degeneration, and axonal loss, or a combination of these factors. Standard T2 acquisition methods are fairly insensitive to GM lesions; however, newer acquisition protocols such as fluid attenuated inversion recovery (FLAIR) and double inversion recovery (DIR) detect GM lesions with greater sensitivity. Brain T2 lesions show weak correlation with clinical disability, which may be due to the compensatory ability of brain tissue, the limitations of the clinical rating scale, the failure of T2-weighted images to characterize diffuse disease, and the presence of spinal cord disease. The strength of this technique lies in its ability to detect all varieties of lesions; however, its inability to explain underlying pathophysiology and resulting disability necessitates the use of other imaging methods.
Figure 2-5. T2 and T1 images taken at 1.5T from a single MS patient. Lesions appear hyperintense with T2 weighting, and as "black holes" with T1 weighting.

A subset of the lesions visualized on the T2-weighted images appears on T1-weighted images. These lesions show up as hypointense regions or "black holes", as depicted in Figure 2-1 on the right. Most T1-hypointense lesions initially present as gadolinium enhancing lesions, and have an approximately 50-50 chance of being transient (lasting 6-12 months) or permanent. \(^{38}\) Unlike their T2 counterparts, T1 lesions reflect underlying pathology. Those that show profound hypointensity and permanence primarily represent irreversible tissue destruction such as demyelination and axonal loss, whereas the transient ones reflect edema and demyelination with subsequent remyelination. \(^{39,40}\) Consequently, chronic persistent T1 lesions are associated with a moderate degree of global brain atrophy, and studies indicate a moderate to strong correlation between whole-brain T1-hypointense lesion load and neurological
impairment. T1-weighted imaging adds additional information to that taken from T2 sequences, but it still lacks specificity about underlying disease processes and diffuse disease.

Both T2 and Gd-enhancing lesions are used to aid the diagnosis of MS. Historically, criteria for diagnosis included two separate clinical attacks to indicate dissemination in space and time. Recently, the International Panel convened and changed the criteria so that now a characteristic change in MRI findings can prove dissemination in time and substitute for a second clinical attack. They also expanded the use of spinal cord lesions in the MRI criteria for dissemination in space. It is likely that future improvements in diagnostic criteria will include more MRI markers of disease as imaging technology develops and the disease processes and markers are better understood.

2.4.2.2 Advanced MRI Techniques

Anatomic imaging performed using conventional MRI methods has had great success in visualizing MS disease, and is used heavily in clinical practice, so much so, that the diagnostic criteria have been adjusted to factor in MRI findings. The benefits of these methods are undeniable, yet there exists a paradox whereby what is seen in the images do not reflect the clinical disability of patients in many cases. Advanced MR techniques reviewed here have helped elucidate the underlying disease processes and in many cases better reflect what is occurring clinically.
Proton MR spectroscopy studies have revealed that brain metabolite concentrations including N-acetylaspartate (NAA), choline (Cho), lactate, lipids, myoinositol (mI), and glutamate/glutamine are altered in MS patients.\textsuperscript{45-47} These spectroscopic findings are believed to reflect specific underlying pathologies. For example, decreased NAA is thought to be due to loss of axonal/neuronal integrity, increased Cho indicates membrane turnover such as occurs in demyelination and remyelination, increased lipid may be due to myelin breakdown or necrosis, and increased mI stems from gliosis. MR spectroscopy is a strong tool for studying MS disease processes not only due to its specificity, but also because it can be used to study diffuse disease in normal appearing tissue, has had success in detecting damage of the GM, and also it has been shown to have a stronger correlation with clinical measures of disability than do conventional MRI measures.\textsuperscript{48,49}

Magnetization transfer imaging (MTI) is another advanced technique that has been applied for MS research. Injuries that occur to CNS structures, like what happens in MS disease processes, cause a decrease in exchange of tissue mobile protons, which can be detected as a decrease in the magnetization transfer ratio (MTR). This decrease is believed to primarily reflect demyelination and axonal loss, but it is also probably affected to some extent by inflammation, gliosis, and edema.\textsuperscript{50} Like spectroscopy and other advanced techniques, the usefulness of MTI is that it can detect damage in overt lesions as well as in
normal appearing tissue.\textsuperscript{51-53} The disadvantage is that the source of the signal change is not well understood and may have many confounding factors, and its ability to detect changes in the grey matter is disputed.\textsuperscript{54,55} Regardless, MTI remains a tool for abnormality measurements and shows moderate to strong correlations with physical disability and cognitive dysfunction.\textsuperscript{56}

Diffusion weighted imaging is sensitive to changes in tissue microstructure such as those that occur as a result of the disease processes in MS. Many recent MS studies have used diffusion-derived parameters to better understand the underlying pathophysiological processes that occur in the brain and spinal cord. An increase in the apparent diffusion coefficient (\(<D>\)) and a decrease in the fractional anisotropy (FA) have been seen in focal white matter lesions; changes that correspond hypothetically to the inflammatory process and loss of axons and organization in the brain tissue.\textsuperscript{57-59} Similar diffusion changes have been seen in white matter that appears normal on anatomic images, which is reflective of the diffuse aspect of the disease.\textsuperscript{60-63} Diffusion and diffusion tensor imaging (DTI) have also been applied in studies of the grey matter, in which it has been able to measure an overall increase in \(<D>\); however, whether these changes are due to lesions that were undetected on anatomic images, microscopic lesions, or other processes is still misunderstood.\textsuperscript{64-66} Advances in diffusion imaging techniques have led to a growing interest in how these metrics correspond to clinical measures of MS including physical disability, disease progression, and cognitive function; however initial studies have shown conflicting results.\textsuperscript{67,68}
T1- and T2-relaxation times as measured by relaxometry have been shown to be sensitive, but not specific for underlying pathology in the brain and spinal cord of MS patients.\textsuperscript{69} Such measures can detect local abnormalities in areas of overt lesions, as well as in diffuse regions that appear normal on anatomical scans. The most common effect of MS in the white matter and grey matter on T1 relaxation is prolongation, which is believed to result from a variety of processes such as edema, demyelination, gliosis, and axonal loss.\textsuperscript{70} Many grey matter regions show hypointensity on T2-weighted images of patients, hypothetically due to an increase in iron deposition seen in MS.\textsuperscript{71} Both these relaxometry methods have been shown to have weak to moderate correlations with clinical disability.\textsuperscript{69,72}

Progressive brain atrophy is a well-recognized result of the MS disease processes, and can be measured through MRI segmentation in a variety of ways. Although the histological basis for deterioration is not fully understood, it is believed to be primarily due to demyelination, axonal loss, and neuronal loss; however, many other factors may confound brain volume measurements including medication effects, fluid status, and inflammation. Atrophy occurs in MS patients of all clinical subtypes, and appears to result from previous inflammatory demyelination-related damage in overt lesions.\textsuperscript{73} Brain atrophy includes loss of tissue in both white matter and grey matter regions, although recent work has shown that the grey matter may be disproportionately affected.\textsuperscript{74}
Brain atrophy has been shown to better correlate with clinical status than conventional MRI lesion measures.\textsuperscript{75}

This brief overview of MRI applications in MS has described many, but not all, of the techniques in clinician's toolbox to aid in diagnosis and monitor disease activity. The use of conventional MRI has far exceeded its original role as a diagnostic aid, and advanced techniques have significantly furthered the understanding of underlying pathophysiology, and help predict patient outcome and therapy effects. Although much work has been done, many holes remain, and the research done for this dissertation was aimed to address these deficits.

2.5 References


7. Smith SM, Zhang Y, Jenkinson M, Chen J, Matthews PM, Federico A, De Stefano N. Accurate, robust, and automate longitudinal and cross-sectional


51. Filippi M, Rocca MA. Magnetization transfer magnetic resonance imaging of the brain, spinal cord, and optic nerve. Neurotherapeutics (2007), 4:401-413.


53. Loevner LA, Grossman RI, Cohen JA, Lexa FJ, Kessler D, Kolson DL.


73. Jasperse B, Minneboo A, de Groot V, Kalkers NF, van Helden PE, Uitdehaag


3.1 Introduction

3.1.1 Application of MR Techniques to Multiple Sclerosis

Conventional MRI techniques such as T1 or T2 weighted images are commonly used in multiple sclerosis (MS) patients to identify white matter lesions, which indicate focal regions of tissue damage and neurodegeneration, and to measure T1 and T2 lesion loads.\textsuperscript{1-3} This information is useful in supporting MS diagnosis and monitoring disease activity and progression, however, it has not been shown to be strongly correlated to disease progression.\textsuperscript{4-7} The information available from standard imaging procedures is not specific enough to identify more diffuse markers of axonal and neuronal injury that appear in the white matter such as inflammation, edema, gliosis, demyelination and remyelination, processes which have a complex inter-relation, and may better reflect a patient’s current disease status. These pathological processes have been the focus of much attention recently and advanced MRI techniques have been applied to the detection and measurement of potential markers for damage in tissue that appears normal using conventional imaging.

One advanced MR method that has been used to look at microscopic changes in normal appearing white matter (NAWM) of MS patients is longitudinal (T1) relaxation estimates. Longitudinal relaxation changes may indicate complex
pathological changes that are not visible in standard imaging. The T1 relaxation times have been shown to be elevated in patients with MS, and correlate with clinical measures of disability.\textsuperscript{8-10} Diffusion tensor imaging (DTI) is a technique that can provide quantitative information about the magnitude and directionality of water diffusion in the brain. Several studies have shown diffusion tensor abnormalities in the NAWM of patients with MS.\textsuperscript{11-15} Previous work in patients with relapsing-remitting MS showed that, in areas of high anisotropy white matter, there is decreased fractional anisotropy due to an increase in transverse diffusion, which could be evidence of Wallerian degeneration.\textsuperscript{14} Animal studies have shown that axon and myelin degeneration can be differentiated using directional diffusivities derived from the diffusion tensor; diffusion parallel to fiber tracks may be used as a marker of axon degeneration, whereas transverse diffusion may indicate myelin degeneration.\textsuperscript{16,17}

\subsection{3.1.2 Project Goal in Patients with Clinically Isolated Syndromes}

In a large proportion of patients with multiple sclerosis (MS), the first presentation of disease comes as a clinically isolated syndrome (CIS) suggestive of MS. Although not all will go on to a diagnosis of MS, many CIS patients may be considered to be at the earliest stages of MS. Studies have shown that atrophy and diffusion abnormalities are apparent in the early stages of disease, however, the point at which the changes are first detectable and their complex relationships are less understood.\textsuperscript{18-21} By studying a group of patients at an average of four months following their first optic neuritis, myelitis, or brain stem
presentation, we hope to assess the earliest manifestation of these brain changes. The aims of this study were two-fold. The first was to investigate whether there are detectable changes in normalized brain volume measurements, longitudinal relaxation estimates of the normal appearing white matter (NAWM), or diffusion tensor metrics of the NAWM in patients presenting with a CIS. The second aim was to determine if relationships exist between the three different measurements, which may help clarify the pathological processes present at the earliest stages of disease.

3.2 Materials and Methods

3.2.1 Patients
Patients presenting with a clinical attack indicative of MS were scanned at, on average, four months (range, 0.3-8.7 months) following optic neuritis, myelitis, or brain stem syndromes. The patient group was made up of 25 patients (19 females and 6 males; mean age 36.6 years; range, 21.6-56.7 years) enrolled in an ongoing study on CIS. Fourteen healthy volunteers with no history of neurological dysfunction served as controls (9 females and 5 males, mean age 36.2 years, range, 24.1 – 46.5 years). The UCSF Committee on Human Research approved the study, and all subjects provided informed written consent.
3.2.2 Image Acquisition

All imaging was performed on a 1.5 T General Electric Medical Systems Signa scanner (General Electric, Milwaukee, WI) with 4 G/cm gradients and a standard quadrature head coil. The imaging protocol included two axial T1-weighted 3D spoiled gradient recalled echo (SPGR) image volumes with different flip angles (40 and 8 degrees), with all other imaging parameters kept identical between the two acquisitions (27/6 ms TR/TE; field of view, 180 x 240 x 186 mm<sup>3</sup>; matrix, 192 x 256 x 124). The imaging examination also included a single shot, multi-repetition echo planar DTI sequence (1.7 x 1.7 x 2.1 mm; TR/TE = 7 s/100 ms; nine averages, b value = 2000 s/mm<sup>2</sup>). Axial T2-weighted spin-echo images (TE/TR = 20/2500 ms, 192x256 matrix, 180x240 mm FOV, 3 mm thick slices, 50 slices) were also acquired to aid in lesion identification.

3.2.3 Image Processing

3.2.3.1 Calculation of Maps

T1 relaxation maps (Figure 3-1b) were calculated using methods described previously<sup>22</sup> based on the ratio of signal intensities from the two T1-weighted SPGR images acquired with different flip angles.
Figure 3-1. Example of T1-weighted imaging and derivatives from a CIS patient. (a) T1 weighted flip 40 3D-SPGR image, (b) calculated T1-estimate map, (c) grey matter mask, and (d) normal-appearing white matter mask output by SIENAX.

The eigenvalues of the diffusion tensor were calculated on a voxel by voxel basis using software developed in our lab. From the eigenvalues, maps were calculated for the mean diffusivity ($D_{av}$), fractional anisotropy (FA), and the
average of the minor eigenvalues ($\lambda_{23} = \lambda_2 + \lambda_3 / 2$). DTI data quality is demonstrated in Figure 3-2.

Figure 3-2. Calculated DTI maps. (a) $\lambda_1$, (b) $\lambda_{23}$, (c) $D_{av}$, and (d) FA.
3.2.3.2 Lesion Identification

For each patient, lesions were manually identified and outlined on the T1-weighted, 40 degree flip SPGR images. Criteria for lesions included lower signal intensity as compared to surrounding normal appearing white matter, a corresponding hyperintense region on T2-weighted images, and area of the hypointense region being greater than 3 mm².

3.2.3.3 Tissue Segmentation & Volume Calculations

Brain segmentation and normalization was performed using Structural Image Evaluation, using Normalization, of Atrophy, single-time-point estimation (SIENAX), a fully automated program developed by the FMRIB Image Analysis Group, Oxford University, United Kingdom, and was downloaded from the site http://www.fmrib.ox.ac.uk/fsl. A complete description of the SIENAX process may be found elsewhere, so only the major steps to brain segmentation and normalization are given here.²³

The first step was done using the brain extraction tool (BET) for which the outputs were the extracted brain and skull images. Next, brain and skull images were registered to the MNI152 brain and skull templates. This constrained registration of the skull image to the MNI152 skull results in a scaling factor that was used for normalization of different brain sizes between subjects. Brain tissue segmentation was performed using FMRIB’s Automated Segmentation Tool (FAST), which segments the brain images into GM, WM, and CSF partial
volume estimate (pve) maps based on a hidden Markov field model. Lesion masks made from the manually drawn ROI’s were used to remove lesions from each of the resulting pve maps. Total NAWM, GM, and CSF volumes were calculated from the pve maps by summing over the specific pve map, multiplying by the voxel volume, and then scaling the volume by the factor determined in the skull normalization to MNI space. The normalized WM volume was calculated as the normalized NAWM volume plus the normalized lesion volume (the lesion volume multiplied by the skull scaling factor). FAST also outputs binary tissue masks based on a thresholding of the pve maps. The GM and WM masks output can be seen in Figure 3-1c and 3-1d.

3.2.3.4 DTI Segmentation
The 3D-SPGR images were aligned to the DTI T2-weighted (b = 0 s/mm²) image using a nonlinear registration program AIR. The resulting alignment parameters were then used to register the NAWM mask to the DTI dataset. The NAWM of the DTI data was further segmented into high, medium, and low-anisotropy NAWM regions using a method based on histogram equalization. The resulting segmentation can be seen below in Figure 3-3. The NAWM mask was overlaid on the fractional anisotropy (FA) map, and the result was histogram equalized. The voxels greater than 75th percentile in the histogram-equalized FA NAWM were kept and constituted the high-anisotropy (HA-NAWM) region. The voxels falling between the 50th and 75th percentiles were considered medium, and below the median considered low anisotropy regions.
3.2.4 Histogram Analysis

Histograms were generated for the NAWM of the T1-estimate maps as well as $D_{av}$, FA, and eigenvalues of the DTI data. Also, for the DTI data, histogram analysis was done on the HA-NAWM regions. The histogram distributions were produced for each individual parameter and compared to controls.

3.2.5 Statistical Analyses

The effect of age on all parameters was removed by calculating age-predicted z-scores. For each parameter, the control data was plotted against age, and a regression line was determined. The regression line gives the predicted value of the parameter at a given age. Then the z-score is calculated as the difference between the measured value and the predicted value, divided by the standard deviation of the control data. All further statistical analyses were performed on
the patient’s calculated z-scores. Group comparisons were made using the two-tailed student’s t-test. To look for relationships, Spearman’s rank sum correlations were calculated for the NAWM T1, NAWM DTI, and HA-NAWM DTI medians versus the normalized tissue volumes. Significance was defined as p<0.05.

3.3 Results

3.3.1 Volume Measurements

Figure 3-4 shows the patient and control z-score data for the normalized volume measures output by SIENAX followed by age correction. A z-score of zero represents the predicted normalized volume with respect to that individual’s age. The control subjects show normal variance around the expected value, whereas the patient group shows a large overlap with normal values, and a few patients exhibited volumes that were more than two standard deviations below the normal age-corrected value. As a group, the CIS patients do not show significantly different nWM (p=0.21), nGM (p=0.33), or nBV (p=0.07) compared to the control group, however, the patient group did tend to have smaller volumes in each of the cases (average z-score = -0.45, -0.37, and -0.94, respectively).
Figure 3-4. Age-corrected z-score plots of the normalized white matter, grey matter, and brain volumes for each of the individual subjects. The dotted lines encompass data points that lie within two standard deviations of the expected values.

3.3.2 T1-Relaxation Time Estimates

The results of the NAWM T1-relaxation time estimate analysis are graphically illustrated in Figure 3-5. The histogram for the CIS group (dashed line) is shifted to the right compared to controls, indicating an increase in the longitudinal relaxation time. The T1-estimate histogram parameters in the NAWM of CIS patients have significantly elevated 25th (p=0.009), 50th (p=0.01), and 75th percentiles (p=0.017), as well as a higher peak position (p=0.005), and mean (p=0.015) compared to control subjects. The peak height (p=0.21) and standard deviation (p=0.83) of the distributions were not significantly different. The plot of the individual z-scores based on histogram medians in Figure 4-5b demonstrates that the histogram medians for all the normal control subjects lie within two
standard deviations of the expected value, whereas six out of 25 patients had medians that were greater than two standard deviations above the predicted value.

**Figure 3-5.** T1-estimate analysis a) Normalized histograms for the NAWM of controls and CIS patients. b) Age-corrected z-score plot for the median T1.

### 3.3.3 DTI

Figure 3-6 shows the histogram analysis and normalized z-score plots for the NAWM and HA-NAWM regions of the $\lambda_1$ and $\lambda_{23}$ maps. The top row in Figure 3-6 (a-b) shows the normalized histograms for the NAWM. In this region, DTI parameter differences are small, and of the eigenvalues, only the 25$^{th}$ percentile of $\lambda_{23}$ was statistically elevated compared to controls ($p<0.05$). The 25$^{th}$ percentile for $D_{av}$ was also significantly elevated ($p<0.05$), an effect presumably driven by the increased $\lambda_{23}$. Breaking the NAWM down further into a region based on high anisotropy was able to distinguish other differences and help elucidate from where the NAWM changes stemmed. The normalized histograms from the HA-NAWM regions are depicted in the middle row of Figure 3-6 (c-d). In
the HA-NAWM none of the histogram parameters for $\lambda_1$ were different from controls. In contrast, the histogram parameters ($25^{th}$, $50^{th}$, $75^{th}$, peak position, mean) for $\lambda_{23}$ were all statistically elevated compared to controls ($p<0.02$). Additionally, because the change in $\lambda_1$ was small, the changes seen in the $\lambda_{23}$ of the HA-NAWM were primarily responsible for increased $D_{av}$ ($25^{th}$, $50^{th}$, $75^{th}$, peak position, mean; $p<0.05$). The age-corrected z-scores for the patient and controls are also shown in the last row of Figure 3-6 (e-f).
Figure 3-6. DTI eigenvalue analysis a) Normalized histogram for NAWM $\lambda_1$, b) Normalized histogram for NAWM $\lambda_{23}$, c) Normalized histogram for HA-NAWM $\lambda_1$, d) Normalized histogram for HA-NAWM $\lambda_{23}$, e) Age-corrected z-score plot for median $\lambda_1$ in the NAWM and HA-NAWM, f) Age-corrected z-score plot for median $\lambda_{23}$ in the NAWM and HA-NAWM.
3.3.4 Correlations

Also of interest are the relationships that exist between the different measures. The Spearman rank correlation coefficients for each of these correlations are listed in Table 3-1. The T1-estimate of the NAWM was not significantly correlated with the nGM, nWM, or nBV for either the CIS patients or control subjects. The DTI parameters did exhibit some significant correlations with the volume measures. The $\lambda_1$ of the NAWM is correlated to nGM volume in controls. In the patient group, the $\lambda_1$ of the HA-NAWM were correlated with nGM and nBV, however not in the NAWM, as was the case in the control subjects. The NAWM and HA-NAWM $\lambda_{23}$ of the CIS group is inversely correlated with nGM volume, which means with an increase in $\lambda_{23}$, nGM volume is lower. The relationship between $\lambda_{23}$ and nGM was not significant in controls. The ADC did not show significant correlation with any of the volume measures in either patients or controls. In the CIS patients NAWM and HA-NAWM FA correlated with the nGM, and also NAWM FA correlated with nBV. The control data did not show any significant correlations between FA and brain volume measures.
<table>
<thead>
<tr>
<th></th>
<th>nWMV</th>
<th></th>
<th>nGMV</th>
<th></th>
<th>nBV</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CIS</td>
<td>CTRL</td>
<td>CIS</td>
<td>CTRL</td>
<td>CIS</td>
<td>CTRL</td>
</tr>
<tr>
<td>T1</td>
<td>0.13</td>
<td>-0.45</td>
<td>-0.35</td>
<td>0.32</td>
<td>-0.13</td>
<td>-0.12</td>
</tr>
<tr>
<td>$\lambda_1$</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NAWM</td>
<td>-0.05</td>
<td>-0.53</td>
<td>0.12</td>
<td>0.60</td>
<td>0.12</td>
<td>0.04</td>
</tr>
<tr>
<td>HA-NAWM</td>
<td>0.18</td>
<td>-0.45</td>
<td>0.51</td>
<td>0.44</td>
<td>0.57</td>
<td>-0.07</td>
</tr>
<tr>
<td>$\lambda_{23}$</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NAWM</td>
<td>0.18</td>
<td>0.07</td>
<td>-0.48</td>
<td>-0.02</td>
<td>-0.4</td>
<td>0.17</td>
</tr>
<tr>
<td>HA-NAWM</td>
<td>-0.14</td>
<td>-0.16</td>
<td>-0.49</td>
<td>0.04</td>
<td>-0.35</td>
<td>-0.08</td>
</tr>
<tr>
<td>$D_{av}$</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NAWM</td>
<td>-0.04</td>
<td>-0.23</td>
<td>-0.37</td>
<td>0.21</td>
<td>-0.20</td>
<td>0.05</td>
</tr>
<tr>
<td>HA-NAWM</td>
<td>-0.05</td>
<td>-0.27</td>
<td>-0.07</td>
<td>0.27</td>
<td>0.01</td>
<td>-0.04</td>
</tr>
<tr>
<td>FA</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NAWM</td>
<td>0.29</td>
<td>-0.49</td>
<td>0.59</td>
<td>0.30</td>
<td>0.62</td>
<td>-0.36</td>
</tr>
<tr>
<td>HA-NAWM</td>
<td>0.18</td>
<td>-0.39</td>
<td>0.70</td>
<td>0.52</td>
<td>-0.40</td>
<td>0.01</td>
</tr>
</tbody>
</table>

Table 3-1. Spearman rank correlations between quantitative imaging parameters and normalized brain measurements. Significant relationships (p<0.05) are shown in bold.

The Spearman’s rank correlation coefficients for the DTI parameters versus the T1-estimates are listed in Table 3-2. In controls, there were no significant relationships between the measured T1-estimate and the DTI parameters. In the CIS patients the $\lambda_1$ of the NAWM significantly correlated with T1 relaxation time estimate, and also the ADC of the HA-NAWM significantly correlated with T1 relaxation time estimate.
### Table 3-2. Spearman rank correlations between T1 and DTI measurements. Significant correlations of p<0.05 are shown in bold.

<table>
<thead>
<tr>
<th></th>
<th>T1-Relaxation</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CIS</td>
<td>CTRL</td>
<td></td>
</tr>
<tr>
<td>$\lambda_1$</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NAWM</td>
<td>0.44</td>
<td>-0.03</td>
<td></td>
</tr>
<tr>
<td>HA-NAWM</td>
<td>0.26</td>
<td>-0.08</td>
<td></td>
</tr>
<tr>
<td>$\lambda_{23}$</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NAWM</td>
<td>0.28</td>
<td>-0.12</td>
<td></td>
</tr>
<tr>
<td>HA-NAWM</td>
<td>0.29</td>
<td>-0.05</td>
<td></td>
</tr>
<tr>
<td>$D_{av}$</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NAWM</td>
<td>0.38</td>
<td>-0.02</td>
<td></td>
</tr>
<tr>
<td>HA-NAWM</td>
<td>0.44</td>
<td>-0.08</td>
<td></td>
</tr>
<tr>
<td>FA</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NAWM</td>
<td>-0.17</td>
<td>-0.01</td>
<td></td>
</tr>
<tr>
<td>HA-NAWM</td>
<td>-0.25</td>
<td>-0.02</td>
<td></td>
</tr>
</tbody>
</table>

3.4 Discussion

3.4.1 Earliest Detectable Measurement of Atrophy in CIS

There are many MRI-detecatable indicators of brain abnormality at this earliest stage of disease. Although the volumes were not statistically different as a group, looking at individuals on the z-score plot of Figure 3-4 shows differences beginning to emerge. This plot and the other parameters' z-score plots brings to light one of the most difficult issues when dealing with a CIS cohort, and that is the great amount of patient heterogeneity. These patients are all at the same stage of diagnosis, one clinically presented attack, but it does not mean that they have all had similar duration of disease. Some patients could have been having attacks for years without noticing, whereas another may have presented clinically on the first attack. The possible difference in disease duration may explain why
some patients had a lot of atrophy at this stage, whereas others did not, and thus may make detection of group differences difficult.

3.4.2 T1 as a Marker of Disease Processes

The T1 of the NAWM in CIS patients was found to be significantly higher than in controls. This agrees with previous findings of elevated T1 in clinically definite MS (CDMS). However, this finding gives additional information to show that this parameter is indicative of disease processes very early-on, perhaps even before being able to be diagnosed by current criteria. The increase in longitudinal relaxation may be indicative of inflammation of the WM away from the site of acute lesions. It could also be due to microscopic lesions that are too small to be detected at the spatial resolution that was used in this study, or the increase could stem from other unknown underlying pathological processes. In the future, it may be interesting and informative to monitor this parameter through time to see if it is related to new lesion occurrence, or if it decreases as lesions heal and inflammation subsides.

3.4.3 DTI as a Marker of Disease Processes

The DTI analyses resulted in a difference detected in the high-anisotropy regions of the white matter, with the changes primarily linked to an increase in $\lambda_{23}$. One of the challenges in using this methodology is that it is difficult to determine whether this change is only occurring in high-anisotropy regions, or if this method is just not sensitive to changes in the lower anisotropy regions. This could be
overcome if it is determined what underlying disease mechanism is responsible for this increase. According to animal data, this parameter increases with breakdown of myelin that occurs after neuron injury. So, this increase could indicate Wallerian degeneration occurring away from the lesion site. Future work may involve looking at specific pathways transected by lesions using DTI tractography, and determining whether the increase in $\lambda_{23}$ is specifically in those, or all over the HA-NAWM, which could come back to inflammatory activity.

3.4.4 Interpretation of Parameter Correlations

The correlations were derived to see if any relationships could be determined to exist between MRI-detectable parameters, and if this information could elucidate what underlying processes these parameters represent. The relationship found between longitudinal and transverse diffusion in the normal-appearing white matter and the nGM volume in CIS is a very interesting finding. Other studies have shown GM differences at the earliest stages of disease.$^{19}$ This finding builds on the previous findings, and suggests that neuronal degeneration occurring from damage caused by lesions may result in neuron cell body death and thus be responsible for GM atrophy.

It was also important to determine if these values were perhaps two indicators of the same underlying cause. Analysis of correlations between T1 and DTI parameters in controls showed no relationship, whereas there was some correlation between longitudinal diffusion and T1-relaxation in patients. While
these two parameters may reflect completely different processes, it is also possible that there is some commonality. This suggests that there may be a complex relationship, and interpretation of results should consider this interaction.

3.5 Conclusion

The quantitative imaging methods used in this study show that brain changes can be detected as early as the first clinically observed episode of disease resembling MS. In addition to conventional imaging methods that are used for diagnosis and disease monitoring, these metrics may prove to be useful diagnostically or as predictors of disease prognosis.

Although slightly decreased, the brain volumes measured in this work were not statistically different from the control group, indicating that atrophy may be slow to accrue with time. This finding is promising in that if disease is caught early and patients are put on disease-modifying therapy, the effects of neurodegeneration may be mitigated. The NAWM of the patient group did demonstrate differences in T1 relaxation and various diffusion parameters. These differences could be markers of neurodegeneration away from lesions, or may be transient markers for inflammatory processes that occur in MS. A better understanding of these changes and their pathophysiological meaning may be better determined from studying at these parameters through time.
3.6 References


Chapter 4: Longitudinal Brain Changes in Patients with Clinically Isolated Syndromes (CIS) Suggestive of MS

4.1 Introduction

4.1.1 MR Indicators of Disease in Clinically Isolated Syndromes

Previous investigations have clearly identified many MRI-detectable brain alterations in patients with multiple sclerosis (MS) including volumetric, T1 and T2 relaxation time, diffusion, spectroscopic, and magnetization transfer changes.\(^1\) Some of these abnormal metrics are present in patients even as early as the first clinical presentation of a clinically isolated syndrome (CIS) suggestive of MS.\(^2-4\) The etiology of these observed differences is multi-factorial, and probably includes many different disease processes including inflammation leading to oligodendrocyte injury, demyelination, axonal compromise with resultant Wallerian degeneration, and gliosis.\(^5\) Although many studies have shown brain abnormalities as a result of disease at a specific timepoint, the temporal profiles of these changes as well as the underlying cause of the changes remains unclear.

4.1.2 Goal of Longitudinal Study

This study was a longitudinal, case-controlled study of the normalized white matter (nWM) volume, normalized grey matter (nGM) volume, and normalized brain parenchymal volume (nBV) as well as the T1-relaxation time and diffusion tensor metrics of the normal appearing white matter (NAWM) in patients with a CIS. This study builds on the prior cross-sectional investigation of CIS patients.
that showed differences in volumes, T1, and diffusion measures at the earliest presentation of disease. The goal of this study was to investigate how MR derived parameters change over time in patients with probable early stage MS to better understand the meaning of these changes in respect to disease processes.

4.2 Materials and Methods

4.2.1 Research Participants

Research participants evaluated for a clinically isolated syndrome suggestive of multiple sclerosis at the University of California, San Francisco Multiple Sclerosis Center were invited to participate in this study and followed for one year. Clinical assessments including the Kurtzke Expanded Disability Status Scale (EDSS) and brain MRI scans were performed upon entry into the study and at the 12 month follow-up for all study participants. CIS was defined as the first-isolated, well-defined neurological event lasting ≥48 hours, and involving the optic nerve, spinal cord, brainstem, or cerebellum. Participants with neurological symptoms and asymptomatic neurological abnormalities with EDSS scores from 0-5.0 were eligible for this study. All MRI analyses were performed without knowledge of treatment assignment. This research investigation was performed with the approval from the University of California, San Francisco Committee on Human Research. Written informed consent was obtained from all study participants.
4.2.2 Image Acquisition

All brain MR images were acquired using a 1.5 Tesla GE scanner (General Electric Medical Systems, Milwaukee, WI) with 4 G/cm gradients and a quadrature head coil. Each MR imaging examination included two axial T1-weighted three-dimensional spoiled gradient echo (3D SPGR) imaging sequences with different flip angles and all other imaging parameters identical (TE/TR = 6/27 ms, flip angles = 8° and 40°, 192x256x124 matrix, 180x240x186 mm FOV). The imaging examination also included a single shot, multi-repetition echo planar DTI sequence (1.7x1.7x2.1 mm; TR/TE = 7 s/100 ms; nine averages, b value = 2000 s/mm²). Brain MR images were acquired within two weeks of initial symptom onset.

4.2.3 Image Post-Processing

4.2.3.1 Lesion Identification

Lesions were identified on the baseline T1-weighted images, and ROIs were manually drawn using in-house software. Lesions were identified on the follow-up exam by editing the ROIs from the baseline scan. The baseline and one-year follow-up exams were registered to one another in order to make same-subject longitudinal comparisons. The T1-weighted image from the one-year follow-up exam was aligned to the baseline image using a 12-parameter affine image transformation (FLIRT, Image Analysis Group, Oxford, UK). The original ROIs were opened on the aligned image from the one-year follow-up exam, and edited
to add new lesions or change the size and/or shape of the old lesions. Lesion ROIs were saved as mask images and transformed back to the original image space for each follow-up examination.

4.2.3.2 MRI-Derived Brain Tissue Parameter Calculation

Brain segmentation, normalization, and volume calculations were performed using SIENAX (Image Analysis Group, Oxford, UK). Lesion masks were created as described, and used to correct for tissue misclassifications by the SIENAX program. The lesion mask over-rode all SIENAX tissue classifications. Normalized tissue volumes were calculated by summing the lesion corrected partial volume estimate maps, and then multiplying the total sum by the brain-scaling factor calculated by the SIENAX program.

T1 relaxation maps at baseline and the follow-up exam were calculated based on the ratio of signal intensities from the two T1-weighted SPGR images at different flip angles using the method described previously.

The eigenvalues of the diffusion tensor were calculated on a voxel by voxel basis using software developed in our lab. From the eigenvalues, maps were calculated for the mean diffusivity \( (D_{av}) \), fractional anisotropy \( (FA) \), and the average of the minor eigenvalues \( \lambda_{23} = \lambda_2 + \lambda_3 / 2 \).
DTI difference maps were produced by subtracting parameters at the one-year follow-up from their baseline value. In order to accurately perform longitudinal comparisons, the images from the two time-points needed first to be reconstructed in the same image space. The baseline b=0 T2-weighted echo planar images were aligned to the MNI152 brain template (Image Analysis Group, Oxford, UK) using a nonlinear 30-parameter image transformation from the Automated Image Registration (AIR) library. The resulting image was used as the alignment target for the follow-up scan. The transformation matrices were used on the DTI maps from each time point, and then the co-registered maps were subtracted.

4.2.3.3 Fiber Tracking

Fiber-tracking was performed on the baseline scan using IDL software (IDL, Research Systems, Boulder, CO) developed in-house, which was based on the fiber assignment by continuous tracking (FACT) method. Fiber tracts that were likely to be affected by MS disease pathology were selected as the starting region for tracking. The mean diffusivity, fractional anisotropy, primary eigenvalue, and the mean of the two minor eigenvalues were measured from voxels through which the tracts passed. A graphical interface allowed visualization of the DTI fiber tracts as well as the corresponding DTI parameter values of the tracts, overlaid on the b=0 echo planar image. In order to measure diffusion parameters of the tract through time, the b=0 T2-weighted images from the follow-up exams were aligned to the baseline image using a nonlinear 30-
parameter image transformation from the AIR library. The transformation matrix was inverted and applied to the tract mask so that it could then be opened in the image space of the follow-up exams and the DTI parameters measured.

4.2.4 Statistical Methods

Histograms were generated for the normal-appearing white matter (NAWM) of the T1-estimate maps as well as $D_{av}$, FA, and eigenvalues of the DTI data. Also, for the DTI data, histogram analysis was done on the high-anisotropy normal appearing white matter (HA-NAWM) regions.

Age-corrected z-scores were calculated for all volume, T1, and DTI parameters to remove any effects of age. All further statistical analyses were preformed based on the patient and control groups' calculated z-scores. Group comparisons were made using the two-tailed student's t-test. To investigate parameter inter-relationships, Spearman’s rank sum correlations were calculated for the NAWM T1 and HA-NAWM DTI medians at baseline versus the change in the normalized tissue volumes over the year of the study. A p-value less than or equal to 0.05 was considered statistically significant.

4.3 Results

4.3.1 Clinical Data

Twenty-one CIS patients were studied over the course of one year. Fourteen control subjects were also scanned for comparison. The participant data can be
found in Table 4-1 below. Of the 21 CIS patients, 9 received disease modifying therapies which included low dose Interferon B-1a [n=4], high dose interferon B-1a [n=1], interferon B-1b [n=1], and glatiramer acetate [n=3]. All patients were free from high-dose steroid therapy during the study period.

<table>
<thead>
<tr>
<th></th>
<th>CIS</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>n (F:M)</td>
<td>21 (15:6)</td>
<td>14 (9:5)</td>
</tr>
<tr>
<td>Average Age [range]</td>
<td>35.7 [21.6 – 51]</td>
<td>36.0 [24.1 – 46.5]</td>
</tr>
<tr>
<td>Disease Duration at Baseline [range]</td>
<td>0.39 years</td>
<td>NA</td>
</tr>
<tr>
<td>EDSS at Baseline</td>
<td>1.26 ± 0.94</td>
<td>NA</td>
</tr>
<tr>
<td>EDSS at 1-Year</td>
<td>1.10 ± 0.90</td>
<td>NA</td>
</tr>
</tbody>
</table>

Table 4-1. CIS patient and control population clinical information.

There were no statistically significant differences between the CIS cohort and their control group in terms of age or proportion of gender. Also, no statistically significant differences were detected between the baseline EDSS and the EDSS measured at the one-year follow-up.

4.3.2 MRI Metrics Measured Over One Year

4.3.2.1 Brain Volume Measurements

Although the patient z-scores were lower than controls at baseline, there were no significant differences between groups. At the end of 12 months, the volume measures remained depressed compared to healthy volunteers; however, at this point the brain volume had decreased to a level significantly different from controls. A graph showing average patient z-scores for the brain volume measures can be seen in Figure 4-1.
Figure 4-1. Mean CIS patient brain volume z-scores for nWM, nGM, and nBV measurements at baseline and the 1-year follow-up exam. The asterisks indicate significant different between the patient group and controls.

4.3.2.2 T1-Relaxation Estimates

One year following the baseline exam, NAWM T1 was still elevated compared to the controls; however, the histogram median shifted back towards normal, and was no longer significantly different from the control values. The graphical representation of NAWM T1-estimate z-scores is displayed in Figure 4-2.
4.3.2.3 DTI Parameters

At the one-year follow-up exam, the transverse diffusion of the HA-NAWM was even more elevated than it was at baseline. In contrast, the parallel diffusion as indicated by Ev1 was slightly elevated, but remained an inaccurate marker for any change in CIS patients compared to controls. The $D_{av}$ was elevated, and the FA was even more decreased after one year than at the baseline scan, and at this point was significantly different from the control group. Figure 4-3 shows the z-scores for the DTI data at baseline and one year later.
4.3.3 Baseline Measures as a Predictor of Volume Change

Spearman rank correlations were calculated for T1-estimates and DTI parameters versus brain volume measurement changes. The resulting correlation coefficients can be seen in Table 4-2. The transverse diffusion at baseline correlated strongly (p<0.02) with the change in white matter volume over one year. Also, the baseline $D_{av}$ correlated with the change in white matter volume as well as the change in overall normalized brain volume.

Figure 4-3. Mean z-scores for the DTI parameters of the HA-NAWM CIS patients at baseline and the 1-year follow-up exam. The (*) indicates significance at p < 0.05, whereas the (**) indicates p < 0.005.
Table 4-2. Spearman rank correlation coefficients for baseline T1 and DTI values versus change in brain volume measurements over one year. The (*) indicates a significant correlation with \( p < 0.05 \), (***) indicates \( p < 0.02 \).

The EvT and \( D_{av} \) correlations with volume change over a year can be seen in Figures 4-4 and 4-5, respectively. In each case, the negative correlation indicates that the larger DTI parameter value (EvT or \( D_{av} \)) at baseline corresponds to a more negative change in volume, or in other words, loss of tissue.
4.3.4 Fiber Tract Specific Changes

The tissue parameters discussed in the previous sections indicated that changes occurred in the brain parenchyma of CIS patients; however, whether these alterations are diffuse or more localized to specific regions of tissue injury is yet to be determined. Several methods to identify more local changes were examined. An example of the difference maps can be seen in Figure 4-6. In this example, change is occurring very locally in the tissues surrounding the ventricles, a region where this patient had at least one lesion, and tracks running through that lesion are likely to be located. These periventricular changes
contrast with the difference map for the control in Figure 4-6(D) where the only
difference registered were the differences around the skull.

Figure 4-6. Top row corresponds to images from a CIS patient, (A) is the T2-
weighted image from baseline, and (B) is the difference map for ev2 over 1-year.
The second row (C) and (D) are the T2 and ev2 counterparts for a healthy
volunteer.
A second method that was used to look at localized changes is demonstrated in Figure 4-7. Fiber tracking was used to determine a pathway that was transected by a lesion, and diffusion parameters along that specific tract were investigated over time. Both views demonstrate a decrease in the amount of white (very high FA) in the corpus callosum over time. Also of interest is the region indicated with the green arrow, which shows decreasing anisotropy over time in a region distal to the lesion (circled in green in the top row).

Figure 4-7. Fractional anisotropy of a fiber tract followed at three-month intervals from baseline scan to one year follow-up. The green circle indicates lesion location, green arrow highlights changes occurring distal to the lesion location. White indicates very high anisotropy scaling through yellow, orange, red, and then to black for very low anisotropy.
4.4 Discussion

4.4.1 Interpretation of Longitudinal Metrics

Volumetric changes have been described in all subtypes of MS.\textsuperscript{11-14} However; many previous studies have failed to consider the influence of normal, age-related changes, causing these results and reported values to be misleading. The age-corrected volumetric results in this study show that there was a decrease in brain volume over the time frame of a single year in the group of CIS patients, and that the decrease was driven by a loss of GM. The WM volume in this patient population as a whole did not decrease in a year, but it remained less than in the group of healthy control subjects. The detection of WM loss as a group may be difficult due to many factors influencing the WM volume measurements including inflammation, hydration status, and medication effects. Controlling for these factors may improve sensitivity of this method to detect changes within a patient, as well as the group, when making longitudinal comparisons.

The T1 relaxation time is another MRI-derived metric that has been shown in previous studies to be elevated not only in lesions, but also in the NAWM of patients with all subtypes of MS.\textsuperscript{15} This work showed that the T1 is elevated in the CIS patient group compared to controls one year following initial presentation, but that it has returned closer to normal values and is not significantly different, as it was at the baseline scan. One hypothesis for this return to more normal values is that T1 may be a marker for inflammation. When
the brain has more fluid as a result of swelling and MS inflammatory activity, it is possible that the T1 relaxation time constant could be elongated as a result. In future studies it may be necessary to control for new lesion activity and use of anti-inflammatory therapies in order to better understand the effects of these factors on the T1 in the NAWM. These T1 changes in this early stage CIS patient group emphasize the dynamic nature of disease in MS, and the challenge of group studies to look at disease pathophysiology.

Diffusion tensor imaging parameters of the NAWM have been shown to be markers of disease activity at baseline in this population of CIS patients, as well as in other studies with different MS patient subtypes. Longitudinal DTI data in this CIS population shows that the transverse diffusion, which may be a marker for Wallerian degeneration in lesion-affected fiber bundles, continued to increase over the span of one year, resulting in a corresponding increase in the mean diffusivity, and decrease in the anisotropy. The challenge with these values from the HA-NAWM continues to be relating the changes that are detected to specific injuries and corresponding disease processes. The initial results with the fiber tracking and difference maps show that localization methods hold promise as a future way of detecting within-patient, region-specific changes in the brain. Future work will benefit from fiber tracking and isolation of lesion-affected tracts to better understand the relationship of diffusion tensor parameters and neurodegeneration as a result of lesion activity.
4.4.2 Predictability of Volume Change

The study of early-stage MS patients immediately following the first episode indicative of MS attack in Chapter 3 discusses the inter-relationship of baseline MRI measures, in particular, the correlation between the baseline diffusion value and GM volume. An increased value in the HA-NAWM transverse diffusion and mean diffusivity correlated to a decreased GM volume. This relationship existing at the earliest presentation may indicate that disease has been present for quite some time prior to the first episode suggestive of MS. Longitudinally, the relationship lies between the baseline diffusion value and change in white matter volume over a year. In this group of patients, an increased transverse diffusion and mean diffusivity at baseline indicated more WM tissue loss measured over one year. These results help support the idea that transverse diffusion, and thus mean diffusivity, are markers for Wallerian degeneration. These relationships at baseline and one year later show the slow time-scale of the neurodegenerative process in the CNS, and support the need for further longitudinal investigation of extended duration.

4.5 Conclusion

Multiple sclerosis is a dynamic disease characterized by episodes of attacks with new lesion formation and inflammation, which is then often followed by periods of decreased inflammation and recovery. Over time, these repeated attacks lead to accumulated damage in the CNS. Even at this early stage of disease, the accrual of damage was demonstrated through brain atrophy detected over a
year, driven by a decrease in the grey matter volume. The continued increase in the transverse diffusion and its relationship to brain volume changes supports the hypothesis that diffusion characteristics of the white matter may be a marker for neuronal degeneration leading to atrophy. Also, it is hypothesized that the return towards normal of the T1 in the NAWM may be indicative of a recovery phase or decrease in inflammation occurring concurrently with the atrophy processes.

The longitudinal nature of this study allowed the investigation of the dynamics of MRI-detectable brain changes that occur with disease progression in early stage MS. Brain atrophy is a well documented signature of disease in MS; however, the underlying pathophysiology of disease that these quantitative MRI techniques may measure is less understood. Further investigations will provide more information on these processes and their relationships to tissue loss and accrual of disability, and may prove useful in creating better treatments for improved patient outcome.

4.6 References


5.1 Introduction

5.1.1 Diffusion Tensor Imaging

Diffusion tensor imaging (DTI) is a non-invasive MR technique for studying tissue microstructure. In the brain, the diffusivity of water is restricted by the presence of semi-permeable membranes such as those of myelinated axons. Due to orderly arrangement of neurons, diffusion of water is restricted transverse to axon bundles and is relatively unrestricted parallel to neural fibers.¹ Many diseases of the central nervous system, including multiple sclerosis (MS) for which the developments within this dissertation have been structured, have disease processes that result in the disruption of brain tissue microstructure. These brain tissue changes result in a difference in the mobility of water, which can then be detected using DTI and give clinically relevant information as to what is occurring physiologically in disease.

5.1.2 Limitations of Current DTI Tools

At this time, clinical DTI is typically acquired using an echo planar imaging (EPI) sequence. While this technique offers speed and high SNR, it suffers from several image artifacts including magnetic susceptibility signal dropouts, N/2 ghosting, and image distortions arising from eddy currents.² Another concern with the EPI technique is that spatial resolution is limited to the number of phase
encode steps that can be acquired within a single-shot acquisition. A second acquisition technique that has been used to collect DT images is based on the single-shot fast spin echo (SSFSE) sequence. This method decreases the distortions that trouble EPI, however, it does have reduced SNR and is susceptible to T2-blurring effects due to its long echotains.\(^2\)

5.1.3 3D DW-SSFP Pulse Sequence

The most common diffusion-sensitive pulse sequences are based on normal spin-echo with a pair of bipolar gradient pulses. Motion that occurs during the interval between gradient pulses results in spoiling of the gradient echo that would occur for stationary spins.\(^1\) The sequence can be made more sensitive to diffusion by increasing the time between gradient pulses, which is the reason for the increased sensitivity in the case of the SSFP sequence. The SSFP method uses a single unbalanced gradient to obtain diffusion sensitivity.\(^4\) Figure 5-1 shows the unbalanced diffusion gradient followed by a 3D Fourier Transform (FT) readout. The steady-state signal is a combination of echoes, where each echo is dependent on its magnetization orientation in the preceding periods. In this method an effective bipolar pair is formed between diffusion gradients separated by multiple TRs. Following a transverse period, magnetization may be stored along the longitudinal axis during which time signal is not lost rapidly to T2 processes, but decays by the much slower T1 processes. This signal pathway allows for a long diffusion time, while minimizing T2 decay. However, this method introduces a new weighting to diffusion imaging since the signal is now
reflective of T1 relaxation that occurs while the magnetization is in the longitudinal plane. The resulting images have diffusion weighting, as well as being T2/T1 weighted.\(^4\)

![Phase navigated 3D SSFP diffusion weighted sequence. The diffusion gradients can be played out on any combination of the three axes.](image)

**Figure 5-1.** Phase navigated 3D SSFP diffusion weighted sequence. The diffusion gradients can be played out on any combination of the three axes.

### 5.1.4 High Field Strength MRI

Moving MR applications, including DTI, to higher field strengths is desirable due to improved sensitivity, higher signal-to-noise ratios (SNR), and better spatial resolution; however, realization of these benefits is limited by technological challenges and safety limitations. The aforementioned EPI and SSFSE DTI methods pose problems when moving to higher field strengths. Both methods include 180° refocusing pulses and thus can be limited by peak B1, which needs to be increased at high field.\(^3\) In SSFSE especially, SAR becomes a concern because of the large number and high flip angle of rf pulses that are used.\(^3\)
case of EPI, the effects of magnetic susceptibility worsen as the field strength is increased. Figure 5-2 illustrates the effects of magnetic susceptibility on EPI image quality at 3T, and a comparable SSFP acquisition; at 7.0 Tesla these effects would be even more dramatic.

![Figure 5-2. Sagittal section taken from the same subject using EPI and SSFP imaging at 3T. Note the geometric distortions especially around the sinuses and down in the spinal cord due to magnetic susceptibility effects in the EPI acquisition.](image)

The SSPP method is an attractive alternative to these techniques due to its inherent sensitivity to diffusion, low flip angle and modest gradient requirements. This study investigated the use of a 3D steady-state free precession (3D-SSFP) diffusion acquisition scheme at 7 Tesla to produce high SNR, high resolution, minimally distorted DT images of the brain.

### 5.2 Methods

#### 5.2.1 Calculation of Sequence Parameters
The DW-SSFP signal is a weighted summation of multiple diffusion-weighted echoes, and thus the signal has a dependence on many factors including $T_1$, $T_2$, $D$, and the flip angle ($\alpha$). Although the signal is a summation of all previous excitations, it has been shown to be dominated by echoes with only two transverse periods, so that the signal attenuation can be approximated by:

$$A_{ssfp} = \frac{M(G\tau)}{M(0)} = \frac{A_D(1 + A_D E_1)(1 - E_1 \cos(\alpha))}{(1 + E_1)(1 - A_D E_1 \cos(\alpha))}$$  \hspace{1cm} \text{Equation 5-1}

where,

$$A_D = \exp(-\gamma G \tau^2 T_R D)$$  \hspace{1cm} \text{Equation 5-2}

$$E_1 = \exp(-\frac{T_R}{T_1})$$  \hspace{1cm} \text{Equation 5-3}

Equation 5-2 is the diffusion attenuation factor for a diffusion gradient of strength $G$ and duration $\tau$, and the expression is independent of $T_2$ since all echoes have the same transverse relaxation. The diffusion attenuation achieved using the SSFP method is different from standard DWI, where the attenuation is given by $A = \exp(-bD)$. In order to get similar attenuation in the SSFP method as in the standard EPI, the set of equations above need to be solved such that the $A_{ssfp}$ is equal to $A_{EPI}$.

5.2.2 Image Acquisition

Scanning was performed on a normal volunteers using a 7 Tesla GE Signa scanner (GE Healthcare, Milwaukee, WI) with 40 mT/m maximum gradient amplitude, and a volume excite, 8-channel receive phased-array head coil (Nova Medical, Boston, MA). Images were collected using a diffusion-weighted 3D-
SSFP sequence. Six gradient directions and a single b~0 image were collected axially in the brain with a 24x24x4.8 cm FOV, a 256x128x16 image matrix, a flip angle of 25°, and 17/34 ms TE/TR; resulting in an imaging time of nine minutes. A low resolution 2D spiral navigator with 24x24 cm, FOV, and a 4x4 image matrix, was collected at k_z=0 prior to each readout to correct motion-induced phase errors in the images.\(^5\)

5.2.3 Image Post-Processing

5.2.3.1 Data Processing and Image Reconstruction

The 3D-DW-SSFP raw data was saved to the scanner and transferred off-line to a Sun Ultra20 workstation for data processing and image reconstruction. First, each individual coil image was reconstructed and corrected for phase errors using the method discussed in the following section. Then the eight phase-corrected coil images were combined using the sum-of-squares. Signal-to-noise (SNR) calculations and attenuation ratios were carried out on the coil-combined images and done using region-of-interest (ROI) analyses on each of the b~0 and diffusion weighted series. For the DTI parameter calculations, mean diffusivity and anisotropy maps were created using an in-house diffusion tensor calculation software program.

5.2.3.2 Motion Correction

Bulk motion that occurs between excitations in a multi-shot diffusion acquisition may cause phase shifts that destructively interfere with one another and cause
artifacts in brain images. Although a number of methods have been proposed to correct these phase errors, this work applied a generalized reconstruction method known as the refocusing reconstruction which corrects for non-rigid motion of the brain based on an estimate of the least-squares solution. The mathematical details of reconstruction are beyond the scope of this dissertation, but are provided for reference in Miller KL, et al, 2003. The basic steps of reconstruction are 1) to reconstruct the low-resolution navigator for each interleave, 2) reconstruct the high-resolution DWI interleave, and 3) multiply the interleave by the phase conjugate of the navigator image. The phase-corrected interleaves can then be summed together to build the complete phase-corrected brain image. This algorithm was originally designed for 2D DW-SSFP images, but was modified to phase correct the 3D data collected as part of this project.

5.3 Results

5.3.1 3D DW-SSFP Pulse Sequence Parameters

In balanced SSFP sequences, and in this case with the diffusion gradient present, TR must be twice the length of TE. Lengthening TR increases the diffusion weighting; however, a longer TE results in a lower image SNR. A compromise with TE/TR = 17/34ms, and the flip angle, $\alpha=25^\circ$, were set based on the high diffusion-weighting requirement and previous work with DW-SSFP in the brain and knee cartilage which showed that these parameters were optimal to achieve significant diffusion weighting. For the initial application of DW-SSFP in the brain, it was desirable to get similar diffusion weighting to that achieved by
standard EPI methods for image comparison purposes. In order to achieve this, Equation 5-1, which corresponds to diffusion attenuation in the SSFP sequence had to be set equal to attenuation achieved in the brain when the b-value is set to 1000 s/mm$^2$. Reverse calculation using the predetermined TR, $\alpha$, and approximations brain T1 and D, and using the gradient maximum of 4.0 G/cm, resulted in $\tau = 6500 \mu$s.

5.3.2 Preliminary 3D DW-SSFP Imaging Results

Figure 5-3 shows the coil-combined, motion-corrected brain images with and without diffusion weighting, and also a map of the calculated mean diffusivity ($D_{av}$). This method produced high quality images with good SNR (SNR=32, non-diffusion-weighted image). The sequence parameters chosen for this acquisition resulted in high diffusion weighting, as can be seen by visually comparing Figure 5-3(a) ($b\sim0$) to the diffusion-weighted counterpart in (b). The effective b-value, $b_{eff}$, which relates the observed signal attenuation to that of an equivalent spin echo acquisition, is approximately 1050 s/mm$^2$. 
Figure 5-3. 7T diffusion weighted imaging with 0.94 x 1.88 x 3 mm resolution.  a) b = 0,  b) $b_{\text{eff}} \sim 1050$ s/mm$^2$,  c) map of $D_{\text{av}}$.

Figure 5-4 shows a single-coil image before and after motion correction. Without diffusion weighting (top row), motion is not a large concern, and correspondingly, the phase correction does not have a large noticeable effect on the resulting image. As the diffusion weighting increases, the effects of motion artifacts increase as well. The correction can decrease the black-hole artifacts as indicated by the red arrowhead in Figure 5-4, as well as improve structure definition as shown with the white arrowhead.
Figure 5-4. Phase error motion correction using efficient refocusing method. a) b=0, uncorrected image, b) b=0, phase-corrected image, c) b=1050 s/mm², uncorrected image, d) b=1050 s/mm², phase-corrected image.

5.4 Discussion

5.4.1 Benefits of DW-SSFP

The 3D DW-SSFP sequence is a promising method for obtaining high-resolution, minimally distorted, diffusion images of the brain at 7 Tesla. This acquisition technique offers many benefits over spin-echo methods, and these become especially important when working at high field strengths. Arguably, the most
significant benefit of using the SSFP method over standard EPI is its insensitivity to magnetic susceptibility effects. Although EPI can and has been used to collect DTI images on high-field systems, there continues to be issues with magnetic susceptibility even when using parallel imaging, resulting in certain areas of the brain that cannot be covered due to significant artifact and signal dropout. An additional benefit of this new method is the reduced RF requirements for SSFP versus spin-echo based methods. Spin-echo acquisitions require 180° pulses that are difficult to achieve uniformly with the existing rf technology, and these large flip angles also contribute to quickly reach the SAR limit for patient imaging. The small flip angles used in DW-SSFP minimize both of these rf issues.

5.4.2 Challenges with 3D DW-SSFP

Despite the proven advantages to the use of steady-state free precession in DWI, several challenges remain to be improved upon before this technique becomes more robust and applicable in a clinical setting. The most pressing issue is that in this technique, as with all multi-shot acquisitions, bulk motion creates phase errors and can quickly destroy image quality. In this work, translational motion was corrected using a spiral navigator; however, more sophisticated correction techniques that deal with complex rotations and non-rigid deformations associated with the cardiac cycle may be required. Research has also been done on SSFP-based acquisition methods that do not require phase navigators so that the need for time-consuming, off-line image post-processing is eliminated.
A second major hurdle in the adoption of DW-SSFP is the complex signal, and relative current lack of a deeper understanding of the diffusion signal that is detected. Because the echo that is measured is a combination of many echoes with different diffusion weightings, the b-value for this method is much more complicated than in the case of EPI imaging. Although this is not particularly important for qualitative imaging where contrast is the most important factor, it does pose a problem for quantitative measures of the differences in slow and fast diffusion components comprising a particular tissue.

5.4.3 Quantitative SSFP-DTI

Quantitative DTI is an important and quickly growing clinically applicable tool in the field MR imaging. When using SSFP to collect DT images, new algorithms, and perhaps new imaging protocols as a whole, have to be developed to solve for $<D>$ due the complexity of the signal as shown in Equations 5-1 through 5-3. In order to accurately calculate the diffusion coefficients, the tissue relaxation times must be known, and it may be necessary to measure them in the same imaging session as the diffusion sequence. Research has been done on quick mapping techniques that can be used in conjunction with DW-SSFP in order to make quantitative SSFP-base DTI MRI a reality.$^9,10$
5.5 Conclusion

The preliminary work and initial images produced using this new acquisition scheme show the promise that diffusion-weighted SSFP imaging techniques lend to the area of high field strength MRI. New pulse sequence development and parameter calculation for the image acquisition was performed in order to achieve these results. Additional work was required in order to improve the image reconstruction for MRI acquisition with a multi-channel head coil, and also modified for best performance when employed with 3D imaging techniques. These developments produced excellent quality images, and highlighted the strengths of this technique compared to highly distorted images that are collected using EPI which is currently the standard acquisition method for diffusion-weighted imaging.

As with many MR advancements, this method does not come without challenges such as motion artifact and difficulty with confounding factors affecting image weighting; however, there are also many advantages to using DW-SSFP. These advantages are more pronounced at high field strengths including a dramatic reduction in artifacts that occur in standard EPI-based diffusion sequences, fewer safety concerns due to lower power and reduced gradient switching requirements, and also the high signal can be used to collect ultra high-resolution images. These advantages result in the ability to cover areas of the brain such as the cortex, and areas near the sinuses and the spinal cord with better image quality than has previously been possible with DW-EPI. These advancements
may allow for a more comprehensive understanding of brain anatomy, and in the
future can be applied to disease to drive further investigations into the processes
responsible for disease pathology.

5.6 References

1. Le Bihan D, Mangin JF, Poupon C, Clark CA, Pappata S, Molko N, Chabriat
   H. Diffusion tensor imaging: concepts and applications. J Magn Reson

2. Xu D, Henry RG, Mukherjee P, Carvajal L, Miller SP, Barkovich AJ, Vigneron
   DB. Single-shot fast spin-echo diffusion tensor imaging of the brain and spine
   with head and phased array coils at 1.5 T and 3.0 T. Magn Reson Imaging

   18(5):519-29.

4. Buxton RB. The diffusion sensitivity of fast steady-state free precession

5. Miller KL, Pauly JM. Nonlinear phase correction for navigated diffusion

6. Miller KL, Hargreaves BA, Gold GE, Pauly JM. Steady-state diffusion-
   398.

   DAC, Nelson SJ, Vigneron DB. High resolution and diffusion 7T MR imaging
   of multiple sclerosis patients. Advanced in Highfield MR workshop, ISMRM,

8. Sigmund EE, Parrish TB. Gradient-Alternated SSFP (GASP) Diffusion
   Imaging. Proceedings of the 13th Annual Scientific Meeting of the
   International Society of Magnetic Resonance in Medicine, p1288. Miami, FL,
   USA, 2005.

9. Deoni SC, Peters RM, Rutt BK. Quantitative diffusion imaging with steady-
Chapter 6: High Resolution Phased-Array MRI at 7 Tesla

6.1 Introduction

6.1.1 High Field MRI

Magnetic resonance imaging was introduced for clinical diagnostic imaging in the early 1980's, and has gone through a rapid series of technological and application-driven advances since that time. Besides the inherent advantages that MR has over other medical imaging technologies, one of the main reasons that it has been able to be successful in so many different applications has to do with the continual technical progress with software and hardware used to drive these systems. A recent area of advancement for MRI is in the realm of increasingly higher field strengths. Although imagers with higher static magnetic field strengths were developed and tested in research labs for the last couple of decades, their application to patient MR studies have been extremely limited.

6.1.2 New Phased-Array Coil Technology

The development of high field strength imaging systems also requires technological advances in coil design. Imaging at 7 Tesla brings advantages in SNR and image contrast and resolution, but these benefits can only be realized if the correct coils exist to capture the images. There are many challenges with high field signal excitation and reception, including RF field homogeneity, high RF power requirement, and coil specific issues such as element coupling, tuning, matching, and cable interactions. These engineering issues have been theoretically and experimentally investigated by several of the leading high field
research groups. Recently, phased array coils at 7T have demonstrated more uniform reception and increased sensitivity to visualize small anatomic structures not previously detected. \textsuperscript{10-12} However, the value of these high resolution 7T MRI techniques have not yet been well established for patient studies.

6.1.3 High Field Imaging: Application to Patients with MS

Multiple sclerosis (MS) is one clinical application that could benefit tremendously from the advances in high-field MRI. MS is a multi-focal, inflammatory, heterogeneous demyelinating condition traditionally regarded as a disorder of the white matter. Postmortem pathologic studies reveal significant involvement of the grey matter affecting principally the cingulate gyrus, frontal, parietal, and temporal lobes. \textsuperscript{13-15} White matter lesions are typically detected easily \textit{in vivo} at the conventional field strength of 1.5T, and with even better resolution at 3T. However, cortical grey matter lesions are poorly detected at these field strengths due to their small size and the technical limitations associated with achieving adequate spatial and contrast resolution. At 7T, the signal-to-noise ratio (SNR) increase improves spatial resolution, allowing for the visualization of smaller anatomical structures, in addition to providing morphological details. The objective of this study was to develop high-resolution 7T MRI techniques using high sensitivity, specialized phased-array coils, with optimized acquisitions for improved grey and white matter differentiation and apply them for the first time for improved visualization of MS lesions.
6.2 Materials and Methods

6.2.1 Research Participants

Healthy, adult subjects were enrolled in the preliminary 7T studies. Following acquisition of initial scans and sequence optimization, adult patients with an established diagnosis of clinically definite multiple sclerosis (CDMS) evaluated by the Multiple Sclerosis Center at the University of California, San Francisco were invited to participate. A total of 23 subjects were enrolled, 17 with CDMS (11 females, 6 males; mean age 43.4 years; range 22-64 years) and 6 healthy controls (2 females, 4 males; mean age 39.0 years; range 27-67 years). The clinical MS subtype for all 17 patients was relapsing-remitting. The concomitant use of disease modifying therapies for MS was permitted. The Committee on Human Research at the University of California, San Francisco, approved the protocol and informed consent obtained from all participants.

6.2.2 Image Acquisition

All patients were scanned on a GE EXCITE 7T scanner (General Electric Healthcare Technologies, Waukesha, WI). Excitation was performed using a commercial volume transmit head-coil with a shielded, high-pass birdcage coil design (NOVA Medical, Wilmington, MA). An in-house, custom-made, 8 channel phased-array surface coil was used for signal reception in nine patients and four controls. A commercially-available, 8-element phased-array receive coil (NOVA Medical, Wilmington, MA) was used in the other eight patients and controls.
A low-resolution 3-plane localizer was followed by a series of gradient echo acquisitions to obtain T2*-weighted images with different resolutions and orientations. Two axial series were acquired, one with a reduced FOV and less gap between slices for higher resolution images. Axial slices were prescribed with the most inferior slice along the anterior commissure to posterior commissure (AC-PC) line and the rest superior to that in order to cover the corpus callosum and above. Sagittal series were collected in five patients. The sagittal series was prescribed with the center slice aligned with the mid-sagittal image from the localizer, and dividing the remaining slices to both the right and left hemispheres of the brain. The sequence parameters for each of these series are shown below in Table 6.1.

<table>
<thead>
<tr>
<th></th>
<th>Axial – 1</th>
<th>Axial – 2</th>
<th>Sagittal</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>TR/TE (ms)</strong></td>
<td>250/15</td>
<td>250/15</td>
<td>250/15</td>
</tr>
<tr>
<td><strong>flip angle</strong></td>
<td>20°</td>
<td>20°</td>
<td>20°</td>
</tr>
<tr>
<td><strong>slice thickness (mm)</strong></td>
<td>2</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td><strong>skip</strong></td>
<td>4</td>
<td>1.5</td>
<td>1.5</td>
</tr>
<tr>
<td><strong>NEX</strong></td>
<td>2</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td><strong>FOV (cm)</strong></td>
<td>22</td>
<td>20</td>
<td>22</td>
</tr>
<tr>
<td><strong>matrix size</strong></td>
<td>1024 x 768</td>
<td>1024 x 768</td>
<td>1024 x 768</td>
</tr>
<tr>
<td><strong>imaging time</strong></td>
<td>6:28</td>
<td>9:38</td>
<td>6:28</td>
</tr>
</tbody>
</table>

*Table 6-1. 7T T2*-weighted GRE Imaging Parameters*

Diffusion-weighted single-shot EPI images were acquired in four patients using a custom sequence with ASSET parallel imaging and incorporating a newly designed high bandwidth fat saturation pulse. The parameters for the DTI sequence were: 25.6 cm FOV, 256x256 matrix, 2-mm slices with no gap, 25
directions, b=1000s/mm², and ASSET R=2; resulting in an acquisition time of approximately 3 minutes. Higher order shimming was performed prior to the acquisition.\textsuperscript{16}

6.2.3 Acquisition Optimization

In addition to very high spatial resolution, the objective of the T2*-weighted GRE sequence was to maximize the contrast resolution between grey matter and white matter in the brain, while maintaining excellent SNR throughout the brain, especially in the cerebral cortex. An in-house custom-built 8-channel phased-array surface coil was compared to a commercially available 8-channel phased-array receive coil to evaluate SNR and to determine which would be better suited for applications in cortical imaging. To determine the imaging parameters that best accomplished tissue contrast, a set of GRE sequences were acquired, each had the same sequence parameters, except the TE was varied from 12-24 ms, at intervals of 3 ms. Contrast to noise (CNR) and SNR were calculated for each of the image sets and the TE that provided the best grey/white differentiation was chosen.

6.2.4 Image Processing

The high resolution, phased-array 7T images were reconstructed on the scanner and then transferred off-line to a Sun Ultra 20 workstation for image processing. High coil sensitivity on the brain periphery resulted in images that are very bright at the edge, and darker in the center of the brain, as illustrated in Figure 6-1A. In
order to compensate for this signal heterogeneity, and achieve a more uniform signal profile, the brain images were masked from the background noise and an edge-filled low-pass filtering algorithm was used, the effects of which are shown in Figure 6-1B. This post-processing facilitates visual inspection and qualitative analysis of the images. All signal-to-noise (SNR) and contrast-to-noise (CNR) calculations were performed via region-of-interest (ROI) analyses of the images before the image processing was applied. For the DTI studies, anisotropy and mean diffusivity maps were created using the diffusion tensor calculation software that was developed in-house.

Figure 6-1. Images in a control subject demonstrating image intensity across a single slice before (A) and after image post-processing (B).
6.3 Results

6.3.1 Optimization

The custom fabricated phased array coil provided a 2.5-fold increase in SNR at the cortex and 30% increase in deep brain parenchymal structures compared to the commercially available 8-channel phased-array coil. The increased sensitivity at the cortex resulted in increased image brightness at the edges, as seen in Figure 6-1A. From the results of the SNR calculation, it was decided to use the custom-built coil in the majority of the scans to capitalize on the sensitivity at the cortex and focus on anatomy in this region.

The results of the SNR and CNR calculations used to determine the optimal TE can be seen in Table 6-2. The numbers show that for a TE of 12 and 15ms the SNR of the grey matter and white matter are similar, with the 15ms TE being less than 2% lower in each case than the 12ms echo time. This minimal change leads to the CNR being approximately the same in both the 12 and 15ms TE case, although the CNR is slightly lower with the increased TE. As the TE increases beyond 15ms the SNR, and corresponding CNR, drop to a greater extent.

<table>
<thead>
<tr>
<th>TE</th>
<th>WM SNR</th>
<th>GM SNR</th>
<th>WM/GM CNR</th>
</tr>
</thead>
<tbody>
<tr>
<td>TE = 12ms</td>
<td>16.4</td>
<td>25.3</td>
<td>8.9</td>
</tr>
<tr>
<td>TE = 15ms</td>
<td>16.1</td>
<td>24.9</td>
<td>8.8</td>
</tr>
<tr>
<td>TE = 18ms</td>
<td>13.7</td>
<td>19.3</td>
<td>5.6</td>
</tr>
<tr>
<td>TE = 21ms</td>
<td>11.8</td>
<td>18.7</td>
<td>6.9</td>
</tr>
<tr>
<td>TE = 24ms</td>
<td>11.0</td>
<td>13.9</td>
<td>2.9</td>
</tr>
</tbody>
</table>

Table 6-2. TE optimization for maximal SNR and CNR in 7T T2*-weighted GRE
In addition to quantitative criteria, the TE decision was also based on the qualitative image inspection by a neurologist and neuroradiologist. Images resulting from the TE optimization experiment can be seen in Figure 6-2. The qualitative and quantitative analyses resulted in 15ms being chosen as the optimal TE for the T2*-weighted GRE sequence.

Figure 6-2. Axial 2DGRE images at 7T were acquired from a volunteer with varying TE to determine optimal T2* weighting for maximum image SNR and CNR between the grey and white matter.
6.3.2 Ultra-High Resolution Anatomic Imaging

Based on the imaging parameters listed in Table 6-1, the spatial resolution was 195x260 µm or 215x286 µm, depending on the FOV for the particular image acquisition series. Figure 6-3 shows several axial slices at different slice locations in a control subject. These images demonstrate that excellent image quality can be obtained even at the ultra-high resolutions done with these imaging parameters. The images in Figure 6-3 illustrate the level of structural detail that it is possible to achieve using the combination of specialized phased-array coils and the optimized GRE acquisition.

![Figure 6-3. Axial 7T T2*-weighted slices from a single healthy volunteer showing excellent quality, high-resolution anatomical images. These images demonstrate good differentiation between the grey and white matter, as well as highlight the appearance of vasculature. In-plane resolution: 215x286 µm.](image)

The T2*-weighted GRE acquisition allowed for excellent grey-white contrast as demonstrated in the axial slices of the control subject in Figure 6-3. The images in Figure 6-4 illustrate the outstanding grey-white contrast at the cortex in an MS patient, as well as the lesion contrast that is achieved with this method. The
patient images also highlight the appearance of vasculature when using this method due to susceptibility-based signal loss in the vessels.

Figure 6-4. Axial 7T T2*-weighted slices in a single MS patient demonstrating multiple foci of abnormality involving the deep white matter structures. Note the enhanced grey matter, white matter, and lesion contrast, in addition to lesion distribution and vasculature. In-plane resolution: 215x286 µm.

White matter lesions were easily detected and better delineated from adjacent structures compared to lower field strengths (1.5 and 3T). The good contrast, in addition to the improvement in spatial resolution, makes differentiation between juxtacortical white matter lesions and cortical lesions possible (Figure 6-5). Figure 6-6 demonstrates the fine detail that can be visualized in a cortical lesion that originates from the pia, and extends through the grey matter to the subcortical white matter.
Figure 6-5. *Left:* Axial T2*-weighted image (195x260 µm) of an MS patient with numerous white matter lesions. *Right:* Zoom of highlighted region demonstrating a sub-cortical WM (blue arrow) and juxtacortical (red arrow) lesion.

Figure 6-6. A) Axial T2*-weighted image (195x260 µm) of an MS patient with a cortical GM lesion. B) Enlarged image of the highlighted selection from Figure 7-6A demonstrating a cortical grey matter lesion.
Figure 6-7 contrasts sagittal images from an MS patient with a sex and age-matched control subject. These images demonstrate the extent of the coil coverage in the superior/inferior direction and also illuminate some deep white matter structural differences. From these images we see that we can obtain excellent quality high-resolution sagittal images with coverage from the top of the brain down through the cerebellum. The red arrowheads in the zoomed-in image from the MS patient highlight the dramatic thinning of the corpus callosum compared to control. The contrast resolution achieved with the T2*-weighted GRE sequence made it possible to view the extent of lesions in the splenium of the corpus callosum in this particular patient.
6.3.3 High Resolution Diffusion Tensor Imaging

Figure 6-8 shows the T2-weighted image, anisotropy, and mean diffusivity maps at multiple slice locations from a DTI scan in a healthy control. Using the optimized sequence, 28 slices can be collected in approximately 3 minutes, covering a large portion of the supratentorial brain. The high-resolution DT-images had good SNR (14.9 for b=0 and 5.8 for b=1000) despite the small voxel size of 2 mm$^3$ and use of parallel imaging. The anterior/posterior distortions from EPI were acceptable with the use of parallel imaging and the high order shimming routine.
Figure 6-8. Multiple slice locations from a 7T DTI scan in a control subject.

DT-images from an MS patient can be seen in Figure 6-9. These results show that the lesions are well visualized on the b=0 image, as well as on the mean diffusivity (MD) and relative anisotropy (RA) maps. The hyperintensity of the lesions on the MD map and the hypointensity on the RA correspond to previous findings that diffusion increases and the anisotropy decreases in MS lesions.
6.4 Discussion

6.4.1 Technical Challenges with Coils

The use of novel phased-array coils in this project significantly increased the SNR at the cortex, and even improved SNR at the center of the head compared to the commercially available head coil. However, the continuous capacitance coil used in this study has technical challenges that need to be addressed before it can be incorporated into routine clinical protocols. One issue is the coupling of coil elements, which limits this coil's use for spectroscopy and diffusion sequences that ideally would be included in patient exams. Although coil switching within a session is possible, it is time-consuming and not necessarily the most comfortable situation for research subjects. Other challenges include limited superior-inferior coverage due to the geometry of the coil, and the adjustment of the coil to different sizes of head.
6.4.2 Contrast and Resolution

The resolutions attainable using the increase in SNR from the ultra-high field strength are much higher than what is achieved in current clinical protocols at 1.5 and 3.0T. In this study, 195 x 260 µm was a typical resolution for the T2*-weighted image series. The clinical anatomical imaging protocols run on 1.5T and 3.0T systems at our institution generally have resolutions of 938 x 938 µm or larger in the case of the former, and 469 x 469 µm in the best case for the latter. It should be noted that although the ultra-high resolutions obtained at 7T could theoretically be done at lower field strength, it is not truly feasible in a clinical scan due to very long scan times necessary to achieve sufficient SNR.

The imaging parameters in the T2*-weighted sequence were optimized for the best CNR between WM and GM. The images in Figure 7-4 show that not only was excellent contrast achieved between grey and white matter, but also, in the case of MS, lesions were well distinguished from the surrounding normal-appearing brain tissue. Another fortuitous result of this optimization was the prominent appearance of the blood vessels and microvasculature due to the increased magnetic susceptibility of the blood at high field strengths causing signal drop-out. The pronounced appearance of vasculature may be useful to various neurological applications of high-field MRI. For example, previous histopathology has shown that MS lesions are often located near or adjacent to blood vessels. Now that the location of both blood vessels and lesions can be
seen *in vivo* using this methodology, the relation between the two may be investigated in future studies.

**6.4.3 Difficulties and Possibilities with High-Field DTI**

The findings in these preliminary high field diffusion studies are complementary with what is seen at lower field strengths. The primary advantage of the increase in field strength is the corresponding increase in signal, which translates into the possibility of higher resolution images or faster scan times. Increase in chemical shift and magnetic susceptibility artifacts come along with the advantageous increase in signal. At lower field strengths EPI is the gold standard for diffusion tensor imaging studies, and this method is very susceptible to image distortions, signal dropout at high field strengths. The use of parallel imaging minimizes these effects, however brain coverage may be limited. Different diffusion methodologies that have been explored may prove to be the best way to continue diffusion research as high field strength MR scanners become more common in clinical imaging.\(^\text{18,19}\)

**6.4.4 High Field MRI in MS**

The introduction of structural neuroimaging in MS has proven to be an invaluable tool for the diagnosis, clinical surveillance, management, and the assessment of therapeutic efficacy in pivotal MS clinical trials. The application of high field MR imaging and advanced techniques provided additional insight into MS pathology compared to conventional and lower field strength imaging metrics by enhancing
lesion morphology and spatial distribution. Post mortem studies have shown some of the benefits to ultra high field imaging, however, the challenge of visualizing in vivo many of the differences is still large.\textsuperscript{20} In addition to the importance of increased resolution demonstrated in this work, other studies have shown the possibility of novel image contrast techniques that may highlight disease pathology better than conventional imaging methods.\textsuperscript{21-24} It will be important in the future to determine what methods capture the most information, and test and implement these on high field systems to merge the resolution benefits with the advantages of the best contrast mechanisms.

Very high field MR imaging of all CDMS patients at 7T was demonstrated to be safe, well tolerated, and provided high-resolution anatomical images allowing for the visualization and differentiation of structural abnormalities localized near or within the cortical layers. Clear involvement of the grey matter was observed in one of the patients at 7T, with improved lesion detail in comparison to imaging at lower field strengths. Grey matter involvement observed in MS is postulated to be the result of retrograde (Wallerian) degeneration from lesions originating in the white matter structures. Recent data, however, suggests a dissociation between cortical demyelination and white matter pathology, suggesting that MS patients have the potential to possess both grey and white matter demyelinating pathology independently.\textsuperscript{25}
6.5 Conclusion

The human brain images produced through this work demonstrate that high field imaging can produce excellent quality images with ultra-high spatial resolution. To achieve these results, significant new technologies had to be developed and employed. New coils designed for improved sensitivity at the cortex were tested and the utility of the gain in SNR was investigated. Also, MR imaging sequences were optimized to produce images of the highest quality possible at this field strength. These developments allowed for exceptional images to be acquired and highlighted what value could come from applying these methods in patient populations.

The preliminary studies in MS patients indicated that very high field MR imaging was well tolerated and provided ultra-high resolution anatomical images allowing for the visualization of structural abnormalities associated with disease. These initial results suggest that scanning MS patients at 7T is not only feasible, but also can detect lesions at a higher spatial resolution than at lower fields. MS lesions were detected in the white matter, and lesions in the cortical grey matter ribbon, rarely viewed with such detail at lower field strengths, were clearly observed. The high sensitivity provided by 7T allowed the detection of diffusivity and anisotropy changes in MS lesions at a better spatial resolution than has previously been reported. These advancements may allow for a more comprehensive understanding of multiple sclerosis, allowing for further investigations into the natural history and the in vivo delicate immune-mediated
systems responsible for the genesis of multiple sclerosis pathology.

6.6 References


Chapter 7: Summary

This dissertation project investigated advanced MR imaging techniques and novel acquisition methods to assess their capabilities and clinical applications in patients with multiple sclerosis. This project had several parts, each of which filled a void in the existing MS research literature. The first part of this project involved looking for the earliest MRI detectable manifestations of disease in patients suffering from a clinically isolated syndrome suggestive of MS. The T1-relaxation measures and DTI measures were found to be sensitive enough to detect abnormalities in the NAWM and HA-NAWM, respectively, as early as initial presentation of a suspected MS episode. The second aspect of this project was to investigate tissue loss in early stage disease, and the relationship between the quantitative MR measures and atrophy, as detected by various brain volume measurements. The brain volume measures in the patient group were not different from controls at presentation; however, DTI measures did correlate with GM volumes as well as the atrophy that occurs over a year, suggesting that DTI may be a marker for Wallerian degeneration occurring due to tissue injury. The third part of this project involved testing a novel diffusion acquisition method aimed at new clinical applications and for use on high-field MR systems. This SSFP technique showed promise in collecting high-resolution images with minimal distortion. The last portion of this dissertation work focused on the clinical utility of ultra-high field MR in MS. This work was designed to collect excellent quality high-resolution images, obtain enhanced image contrast, and to
visualize lesions that were not previously identifiable on images from lower field strength systems. As a result of this dissertation project, more has been learned about the earliest tissue damage that occurs in probable MS, as well as the possible underlying mechanisms for this damage. Also new techniques were developed to further investigate and visualize disease using MR systems.

This dissertation project has shown the value of quantitative MR imaging techniques, image processing tools, and new high-field systems as non-invasive methods for studying the human brain and disease therein. Although individually these techniques are each sensitive to brain alterations caused by disease, there is still a limited understanding of the specificity of these changes and how, or if, they are inter-related with one another. Despite the fact that additional research is required in order to better understand MS, this body of work advanced our knowledge in relating MR metrics to early stage underlying disease processes, following longitudinal MS changes, and improving characterization over what is seen in conventional anatomic imaging.
UCSF Library Release

Publishing Agreement

It is the policy of the University to encourage the distribution of all theses and dissertations. Copies of all UCSF theses and dissertations will be routed to the library via the Graduate Division. The library will make all theses and dissertations accessible to the public and will preserve these to the best of their abilities, in perpetuity.

Please sign the following statement:

I hereby grant permission to the Graduate Division of the University of California, San Francisco to release copies of my thesis or dissertation to the Campus Library to provide access and preservation, in whole or in part, in perpetuity.

[Signature]
Author Signature

9/4/08
Date