INFORMATION TO USERS

This manuscript has been reproduced from the microfilm master. UMI films the text directly from the original or copy submitted. Thus, some thesis and dissertation copies are in typewriter face, while others may be from any type of computer printer.

The quality of this reproduction is dependent upon the quality of the copy submitted. Broken or indistinct print, colored or poor quality illustrations and photographs, print bleedthrough, substandard margins, and improper alignment can adversely affect reproduction.

In the unlikely event that the author did not send UMI a complete manuscript and there are missing pages, these will be noted. Also, if unauthorized copyright material had to be removed, a note will indicate the deletion.

Oversize materials (e.g., maps, drawings, charts) are reproduced by sectioning the original, beginning at the upper left-hand corner and continuing from left to right in equal sections with small overlaps. Each original is also photographed in one exposure and is included in reduced form at the back of the book.

Photographs included in the original manuscript have been reproduced xerographically in this copy. Higher quality 6" x 9" black and white photographic prints are available for any photographs or illustrations appearing in this copy for an additional charge. Contact UMI directly to order.

UMI

A Bell & Howell Information Company
300 North Zeeb Road, Ann Arbor MI 48106-1346 USA
313/761-4700 800/521-0600

**A Study of Water Mobility in Human Tissue In
Vivo:
Apparent Diffusion Coefficients in Brain and
Leg Muscle**

by

Lawrence L. Gates B.Sc., M.Sc.

A thesis submitted to the
Faculty of Graduate Studies and Research
in partial fulfillment of the requirements
for the degree of
Doctor of Philosophy

Ottawa-Carleton Institute for Physics
Department of Physics, Carleton University
Ottawa, Ontario, Canada
August, 1997

©copyright
1997, Lawrence Gates

National Library
of Canada

Acquisitions and
Bibliographic Services

395 Wellington Street
Ottawa ON K1A 0N4
Canada

Bibliothèque nationale
du Canada

Acquisitions et
services bibliographiques

395, rue Wellington
Ottawa ON K1A 0N4
Canada

Your file Votre référence

Our file Notre référence

The author has granted a non-exclusive licence allowing the National Library of Canada to reproduce, loan, distribute or sell copies of this thesis in microform, paper or electronic formats.

The author retains ownership of the copyright in this thesis. Neither the thesis nor substantial extracts from it may be printed or otherwise reproduced without the author's permission.

L'auteur a accordé une licence non exclusive permettant à la Bibliothèque nationale du Canada de reproduire, prêter, distribuer ou vendre des copies de cette thèse sous la forme de microfiche/film, de reproduction sur papier ou sur format électronique.

L'auteur conserve la propriété du droit d'auteur qui protège cette thèse. Ni la thèse ni des extraits substantiels de celle-ci ne doivent être imprimés ou autrement reproduits sans son autorisation.

0-612-26852-7

Canada

The undersigned recommend to
the Faculty of Graduate Studies and Research
acceptance of the thesis

**“A Study of Water Mobility in Human Tissue In Vivo:
Apparent Diffusion Coefficients in Brain and Leg Muscle”**

submitted by **Lawrence L. Gates, B.Sc., M.Sc.**

in partial fulfillment of the requirements for

the degree of Doctor of Philosophy

Chair, Department of Physics

Thesis Supervisor

External Examiner (Ph.D.)

CARLETON UNIVERSITY

Date: August 29, 1997

Abstract

Water diffusion in human tissue was investigated in this thesis. Tissue structures and fluids in the body have a pronounced effect on the mobility of water at the microscopic level. They restrict the motion of water molecules which would otherwise move according to free diffusion. The result is that the diffusion coefficient appears to be smaller. A comparison of the measured apparent diffusion coefficient, D_a , with the *unrestricted* coefficient, D , can yield information on membrane permeability, cell size and orientation. This thesis presents results of D_a vs. diffusion time in human white matter in vivo and human skeletal muscle (soleus) in vivo. The range of diffusion times investigated in white matter is 13.7 ms - 1020 ms and in the leg muscle it is 4.0 ms - 50 ms. A double Maxwell gradient coil, specifically designed for diffusion measurements, was built and its electrical implementation on a conventional clinical MR imager is presented.

From current analytical equations, the diffusion profiles in the white matter reveal that the membrane permeability of the axons falls within lower and upper limits of $P=0.002$ cm/s and $P=0.019$ cm/s. However, this thesis also presents a Monte Carlo analysis and its results suggest a prominent mechanism affecting water mobility is missing from these theoretical equations. It is found that fluid motion at the cellular level (probably originating from cerebrospinal fluid pressure) is most likely a prominent factor affecting the motion of water molecules. The experimental findings in this thesis also reveal that the diffusion anisotropy of white matter is removed at low diffusion times. In the skeletal muscle, this work has found a very small restricted diffusion effect in the longitudinal direction of muscle fibre, probably due to the Z-line of the sarcomere.

Diseases such as stroke and multiple sclerosis may benefit from clinical applications of diffusion imaging. More information about the basic processes of water diffusion in tissue is needed before the full potential of diffusion MRI can be realized.

Acknowledgements

My first thank you goes to my wife, Janey, for her constant support during this project, for her understanding of my long and bizarre work hours, for helping me focus and for giving me a different angle to view things.

Special credit and thanks goes to my supervisor, Ian Cameron, for his involvement and guidance in this research. His influence has directed this study into one of the more exciting branches of MR research.

It was a pleasure to work with Cathy MacGillivray, a Masters student with whom I shared an office for 3 years.

Thank you's to my Ph.D. committee members Paul Johns, Serge Desgreniers, and John Saunders for their valuable suggestions and comments.

In the Department of Radiology at the Ottawa General Hospital, Dr. Len Avruch has helped me understand the physiology and anatomy of the brain. The Siemens engineering technician, Norm Burgstaller, has been both a source of information to draw from and a pleasure to work with. I thank Anne Ashley for helping me identify some useful imaging packages that highlight white matter. I also thank Marg Crosbie, who not only is a pleasure to work with, but has used her remarkable ability to cut through red tape to help me reach deadlines.

At Carleton University, I thank Bog Jarosz, Bob Clarke, Giles Santyr, George Curley, Gary Findlay, and Jim Sliwka for their interest in my project and their insights. At the Science Technology Centre, I thank Larry Boissoneault for his helpful suggestions. I thank Bill Jack for his generous help with my computer needs.

I wish to thank my fellow graduate students Ruth Brown, Ted Lawrence, and Jian-Su Wei for sharing many hours on course problems. For their insightful conversations, I acknowledge also Doru Kaytar, Daryoush Sheik-Bagheri, Greg Cron, Matthew Jones, Sherry Towers, Julia Wallace, Alan Dekok, Kevin Lenton,

Miller MacPherson, and most of all Mike Boyce, who has been invaluable as a computer resource person and someone to bounce my ideas off of.

Many thanks to my friend Joanna Tsoutouras for going in the magnet so often.

I thank also Doug Salhani, for his inspirational comments and Pat Kalyniak, who helped me bring this thesis to a close.

The funding of this research by grants from the Natural Sciences and Engineering Research Council (NSERC) is appreciated. In addition, the personal financial support provided by an Ottawa General Hospital Studentship is gratefully acknowledged.

Contents

Abstract	iii
Acknowledgements	iv
1 Introduction	1
1.1 History of MRI	2
1.1.1 Chronicle of Diffusion Measurements in the Brain	3
1.2 NMR Characteristics	7
1.3 Diffusion	7
1.4 Restricted Diffusion	9
1.5 Experiments on Human Brain White Matter In Vivo	13
1.6 Experiments on Human Leg Muscle In Vivo	14
2 Theory	16
2.1 Nuclear Magnetic Resonance	16
2.1.1 Quantum Mechanical Description	21
2.1.2 Equilibrium Distribution of Spin States	28
2.2 Diffusion: Fick's Law and the Einstein Equation	29
2.3 Diffusion and Experimental NMR	33
2.4 The Bloch Equations	36
2.5 More on Diffusion Sequences	41
2.6 Tissue Anisotropy and the Diffusion Tensor	45
2.7 Restricted Diffusion	48
2.8 Gradient Coils	53

3	Materials and Methods	61
3.1	Diffusion Sequences	62
3.1.1	PGSE and VOI Selection	62
3.1.2	AGPS	67
3.1.3	STEAM	69
3.2	Measurement Protocol for White Matter	71
3.3	Phantoms	74
3.4	Gradient Coil Design	76
3.4.1	Coil Calibration	86
3.5	Measurement Protocol for Leg Muscle	88
3.6	Data Analysis Programs and Error Computation	91
3.7	Monte Carlo Simulations	94
4	Results and Analysis	103
4.1	White Matter Data	103
4.2	Phantom Results	113
4.3	Monte Carlo Results	118
4.4	Gradient Coil Calibration	131
4.5	Leg Muscle Data	132
5	Discussion and Further Results and Analysis	143
5.1	White Matter Experiments	143
5.1.1	The 1-Compartment Model of Tanner	146
5.1.2	The 2-Compartment Model of Szafer	147
5.1.3	Monte Carlo Models	148
5.1.4	Consideration of the White Matter Fibre Orientation	154
5.1.5	Analysis of the Signal Decay in Human White Matter	157
5.1.6	T_2 Effects on the Apparent Diffusion Coefficient	163
5.1.7	Monte Carlo Simulation of the Signal Decay	165
5.1.8	2-D Monte Carlo With a Flow Channel	168

5.2	Leg Muscle Experiments	173
6	Conclusions	181
6.1	Human White Matter	181
6.2	Human Leg Muscle	184
7	Future Directions	187
7.1	Clinical Studies for Multiple Sclerosis	187
7.2	Linescan Imaging and Anisotropy Removal	188
A	Calculation of Field Derivatives	190
B	Details of the ‘phase Monte Carlo’ Algorithm	193
	Bibliography	195

List of Tables

1.1	Summary of literature D_q 's.	7
1.2	Table of NMR Nuclei	9
3.1	Table of sequences 1	101
3.2	Table of sequences 2	102

List of Figures

1.1	D_a vs. $T_d^{1/2}$ from non-tensor/tensor methods. Literature only.	8
2.1	Precession of the magnetic moment about a static field $\mathbf{B}_0$	18
2.2	Tipping the magnetization into the x-y plane.	20
2.3	Zeeman splitting.	22
2.4	Conservation of mass diagram.	31
2.5	Diagram of PGSE sequences.	35
2.6	Diagram of diffusion sequences.	44
2.7	Particle in a bound medium. Velocity correlations.	47
2.8	Schematic D_a and Diffusion distance vs. SQRT(diffusion time) . . .	50
2.9	Regularly spaced 1-D barriers	52
2.10	Coordinate system for B_z from a current loop.	54
2.11	Maxwell Coil	55
2.12	Parameters of a double Maxwell gradient coil.	57
3.1	Timing of a PGSE sequence.	63
3.2	Creation of the ROI via orthogonal slices	65
3.3	Relationship among time, frequency and the absolute domain	67
3.4	AGPS sequence	68
3.5	STEAM sequence	70
3.6	Schematic diagram of white matter fibres. Macroscopic.	73
3.7	Schematic diagram of the dialysis filter.	76
3.8	Design of gradient coil.	79

3.9	Design of coil holder.	80
3.10	Switching circuit.	83
3.11	Entire circuit diagram.	85
3.12	Transverse image of Soleus Muscle.	90
3.13	Artifacts from averaging in complex space.	92
3.14	Computer model of 2-D white matter.	97
3.15	Computer model of 2-D leg, and tortuosity simulations.	98
3.16	Flow channel computer model for leg data simulations.	100
4.1	Frequency domain profiles and attenuation plot for white matter.	104
4.2	Diffusion profiles for $T_d = 13.7$ ms to $T_d = 37$ ms.	107
4.3	D_a vs. $T_d^{1/2}$ for 1 human volunteer.	111
4.4	D_a vs. $T_d^{1/2}$. Average of 6 volunteers.	112
4.5	D vs. $T_d^{1/2}$ for water and acetone at 22°C	115
4.6	b -value attenuation plot for water and acetone at $T_d = 50.6$ ms.	116
4.7	Dialysis phantom results for water.	117
4.8	Convergence of Monte Carlo by iterations.	119
4.9	Convergence of Monte Carlo with step size.	120
4.10	Monte Carlo results for $a = 5 \mu\text{m}$ and $a = 20 \mu\text{m}$	121
4.11	Monte Carlo results on white matter.	122
4.12	Monte Carlo comparison with Szafer's data.	124
4.13	Monte Carlo comparison with Tanners equation.	126
4.14	Monte Carlo and Tanners equation, varying a	127
4.15	Monte Carlo and Mica stack data.	129
4.16	Monte Carlo and dialysis filter data.	130
4.17	Coil Calibration. G vs. I	133
4.18	Experimental gradient coil error map.	134
4.19	Theoretical gradient coil error map.	135
4.20	Frequency domain profiles and attenuation plot for leg muscle.	137
4.21	Water phantom diffusion weighted profiles. Using leg coil.	138

4.22	Diffusion profiles for $T_d = 4$ ms to $T_d = 50$ ms.	140
4.23	D_a vs. T_d for 1 and 7 volunteers in human leg muscle.	141
4.24	D vs. T_d for water and acetone.	142
5.1	1-D and 2-D simulations using extrapolated data.	149
5.2	Monte Carlo simulations, varying P	151
5.3	Monte Carlo simulations, varying f	152
5.4	Monte Carlo simulations, varying a	153
5.5	Adjusted D_a vs. $T_d^{1/2}$, corrected for parallel fibre component	156
5.6	Adjusted central-AP/posterior-RL data and MC simulation.	158
5.7	33-point decay curves for $T_d = 513$ ms in white matter.	160
5.8	33-point decay curves for $T_d = 50$ ms in white matter.	162
5.9	D_a vs. TE in human white matter in vivo.	166
5.10	Monte Carlo flow geometry for the head data.	170
5.11	Monte Carlo flow data and head experimental data.	171
5.12	Schematic diagram. Structure of skeletal muscle.	174
5.13	1-D and 2-D Monte Carlo fits to the leg data.	176
5.14	Flow Monte Carlo data with experimental data	179

Chapter 1

Introduction

Magnetic Resonance Imaging (MRI) is revolutionizing clinical diagnostic radiology. Sharp, detailed images of the internal human body provide radiologists with unprecedented soft tissue contrast. This versatile tool can noninvasively evaluate tissue anatomy, pathology, metabolism, and flow. There are also numerous [1, 2, 3, 4, 5] innovative new applications under development which may prove clinically useful in the future.

The ability of MRI to detect diffusion of molecules in the human body may be of enormous clinical benefit. Diseases such as multiple sclerosis (MS) almost certainly affect the diffusion of water in brain tissue. Stroke is known to significantly affect the diffusion coefficient of water in brain [6]. It may be possible, then, to use diffusion sensitive MRI scans to achieve early detection of these pathologies. With early detection, aggressive therapy [7, 8] to reverse or prevent the damage could be an option.

More information about the basic processes of water diffusion in tissue is needed before the full potential of diffusion MRI can be realized. The purpose of this work is to obtain a better knowledge of water diffusion in vivo, so that a foundation for clinical protocols may be laid for specific pathologies with a better

understanding of the information that they contain.

1.1 History of MRI

The concept that certain atomic nuclei have magnetic moments was first postulated by W. Pauli in the 1920's, but it was not until 1939 that the effect of resonance in nuclear magnetic moments was first observed in particle beams by Rabi *et al.* [9]. In 1946, the effect was demonstrated in bulk matter by the groups of Bloch at Stanford [10] and Purcell at Harvard [11]. Both Bloch and Purcell received the Nobel Prize for elucidating the phenomenon of nuclear magnetic resonance (NMR) in solids and liquids [12].

The sensitivity of NMR to diffusion was first discovered in 1950 by Hahn [13] and techniques for measuring the self diffusion coefficient using NMR soon followed this discovery [14, 15]. In 1965 the PGSE (Pulsed Gradient Spin Echo) technique, which was an important improvement over the previous methods, was introduced by Stejskal and Tanner [16]. The most commonly used techniques for measuring diffusion today are still closely related to this original sequence. The concept of obtaining diffusion images was first suggested by Mansfield [17] in 1982, but was not implemented until 1985 by several researchers [18, 19, 20] independently.

Until 1972, the clinical potential of magnetic resonance (MR) was limited because there was no procedure to provide spatial localization of the signal. Lauterbur solved this problem by using magnetic field gradients and mathematical methods that had previously been developed for X-ray computed tomography (CT) [21] to reconstruct the first proton nuclear spin density map using NMR [22]. A key element in the growth of tomographic techniques has been the availability and improvement of high speed computers. Even today, the advances made in computer

power allow some MRI clinical protocols (such as maximum intensity projection (MIP) angiography) to be feasible whereas a few years ago they were not.

In 1973, Mansfield and Grannell [23] demonstrated the Fourier relationship between the spin density and the NMR signal acquired in the presence of a small magnetic field gradient. Today, Fourier transform techniques have replaced reconstructive tomography as the method used to provide spatial localization in the image. There have been many technical innovations, such as 2 dimensional (2D) single slice acquisition using selective irradiation, 2D multislice acquisition, 3 dimensional (3D) data acquisition, fast scanning [24] with gradient echo and echo planar imaging, angiography, and image enhancement with injections of contrast agents, to name a few.

1.1.1 Chronicle of Diffusion Measurements in the Brain

The first diffusion measurements in the human brain in vivo were by Le Bihan *et al.* [18] in 1985. Since then many researchers such as Turner [25], Moseley [6], Wesbey [26], Thomsen [27], Henriksen [27], Chenevert [28], and more recently Gore [7], Szafer [7], Bassar [29], Pierpaoli [30], Horsfield [31], Latour [32], Henkelman [33], Allen [34], Larsson [35], and Hajnal [36], have studied the effects of water diffusion in humans in vivo. Unfortunately, the results from these studies vary immensely. The apparent diffusion coefficient, D_a , measured from these works varies from $(0.15 - 1.8) \times 10^{-3} \text{mm}^2/\text{s}$. The main reason for such a large variation is the anisotropy of the white matter fibres to water mobility, explained in more detail in Sections 1.4 and 2.6. However, even when white matter directionality is taken into account, there is still a wide range of values for D_a . The reported values for D_a range from $(0.15 - 1.1) \times 10^{-3} \text{mm}^2/\text{s}$ when the diffusion is perpendicular to

the white matter fibres ($D_{a\perp}$), and when it is parallel ($D_{a\parallel}$), the values range from $(0.8 - 1.8) \times 10^{-3} \text{mm}^2/\text{s}$. While most studies confirm the anisotropy [37] of the white matter fibres by reporting $D_{a\parallel} > D_{a\perp}$, there is no clear agreement on the magnitude of D_a .

There are many possible reasons for the discrepancy among the studies. The most confounding factor is bulk motion, such as patient table vibration, contributions from cerebrospinal fluid (CSF) and blood pulsations, and unintentional motions of the volunteer (swallowing, coughing, etc.). Bulk motions such as these alter the phase in the signal, which leads to drastic artifacts in the image because the 2D fast Fourier transform (FFT) algorithm depends on the phase of the signal to reconstruct the image. To avoid this, some recent studies have used ‘fast’ imaging sequences to acquire the data in under a second [38], while others have introduced a ‘navigator echo’ technique to measure the phase offset due to motion, then correct for it during post processing [39, 40]. Some fast sequences such as echo planar imaging (EPI) are extremely demanding on gradient hardware and many sites are not capable of running these sequences. EPI sequences are also extremely sensitive to susceptibility variations within the patient. The resulting images are strongly distorted. This does not pose a problem for most regions of the brain but it can limit the usefulness of EPI sequences in many other parts of the body.

Cardiac gating is sometimes used to eliminate the effects of blood pulsations. Each scan is taken at the same point on the cardiac cycle, so that the phase offset is identical for each line and acquisition. However, cardiac gating can also introduce new problems since variations in a person’s heart rate can lead to small but significant signal intensity changes.

Linescan imaging [41, 42] is emerging as a popular method to avoid the 2D-FFT algorithm completely, and thus avoid bulk motion artifacts. The diffusion sequences used in this work are similar to the linescan techniques, except we only measure one line, or column. Thus, our data are not corrupted by the effects of bulk motion such as table vibrations and head movements, but it is still sensitive to ‘relative’ motions within the region of interest, which may arise from such things as CSF pulsations or blood perfusion.

Another explanation for the large variation in the value of D_a reported in the literature is that many diffusion studies are primarily concerned with demonstrating that the contrast in a diffusion image is from different values of $D_{a\parallel}$ and $D_{a\perp}$ in white matter, and D_a in grey matter. The accuracy of the magnitude of D_a seems to be of secondary concern. Incredibly, many studies [43, 44, 45], even recent ones [46, 47, 48, 49, 50], use only two points on the decay curve to obtain a value for D_a (see Fig. (4.1)), and it is rare to find a paper that has more than four points on the decay curve. The reason for this is that the purpose of such studies is to demonstrate new ‘techniques’, and/or they want to minimize acquisition time. A short acquisition time is an important consideration if diffusion imaging is to be used clinically.

Many studies fail to report the diffusion time of their sequence [36, 42, 45, 51, 52, 53]. This further exacerbates a comparison of the literature data since it is very likely that D_a depends on the time allowed for diffusion in the sequence. It is well known that diffusion within a system of semi-permeable barriers (such as white matter axons) yields apparent diffusion coefficients that depend on the time allowed for diffusion [7, 32, 54, 55]. It is reasonable then, to expect a time dependence for D_a in the human brain, since the myelin sheath of the axons may play a role as a

restricting structure. It wasn't until recently that Horsfield and McDonald [56] first experimentally demonstrated this time dependence in white matter. Later, Gates and Cameron [57], Horsfield, Lai, Barker, Turner *et al.* [58], and Le Bihan [37] all report an experimentally observed time dependence. One of the hypotheses of this thesis is that the myelin sheath and/or the axon membrane will restrict the mobility of water, and therefore cause a decline in D_a with increasing diffusion time.

In general, a tensor must be used to characterize diffusion. However, most MRI studies of diffusion, including this work, measure only one or two components of the tensor. The few works that report the eigenvalues of the diffusion tensor measured in human white matter appear [30, 59] to obtain values for $D_{a\parallel}$ and $D_{a\perp}$ that are at the high and low end, respectively, of the literature range. Unfortunately, tensor diffusion studies are costly in acquisition time since six measurements must be performed to fill in all elements of the tensor (see Section 2.6), therefore most tensor studies have few data points on the attenuation curve. The major thrust behind tensor diffusion imaging is to remove the anisotropy of the white matter fibres by computing an image based on the trace of the tensor, so again, accuracy in D_a is not the paramount issue.

In Table 1.1, we attempt to summarize the literature data by breaking them into categories which reflect the topics covered in this section: values for $D_{a\parallel}$, $D_{a\perp}$, and the trace of the diffusion tensor, $\text{Tr}(D_a)/3$, are averaged for those studies where the diffusion time is short (20-40 ms), medium (50-100 ms), and long (100-600 ms). All values are for human brain in vivo. In Fig. 1.1(a), we plot literature averages for $D_{a\parallel}$ and $D_{a\perp}$ as a function of diffusion time for non-tensor studies. In Fig. 1.1(b), we show corresponding values from studies that measure the full

diffusion tensor.

Table 1.1: Summary of literature data for D_a in human white matter in vivo. The units for all values are $10^{-3}\text{mm}^2/\text{s}$. The number of papers, N , from which the results are averaged, are given.

Grey matter: 1.09 ± 0.13 ($N=9$)			
White matter	$T_d = (20 - 40 \text{ ms})$	$T_d = (50 - 100 \text{ ms})$	$T_d = (100 - 600 \text{ ms})$
$D_{a }$	1.30 ± 0.15 ($N=5$)	1.18 ± 0.10 ($N=9$)	1.23 ± 0.18 ($N=3$)
$D_{a\perp}$	0.67 ± 0.10 ($N=7$)	0.60 ± 0.06 ($N=10$)	0.45 ± 0.16 ($N=3$)
$\text{Tr}(D_a)/3$	0.85 ± 0.08 ($N=3$)	0.80 ± 0.02 ($N=3$)	0.78 ± 0.23 ($N=2$)

1.2 NMR Characteristics

Hydrogen is the atom most commonly used in NMR imaging, and in this study we assume that our NMR signal comes from the hydrogen atoms in water molecules, due to the abundance of water in the human body. Hydrogen has the largest gyro-magnetic ratio, γ , (a quantity which depends on the size and shape of a nucleus) and also has the highest sensitivity for the NMR signal. Any nucleus with an odd number of either protons or neutrons can, in theory, produce an NMR signal. Table 1.2 shows various nuclei that can be studied with NMR along with their characteristics. Hydrogen also has a very high natural abundance. This, combined with its sensitivity, makes images obtained from hydrogen far superior in quality to images obtained using the other nuclei in Table 1.2.

1.3 Diffusion

The NMR method employed in this work to measure diffusion is in some ways, superior to tracer diffusion studies [60]. The central feature in tracer methodology involves the injection of a ‘labelled’ sample involved in a biological process. The

(a)

(b)

Figure 1.1: D_a vs. $T_d^{1/2}$ from various works by other authors. The data in (a) are from studies that do not use the tensor method approach. The data in (b) are from studies that measure the full diffusion tensor. The data are for D_a in human white matter in vivo, but not necessarily from the same region of the brain. The error in the data is the standard deviation of the mean from several papers, or it is the error reported in the study. For the points without error bars, only 1 reference could be found and that paper did not report an error.

Table 1.2: MR Characteristics of various elements.

Nucleus	γ (MHz/T)	Natural Isotopic Abundance	Relative Sensitivity	Spin
		(%)		
^1H	42.576	99.985	1	1/2
^2H	6.536	0.015	0.0096	1
^3He	32.432	0.0001	0.44	1/2
^{13}C	10.705	1.108	0.016	1/2
^{14}N	3.076	99.635	0.001	1
^{15}N	4.315	0.365	0.001	1/2
^{17}O	5.772	0.037	0.029	3/2
^{19}F	40.055	100	0.834	1/2
^{23}Na	11.262	100	0.093	3/2
^{31}P	17.236	100	0.066	1/2
^{33}S	3.266	0.74	0.0023	3/2
^{39}K	1.987	93.08	0.0005	3/2
^{129}Xe	11.776	26.44	0.0212	1/2

labelled sample is usually a radioactive, tritium labelled amino acid [61] or a ^{11}C synthesized molecule [62]. At least in liquids, the tracer's diffusion coefficient does not necessarily correspond to that of the major chemical species [63]. Furthermore, the NMR technique is noninvasive, so our 'sample' is not contaminated by tracer material.

1.4 Restricted Diffusion

It is really the *restricted* diffusion coefficient that is of biological interest. Cell membranes and other constituents of tissue limit the mobility of water. They hinder the motion so that, after a period of time, the molecules do not move as far as they otherwise would in a free system. The result is that the diffusion coefficient

appears to be smaller. A comparison of the measured apparent diffusion coefficient, D_a , with the *unrestricted* coefficient, D , can yield information on membrane permeability, cell size and orientation, and possibly the rate of fluid transport.

In human brain, water molecules diffuse in intracellular space, extracellular space, in the cerebrospinal fluid and within blood vessels. Within intra/extracellular space, tortuous pathways for the random motion exist, caused by structures such as the myelin sheath, the axonal membrane, cellular organelles, and large molecules within the cytoplasm.

It is of great interest to obtain diffusion coefficient measurements of water for very short diffusion times, T_d (~ 8 ms). The diffusion range of water for these short times is comparable with the dimensions of most biological cells, enabling one to probe the tissue microstructure [64]. By comparing the diffusive behaviour in the tissues at short diffusion times with those at long diffusion times, the effect of cell membranes, internal cell structures, and active transport mechanisms may be assessed. The fundamental processes of water mobility within cells are not well understood. Processes such as active and passive transport, membrane permeability, cellular organelle transport, and cytoplasm tortuosity all may play roles in the molecular displacement that is measured with MRI sequences.

There are many studies [65, 66, 67] of the diffusion coefficient in human tissue *in vivo*, as well as theoretical works which model tissue structure and the behaviour of the water molecule within it [7, 32, 33, 54, 68]. Most experimental works are of diffusion in human brain [28, 31, 64, 69]. The anisotropy of the diffusion coefficient in white matter is thought to result from restriction of the myelin sheath [64, 34]. This may ultimately lead to new information about the pathogenesis of demyelination in multiple sclerosis plaques [35]. It has also been shown that stroke greatly

alters the diffusion coefficient in the injured area of the brain [6, 7]. In the leg tissue, peripheral vascular disease and the effects of diabetes cause hydration [65] or swelling of the tissue. Characterizing the diffusion coefficients in healthy leg muscle will give reference values for future investigations of these pathologies. A better understanding of water diffusion in cells is needed before its full potential can be put to clinical use.

NMR techniques designed to measure the diffusion coefficient rely on the net average displacement of molecules, $\langle r \rangle$, to attenuate the NMR signal. The relative attenuation, $M/M(0)$, is used to evaluate the diffusion coefficient from the equation

$$\frac{M(b)}{M(0)} = \exp(-bD), \quad (1.1)$$

where b is the 'gradient factor', a measure of the sensitivity of the NMR sequence to diffusion [52]. Eq. (1.1) is derived, and the variable, b , is defined later in Section 2.4. D is the diffusion coefficient for a free homogeneous medium. However, in brain tissue, many structures can act as barriers to the displacement of the water [64]. Compartmentalization of intra and extracellular fluid may also result in a very inhomogeneous system. Under these circumstances, we must replace D in Eq. (1.1) with the apparent diffusion coefficient (D_a). D_a then, is expected to decrease in systems where diffusion is hindered [44, 55] since the water molecules cannot diffuse as far.

Studies of D_a for water in human brain tissue have revealed different values for white and grey matter [36], as well as anisotropy within the white matter [28]. D_a values measured parallel to the white matter fibres ($D_{a\parallel}$) are in general, larger [28, 31, 57, 70] than the values when the diffusion is measured perpendicular to the fibres ($D_{a\perp}$). A suggested explanation [28, 64] for the anisotropy is that the myelin

sheath impedes the motion of water molecules across it, either by providing a complex pathway through the many layers of membrane that make up the sheath, or by simply being a semi-permeable membrane to water. The axonal membranes and neurofibrils are also structures that could hinder motion [34] across the fibres, yet have little effect on motion longitudinally. Active transport mechanisms may also explain the anisotropy. Axonal transport along the fibre direction could elevate the observed $D_{\alpha\parallel}$.

In this thesis, we show that the anisotropy, $D_{\alpha\parallel}/D_{\alpha\perp}$, increases dramatically when the time allowed for diffusion increases. This is because more water molecules, both intra and extracellular, have time to encounter barriers to diffusion.

The anisotropy in white matter can also be exploited to determine the fibre tract orientation [29, 71, 72]. The diffusion tensor, $\bar{\mathbf{D}}$, in an anisotropic system will have its off diagonal elements equal to zero only when the laboratory frame (i.e. the axes of the diffusion sensitizing gradients) coincides with the ‘fibre frame’ [73]. When this is the case we can write

$$\bar{\mathbf{D}} = \bar{\mathbf{D}}' = \begin{pmatrix} D'_{xx} & 0 & 0 \\ 0 & D'_{yy} & 0 \\ 0 & 0 & D'_{zz} \end{pmatrix}. \quad (1.2)$$

where $\bar{\mathbf{D}}'$ is the diffusion tensor expressed in the fibre frame of reference. In general, this will not be the case, and $\bar{\mathbf{D}}$ must be diagonalized to the form in Eq. (1.2). The eigenvector of $\bar{\mathbf{D}}'$ which corresponds to the largest diagonal element will give the fibre tract orientation.

Although determining $\bar{\mathbf{D}}'$ is a good way to measure D'_{xx} and D'_{yy} , it is costly in acquisition time and post processing: six measurements [29, 74] must be taken to determine $\bar{\mathbf{D}}$. In this study, specific locations in the corona radiata have been

targeted because the predominant fibre orientation in these regions is already known [28, 31, 36]. Our sequences are sensitized to diffusion either parallel or perpendicular to the predominant fibre orientation and thus, our laboratory frame coincides with the fibre frame. The D_a values reported here therefore correspond to either D'_{xx} or D'_{yy} in Eq. (1.2).

1.5 Experiments on Human Brain White Matter In Vivo

It is important to increase our knowledge of water transport in human tissue since diffusion weighted MR imaging (DWI) shows some promising clinical applications. It has been shown that D_a for brain tissue drops by as much as 30% within minutes [75, 76] of an ischemic injury. DWI then, is an imaging modality that can reveal the affected area in acute stroke long before any other imaging technique [8, 77]. This may have a major impact on the development of stroke therapies.

If indeed the myelin sheath plays a role in the anisotropy seen in human white matter, then our understanding of demyelinating diseases, such as multiple sclerosis, could benefit from studies of diffusion in human white matter [35].

In this study, we measure D_a in human white matter in vivo for a very large range of diffusion times, T_d (13.7 ms - 1020 ms). We obtain both $D_{a\perp}$ and $D_{a\parallel}$ for the same locations in the white matter. In order to effectively assess the behaviour of D_a over such a large range, we break up the range into three sections: (13.7 ms - 50 ms), (50 ms - 93 ms), and (170 ms - 1020 ms). For each section, we use a different NMR sequence (see Section 3.1) so as to optimize signal for that particular range. This technique has its drawbacks, which are discussed later in Chapter 5.

but it is the only way to obtain D_a over this large range, which, to our knowledge, has not been covered in a single study before [37, 56, 58, 64, 69]. We analyze our profiles for D_a vs. $T_d^{1/2}$ using analytical equations and Monte Carlo simulations. The analytical equations give upper and lower limits of the membrane permeability of the axon of $P=0.002$ cm/s and 0.019 cm/s, respectively. We find, however, that the Monte Carlo simulations reveal that human brain white matter is more complex than a simple two compartment system of intracellular and extracellular spaces. We find that our simulated model needs an extracellular ‘flow’ component added in order to obtain agreement with the experimental results. The implications of this will be discussed in Chapter 5.

1.6 Experiments on Human Leg Muscle In Vivo

In order to sensitize MRI sequences to diffusion at short diffusion times, large amplitude gradients are required [51]. The maximum gradient strength on most clinical scanners is 10-15 mT/m, which limits the diffusion time to ≥ 14 ms in practice [78]. In this work, we constructed a double Maxwell gradient coil for larger diffusion gradients and we show an inexpensive method to interface the coil to a clinical scanner.

Due to several practical constraints, it was decided not to build this coil large enough that it could be used for studies of diffusion in the brain. Instead, we decided to use it to study diffusion in calf muscle tissue. Muscle is also a highly oriented tissue and it was felt to be a good model system for studying anisotropic restricted diffusion in vivo.

We were only able to find one previous study of diffusion in human leg muscle, by Chenevert *et al.* [67]. Their data give $1.3 \times 10^{-3} \text{mm}^2/\text{s}$ as the apparent diffusion

coefficient for human calf muscle.

Diffusion coefficients in human forearm and finger muscle have been obtained by Morvan *et al.* [79] ($D_a = 1.71 \pm 0.05 \times 10^{-3} \text{mm}^2/\text{s}$) and Damion *et al.* [65] ($D_a = 1.15 \pm 0.05 \times 10^{-3} \text{mm}^2/\text{s}$) respectively. In rat and frog muscle, there have been several studies [80, 81, 82, 83] that report diffusion coefficients in the range $1.01 - 1.56 \times 10^{-3} \text{mm}^2/\text{s}$.

With the gradient coil that we built, we have measured novel data on the diffusion coefficient in human muscle in vivo, and we show that the current theoretical models do not support the experimental findings. However, similar to our findings in the white matter, when processes mimicking active transport and/or pseudo-flow are incorporated into the models, better agreement emerges.

Chapter 2

Theory

2.1 Nuclear Magnetic Resonance

Electrons, protons, and neutrons have an internal angular momentum, like a ball spinning on its own axis. In nuclear magnetic resonance, the measured signal is generated by the magnetic dipole moment, μ , of the nucleus. μ is related to the spin angular momentum, J by [84]

$$\mu = \gamma J, \quad (2.1)$$

where $\gamma = 2.675 \times 10^5 \text{mT}^{-1}\text{s}^{-1}$ is the nuclear gyromagnetic ratio for protons.

When a proton is placed inside a homogeneous magnetic field, B_o , its magnetic moment will tend to align itself with the field [85] with an angle, θ , between its axis and the field, B_o , as shown in Fig. 2.1(a). Due to the spin angular momentum, the dipole moment will precess about B_o , much like a gyroscope. In Fig. 2.1, the torque that causes μ to precess is exerted by B_o and is given by [84]

$$\tau = \mu \times B_o.$$

and

$$|\boldsymbol{\tau}| = \gamma J B_o \sin \theta. \quad (2.2)$$

Also, from Newton's second law of motion, we have

$$d\mathbf{J} = \boldsymbol{\tau} dt,$$

and

$$|d\mathbf{J}| = dJ = \gamma J B_o \sin \theta dt. \quad (2.3)$$

Thus, during an infinitesimal time, dt , the angular momentum will change by an amount $d\mathbf{J}$, and $\boldsymbol{\mu}$ will rotate through an angle $d\phi$. It can be seen from Fig. 2.1(b) that $d\phi$ is given by

$$\begin{aligned} d\phi &= \frac{dJ}{J \sin \theta} \\ &= \frac{\gamma J B_o \sin \theta dt}{J \sin \theta} \\ &= \gamma B_o dt. \end{aligned} \quad (2.4)$$

The frequency of precession of the proton is therefore given by

$$\frac{d\phi}{dt} = \omega = \gamma B_o. \quad (2.5)$$

Eq. (2.5) is the Larmor equation [60]. It states the fundamental relationship of NMR: that the proton resonant frequency is directly proportional to the magnetic field.

In our macroscopic sample, the bulk magnetization $\mathbf{M}$ may be taken as the sum of the individual magnetic moments [86]. Thus, from Eq. (2.1) we see that the macroscopic angular momentum is given by $\mathbf{M}/\gamma$. Since torque is the time rate of change of angular momentum, we can use Eq. (2.2) to get

Figure 2.1: Precession of the magnetic moment about a static field B_0 .

$$\frac{d\mathbf{M}}{dt} = \gamma \mathbf{M} \times \mathbf{B}. \quad (2.6)$$

If we observe the spin in a reference frame rotating with the spin precession, then the expression for $\frac{d\mathbf{M}}{dt}$ (in the laboratory frame) will include time derivatives of the rotating frame unit vectors. Thus [84]

$$\frac{d\mathbf{M}}{dt} = \left(\hat{x}' \frac{dM_{x'}}{dt} + \hat{y}' \frac{dM_{y'}}{dt} + \hat{z}' \frac{dM_{z'}}{dt} \right) + \left(M_{x'} \frac{d\hat{x}'}{dt} + M_{y'} \frac{d\hat{y}'}{dt} + M_{z'} \frac{d\hat{z}'}{dt} \right). \quad (2.7)$$

where $\hat{x}'$, $\hat{y}'$, $\hat{z}'$ are the unit vectors along the x' , y' , z' axes of the rotating frame, respectively, and $M_{x'}$, $M_{y'}$, $M_{z'}$ are the corresponding components of magnetization along these directions. For a reference frame rotating at angular frequency ω [84],

$$\frac{d\hat{x}'}{dt} = \boldsymbol{\omega} \times \hat{x}', \quad (2.8)$$

and similarly for $\hat{y}'$ and $\hat{z}'$. Then Eq. (2.7) is

$$\frac{d\mathbf{M}}{dt} = \frac{\partial \mathbf{M}}{\partial t} + (\boldsymbol{\omega} \times \mathbf{M}), \quad (2.9)$$

where the notation $\frac{\partial}{\partial t}$ is used here to designate a time derivative in the rotating frame of reference. Substituting this expression into Eq. (2.6) we get

$$\frac{\partial \mathbf{M}}{\partial t} = \gamma \mathbf{M} \times (\mathbf{B} + \boldsymbol{\omega}/\gamma). \quad (2.10)$$

Eq. (2.10) says that in the rotating frame, the magnetization precesses about an effective field $\mathbf{B} + \boldsymbol{\omega}/\gamma$. Thus, entering the rotating frame with a frequency $\boldsymbol{\omega} = \gamma \mathbf{B}$ is equivalent to adding a fictitious field $\boldsymbol{\omega}/\gamma$. If the field $\mathbf{B} = B_0 \hat{z}$ is our main NMR field, choosing $\boldsymbol{\omega} = -\gamma \mathbf{B}_0$ will cause the effective field in the rotating frame to vanish. Eq. (2.10) is then $\frac{\partial \mathbf{M}}{\partial t} = 0$, which means that the magnetization in the rotating frame is a static vector.

We will now consider the effect of a pulse of radio frequency (rf) radiation on the spin system. If we were to apply a magnetic field $\mathbf{B}_1$, which is perpendicular to $\mathbf{B}_0$, and rotates about $\hat{z}$ with frequency $\boldsymbol{\omega} = \omega_0 = -\gamma \mathbf{B}_0$, the effective field in Eq. (2.10) would no longer vanish and we would get [87]

$$\frac{\partial \mathbf{M}}{\partial t} = \gamma \mathbf{M} \times \mathbf{B}_1. \quad (2.11)$$

For $\mathbf{B}_1$ applied along $\hat{x}'$ and an initial condition $\mathbf{M}(0) = M_0 \hat{z}'$, the solution to this equation is [84]

$$\begin{aligned} M_{x'} &= 0 \\ M_{y'} &= M_0 \sin \omega_1 t \\ M_{z'} &= M_0 \cos \omega_1 t, \end{aligned} \quad (2.12)$$

Figure 2.2: The effect of a $\pi/2$ or 90° pulse is to tip the magnetization into the x-y plane. (a) shows the rotating reference frame with axes x' , y' , z' and (b) shows the laboratory frame with axes x , y , z .

where $\omega_1 \equiv -\gamma B_1$. The magnetization precesses about $\hat{x}'$ for the duration of the B_1 pulse. If the rf pulse is on for a time t such that $\omega_1 t = \pi/2$, then the magnetization lies along $\hat{y}'$ after the rf pulse. In the laboratory frame, the effect of the $\pi/2$ or 90° pulse is to tip the magnetization from $\hat{z}$ into the x-y plane, as shown in Fig. 2.2. In Fig. 2.2(b), the helical trajectory of the magnetization vector in the laboratory frame is shown.

If we apply a 180° pulse (i.e. $\omega_1 t = \pi$), then from Eq. (2.12) we see that the magnetization will be along $-\hat{z}'$. Thus a 180° pulse flips the magnetization from $+\hat{z}'$ to $-\hat{z}'$. We will demonstrate this using quantum mechanics in the next section.

If the rf radiation is not applied on resonance, then Eq. (2.10) becomes

$$\frac{\partial \mathbf{M}}{\partial t} = \gamma \mathbf{M} \times (\mathbf{B}_1 + \Delta \mathbf{B}_o), \quad (2.13)$$

where $\Delta \mathbf{B}_o = \Delta B_o \hat{z}$ is the field offset from resonance. The magnetization will

thus precess about the effective field $B_1 + \Delta B_0$.

2.1.1 Quantum Mechanical Description

The NMR signal comes from nuclei which have internal angular momentum, like a ball spinning on its own axis. The magnetic dipole moment, μ , can be viewed as arising from the current loop formed by the rotating positive charge on the spinning nucleus [88]. In MRI, the signal usually comes from hydrogen nuclei which consist of a single proton.

The magnetic moment of the proton has an intrinsic spin angular momentum, described by a vector operator [89]

$$\mathbf{S} = S_x \hat{x} + S_y \hat{y} + S_z \hat{z}. \quad (2.14)$$

One of the properties of all spin 1/2 particles is that they have only 2 eigenstates, say $|z+\rangle$ and $|z-\rangle$. A measurement of the component of the angular momentum along $\hat{z}$ would give a value of either $+\hbar/2$ or $-\hbar/2$, the eigenvalues of $|z+\rangle$ and $|z-\rangle$, respectively [89]. Hence

$$\begin{aligned} S_z |z+\rangle &= \hbar/2 |z+\rangle \\ S_z |z-\rangle &= -\hbar/2 |z-\rangle. \end{aligned} \quad (2.15)$$

These two eigenstates form a complete basis for the description of the proton spin [89]. That is, any state of the spin, $|w\rangle$, must be expressible as a linear combination of these two states, i.e.

$$|w\rangle = c|z+\rangle + d|z-\rangle, \quad (2.16)$$

where c and d are [87] complex numbers such that $|c|^2 + |d|^2 = 1$.

Figure 2.3: The energy levels of a proton are split by the presence of a magnetic field.

The interaction energy of the magnetic dipole moment, μ , in a magnetic field B_o , is [60] $-\mu \cdot B_o$. When B_o is along the $\hat{z}$ direction, the Hamiltonian operator becomes [86]

$$\mathcal{H}_{spin} = -\mu_z B_o = -\gamma S_z B_o. \quad (2.17)$$

Thus, $\mathcal{H}_{spin}$ will have energy eigenvalues related to the eigenvalues of S_z by a proportionality constant $-\gamma B_o$. The energy eigenvalues are thus [84]

$$\begin{aligned} E_{+\frac{1}{2}} &= -\gamma \frac{\hbar}{2} B_o. \\ E_{-\frac{1}{2}} &= +\gamma \frac{\hbar}{2} B_o. \end{aligned} \quad (2.18)$$

for spins in the $|z+\rangle$ and $|z-\rangle$ states, respectively. The energy difference between the two states is $\Delta E = \gamma \hbar B_o$. This is the Zeeman splitting, represented

in Fig. 2.3. In order to excite a transition from one state to another, a quantum of energy

$$\Delta E = \hbar\omega = \gamma\hbar B_o \quad (2.19)$$

is needed. Here, ω is the angular frequency of the applied radiation. Thus, the absorption frequency, $\omega = \gamma B_o$, is identical to the frequency of precession [84].

Consider the spin in our NMR experiment, subject to a homogeneous magnetic field, $\mathbf{B}_o$ along $\hat{z}$. The classical picture starts with the spin aligned parallel to the field. An rf excitation pulse, described in Section 2.1, tips the magnetization from the $\hat{z}$ direction into the x-y plane. It is the rotating magnetic field associated with the magnetization that induces the signal from which all data from a magnetic resonance imager are obtained. Even when the macroscopic magnetization lies in the x-y plane, $|x_{\pm}^{\pm}\rangle$ and $|y_{\pm}^{\pm}\rangle$, the eigenstates of S_x and S_y , can be described completely by a combination of $|z+\rangle$ and $|z-\rangle$, or, by Eq. (2.16). In fact, it can be shown that [90]

$$\begin{aligned} |x_{\pm}^{\pm}\rangle &= \frac{1}{\sqrt{2}}|z+\rangle_{\pm} \pm \frac{1}{\sqrt{2}}|z-\rangle_{\pm} \\ |y_{\pm}^{\pm}\rangle &= \frac{1}{\sqrt{2}}|z+\rangle_{\pm} \pm \frac{i}{\sqrt{2}}|z-\rangle_{\pm}. \end{aligned} \quad (2.20)$$

Also unlike the classical picture, where the spin pointing up is merely in the opposite direction to the spin pointing down, the $|z+\rangle$ and $|z-\rangle$ are orthogonal vectors, that is [90],

$$\langle -z | z+ \rangle = 0, \quad (2.21)$$

which must be the case if $|z+\rangle$ and $|z-\rangle$ are to form a complete basis. The situation is formally analogous to photon polarization, where spin ‘up’ and ‘down’

are like linear polarization in directions perpendicular to each other [89]. The $|x_{\pm}^{\pm}\rangle$ and $|y_{\pm}^{\pm}\rangle$ eigenstates are analogous to right and left circularly polarized light.

Let us further illustrate the quantum behaviour of the proton spin by considering the effect of a $\pi/2$ rf pulse applied to a proton initially in the $|z+\rangle$ state within our MRI magnet. As in Section 2.1, let us apply a rotating radio frequency magnetic field to the spin. Since the spin precesses about $\hat{z}$ with frequency ω_o , if we apply a magnetic field

$$B_x = B_1 \cos \omega t, \quad B_y = -B_1 \sin \omega t. \quad (2.22)$$

where $\omega \approx \omega_o$, in the rotating frame of the spin the field B_1 appears not to be rotating at all. Effectively then, the spin feels only the weak field B_1 and precesses about it at $\omega_1 = \gamma B_1$. In the laboratory frame where the spin experiences both B_o and B_1 , the resulting precession is more complex.

The Hamiltonian of the spin in this case becomes

$$\mathcal{H}_{spin} = -\gamma \mathbf{B} \cdot \mathbf{S} = -\gamma [B_o S_z + B_1 [\cos \omega t S_x - \sin \omega t S_y]]. \quad (2.23)$$

and the Schrodinger equation for the spin is [90]

$$i\hbar \frac{d|\Psi(t)\rangle}{dt} = \mathcal{H}_{spin} |\Psi(t)\rangle. \quad (2.24)$$

where $|\Psi(t)\rangle$ is the time dependent spin state. Finding a solution to this equation in the laboratory frame of reference is not easy. However, the problem can be simplified considerably by transforming to a frame of reference that rotates about the $\hat{z}$ axis with an angular frequency, Ω [89]. To obtain the wave function for the

spin state in the rotating frame. $|\Psi'(t)\rangle$, we must apply the rotation operator [89] $e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t}$ to $|\Psi(t)\rangle$ to get:

$$|\Psi(t)\rangle = e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} |\Psi'(t)\rangle. \quad (2.25)$$

where, for $\mathbf{\Omega} = -\Omega\hat{z}$, $|\Psi(t)\rangle$ rotates about $\hat{z}$ with angular velocity $-\Omega$. Then,

$$\frac{d|\Psi(t)\rangle}{dt} = \frac{-i}{\hbar} \mathbf{\Omega} \cdot \mathbf{S} e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} |\Psi'(t)\rangle + e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} \frac{d|\Psi'(t)\rangle}{dt}. \quad (2.26)$$

and Eq. (2.24) becomes

$$\begin{aligned} i\hbar \frac{d|\Psi'(t)\rangle}{dt} &= \left[e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} \mathcal{H}_{spin} e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} - \mathbf{\Omega} \cdot \mathbf{S} \right] |\Psi'(t)\rangle \\ &= \left[-\gamma e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} [B_o S_z + B_1 (\cos \omega t S_x - \sin \omega t S_y)] e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} + \right. \\ &\quad \left. \Omega S_z \right] |\Psi'(t)\rangle \\ &= \left[-\gamma \left[B_o S_z + B_1 (\cos \omega t e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} S_x e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} - \sin \omega t e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} S_y \right. \right. \\ &\quad \left. \left. e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} \right] + \Omega S_z \right] |\Psi'(t)\rangle. \end{aligned} \quad (2.27)$$

Now, the effect of the term $e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} S_x e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t}$ is just to rotate S_x about $\hat{z}$ by $+\Omega t$ [89] (and similarly for $e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t} S_y e^{\frac{i}{\hbar}\mathbf{\Omega} \cdot \mathbf{S} t}$). Thus

$$\begin{aligned} i\hbar \frac{d|\Psi'(t)\rangle}{dt} &= [-\gamma [B_o S_z + B_1 [\cos(\omega - \Omega)t \cdot S_x - \sin(\omega - \Omega)t \cdot S_y]] + \Omega S_z] |\Psi'(t)\rangle \\ &= [(\Omega - \omega_o) S_z - \gamma B_1 [\cos(\omega - \Omega)t \cdot S_x - \sin(\omega - \Omega)t \cdot S_y]] |\Psi'(t)\rangle. \end{aligned} \quad (2.28)$$

By convention, the frequency of the rotating frame, Ω , is always chosen to be equal to the frequency of the rf pulse, ω , which is not necessarily equal to ω_o , the

precessional frequency. Following this convention and setting $\Omega = \omega$. Eq. (2.28) reduces to

$$i\hbar \frac{d|\Psi'(t)\rangle}{dt} = [(\omega - \omega_o)S_z - \omega_1 S_x] |\Psi'(t)\rangle, \quad (2.29)$$

where $\omega_1 \equiv \gamma B_1$.

As expected, transforming to a rotating frame results in a static Hamiltonian. This corresponds to a coupling of the spin with an effective static field

$$\mathbf{B}_{eff} = \frac{1}{\gamma} [\omega_1 \hat{x}' - (\omega - \omega_o) \hat{z}']. \quad (2.30)$$

and the spin will precess about $\mathbf{B}_{eff}$ at a frequency $\gamma \mathbf{B}_{eff}$. On resonance ($\omega = \omega_o$), we have $\mathbf{B}_{eff} = \frac{\omega_1}{\gamma} \hat{x}' = B_1 \hat{x}'$, so in the rotating frame the spin will precess around the $\hat{x}'$ -axis.

Eq. (2.29) clearly has the solution [89]

$$|\Psi'(t)\rangle = e^{\frac{i}{\hbar} [(\omega - \omega_o)S_z - \omega_1 S_x] t} |\Psi'(0)\rangle, \quad (2.31)$$

and on resonance,

$$|\Psi'(t)\rangle = e^{\frac{i}{\hbar} \omega_1 S_x t} |\Psi'(0)\rangle. \quad (2.32)$$

If we substitute this into Eq. (2.25), we get

$$|\Psi(t)\rangle = e^{\frac{i}{\hbar} \omega_o S_z t} e^{\frac{i}{\hbar} \omega_1 S_x t} |\Psi(0)\rangle \quad (2.33)$$

since $|\Psi(0)\rangle = |\Psi'(0)\rangle$. This is the solution to Eq. (2.24). Since the exponential factors in this expression are also rotation operators [89] we can see from this equation that the spin state in the laboratory frame precesses very rapidly about the z-axis while simultaneously precessing about the x-axis at a much slower rate.

We can now make use of this result to compute the probability that a spin, initially in the state $|z+\rangle$, has flipped to $|z-\rangle$ as a result of the rf pulse. We begin by setting

$$|\Psi(0)\rangle = |z+\rangle. \quad (2.34)$$

The amplitude for finding the system in state $|z-\rangle$ at time t is

$$\langle -z | \Psi(t) \rangle = \langle -z | e^{\frac{i}{\hbar} \omega_o t S_z} e^{\frac{i}{\hbar} \omega_1 t S_x} | z+ \rangle, \quad (2.35)$$

which reduces to [89]

$$\langle -z | \Psi(t) \rangle = e^{-i\omega_o t/2} \langle -z | \left(\cos \frac{\omega_1 t}{2} + i \sigma_x \sin \frac{\omega_1 t}{2} \right) | z+ \rangle, \quad (2.36)$$

where $S_z \equiv \frac{\hbar}{2} \sigma_z$ and $S_x \equiv \frac{\hbar}{2} \sigma_x$ and σ_x, σ_z are the Pauli spin matrices $\boldsymbol{\sigma} = (\sigma_x, \sigma_y, \sigma_z)$ given by [89]

$$\sigma_x = \begin{pmatrix} 0 & 1 \\ 1 & 0 \end{pmatrix}, \quad \sigma_y = \begin{pmatrix} 0 & -i \\ i & 0 \end{pmatrix}, \quad \sigma_z = \begin{pmatrix} 1 & 0 \\ 0 & -1 \end{pmatrix}. \quad (2.37)$$

Continuing with Eq. (2.36) we get

$$\begin{aligned} \langle -z | \Psi(t) \rangle &= i e^{-i\omega_o t/2} \langle -z | \sigma_x | z+ \rangle \sin \frac{\omega_1 t}{2} \\ &= i e^{-i\omega_o t/2} \sin \frac{\omega_1 t}{2}. \end{aligned} \quad (2.38)$$

The probability that the spin has flipped by time t , shown in Fig. 2.2, is therefore

$$P_{\downarrow}(t) = |\langle -z | \Psi(t) \rangle|^2 = \sin^2 \frac{\omega_1 t}{2} = \frac{1}{2} (1 - \cos \omega_1 t). \quad (2.39)$$

As expected, a 180° pulse ($\omega_1 t = \pi$) flips the spin with certainty [89]. A 90° pulse, however, would flip the spin with a probability equal to $\frac{1}{2}$. Since $|z+\rangle$ and

$|z-\rangle$ are the only two possible states [89], then the spin will remain in the $|z+\rangle$ state with a probability equal to $\frac{1}{2}$ as well. Since the $|x_{\pm}\rangle$ and $|y_{\pm}\rangle$ eigenstates in Eq. (2.20) are just linear combinations of $|z+\rangle$ and $|z-\rangle$, the statement that a spin initially along $|z+\rangle$ has turned with certainty onto the x-y plane is the same as saying the spin is ‘up’ with a probability equal to $\frac{1}{2}$ and ‘down’ with a probability equal to $\frac{1}{2}$. The ‘mixing’ of the $|z+\rangle$ and $|z-\rangle$ states to define $|x_{\pm}\rangle$ and $|y_{\pm}\rangle$ states, resulting in a net magnetic moment in the x-y plane, is possible only because the spin 1/2 space is a complex 2-dimensional vector space [90].

2.1.2 Equilibrium Distribution of Spin States

An rf pulse applied to a ‘sample’ of protons will excite transitions from $|z+\rangle$ to $|z-\rangle$ and from $|z-\rangle$ to $|z+\rangle$ with equal probability. In order for there to be a net transition, or a net bulk signal, there must initially be a difference in population between the two states.

At thermal equilibrium, the spin populations of the Zeeman energy levels will be given by the Boltzmann distribution. If there are $n_{\uparrow}$ spins in the $|z+\rangle$ state and $n_{\downarrow}$ spins in the $|z-\rangle$ state, then [84]

$$\begin{aligned}\frac{n_{\downarrow}}{n_{\uparrow}} &= \exp(-\Delta E/k_B T) \\ &= \exp(-\gamma \hbar B_o/k_B T).\end{aligned}\tag{2.40}$$

Here, k_B is the Boltzmann’s constant and T is the absolute temperature. Since the total number of spins is

$$n = n_{\uparrow} + n_{\downarrow},\tag{2.41}$$

the difference in population is given by

$$n_{\uparrow} - n_{\downarrow} = n \frac{\{1 - \exp(-\gamma \hbar B_o / kT)\}}{\{1 + \exp(-\gamma \hbar B_o / kT)\}}. \quad (2.42)$$

In a magnetic field of 1.5 T and for a body temperature $T=310$ K, the fractional excess of spins in the low energy state is only $\sim 5 \times 10^{-6}$. The bulk magnetization is large enough to be measured, however, due to the enormous value of n . Still, the signal to noise ratio (S/N) in NMR becomes a problem when the volume of interest is less than 1 mm^3 or so. The small fractional excess of spins arises because the Zeeman energy, $\gamma \hbar B_o$, is small compared with the thermal energy quanta, which are of magnitude $k_B T$ [84].

2.2 Diffusion: Fick's Law and the Einstein Equation

Diffusion is the result of the random, thermal motion of molecules. Each molecule has a characteristic mean thermal speed, or drift velocity [91]. Eventually, the molecule will travel, on average, a distance equal to its mean free path before it collides with a neighbouring molecule. This motion, also known as Brownian motion, arises due to random collisions with other molecules in the system [92].

Consider a system of water molecules in a magnetic field $\mathbf{B}_o = B_o \hat{z}$, where the hydrogen nuclear spin magnetic moments are all precessing about $\mathbf{B}_o$ with the same frequency and phase. Suppose we managed to cause n_1 molecules per unit volume to be out of phase with the remaining molecules. Our system would then have n_1 labelled molecules per unit volume¹. Further, let the distribution of these molecules be nonuniform, so that we have a nonequilibrium system, where n_1 is a

¹Here, we are assuming that the phase of the molecules is not affected by the local magnetic fields at the atomic level.

function of position. This system will tend to increase its entropy by a net motion of labelled molecules, making the concentration n_1 more uniform. If we denote the flux of the labelled molecules by J_f , and define the flux as the net number of labelled molecules crossing a unit area of a plane per unit time, then we expect that J_f should be proportional to the concentration gradient, or

$$J_f = -D \frac{\partial n_1}{\partial z}, \quad (2.43)$$

where D is the proportionality constant, which we define as the diffusion coefficient. If $\frac{\partial n_1}{\partial z} = 0$, then the distribution of labelled molecules is uniform and there is no net flux. If $\frac{\partial n_1}{\partial z} < 0$, the flow of labelled molecules is in the $\hat{z}$ direction so as to equalize the concentration. Thus the minus sign in Eq. (2.43) is introduced to make D a positive quantity. Eq. (2.43) is Fick's law, which describes the self diffusion of molecules in gases, liquids, or isotropic solids [92].

We can also apply the 'conservation of mass' principle, shown in Fig. 2.4. The number of molecules in the middle slab will be $An_1(z)dz$, where A is the area of the plane and dz is the thickness of the slab. The change in this number per unit time will be equal to [the net number of labelled molecules entering the slab per unit time] minus [the net number of labelled molecules leaving the slab per unit time]. In symbols, this is [92]

$$\begin{aligned} \frac{\partial}{\partial t}(n_1 A dz) &= AJ_f(z) - AJ_f(z + dz) \\ \frac{\partial n_1}{\partial t} dz &= J_f(z) - \left[J_f(z) + \frac{\partial J_f}{\partial z} dz \right] \\ \frac{\partial n_1}{\partial t} &= -\frac{\partial J_f}{\partial z}. \end{aligned} \quad (2.44)$$

Combining this with Eq. (2.43), we obtain the diffusion equation

Figure 2.4: Illustration of the conservation of the number of molecules during diffusion.

$$\frac{\partial n_1}{\partial t} = D \frac{\partial^2 n_1}{\partial z^2}. \quad (2.45)$$

The solution to this equation for a boundary condition $n_1(z) \rightarrow 0$ as $z \rightarrow \infty$ and with an initial condition $n_1(z, t = 0) = \delta(z - z_o)$ is [93]

$$n_1(z, t) = (4\pi Dt)^{-1/2} \exp \left[-(z - z_o)^2 / 4Dt \right], \quad (2.46)$$

which is a Gaussian distribution centred at z_o , the position of the molecules at time zero. Eq. (2.46) represents the spreading of the number density as a function of time. However, for a single molecule originally at z_o , the probability, $P(z, t)$, of finding it at z at some later time, t , follows the same Gaussian distribution [52]. $P(z, t)$ then, depends only on the net displacement $(z - z_o)$, and not on the initial position z_o . It is this net displacement which is actually measured in a diffusion NMR experiment. That is, the net average displacement of a very large number of spins is measured.

In a free, homogeneous medium, each molecule will diffuse according to Eq. (2.46). However, it is the average squared displacement, $\overline{(z - z_o)^2}$, that is used to deter-

mine the diffusion coefficient experimentally. From the definition of an average [92], we have

$$\overline{(z - z_o)^2} = \frac{1}{N_1} \int_{-\infty}^{\infty} (z - z_o)^2 n_1(z, t) dz, \quad (2.47)$$

where N_1 , the total number of particles in the system, is defined by [92]

$$N_1 = \int_{-\infty}^{\infty} n_1(z, t) dz. \quad (2.48)$$

For simplicity, we can take $z_o = 0$ and multiply the diffusion equation, Eq. (2.45), by z^2 and integrate over z . This gives

$$\int_{-\infty}^{\infty} z^2 \frac{\partial n_1}{\partial t} dz = D \int_{-\infty}^{\infty} z^2 \frac{\partial^2 n_1}{\partial z^2} dz. \quad (2.49)$$

The left side gives [92], using Eq. (2.47)

$$\int_{-\infty}^{\infty} z^2 \frac{\partial n_1}{\partial t} dz = \frac{\partial}{\partial t} \int_{-\infty}^{\infty} z^2 n_1 dz = N_1 \frac{\partial}{\partial t} (\overline{z^2}). \quad (2.50)$$

The right side of Eq. (2.49) can be simplified by using integration by parts,

$$\begin{aligned} \int_{-\infty}^{\infty} z^2 \frac{\partial^2 n_1}{\partial z^2} dz &= \left[z^2 \frac{\partial n_1}{\partial z} \right]_{-\infty}^{\infty} - 2 \int_{-\infty}^{\infty} z \frac{\partial n_1}{\partial z} dz \\ &= 0 - 2[z n_1]_{-\infty}^{\infty} + 2 \int_{-\infty}^{\infty} n_1 dz \\ &= 0 + 2N_1, \end{aligned} \quad (2.51)$$

since we know by its Gaussian nature that n_1 and $\partial n_1 / \partial z \rightarrow 0$ as $|z| \rightarrow \infty$. Thus, by inserting Eqs. (2.50) and (2.51) back into Eq. (2.49), we find

$$\frac{\partial}{\partial t} (\overline{z^2}) = 2D. \quad (2.52)$$

or

$$\overline{z^2} = 2Dt. \quad (2.53)$$

Eq. (2.53) is the Einstein equation [52] for diffusion in one dimension, which we will use to obtain the diffusion coefficient experimentally. The Einstein equation holds only for a free and homogeneous system. For our diffusion measurements in human brain and leg muscle, the system is neither free nor homogeneous. However, for systems such as this, Eq. (2.53) is used to define an apparent diffusion coefficient (D_a) which is very useful in interpreting these results.

Eqs. (2.46) and (2.53) were derived for diffusion in a 1-D system. They are, however, the correct formulas to use in our NMR experiments because we sensitize the measurements to diffusion in one direction only, i.e. the direction of the diffusion sensitizing gradient, G_D . For a 3-D system, the equation for the probability distribution of a molecule is [52]

$$P(\mathbf{r}, t) = (4\pi Dt)^{-3/2} \exp \left[-(\mathbf{r} - \mathbf{r}_0)^2 / 4Dt \right], \quad (2.54)$$

where $P(\mathbf{r}, t)$ is the probability of finding the molecule, initially at $\mathbf{r}_0$, at a position $\mathbf{r}$ at some later time t . The 3-D Einstein equation [52] (setting $\mathbf{r}_0 = 0$) is

$$\overline{r^2} = 6Dt. \quad (2.55)$$

2.3 Diffusion and Experimental NMR

The most widely used NMR sequence to measure diffusion is the simple bipolar pulsed gradient spin echo sequence (PGSE), first proposed by Stejskal and Tanner [16]. The PGSE sequence, shown in Fig. 2.5, has two diffusion sensitization gradients, G_D , of amplitude G , duration δ and separation Δ . The first G_D is placed

between the 90° and 180° rf pulses. It dephases the spins because it introduces a phase shift ϕ_1 , which depends on the spin position z [52].

$$\phi_1 = \gamma \int_0^\delta G z_1 dt = \gamma \delta G z_1. \quad (2.56)$$

The 180° pulse flips the spins so that ϕ_1 becomes $-\phi_1$ and the effect of the second G_D is to rephase the spins. The second G_D will produce a phase shift

$$\phi_2 = \gamma \int_\Delta^{\Delta+\delta} G z_2 dt = \gamma \delta G z_2, \quad (2.57)$$

where z_2 is the spin position during the second pulse. The net dephasing is thus

$$f(\phi) = \phi_2 - \phi_1 = \gamma \delta G (z_1 - z_2). \quad (2.58)$$

We can see from Eq. (2.58) that, for static spins (i.e. no diffusion), $z_1 = z_2$, and the gradients provide no net dephasing, and therefore, no net attenuation of the signal. For diffusing spins, however, the G_D 's provide an extra attenuation of the signal amplitude which depends on the spin history during the time interval, Δ , between the pulses. Each spin in the bulk sample will have a different motion history from diffusion. If we sum over all spins, k , the total magnetization obtained is [52]

$$\frac{M}{M(0)} = \sum_k \exp(i f_k(\phi)). \quad (2.59)$$

For a free homogeneous system, we know the probability of finding a spin, originally at z_1 , at z_2 after a time interval Δ , is (from Eq. (2.46))

Figure 2.5: The pulsed gradient spin echo (PGSE) sequence tags the spins by dephasing with the first diffusion gradient pulse. The second diffusion gradient untags the spins by rephasing. Depending on the net displacement during the interval, incomplete rephasing will occur, resulting in an attenuation of the signal. The timing diagram labelled RF shows the rf pulses and NMR signal, as well as the readout and readout compensation gradients. The timing diagram labelled GR represents the diffusion sensitizing gradients.

$$P(z_2, z_1, \Delta) = (4\pi D\Delta)^{-1/2} \exp \left[-(z_1 - z_2)^2 / 4D\Delta \right]. \quad (2.60)$$

Thus, we know the phase distribution of Eq. (2.59), and it becomes [52]

$$\begin{aligned} \frac{M}{M(0)} &= \frac{1}{\sqrt{4\pi D\Delta}} \int_{-\infty}^{\infty} \int_{-\infty}^{\infty} \exp(i\gamma\delta G(z_1 - z_2)) \exp \left[-(z_1 - z_2)^2 / 4D\Delta \right] dz_1 dz_2 \\ &= \exp(-(\gamma\delta G)^2 D\Delta). \end{aligned} \quad (2.61)$$

Eq. (2.61) represents the basis for diffusion measurements using NMR [94]. Since γ , δ , and Δ are all known constants or parameters, the diffusion coefficient, D , can be calculated from a least squares fit to the experimental data, $M(G)$ (for several values of the amplitude G). The duration Δ , represents the time during which irreversible attenuation due to diffusion occurs. This interval, then, is the time allowed for diffusion. There are, however, two restrictions to Eq. (2.61) which must be addressed in this work. First, δ is not negligible compared with Δ , so diffusion also occurs during the gradient pulse. Also, there are unavoidable imaging gradients in the sequence which will contribute to diffusion dephasing. For a rigorous treatment of these effects, the Bloch equations for a general pulse sequence must be solved [52].

2.4 The Bloch Equations

In 1946 Felix Bloch proposed a set of very simple equations derived from phenomenological arguments. He proposed that, in a static field $B_o \hat{z}$, the trend of the magnetization toward its equilibrium value $M_o = \chi_o B_o$, can be described by the equation [95]

$$\frac{dM_z}{dt} = -(M_z - M_o)/T_1, \quad (2.62)$$

where T_1 is the longitudinal relaxation time. Also, when the magnetization is tipped into the x-y plane, by an rf pulse for instance, the distribution of local fields at the microscopic level causes the transverse magnetization to decay at a rate represented by the equation [95]

$$\frac{dM_x}{dt} = -M_x/T_2, \quad \frac{dM_y}{dt} = -M_y/T_2, \quad (2.63)$$

where T_2 is the transverse relaxation time. Bloch also proposed that the equations describing the relaxation and diffusion can be combined with the equation of motion for free spins, Eq. (2.6), so that the Bloch equation with diffusion becomes

$$\frac{d\mathbf{M}}{dt} = \gamma \mathbf{M} \times \mathbf{B} - \frac{M_x \hat{x} + M_y \hat{y}}{T_2} - \frac{M_z - M_o}{T_1} \hat{z} + \nabla \cdot (\bar{\mathbf{D}} \nabla \mathbf{M}), \quad (2.64)$$

where the extra term, $\nabla \cdot (\bar{\mathbf{D}} \nabla \mathbf{M})$, represents the contribution of diffusion to the rate of change of the magnetization [95]. In Eq. (2.64), $\bar{\mathbf{D}}$ is a tensor (see Sections 1.4 and 2.6).

For MRI sequences, the spins are subjected to a linear gradient $\mathbf{G}(t)$ superimposed on the main static field $B_o \hat{z}$, so that

$$\mathbf{B}(\mathbf{r}, t) = (0, 0, \mathbf{r} \cdot \mathbf{G}(t) + B_o), \quad (2.65)$$

where $\mathbf{r}$ is the displacement vector.

Considering only the transverse component of the magnetization, we set $m = M_x + iM_y$. If we expand Eq. (2.64) for $\mathbf{B}(\mathbf{r}, t)$ as given by Eq. (2.65), we find that [94, 95]

$$\frac{dm}{dt} = -i\omega_o m - \frac{m}{T_2} - i\gamma \mathbf{r} \cdot \mathbf{G}(t)m + \nabla \cdot (\bar{\mathbf{D}} \nabla m). \quad (2.66)$$

To remove the effects of transverse relaxation and Larmor precession, we can introduce $\psi(t)$ through

$$\begin{aligned} \psi(t) &\equiv m e^{(i\omega_o + 1/T_2)t} \\ &= (M_x + iM_y) e^{(i\omega_o + 1/T_2)t}. \end{aligned} \quad (2.67)$$

The equation of motion for $\psi(t)$ then becomes [95]

$$\frac{d\psi}{dt} = -i\gamma \mathbf{r} \cdot \mathbf{G}(t)\psi + \nabla \cdot (\bar{\mathbf{D}} \nabla \psi). \quad (2.68)$$

In order to solve this equation it will prove useful to first consider the situation where there is no diffusion. In this case we have

$$\frac{d\psi_{D=0}}{dt} = -i\gamma \mathbf{r} \cdot \mathbf{G}(t)\psi_{D=0}. \quad (2.69)$$

which has a solution

$$\begin{aligned} \psi_{D=0}(t) &= \psi_{D=0}(0) \exp \left(- \int_0^t i\gamma \mathbf{r} \cdot \mathbf{G}(t') dt' \right) \\ &= \psi_{D=0}(0) \exp \left(-i\mathbf{r} \cdot \left[\gamma \int_0^t \mathbf{G}(t') dt' \right] \right). \end{aligned} \quad (2.70)$$

By introducing the vector

$$\mathbf{k}(t) = \gamma \int_0^t \mathbf{G}(t') dt'. \quad (2.71)$$

Eq. (2.70) becomes

$$\psi_{D=0}(t) = \psi_{D=0}(0) \exp(-i\mathbf{r} \cdot \mathbf{k}(t)). \quad (2.72)$$

Now consider Eq. (2.68) with $D \neq 0$. If we assume that the only effect of diffusion is to attenuate the amplitude of the magnetization, (or ψ) then we should be able to generate a solution to Eq. (2.68) by multiplying Eq. (2.72) by an attenuation factor, $A(t)$, which we consider to be purely real. Then, our solution will be of the form

$$\psi(t) = \psi_{D=0}(0)A(t)\exp(-i\mathbf{r} \cdot \mathbf{k}(t)). \quad (2.73)$$

From Eq. (2.67), we know that $\psi_{D=0}(0) = M_x + iM_y$. However, since we can choose the phase of the NMR detector arbitrarily in the x-y plane, we can, for simplicity, let $\psi_{D=0}(0) = M_x + iM_y \rightarrow M_x = M_o$. Thus, Eq. (2.73) becomes

$$\psi(t) = M_o A(t) \exp(-i\mathbf{r} \cdot \mathbf{k}(t)). \quad (2.74)$$

Also, we combine $M_o A(t)$ and define $M(t) \equiv M_o A(t)$. Thus, our trial solution is now [94]

$$\psi(t) = M(t) \exp(-i\mathbf{r} \cdot \mathbf{k}(t)). \quad (2.75)$$

If we now substitute this into Eq. (2.68), we get, after some simplification [94],

$$\frac{dM(t)}{dt} = -M(t) \mathbf{k}(t) \cdot \bar{\mathbf{D}} \cdot \mathbf{k}(t), \quad (2.76)$$

which has the solution

$$M(t) = M(0) \exp \left(- \int_0^t \mathbf{k}(t') \cdot \bar{\mathbf{D}} \cdot \mathbf{k}(t') dt' \right). \quad (2.77)$$

If we consider only isotropic diffusion, then the tensor $\overline{\mathbf{D}}$ reduces to the diffusion coefficient, D , which is a scalar quantity, and we obtain [52]

$$\begin{aligned} M(t) &= M(0)\exp\left(-D\int_0^t k^2(t')dt'\right) \\ &= M(0)\exp\left(-D\gamma^2\int_0^{TE}\left(\int_0^t G(t')dt'\right)^2 dt\right) \\ &= M(0)\exp(-bD). \end{aligned} \quad (2.78)$$

Here, we have introduced the 'gradient factor', b , defined by

$$b = \gamma^2 \int_0^{TE} \left(\int_0^t G(t')dt'\right)^2 dt, \quad (2.79)$$

which can be solved by computer for various NMR sequences and diffusion gradient pulse profiles. Eq. (2.78) represents the magnetization attenuation due to diffusion induced spin dephasing. We obtain a similar equation using the apparent diffusion coefficient in an anisotropic system if our magnetic field gradient is applied along only one direction.

From Eq. (2.78), it is apparent that the diffusion coefficient can be determined by measuring $\ln[M(t)/M(0)]$ as a function of b . The b factor is thus a measure of the sensitivity of the sequence to diffusion. Since virtually all MRI sequences use gradients, diffusion attenuation occurs in all sequences, however, in most cases the effect is quite small.

When only the G_D 's are present and $\delta \ll \Delta$, Eq. (2.78) reduces to Eq. (2.61), as expected. If we do not insist that $\delta \ll \Delta$, Eq. (2.79) can be solved for rectangular pulses, as in Fig. 2.5, to give [16] $b = (\gamma\delta G)^2(\Delta - \delta/3)$. Comparing this with Eq. (2.61), we see that the diffusion time is $\Delta - \delta/3$ when δ is not negligible. In practice, most clinical imagers have low amplitudes for their gradients, thus,

diffusion sequences must leave the G_D pulses on for a relatively long duration in order to have a large enough b factor.

2.5 More on Diffusion Sequences

The pulsed gradient spin echo technique, mentioned in Section 2.3, is the most widely used sequence to measure diffusion by NMR [94]. A step by step analysis of Fig. 2.5, which shows this sequence, is instructive at this point. First, the 90° radio frequency pulse tips the spins onto the x-y plane (as described earlier in Section 2.1 and Fig. 2.2). All of the spins are in phase immediately following the pulse. This results in a non-zero magnetization in the x-y plane which generates an NMR signal. The strength of this signal begins to decay after the pulse due to T_1 and T_2 relaxation effects. Next, the first of two diffusion gradient pulses dephase the net signal of the spins by the process described in Section 2.3. By this same process, the inhomogeneity of the main field B_0 , will also cause dephasing. The resulting attenuation from the B_0 inhomogeneity, plus the attenuation due to T_2 , is said to be due to the relaxation time T_2^* [96]. The effects of field inhomogeneities, thankfully, are reversed following the 180° pulse, so PGSE sequences are not affected by this. The 180° pulse flips the spins such that the component of the spin along $\hat{x}$ now points along $-\hat{x}$. The second G_D , which is identical to the first, should cause the same net phase change, except that this time, it will be a rephasing of the spins, as shown in Fig. 2.5. There will be incomplete rephasing if the area under the 2^{nd} G_D is not identical to the area under the 1^{st} G_D , or if the spins have moved during the interval Δ . The same behaviour of dephasing/rephasing occurs between the imaging gradients labelled ‘readout compensation’ and ‘readout’. The point where the area under the readout gradient equals the area under the compensation

gradient is where the peak of the echo will form.

The readout gradient also provides spatial information. A Fourier transform applied to the echo will give a frequency distribution of the signal, which is also a spatial distribution because

$$f = \frac{\gamma}{2\pi}(\mathbf{r} \cdot \mathbf{G} + B_0). \quad (2.80)$$

from Eqs. (2.5) and (2.65). Thus, the Fourier transform yields a 1-D projection along the direction of the readout gradient. The projection is also diffusion weighted because of the G_D 's, and when several projections are taken with varying amplitudes of G_D , a 1-D projection of diffusion coefficients can be generated.

There are several limitations to the PGSE sequence. One is the decay of the signal due to T_2 relaxation, which limits Δ to at most 200 ms [51]. For longer values of Δ the signal is extremely small. The other is the presence of the 180° pulse, which in practice, limits the minimum Δ for reasonable values of b . What constitutes a reasonable range of b values is somewhat subjective because it depends on what percentage attenuation of the signal is deemed 'good enough' for a measurement of D_a , a judgement which also depends on the noise level for a particular experiment. It has been suggested [52] that an attenuation of 20 - 30% is considered adequate.

For diffusion times longer than ~ 100 ms, the signal in a PGSE sequence will have decayed significantly because of T_2 relaxation, and for even longer times, the lack of signal remaining at the end of the sequence makes it impossible to obtain sound diffusion coefficients. However, this is not as much of a problem for stimulated echo acquisition mode (STEAM) sequences [52]. For this reason, a STEAM sequence, such as the one shown in Fig. 2.6, should be used when longer

diffusion times are desired.

The echo generated from a STEAM sequence is called a stimulated echo rather than a spin echo [12], for historical reasons. The stimulated echo is generated by three 90° rf pulses, separated by time intervals τ_1 and τ_2 . The signal decays due to T_2 and T_1 during the τ_1 interval, as in the PGSE sequence, however the 2nd 90° pulse tips half the magnetization along the longitudinal ($\pm\hat{z}$) axis, while the other half remains unaffected in the x-y plane and is later dephased by the crusher gradient. The signal, while being stored longitudinally for the interval τ_2 decays only via T_1 relaxation [52], and is also insensitive to gradient pulses or field inhomogeneities since it no longer has x-y components. Since T_1 is usually much longer than T_2 in biological tissues [12], longer evolution times can be achieved with a STEAM sequence than with a spin echo sequence without the usual signal penalty due to T_2 decay. The 3rd rf pulse returns the stored magnetization to the x-y plane, where an echo is formed after rephasing by the 2nd gradient pulse and the readout gradient. As with the PGSE sequence, the STEAM sequence is not affected by dephasing caused by magnetic field inhomogeneities. Unfortunately, half the signal is sacrificed in this sequence that a spin echo sequence would retain.

When short diffusion times are desired, the alternated gradient pulse sequence (AGPS) sequence will always give shorter diffusion times than the PGSE sequence [52] because the 180° rf pulse has been removed from the diffusing interval. The major drawback is that the b factor is reduced considerably. This drawback is partially alleviated by placing a second gradient pulse pair on the opposite side of the 180° pulse, doubling the b factor. As well, several more gradient pulse pairs could be placed in the sequence, forming a pulse train on each side of the 180° rf pulse. The number of pulse pairs that can be placed in the sequence is limited.

Figure 2.6: Three diffusion sequences used in this work. The STEAM, PGSE and AGPS sequences are the optimum sequences to use when long, medium, and short diffusion times are desired, respectively.

however, because the longer resulting echo time, TE , will cause more decay of the signal by T_2 relaxation. Therefore this sequence has either the drawback of lower b factors or a low signal to noise ratio. The sequence with the smallest diffusion time will be reached when both the b factor and the signal to noise ratio have been decreased to their practical limit. This sequence has no other advantage save the ability to measure diffusion coefficients for low diffusion times.

2.6 Tissue Anisotropy and the Diffusion Tensor

In human white matter, measurements of the diffusion coefficient have revealed different values depending on the direction in which the diffusion was measured [28, 69]. It has been suggested [28, 64] that this anisotropy arises due to the myelin sheath around the axons in white matter. The myelin sheath impedes the motion of the water molecules, thereby decreasing the apparent diffusion coefficient in a direction perpendicular to the fibre axis, $D_{\perp}$, while parallel to the fibre axis, where there is no hinderence by the myelin sheath, the apparent diffusion coefficient, $D_{\parallel}$, is larger.

In situations such as this, the diffusion can no longer be characterized by a single parameter, the diffusion coefficient. Instead it becomes necessary to use the diffusion tensor, $\overline{\mathbf{D}}$, which can be written as:

$$\overline{\mathbf{D}} = \begin{pmatrix} D_{xx} & D_{xy} & D_{xz} \\ D_{yx} & D_{yy} & D_{yz} \\ D_{zx} & D_{zy} & D_{zz} \end{pmatrix}. \quad (2.81)$$

In translational motion theory, each element of the diffusion tensor is the spectrum of the velocity correlation function [86], or

$$D_{\alpha\beta}(\omega) = \frac{1}{2} \int_{-\infty}^{\infty} \overline{v_{\alpha}(0)v_{\beta}(t)} \exp(i\omega t) dt. \quad (2.82)$$

where t is the diffusion time, ω is the Fourier conjugate of t , α and β may take each of the Cartesian directions x , y , z , and $\overline{v_{\alpha}(0)v_{\beta}(t)}$ is the velocity correlation function. The value of an off diagonal element ($\alpha \neq \beta$) will depend on how strongly correlated one velocity component at time $t = 0$ is with a different velocity component at some later time t . Fig. 2.7 demonstrates conditions under which the diagonal element is non-zero and zero, depending on the orientation of the fibre axis relative to the laboratory frame. In Fig. 2.7(a), diffusive motion of the $\hat{y}$ -velocity component is hindered and redirected by the wall of the fibre such that an $\hat{x}$ -velocity component is produced. In this case, it can be shown that $\overline{v_y(0)v_x(t)}$ will be non zero. For Fig. 2.7(b), the fibre wall cannot redirect the particle while it is diffusing with velocity $v_y(t)$, so $\overline{v_y(0)v_x(t)}$ vanishes.

In MRI, the laboratory gradients are not necessarily applied either parallel or perpendicular to the long axis of the neuron. In this case, the diffusion tensor will, in general, have non-zero off diagonal elements. Only when the ‘fibre frame’ corresponds to the laboratory frame will the diffusion tensor be diagonal [29]. Recently, several researchers [30, 72] have used diffusion tensors as a method to map the axonal fibre orientation in the brain. In order to do this, all nine elements of $\overline{\mathbf{D}}$ need to be determined. However, since $\overline{\mathbf{D}}$ is Hermitian, only six measurements are required [59]. Once the tensor elements have been measured, the tensor can be transformed into the ‘fibre frame’ by diagonalizing it using the appropriate rotational operator, $\mathbf{R}$. The diffusion tensor in the ‘fibre frame’, $\overline{\mathbf{D}}'$, can then be written as [29]

$$\overline{\mathbf{D}}' = \mathbf{R}^{-1} \cdot \overline{\mathbf{D}} \cdot \mathbf{R}$$

Figure 2.7: A particle diffusing with a $\hat{y}$ -velocity component within a bound medium, such as within a white matter fibre. In (a) the long axis of the fibre does not coincide with the x or y axes of the laboratory frame and thus the fibre wall will redirect the particle. In (b) there is no such redirection of $\hat{y}$ -velocity components. The redirection in (a) results in a correlation of $v_y(0)$ with $v_x(t)$, which gives non-zero off diagonal elements of the diffusion tensor. In (b) the diffusion tensor is diagonal.

$$= \begin{pmatrix} D'_{xx} & 0 & 0 \\ 0 & D'_{yy} & 0 \\ 0 & 0 & D'_{zz} \end{pmatrix}. \quad (2.83)$$

The orientation of the axonal fibre can now be determined from Eq. (2.83) by identifying the largest of the eigenvalues along the diagonal. This component is equal to $D_{\parallel}$ and its corresponding eigenvector gives the orientation of the axonal fibre relative to the laboratory frame. The other two eigenvalues of $\bar{\mathbf{D}}'$ characterize the diffusion perpendicular to the fibre direction.

The trace of the diffusion tensor can also be used as a signature in MR images. Because of the anisotropy in white matter, there can be considerable variations in the apparent diffusion coefficient from patient to patient depending on the measurement direction and the location within the brain. The trace of the diffusion tensor should remove these variations since the trace is invariant upon rotation. This is often advantageous, since contrast changes caused by anisotropy can frequently mask diseased tissue [59].

2.7 Restricted Diffusion

Restricted diffusion occurs when the medium is confined by borders which are semi-permeable or non-permeable [52]. When the diffusing molecules reach these barriers, they are either reflected back into the medium or they permeate through the barrier. Barriers of this type may be cell walls, myelin sheaths, cellular organelle membranes etc. Other types of barriers such as macromolecules and neurofilaments may simply force the diffusing pathway to be of a tortuous nature. Under such conditions, the diffusion distance does not obey the Einstein relation of Eq. (2.53). The diffusing distance, and therefore the apparent diffusion coefficient

cient. will be lower than it otherwise would be in a free, homogeneous medium [94].

A representation of restricted diffusion is shown in Fig. 2.8. For short diffusion times, T_d , most of the molecules do not have time to reach the barriers, and thus the apparent diffusion coefficient will be close to the free, unrestricted value. When the diffusion time is long enough that the diffusion distance is on the order of the barrier spacing, or the cell size, the apparent diffusion coefficient decreases dramatically until, for very long diffusion times, it reaches an asymptotic value [52]. It is theoretically possible to evaluate the size of the bounded region (i.e. the cell size) from an experimental curve such as in Fig. 2.8, as well as the permeability of the barrier. This is of great interest as a tool to probe tissue microstructure or to determine permeability values of cell membranes.

Several theoretical models have been developed [7, 54] which deal with restricted diffusion. A 1-dimensional system partitioned by regularly spaced barriers, separated by a distance a , with permeability P , and with a constant diffusion coefficient $D(0)$ in the intervening spaces, is shown in Fig. 2.9. Tanner [54] has presented the following analytical solution for D_a as a function of diffusion time for this simple system:

$$D_a(T_d) = \frac{1}{2T_r} \sum_j (I_j/A_j) \exp(-\beta_j^2 T_r), \quad (2.84)$$

where

$$\begin{aligned} A_j &\equiv \frac{1}{2} \sum_k (c_{jk}^2 + d_{jk}^2) + \frac{1}{\beta_j} \sin(\beta_j/2) \cos(\beta_j/2) \sum_k (c_{jk}^2 - d_{jk}^2), \\ I_j &\equiv \frac{c_{j0}}{2\beta_j^4} \sin \beta_j [b_j^2 \sin \beta_j - 2(\sin \beta_j - \beta_j \cos \beta_j)] \sum_k c_{jk} - \frac{d_{j0}}{\beta_j^3} \sin \beta_j \\ &\quad \times [\sin \beta_j - \beta_j \cos \beta_j] \sum_k c_{jk}^k + \frac{c_{j0}}{\beta_j^2} \sin^2 \beta_j \sum_k c_{jk} k^2 - \frac{d_{j0}}{2\beta_j^4} [\sin \beta_j - \beta_j \cos \beta_j]^2 \end{aligned}$$

Figure 2.8: (a) Dependence of the diffusion distance on the diffusion time. For a free and homogeneous solution, molecules obey the Einstein relation $\langle r \rangle = \sqrt{6DT_d}$ and the diffusing distance follows the straight line shown. If there is restriction of the diffusion by barriers, the diffusing distance will level off as shown in the bottom curve. (b) The resulting apparent diffusion coefficient vs. $\sqrt{T_d}$ corresponding to the curves in (a). The bottom curve shows D_a decreasing due to restriction from the barrier, while in the absence of restriction, D_a is, of course, constant, as shown by the top line.

$$T_r \equiv D(0)T_d / a^2. \quad (2.85)$$

and c_{jk} , d_{jk} , β_j and b_j are constants constrained in various ways by boundary conditions. The index k refers to the various spaces between the barriers and the index j refers to the trigonometric terms of the series expansion in the solution.

Although this is an analytical solution, it has not proven to be particularly useful. Computation times are very long since many terms in the series expansion are required for the solution to converge. It also cannot easily be generalized to more realistic systems.

A more useful equation given by Tanner [54] for the asymptotic apparent diffusion coefficient, $D_a(\infty)$, is

$$D_a(\infty) = \left[\frac{1}{D(0)} + \frac{1}{aP} \right]^{-1}. \quad (2.86)$$

Eq. (2.86) is potentially useful in obtaining the cell diameter or cell wall permeability from NMR measurements of $D(0)$ and $D_a(\infty)$.

A 2-dimensional model has also been considered by Szafer, Zhong and Gore [7]. This model is more relevant to measurements in human white matter because, as well as being 2-D, it also has two compartments which can be associated with the intracellular and extracellular spaces. Each compartment also has its own 'free' diffusion coefficient, which we will denote D_I and D_E , respectively. The expression for $D_a(\infty)$ is derived in a manner analogous to computing the equivalent resistance of a network of resistors in series and parallel. Two expressions relevant to diffusion across white matter fibres are obtained: $D_a^{PS}(\infty)$ (Parallel-Series) and $D_a^{SP}(\infty)$ (Series-Parallel):

Figure 2.9: A one dimensional system of regularly spaced planar barriers of permeability P . The analytical solution for the resulting asymptotic value of the diffusion coefficient is $D_a(\infty) = \left[\frac{1}{D(0)} + \frac{1}{aP} \right]^{-1}$.

$$D_a^{PS}(\infty) = \frac{\sqrt{f}}{\frac{\sqrt{f}}{D_c} + \frac{1-\sqrt{f}}{D_E}} + (1 - \sqrt{f})D_E. \quad (2.87)$$

$$D_a^{SP}(\infty) = \left[\frac{\sqrt{f}}{\sqrt{f}D_c + (1 - \sqrt{f})D_E} + \frac{(1 - \sqrt{f})}{D_E} \right]^{-1}. \quad (2.88)$$

where

$$D_c = \left[\frac{2}{aP} + \frac{1}{D_I} \right]^{-1}. \quad (2.89)$$

and f is the cellular volume fraction. Eqs.(2.87) and (2.88) are perhaps a better indicator of water mobility in human white matter.

A Monte Carlo simulation of the model just described would also predict coefficients for intermediate values of diffusion times, which would be practical for assessing tissue characteristics from experimental data. In the absence of feasible, analytic techniques to determine D_a for intermediate values of diffusion times, this should be a very powerful technique. We have decided to use Monte Carlo techniques for the modelling of the data presented in this thesis. See Section 3.7 for more details on the Monte Carlo simulations.

2.8 Gradient Coils

Sensitizing NMR sequences to diffusion requires large amplitude gradients [51]. Although large b factors can be obtained by applying weak gradients for a longer time, for diffusion measurements at low diffusion times the gradient duration must be short. The larger the amplitude of the gradient, the shorter the diffusion time can be made. Since clinical MR imagers often have maximum gradient amplitudes of 10-15 mT/m, diffusion experiments for diffusion times lower than ~ 50 ms are difficult. In order to explore this region of the diffusion behaviour, higher amplitude gradient coils must be built.

The most efficient gradient coil design uses wire loops with cylindrical symmetry [97]. For a single current loop, the magnetic field along the axis is derived in many physics textbooks [98, 99] and is

$$B_z(z) = \frac{\mu_o I a^2}{2(a^2 + z^2)^{3/2}}, \quad (2.90)$$

where a is the radius of the loop, I is the current through it, z is the position along the axis, and $\mu_o = 4\pi \times 10^{-7} \text{ N/A}^2$ is the magnetic permeability of free space.

For points M_p , not necessarily on the z -axis, the magnetic field for the current loop in Fig. 2.10 can be written as [100]

$$B_z(\rho, \theta) = \sum_{n=0}^{\infty} \frac{1}{n!} \left. \frac{d^n B_z(z)}{dz^n} \right|_{z=0} \rho^n P_n(\cos \theta), \quad (2.91)$$

where the $P_n(\cos \theta)$ are the Legendre polynomials, ρ is the distance of point M_p from the origin of the coordinate system, and θ is shown in Fig. 2.10.

The simplest gradient coil is made from two loops each having N turns, radius

Figure 2.10: The coordinate system for describing the B_z component of the magnetic field from the circular current loop.

Figure 2.11: Two coaxial current loops with N turns and radii a separated by a distance l .

a , and carrying a current, I , as shown in Fig. 2.11. The currents in the two loops are in opposite directions relative to each other. For this configuration, the magnetic field along their common axis, $\hat{z}$, is just the sum of the contributions from the individual loops and is given by

$$B_z(z) = \frac{\mu_0 N I a^2}{2} [L_+ - L_-], \quad (2.92)$$

where

$$L_+ = \left[a^2 + \left(z + \frac{l}{2} \right)^2 \right]^{-3/2} \quad L_- = \left[a^2 + \left(z - \frac{l}{2} \right)^2 \right]^{-3/2}. \quad (2.93)$$

For two coils such as those shown in Fig. 2.11, the magnetic field, $B_z(\rho, \theta)$, can again be determined using Eq. (2.91), however, in this case the derivatives of $B_z(z)$ are calculated using Eq. (2.92).

In MRI, the goal is to design coils that have gradients as linear in z as possible.

If we expand Eq. (2.91) we get

$$\begin{aligned}
 B_z(\rho, \theta) &= B_z(0) + \left. \frac{dB_z}{dz} \right|_{z=0} \rho P_1(\cos \theta) + \dots \\
 &= B_z(0) + \left. \frac{dB_z}{dz} \right|_{z=0} \rho \cos \theta + \dots \\
 &= B_z(0) + \left. \frac{dB_z}{dz} \right|_{z=0} z_M + \dots,
 \end{aligned} \tag{2.94}$$

where z_M is the z -coordinate of the point M_p . Thus, if the $n \geq 2$ terms of the expansion are exactly zero, we have an ideal design. For even n , $\left. \frac{d^n B_z(z)}{dz^n} \right|_{z=0}$ vanishes by symmetry, however, the odd order terms do not. For $n = 3$, we have, when setting $l/2 = L$ in Eq. (2.93),

$$\left. \frac{d^3 B_z}{dz^3} \right|_{z=0} = \frac{\mu_o N I a^2}{2} \left[\frac{30L(3a^2 - 4L^2)}{(a^2 + L^2)^{9/2}} \right] \tag{2.95}$$

This term can be set to zero by positioning the coils such that $L = \frac{\sqrt{3}}{2}a$. This arrangement is known as a Maxwell pair [97] and is linear in z to 5th order.

This design, due to its ease of construction and current efficiency, has remained popular despite the emergence of z -gradient coil designs offering better field characteristics [101]. Higher linearity can be achieved by introducing more coil pairs. With N pairs, one has $3N$ independent parameters with which higher order derivatives can be cancelled. For a two coil pair, or a double Maxwell arrangement, a linearity of up to order z^9 can be achieved. As shown in Fig. 2.12, such a coil can be completely characterized by the six parameters I_1 , I_2 , R_1 , R_2 , L_1 , and L_2 .

The ‘optimum’ coil parameters can be calculated for a double Maxwell pair using a similar procedure to that presented above for the single Maxwell pair. However, in this case, practical choices of the maximum current, physical length, and bore access size must be made before solving for the optimum values. Then,

Figure 2.12: Parameters of a double Maxwell gradient coil. The parameters are optimized by forcing the first 3 odd nonlinear terms in the expansion of $B_z(z)$ to vanish. Other parameters set the gradient amplitude and the bore size.

three parameters remain to cancel the derivatives $\frac{d^3 B_z}{dz^3}$, $\frac{d^5 B_z}{dz^5}$, and $\frac{d^7 B_z}{dz^7}$. After setting $X_1 = L_1/R_1$ and $X_2 = L_2/R_2$, we must solve the following system [97]

$$\begin{aligned}
\left. \frac{d^3 B_z}{dz^3} \right|_0 &= \frac{15\mu_o N_1 I_1 X_1 (3 - 4X_1^2)}{R_1^4 (1 + X_1^2)^{9/2}} + \frac{15\mu_o N_2 I_2 X_2 (3 - 4X_2^2)}{R_2^4 (1 + X_2^2)^{9/2}} = 0 \\
\left. \frac{d^5 B_z}{dz^5} \right|_0 &= \frac{315\mu_o N_1 I_1 X_1 (-8X_1^4 + 20X_1^2 - 5)}{R_1^6 (1 + X_1^2)^{13/2}} + \frac{315\mu_o N_2 I_2 X_2 (-8X_2^4 + 20X_2^2 - 5)}{R_2^6 (1 + X_2^2)^{13/2}} \\
&= 0 \\
\left. \frac{d^7 B_z}{dz^7} \right|_0 &= \frac{2835\mu_o N_1 I_1 X_1 (-64X_1^6 + 336X_1^4 - 280X_1^2 + 35)}{R_1^8 (1 + X_1^2)^{17/2}} \\
&+ \frac{2835\mu_o N_2 I_2 X_2 (-64X_2^6 + 336X_2^4 - 280X_2^2 + 35)}{R_2^8 (1 + X_2^2)^{17/2}} = 0. \tag{2.96}
\end{aligned}$$

We can eliminate the unknowns I_1/I_2 , and R_1/R_2 and obtain X_2 as a solution of the eighth order polynomial equation [97]:

$$F(X_1)(3 - 4X_2^2)(-64X_2^6 + 336X_2^4 - 280X_2^2 + 35) = (-8X_2^4 + 20X_2^2 - 5)^2. \tag{2.97}$$

where, for a set value of X_1 , $F(X_1)$ is a known value, given by

$$F(X_1) = \frac{(-8X_1^4 + 20X_1^2 - 5)^2}{(-64X_1^6 + 336X_1^4 - 280X_1^2 + 35)(3 - 4X_1^2)}. \tag{2.98}$$

A computer program can be used to compute X_2 for a given value of X_1 . This method is used in Section 3.4 to calculate the coil parameters for the double Maxwell gradient coil that was built for this project.

Eqs. (2.97) and (2.98) are however, based on loops with infinitesimal wire thickness. In general, gradient designs will have more than one turn per loop, with each turn having a finite thickness. As such, cancellation of the harmonics in Eq. (2.96) may not be complete for each turn. In order to verify the linearity of the final design, a computer can be used to calculate the contribution to $B_z(\rho, \theta)$ from

each individual turn using Eq. (2.91). It has been shown, however, that Eq. (2.91) can be written as [102, 103]

$$B_z(y, z) = \frac{\mu_o I k}{4\pi (ay)^{1/2}} \left[J_1 + \frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} J_2 \right], \quad (2.99)$$

where

$$k = \left(\frac{4ay}{(a + y)^2 + z^2} \right)^{1/2}. \quad (2.100)$$

and J_1 and J_2 are the elliptical integrals of Legendre: [102]

$$\begin{aligned} J_1 &= \int_0^{\pi/2} \frac{d\Upsilon}{(1 - k^2 \sin^2 \Upsilon)^{1/2}} \\ J_2 &= \int_0^{\pi/2} (1 - k^2 \sin^2 \Upsilon)^{1/2} d\Upsilon, \end{aligned} \quad (2.101)$$

where Υ is a dummy integration variable. The advantage of Eq. (2.99) is that it is much easier to solve with a computer than Eq. (2.91). It is therefore advisable that the last stages of optimization utilize Eq. (2.99). This will also facilitate further improvements to the gradient homogeneity by slight shifts of the current loops.

A final remark concerning optimization for gradient coils: it is more convenient to compute $\frac{dB_z}{dz}$ than B_z . The procedure of optimization is then simply to minimize the variation, ΔQ , of the field gradient, which in this case is

$$\Delta Q = \frac{\Delta G_z}{G_z(o)} = \frac{Max(G_z) - Min(G_z)}{G_z(o)}. \quad (2.102)$$

The derivative $G_z = \frac{dB_z}{dz}$, of Eq. (2.99) is given in Appendix A.

Conventional Maxwell coils have the benefit of design simplicity, however the method just presented, which was developed by Saint-Jalmes *et al.* [97], enables a double Maxwell coil to be designed with only minimal extra complexity. The

advantage of the two coil pair design is the more efficient use of the power applied to the coil. The same field linearity is obtained with smaller diameter coils and since the field strength is proportional to $1/R_o^3$, this means that the same field amplitude can be achieved with less electrical power. When switching power is considered, instead of dc, the improvement in efficiency is even greater [97]. For applications to diffusion gradients, the increase in efficiency translates to an increase in the field amplitude G , allowing shorter pulses in the sequences for equivalent b -factors, thus permitting the diffusion time of the sequence to be made shorter. A high performance gradient system is therefore critical when experiments at low diffusion times are needed.

Chapter 3

Materials and Methods

A Siemens SP4000 Magnetom Imager with a main static field of 1.5 Tesla was used for the measurements in this work. The imager has imaging gradients with a maximum strength of 10 mT/m for each Cartesian direction $\hat{x}$, $\hat{y}$, and $\hat{z}$. These gradients were used as the diffusion gradients for the experiments on the white matter of the brain. For the experiments on the leg muscle tissue, a custom made double Maxwell coil was built for the diffusion gradients. Two different rf coils were used for the work presented in this thesis: a 50 cm diameter whole body coil was used for leg muscle measurements, and a 30 cm diameter head coil was used for the white matter measurements. The timing of a sequence of gradients, rf excitation, and signal acquisition is administered by the NUMARIS[®] software. NUMARIS[®] receives the sequence timing information from an external sequence file. We have developed several sequence files for diffusion measurement. All sequence files are either PGSE, AGPS, or a modified STEAM sequence.

3.1 Diffusion Sequences

3.1.1 PGSE and VOI Selection

The diffusion sequences used in this work are operated from the spectroscopy menu of the NUMARIS[®] software, however, the sequence is really a 1-dimensional imaging sequence (i.e. an imaging sequence without phase encoding). Fig. 3.1 shows the timing of the PGSE sequence in detail. The first gradient pulse turned on is G_{+z} . The gradient pulses rise linearly to a specified constant amplitude which they maintain for a certain period of time before they fall linearly to zero. A 90° pulse is applied as soon as the G_{+z} profile reaches its plateau. The 90° pulse is an rf wave that has the profile of a five lobed sinc function. Thus, its frequency spectrum is a 'hat' function. The width of the 'hat' function, or the range of frequencies the rf pulse is able to excite, Δf , is determined by the duration of the pulse. Given Δf , it is easy to determine the amplitude of G_{+z} necessary to select any desired slice thickness Δz , by rearranging the Larmor equation in Eq. (2.5),

$$G_{+z} = \frac{2\pi\Delta f}{\gamma\Delta z} \quad (3.1)$$

where we have used $B_o \rightarrow B_o + G_{+z} \cdot z$ in Eq. (2.5).

The purpose of the 90° pulse and the G_{+z} gradient is to excite the spins within a slice of thickness Δz in the x-y plane. The G_{-z} gradient compensates for spin dephasing that occurs during the G_{+z} gradient; the spins will dephase according to Eq. (2.56). The NMR signal amplitude would suffer severely without the G_{-z} compensation gradient.

The G_{+x} gradient similarly selects a slice in the z-y plane, perpendicular to the slice selected by G_{+z} . Also, G_{-x} compensates for spin dephasing caused by G_{+x} .

Figure 3.1: Timing of a PGSE sequence. The temporal profiles are shown for the radiofrequency (rf) pulses, the magnetic field gradients G_x , G_y , G_z , and the analog to digital converter (ADC). This particular PGSE sequence has an echo time, TE , of 110.6 ms.

Theoretically the dephasing occurring before the centre of the 180° pulse should cancel the dephasing occurring after the centre of the 180° pulse. We have found, however, that the net dephasing is small, but non-zero. A perfectly acceptable S/N is obtained without G_{-x} , however, we have included it to optimize S/N and to eliminate the possibility of artifacts arising from its absence. The dephasing is probably due to imperfect 180° pulses. For some of the spins, the tip angle will be a few degrees greater or less than 180° , and the edges of the slice profile will include spins whose tip angles are from 0° to 180° . This is a well known limitation of rf pulses in MRI.

After the 90° pulse is applied, there will be some recovery of the signal back to the $+\hat{z}$ -axis. This signal will be tipped 180° by the next rf pulse, but due to the imperfection, some spins will have a small component in the x-y plane. This component, in the absence of gradients, will behave like a free induction decay and will not be completely extinguished until the read gradient dephases it after a few microseconds. The presence of this small signal on the ADC, however, seriously corrupts the profile in the frequency domain. This problem occurs only when the diffusion gradient is zero: when $G_{D2} \neq 0$ the component is completely dephased by the diffusion gradient. To avoid this artifact problem, we do not use a 0 mT/m diffusion gradient for the PGSE or STEAM sequences designed for white matter measurements. For the AGPS sequences, the diffusion gradients fail to dephase this small component at any gradient amplitude. The solution to this problem is discussed in Section 3.1.2.

In imaging sequences, the slice selection gradients, which are on during the 90° and 180° pulses, are along the same axis, resulting in the same slice being selected. We, however, have used two perpendicular slices to define our volume of

Figure 3.2: The region of interest (ROI) is the column (shaded), created by the intersection of the two slices.

interest, which is the column created by the intersection of the two slices, shown in Fig. 3.2. We apply frequency encoding along the column using the ‘readout’ gradient in Fig. 3.1. This results in spatial localization along the column, which makes our resulting profile essentially a 1-dimensional image.

Spatial localization, the ‘readout’ gradient and the ‘readout compensation’ gradient are discussed in Section 2.5. The diffusion gradients G_{D1} and G_{D2} are explained in Section 2.3.

All measurements of the NMR signal are obtained in quadrature [60] from a circularly polarized coil (the head or body coil). A circularly polarized coil is essentially two pick up coils physically oriented perpendicular to each other, with axes in the x-y plane. The signal on one coil is phase shifted 90° from the

signal on the other. These two signals, shown in Fig. 3.3, are designated ‘real’ and ‘imaginary’. The receiver is gated on only during the readout gradient. The signals picked up by the rf coils pass through several amplification and filtering stages before reaching the analog to digital converter (ADC). These raw data are then Fourier transformed to give the spatial localization of the signal (see Section 2.5). The modulus is then calculated from the real and imaginary signals to obtain the 1-dimensional image for the sample. Acquisitions were averaged in the complex frequency domain for the white matter measurements, while for the leg measurements the moduli were calculated before averaging was done (see Section 3.5 for more details).

The crusher gradients G_x -crusher, G_y -crusher and G_z -crusher are placed after the data acquisition period to quench any coherent signals that might still remain. After the crusher gradients have turned off, there is a time period during which no rf or gradient pulses are active. During this time the system relaxes back toward equilibrium. The total time between the start of one acquisition and the next, known as the repetition time, TR, was 1500 ms for all the diffusion sequences.

We have set the number of data points, or matrix size, to 1024 for all the diffusion sequences. The dwell time, τ_{dw} , or the data sampling interval was either $8 \mu s$ or $16 \mu s$, depending on the sequence. Tables 3.1 and 3.2 list all of the diffusion sequences used and the parameters used for each. The duration of the ADC was $1024 \cdot \tau_{dw}$.

The Nyquist [86] theorem states that the maximum frequency observable in a digitized data set is $\frac{1}{2 \cdot \tau_{dw}}$. The Nyquist theorem thus places a detection bandwidth restriction on the data. Frequencies above this upper limit will be aliased, or folded back onto the observable frequency range. For quadrature measurements,

Figure 3.3: (a) Fourier relationship between the raw NMR data in the time domain, and (b) the frequency domain after a fast fourier transform (FFT) and (c) the modulus value of the real and imaginary data gives the final signal.

the sign of the frequency can be determined [86], which means spectral components between $\pm \frac{1}{2 \cdot \tau_{du}}$ can be distinguished. The detection bandwidth is then $1/\tau_{du}$. A bandpass filter equal to (or less than) the detection bandwidth is used to eliminate unwanted aliasing, in particular noise aliasing, which limits the noise power.

3.1.2 AGPS

The alternated gradient pulse sequence (AGPS) was developed to obtain the shortest diffusion time possible with the available gradient coil hardware. A discussion of its advantages and disadvantages is given in Section 2.5. An example of the AGPS sequence (for $T_d=13.7$ ms) is given in Fig. 3.4. The dephasing diffusion gradients are G_{+D1} , G_{+D2} , G_{+D3} and G_{+D4} and the rephasing diffusion gradients are G_{-D1} , G_{-D2} , G_{-D3} and G_{-D4} , corresponding to the 4 gradient pulse pairs in this sequence. As with the PGSE sequence, the slice select gradients G_{+z} and G_{x1} have compensation gradients G_{-z} and G_{x2} , respectively. However, in this case the

Figure 3.4: Timing of the AGPS sequence. The temporal profiles are shown for the radiofrequency (rf) pulses, the magnetic field gradients G_x , G_y , G_z , and the analog to digital converter (ADC). The diffusion time for this sequence is 13.7 ms.

compensation gradients occur later in the sequence timing. G_{x2} is positive because G_{x1} was purposely turned on a bit earlier than needed for slice selection. This is because G_{x2} has a second purpose other than compensation: it also quenches the free induction decay induced by the imperfection of the 180° pulse.

In the PGSE sequence, the 2nd diffusion gradient quenches the small free induction decay component after the 180° pulse, however, in the AGPS sequence both dephasing and rephasing diffusion gradients are present after the 180° pulse.

thus the small free induction decay generated by the 180° pulse is rephased after G_{D4} even when large gradient amplitudes are used. Thus, another gradient, G_{x2} , must be introduced to dephase this small signal. It is important that G_{x2} , as well as any other gradients, not be on concurrently with the diffusion gradients. This would tilt the orientation of the diffusion sensitization and result in cross terms in the diffusion tensor in Eq. (2.81).

Fig. 3.4 also shows the ‘readout compensation’, ‘readout’, and crusher gradients for each axis.

3.1.3 STEAM

The stimulated echo acquisition mode (STEAM) sequence, also discussed in Section 2.5, is shown in detail in Fig. 3.5. This sequence was used for white matter measurements for long diffusion times ranging from $T_d = 170 - 1020$ ms. It has 3 slice select gradients G_{x1} , G_{z2} and G_{z3} , where the z -gradients select the same slice and both have the same compensation gradient G_{z1} . G_{x2} is the compensation gradient for G_{x1} . In this sequence, each 90° pulse will invoke a free induction decay, which, for the 2^{nd} and 3^{rd} pulses, we want quenched. As in the PGSE sequence, G_{D2} quenches the signal from the 3^{rd} pulse (G_{x2} helps as well) and the 2^{nd} rf pulses’ free induction decay is quenched by the crusher gradients, G_{xt} -crusher, G_{yt} -crusher and G_{zt} -crusher. These gradients do not affect the diffusion attenuation because they are on during the interval τ_2 , when the magnetization that eventually leads to the signal is not in the x-y plane. These gradients also serve to quench small spin-echoes that can arise among the pulses due to their imperfection. They are not included in the calculation of b in Eq. (2.79).

Similar to the PGSE and AGPS sequences, the STEAM sequence has the ‘read-

Figure 3.5: Timing of the STEAM sequence. The temporal profiles are shown for the radiofrequency (rf) pulses, the magnetic field gradients G_x , G_y , G_z , and the analog to digital converter (ADC).

out compensation', 'readout', and crusher gradients at the end of the sequence. A STEAM sequence is traditionally a spectroscopy sequence without the 'readout compensation', 'readout' or diffusion gradients. In such a sequence, the ADC is turned on immediately after the G_{z3} gradient and the peak of the echo occurs at the beginning of the ADC. In our modified STEAM sequence, the stimulated echo forms in the centre of the ADC, as in the PGSE and AGPS sequences.

3.2 Measurement Protocol for White Matter

All volunteers in this study were normal and healthy. Each volunteer consented to the study and a detailed questionnaire and information sheet were given to each. After all questions were answered, each volunteer signed an 'Ottawa General Hospital Consent for Medical Research' form approved by the Research Ethics Board of the Ottawa General Hospital.

Volunteers were placed supine, head first into the magnet, using the standard Siemens head coil. The volunteer alignment was such that a position roughly 2 cm from the eyebrows (towards the forehead) was placed at the isocentre of the magnet. The magnet's automatic coil tuner was engaged and the operator (myself) exited the magnet room to the console room. From there, a standard spin echo imaging sequence was used to obtain sets of transverse, sagittal and coronal scout images. Each set consisted of 9 slices which were each 10 mm thick. The imaging sequence was called SE_15B130 and we used TR=600 ms, and TE=15 ms. The images were obtained to ascertain the location of the corona radiata in the left cerebral hemisphere, where we wished to place our column.

The corona radiata of the brain was chosen because in this region the orientation of the white matter fibres [28, 31, 36] is well known (see Fig. 3.6). The

white matter fibres are predominantly parallel to the anterior to posterior direction (AP) in the frontal and posterior sections, while in the central region, the fibres emanating from the corpus callosum result in a predominantly right to left (RL) orientation. We place the column as shown in the diagram so as to capture both predominant orientations. Thus, our measurement should include data for diffusion parallel and data for diffusion perpendicular to the white matter fibres [28]. Half of the sequences have diffusion sensitization parallel to the column (AP) and the other half have sensitization perpendicular to the column.

Once the coordinates for the column placement are determined from the scout images, the diffusion sequences are run. For the long diffusion times, $50.6 \text{ ms} \leq T_d \leq 1020 \text{ ms}$, a program called DIFF.PRO, which was written in the NUMARIS[®] command language, is used to automatically run 18 sequences: 9 for the 'AP' direction and 9 for the 'RL' direction. DIFF.PRO queries the operator for the $\hat{x}$, $\hat{y}$ coordinates of the column, and via FORTRAN subprograms, rewrites the slice selection offset into the NUMARIS[®] sequence files. The imaging and column placement takes approximately 15 minutes, and the diffusion sequences run for 50 minutes. This means that the volunteer is in the magnet for a total of about 1:05 hours. During this time the volunteer must remain as motionless as practically possible. The raw data are in the frequency domain, as in Fig. 3.3(b), because we have chosen the 'FFT on' option in the NUMARIS[®] menu for all of the sequences. This means that NUMARIS[®] does a fast fourier transform on the data before writing them to the permanent data file.

For the short diffusion times, $13.7 \text{ ms} \leq T_d \leq 50.6 \text{ ms}$, a different NUMARIS[®] command language program, BIPOLAR.PRO, was written to automatically run the sequences. The AGPS sequences used here are more sophisticated than the

Figure 3.6: Schematic diagram of the column placement and white matter fibres of the corona radiata and emanations of the corpus callosum, demonstrating the dominant orientation as AP (parallel to the column) in the anterior frontal and posterior parietal lobes. Greater RL (perpendicular to column) orientation is seen in the central region.

PGSE or STEAM sequences (because they were written later). The difference is in the way acquisitions are obtained. The AGPS sequences first run 10 measurements, one for each of the 10 different gradient amplitudes, ranging from 0.7 mT/m to 9.7 mT/m. When this is complete, the cycle is repeated 20 times, so that the results for each gradient amplitude are measured 20 times. The PGSE and STEAM sequences use 10 acquisitions, but each gradient amplitude is measured one after the other, that is, 10 measurements of 2.5 mT/m, 10 measurements of 3.5 mT/m, etc... until the last gradient amplitude of the last sequence is finished. The results should be the same regardless of the order of acquisition. We state the methodology here for completeness. There is more dead-time in the PGSE and STEAM protocols because NUMARIS[®] must 'load in' a new sequence file for each gradient amplitude, unlike the AGPS sequence files.

3.3 Phantoms

In order to check the reliability of our diffusion sequences and protocol, we have measured the diffusion coefficients of water and acetone, using the same protocol described in Section 3.2. For water measurements, we used a 17 cm diameter plastic sphere containing water doped with CuSO_4 . This phantom is specifically designed by Siemens for quality assurance on the imager. The CuSO_4 is present to reduce the T_1 of the water, and has a negligible effect on the diffusion coefficient. For the acetone measurements, a one litre sealed glass bottle of 99% pure acetone, from Aldrich Chemical Company was used. For the sequences used for leg muscle measurements, a large plastic pop bottle (31 cm in height, 10 cm in diameter) was filled with tap water and doped with CuSO_4 . The bottle was inserted into the bore of the gradient coil for the water measurements. The same acetone bottle

described above was used for the acetone measurements.

We have also used a medical dialysis filter as a phantom for white matter axons. The filter was a Baxter Cellulose Acetate Hollow Fiber Dialyzer, model CA-170. It consists of long, thin, hollow fibres (200 μm inner diameter, 15 μm wall thickness) made of cellulose acetate. The fibres are packed inside a plastic cylindrical container, but not too tightly since the outside environment is designed for a saline solution to be circulated. A schematic diagram of the dialysis filter is shown in Fig. 3.7.

In our diffusion experiment, the inside of the fibres is filled with undoped tap water. We filled it by connecting a siphon hose from inlet A in Fig. 3.7 to a reservoir (a big bucket). When the siphon was started, water flowed from the reservoir into the filter, through the fibres and out of outlet B. After about 10 minutes of flow, we believed that all of the air had been flushed out of the fibres and that they were filled with water. The siphon was disconnected and the filter was immediately immersed in a water bath so that inlet A and outlet B were submerged, thus preventing air from reentering the fibres.

Although the filter was designed for blood toxins to permeate the fibre walls, this occurs only when the fluid is under osmotic pressure. For example, the permeability of water through the fibres is 8.3 ml/hour under 100 mmHg pressure [104]. Under our experimental conditions, the water is not under pressure, and so the permeability of the fibres would be effectively zero. This was visually confirmed after the experiment since after several weeks no water was seen in the outside environment of the filter.

Water was kept out of the outside environment, and a siphon tube was connected to inlet C and outlet D in Fig. 3.7 to prevent accidental entry of water from

Figure 3.7: Schematic diagram of the Baxter Cellulose Acetate Hollow Fiber Dialyzer, model CA-170. The filter's fibres were filled with tap water, while air was kept in the outside environment. Diffusion measurements were obtained for sensitization parallel and perpendicular to the fibres.

the water bath. The entire water bath and filter were placed in the imager and the same protocol as in Section 3.2 was used to measure the diffusion coefficients. Diffusion sensitization was measured parallel and perpendicular to the fibres. This phantom was not used for the leg sequences since the phantom is too large to fit inside the leg gradient coil for diffusion sensitization perpendicular to the fibres.

3.4 Gradient Coil Design

In order to sensitize MRI sequences to diffusion at short diffusion times, large amplitude gradients are required [51]. As mentioned in Section 2.8, the maximum

gradient strength on clinical scanners is often only 10 mT/m, which limits the diffusion time to ≥ 14 ms in practice [78]. In order to measure diffusion coefficients at lower diffusion times, we have designed and built a double Maxwell gradient coil along with an inexpensive method for interfacing the coil to the clinical scanner. With this coil, we have measured novel data for the diffusion coefficient of water in human muscle in vivo.

The design of the double Maxwell gradient coil is shown in Fig. 3.8. It follows the strategy developed by Saint-Jalmes *et al.* [97], as described in Section 2.8. For a minimum bore access size (i.e. large enough for most human legs), we chose $R_2 = 8.5$ cm and we chose $L_2 = 12.5$ cm as a convenient length for the coil. (See also Fig. 2.12.) Since these choices give us $N_2 = \frac{L_2}{R_2} = 1.47$, we can solve Eqs. (2.97) and (2.98) for N_1 using a computer. When this is done we find $N_1 = \frac{L_1}{R_1} = 0.48$. We set $L_1 = 6.5$ cm so as to have a reasonably large useful volume from our coil. This gives us $R_1 = 13.5$ cm. Once we have values for R_1 , R_2 , L_1 , and L_2 , we can use Eq. (2.96) to get $N_1 I_1 / N_2 I_2 = 0.71$. We wanted to obtain a gradient amplitude of about 100 mT/m when 12 Amps were used. From Eq. (2.96), we find that, if $I_1 = I_2$, we must have $N_1 = 61$ and $N_2 = 86$ in order to get the desired gradient amplitude.

The coil design was further optimized by computing $\frac{dB_z}{dz}$ for each turn of the coil, and summing the contributions. Each turn of wire, due to its finite thickness, was somewhat displaced from the ideal values of R_1 , R_2 , L_1 and L_2 computed above. Therefore, the values did not exactly satisfy Eqs. (2.97) and (2.98) with the result that each turn-pair produced non-zero higher order harmonics. These errors were partially alleviated by fine spacing adjustments of R_1 , R_2 , L_1 and L_2 . This is illustrated in Fig. 3.8. A computer program which computes the contribution

from each turn using the derivative, $\frac{dB_z}{dz}$, of Eq. (2.99) (given in Appendix A) was used to 'fine tune' the final design parameters, which are shown in Fig. 3.8.

The resistance of the coil was measured with an HP-3478A precision multi-meter, and we obtained a value of $1.55 \pm 0.07 \, \Omega$. The inductance of the coil was measured with an LCR bridge, model LCR-740, Leader Electronics Corp., and we obtained a value of $8.64 \pm 0.05 \, \text{mH}$. The capacitance of the coil was below the threshold of the LCR meter's range ($< 10 \, \text{pF}$).

The coil was built (except for the wire winding) by the Science Technology Centre at Carleton University and was machined from a single cylinder of polyethylene. The wire was carefully wound by hand onto the polyethylene cylinder in the appropriate locations using #14 gauge resin coated copper wire with a measured diameter of $1.70 \pm 0.05 \, \text{mm}$. The winding was held in place by glueing the wires with Scotch-Cast epoxy. Epoxy was applied to each layer of winding, where a layer consisted of 9 turns for the outer pair and 8 turns for the inner pair. A final, thick coating of epoxy was applied when the windings were finished. The winding ends meet in the centre of the coil where they can be connected to standard ac cords and cables (although dc current was used) via a 3-pin male receptacle (Switchcraft, Electrosonic Cat. No. EAC310) mounted onto the coil and soldered to the ends of the wires.

In order to stabilize the gradient coil on the Siemens' patient table, a holder for the coil was designed which has small plugs on the bottom that insert directly into two existing holes on the patient table, see Fig. 3.9. The existing holes are to secure various Siemens rf coils in the same way. The gradient coil is held in the holes by its own weight (40 lbs., including the holder), and the weight of the volunteer or phantom.

Figure 3.8: Side view of the gradient coil design. The coordinates of the coils are optimized by a computer, taking each turn into account. The coil is made from a single piece of high density polyethylene. The copper wires are epoxied into place. All dimensions shown are in cm.

Figure 3.9: All the pieces making up the coil holder, and the coil (or former) itself. The holder is made from 20 different pieces, (excluding screws) all from $\frac{3}{8}$ " sheet polyvinylchloride (PVC), except the plugs, which are polyethylene. The yokes and bottom supports are permanently fixed to the plank and top stabilizer bar by epoxy and a tight-fit design. The holder has four removable parts: the plank with its yokes and supports, the top bar with its yokes and the two side stabilizer bars.

The volunteer is positioned supine and feet first into the magnet. The volunteer's leg is inserted into the bore of the gradient coil until the calf muscle is in the centre of the coil. Foam padding is then inserted around the volunteer's leg to keep it in position. The coil and volunteer are then inserted into the imager in the usual way. Once positioned in the imager, the electrical cord from the coil is connected to the electrical cable from the Techron 7570 power supply through the back of the magnet bore.

The existing body coil on the imager, which has a birdcage design [105] with a 50 cm field of view, is used as the rf transmit and receive coil. Usually, the rf coil is placed inside the gradient coil [106] to improve the filling factor and eliminate rf coupling which may shield the sample from the rf coil. In our set up, however, the gradient coil is placed inside the rf coil. This has some advantages and some disadvantages. First, the radius of the coil can be made smaller than would otherwise be possible since allowances for an rf coil insertion need not be made. Since the field gradient is proportional to $1/R_g^3$, the smaller radius means that higher gradient amplitudes are achievable for a given amount of input power. This is an important consideration when short diffusion time sequences are used. As well, the design of an rf coil to fit inside the gradient coil is expensive and non-trivial. Using the existing Siemens rf coil eliminates this need. The price paid for this arrangement is a decrease in the signal to noise ratio, S/N , due to the decreased filling factor. Our implementation thus sacrifices S/N in favour of larger gradient amplitudes and the capability of making diffusion measurements at shorter diffusion times.

Also, coupling of the gradient coil with the rf coil may result in a drastic NMR signal loss due to pick up from the gradient coil. This problem is almost completely

alleviated by the switching circuit shown in Fig. 3.10. This simple circuit, made with readily available, inexpensive parts, effectively provides a high impedance to the rf, while maintaining a low impedance to the amplifier's dc pulse. The circuit eliminates the rf pick up without compromising coil inductance and power efficiency.

The workhorses of this circuit are the TRIACS, which are back to back diodes with a gate lead. When a voltage is applied across the gate (typical minimum= 1.4 V), the diodes are in a conducting state. The conducting state is maintained until the current going through the diodes drops below a certain threshold (typically 30 mA). The gate voltage need not be continuously applied to maintain a conducting state. The TRIACS make the circuit function like a solid state switch, with much better performance than commercial solid state relays (SSR), like the D1D40 SSR used here, because the TRIACS are in a blocking state (high impedance) even at high rf frequencies. The D1D40 SSR does not effectively block rf when it is in the off state.

When a diffusion gradient from the coil is required in the sequence, the sequence file turns on the OSC0 sync-bit on the Siemens imager. This sync-bit is normally used to trigger an oscilloscope when viewing the TTL output for the Siemens' gradients and rf coils. We use the sync bit here to trigger a DG535 Pulse Generator (Stanford Research Systems), which we have programmed to give a pulse profile input to the Techtron 7570 amplifier. The 7570 amplifies the profile 5.75 times, in constant current mode. The OSC0 signal is also used to turn the SSR on, after it has been suitably inverted by an SN74LS04 Hex Inverter. The Inverter is powered from an inexpensive, commercial AC-DC adapter as shown in Fig. 3.11.

Figure 3.10: Schematic diagram of the circuit used to power the gradient coil. TRIAC 1 is a 600 V, 25 A TIC263M. TRIAC 2 is an 800 V, 25 A MAC223A10 and the SSR is a Crydom D1D40.

The TRIACS, which are initially in the off state, force the amplified signal to go through their gates. A positive pulse will go via the resistors R2 and R3, while a negative pulse current is forced through resistors R1 and R4 by the MUR415 diodes. A separate pathway to the gates for negative and positive currents is used because the minimum gate trigger voltage is different depending on the voltage polarity. Separating the pathways allows us to maximize gate resistance, which minimizes the chance of a false trigger. The constant current mode of the amplifier results in a high initial voltage at the junction A, which is necessary since there must be an adequate gate voltage after the large voltage drop across the resistors. Large resistances are used for R1, R2, R3 and R4 to prevent unwanted rf pick up voltages from falsely triggering the TRIACS. These large resistances in series with the TRIACS' gates effectively drop the rf pick up voltage to a level lower than the minimum gate trigger voltage (typically: 0.2 V). Thus the TRIACS remain in the off state, putting the gradient coil in series with a large resistance ($> 1 \text{ M}\Omega$), which disables its ability to pick up rf.

The NMR signal is seriously attenuated in the absence of this circuit because the gradient coil picks up the rf transmission, effectively shielding the sample from the rf pulse. Contrary to our expectations, the rf pick up is not impeded by the 7570 or the D1D40 SSR. Also, the many commercial rf chokes that were tried in the circuit had no noticeable effect.

The TRIACS turn on approximately $10 \text{ }\mu\text{s}$ after the gate voltage is applied, allowing the amplifier's current to pass through the TRIACS, the SSR and of course, the gradient coil. When the gradient pulse is turned off, and 2 ms is allowed for the fall time, the TRIACS should turn off. However, this will not necessarily be true since the 'holding' current of the TRIACS is only $\approx 30 \text{ mA}$.

Figure 3.11: Schematic diagram of the circuits used to run the diffusion sequences for the leg muscle. SW1 and SW2 are manual switches that are turned on before the gradient coil is used. The values of the resistors are $R1 = 2.7 \text{ k}\Omega$, $R2 = 3.9 \text{ k}\Omega$, $R3 = 5.0 \text{ k}\Omega$, and $R4 = 3.8 \text{ k}\Omega$.

There may still be a small amount of current coming from the 7570. A 30 mA baseline current can be established by a mere 5 mV of input from the DG535. A holding current may also be lingering in the circuit from small eddy currents established by the gradient pulse. The purpose of the D1D40 SSR is to turn off this holding current. When the SSR switches off, the current through the TRIAC'S is zero and they stay in the off state until the next gradient pulse is applied.

For negative gradient pulses, the signal from the DG535 Pulse Generator must come from a different channel than the positive pulse signal. From Fig. 3.11, the DG535's 'AB' channel output is sent through a 741 operational amplifier set up as a voltage follower. The signal is then sent to a 2nd 741 set up as a voltage adder. The 'AB' and 'CD' channels are never on at the same time so the voltage adder simply serves to merge the two channels onto one input line to the 7570. The voltage follower also acts as a buffer between the 'AB' and 'CD' channels. Without this, pulse reflections between channels would arise along the BNC cable, seriously distorting the pulse profile. Note that the 'AB' signal is inverted twice because it passes through 2 inverting 741's, while the 'CD' signal is inverted once. The 'AB' channel is therefore used for positive pulses and the 'CD' channel is used for negative pulses. The back to back diodes along this line are simply an extra precaution: they ensure that any small baseline offset from the 741's does not reach the 7570.

3.4.1 Coil Calibration

The calibration of the gradient coil was performed in two steps. A careful calibration was first done at the center of the coil. This information was then used in the calibration of off centre positions to confirm the homogeneity and linearity of the

gradients.

The gradient coil was initially calibrated using a standard PGSE sequence and a 2 cm³ water phantom. The slice selection gradients were turned off so the entire phantom volume was irradiated. Great care was taken to place the cubic phantom in the centre of the gradient coil. Using a ruler, we estimated the positioning error to be $\sigma_x = \sigma_y = \sigma_z = \pm 0.5$ mm. The gradient coil to be calibrated was used as the read gradient in the sequence. Thus, upon FFT of the signal, a 1-dimensional projection of the phantom was obtained, as described in Section 2.5. The gradient amplitude, G_z , was determined from the width of the profile, Δf , since from Eq. (2.80),

$$G_z(r, z) = \frac{2\pi\Delta f}{\gamma\Delta z}, \quad (3.2)$$

where r and z are the radial and axial positions inside the coil, respectively, and Δz , the length of the water phantom, is 19.35 ± 0.05 mm. This NMR technique gives a very accurate calibration of the gradient field, $G_z(r, z)$, for $r = z = 0$.

For the calibration of off centre positions, we used a different technique because the method described above is time consuming and has a resolution of only 2 cm (i.e. the size of the phantom). We used the accurate value of $G_z(0, 0)$ obtained from the method described above to calibrate the entire gradient volume by measuring the diffusion coefficient of water for all points $G_z(r, z)$. We used a PGSE sequence similar to the sequences used for leg muscle measurements (see Fig. 3.1) to obtain a diffusion profile on a large water phantom that filled the entire coil volume. The profile was that of a 10mm×10mm column along the coil's $\hat{z}$ -axis, positioned at 8 different radial positions ranging from 0 mm to 70 mm. The resolution of the calibration obtained with this technique is 10 mm in the radial direction and 1.5

mm along the coil's axis.

The calibration procedure involved measuring the magnetization, M , for 12 different currents, I , through the gradient coil and generating plots of $\ln[\frac{M}{M(0)}](r, z)$ vs. $b(0, 0)$, where $b(0, 0)$ are the gradient factors produced at the isocentre of the gradient coil for the corresponding current settings. Since these curves showed no visible deviation from a straight line we used a least squares routine to fit them to the equation

$$\ln[\frac{M}{M(0)}](r, z) = -D_m b(0, 0), \quad (3.3)$$

where $-D_m$ is the slope.

From Eq. (2.78) we also know that we can write

$$\ln[\frac{M}{M(0)}](r, z) = -Db(r, z), \quad (3.4)$$

where $b(r, z)$ is the gradient factor at the point (r, z) and D is the self diffusion coefficient of water.

If we combine Eqs. (3.3) and (3.4), we can easily show that

$$b(r, z) = b(0, 0) \frac{D_m}{D} \quad (3.5)$$

from which we can calculate reliable values of $b(r, z)$. Once we know $b(r, z)$ we can determine $G_z(r, z)$ using the definition of b given in Eq. (2.79).

3.5 Measurement Protocol for Leg Muscle

The volunteer is positioned in the magnet supine and feet first, as described in Section 3.4. A similar procedure to that in Section 3.2 was used to image the

leg so that the coordinates for column placement could be obtained. The same imaging sequence was used. Our $20\text{mm} \times 20\text{mm}$ column was placed along the large soleus muscle in the leg. Fig. 3.12 shows a transverse slice of a human leg with the column outline superimposed to show its placement. The length of the column is parallel to the diffusion gradient, along the z-axis. The soleus muscle is composed of bundles of muscle fibres that are inclined roughly 20° [107] relative to the tibia, which we have placed parallel to the z-axis. The soleus muscle was chosen because its fibre direction is very close to being parallel with the z-axis and also because it is the largest of the leg muscles in the ‘calf’ area of the leg. Our measurements thus provide diffusion data in the muscle cells for a direction inclined $\approx 20^\circ$ to the long cell axis.

A FORTRAN program is used to achieve proper column placement once the coordinates of the column are determined. When a sequence runs, it is set up to write the raw data as the time domain signal (see Fig. 3.3(a)): one file for each acquisition. We take 10 acquisitions of 12 b -values, for a total of 120 files per sequence. There are 7 diffusion sequences, which are PGSE’s for $T_d \geq 9$ ms, and AGPS’s for $T_d \leq 6$ ms, as shown in Fig. 2.6. Each file is then Fourier transformed and the modulus is calculated. Then the 10 acquisitions are averaged.

Averaging of the acquisitions was performed in the absolute frequency domain to eliminate false attenuation from phase variations. The phase changes from shot to shot in the time domain signal can arise from a slight mismatch of the diffusion gradients [52]. The mismatch causes a negligible change in the b -factor and the amplitude of a single acquisition, however, when several acquisitions with slight phase shifts are averaged in complex space, a significant attenuation of the true amplitude results. This is not as much of a problem for the measurements in the

Figure 3.12: A transverse image of a human leg. The column of interest, which runs into the plane, is shown placed on the soleus muscle. This image has a slice thickness of 10 mm.

white matter described in Section 3.2 because the Siemens' gradients which were used, are very reproducible from shot to shot. Fig. 3.13 demonstrates the difference between averaging in complex space and absolute frequency space.

The order of acquisition for each b -value is the same as for the PGSE and STEAM sequences used to obtain the white matter data described in Section 3.2. However, here, there is not a separate sequence file for each gradient amplitude because the gradient amplitudes are programmed into the DG535 Pulse Generator. Therefore, whenever a set of acquisitions are about to start, an alert operator must manually change the amplitude of the DG535 signal using the DG535 front panel buttons. Fortunately, NUMARIS® can be programmed to delay a set of acquisitions, which allows a few seconds of leeway for the operator.

3.6 Data Analysis Programs and Error Computation

Many publications on diffusion measurements [28, 44, 77, 108], even recent [73, 59, 109] ones, fail to compute b -values using computer integration of Eq. (2.79), and instead use the approximation $b = (\gamma G \delta)^2 (\Delta - \delta/3)$. Although this approximation may be valid in some cases, it is surprising that simple computer integration algorithms are not used more often.

We have developed a computer program, DPRO.FOR, that performs many diffusion related calculations, including the integration of Eq. (2.79) for any gradient pulse profile. The main use of DPRO is to obtain a diffusion coefficient profile from a series of profiles that are diffusion weighted by different values of the G_D amplitude. Depending on the protocol, 6 - 12 different amplitudes of G_D are used, and each value corresponds to a b -value. According to the options specified by

Figure 3.13: Demonstration of artifacts that can arise from shot to shot phase variations in the complex signal. Two acquisitions of the PGSE sequence COIL.TD14 on a water phantom are shown in Acqu.1 and Acqu.2. There is a very slight temporal shift between these two acquisitions, which, if averaged in complex space as in (a), results in an artifact in the resulting FFT, shown in (c). To avoid this problem, the two acquisitions are individually FFT'ed to give FFT.1 and FFT.2, which are then averaged to give an artifact free profile in (b).

the user. DPRO will compute the diffusion coefficient by performing a weighted linear regression to the $\ln[M(t)/M(0)]$ data (see Eq. (2.78)). If desired, DPRO will automatically compute the baseline value and compensate M accordingly, or it can accept user input for the location of the baseline from which to compute its value.

The formula for the slope of $\ln[M(t)/M(0)]$ vs. b , using the weighted linear regression method is well known [110], and it is

$$\text{slope} = -D_a = \frac{1}{\Delta_s} \left[\sum_{j=1}^N \frac{1}{\sigma_{y_{M_j}}^2} \sum_{j=1}^N \frac{b_j y_{M_j}}{\sigma_{y_{M_j}}^2} - \sum_{j=1}^N \frac{b_j}{\sigma_{y_{M_j}}^2} \sum_{j=1}^N \frac{y_{M_j}}{\sigma_{y_{M_j}}^2} \right]. \quad (3.6)$$

Where

$$\Delta_s = \sum_{j=1}^N \frac{1}{\sigma_{y_{M_j}}^2} \sum_{j=1}^N \frac{b_j^2}{\sigma_{y_{M_j}}^2} - \left(\sum_{j=1}^N \frac{b_j}{\sigma_{y_{M_j}}^2} \right)^2. \quad (3.7)$$

The error on D_a using this method is computed by

$$\sigma_{D_a} = \left[\frac{1}{\Delta_s} \sum_{j=1}^N \frac{1}{\sigma_{y_{M_j}}^2} \right]^{1/2}. \quad (3.8)$$

where $y_{M_j} = \ln[M_j(b)]$, the error $\sigma_{y_{M_j}} = \sigma_{M_j}/M_j$, and the error σ_{M_j} is set equal to half the root mean square noise level in the baseline of the profile.

In diffusion analysis, it is always desirable to check the signal attenuation by plotting $\ln[M(t)/M(0)]$ vs. b . Such a plot will confirm the linear relationship of the data points, as predicted by Eq. (2.78). It will also confirm that the signal attenuation has not dropped to the baseline level. Any data points whose value is within the noise level should be discarded. DPRO will produce this attenuation plot upon the user's request and the user must specify the location (the position on the diffusion profile) from which the data are to be taken.

DPRO has another useful feature when averaging of hundreds of files is necessary. For multiple acquisitions using our gradient coil, as mentioned in Section 3.5,

12 sets of 10 files must be averaged before a diffusion profile can be computed. On a volunteer for leg muscle measurements, 7 diffusion sequences, with different diffusion times, are run to obtain a D_α vs. T_d profile similar to Fig. 2.8b. Therefore, a total of 84 sets of 10 files must be averaged. DPRO has options to be selected where it will average any number of sets of files in the absolute domain or the complex domain. DPRO, with its user-friendly features, is an indispensable tool for diffusion analysis.

3.7 Monte Carlo Simulations

Two of the theoretical models for water diffusion in human white matter, presented in Section 2.7, give an analytical value for the asymptotic behaviour of D_α . Since the asymptotic D_α can be obtained experimentally, these models can also be used to give a close estimate of other parameters of the white matter axons, such as membrane permeability. They do not predict, however, the behaviour of D_α for intermediate values of T_d .

The other theoretical model discussed in Section 2.7, that due to Tanner [54], allows us, in principle, to calculate D_α for any value of T_d . Unfortunately, results obtained with this approach have proven to be both difficult to compute and unreliable because the equations involved converge very slowly.

This situation motivated us to use Monte Carlo simulations to explain our experimental results. We have found that all of our experimental data can be simulated using a Monte Carlo algorithm on a computer with any geometry for which there is a known analytical solution. We have chosen to use the geometries used by Szafer *et al.* [7] in their theoretical models discussed in Section 2.7.

The term ‘Monte Carlo’ refers to the use of a random number generator in

the computer program to simulate diffusion steps. In the computer simulation, the particle takes a random walk consisting of steps of random size and direction. Since the probability of a given displacement in bulk water follows a Gaussian distribution, a Gaussian random number generator [111] was used to determine the steps. We have written Monte Carlo programs to simulate diffusion in the 1-dimensional geometry shown in Fig. 2.9, a 2-dimensional geometry as studied by Szafer, Zhong and Gore [7] and mentioned in Section 2.7, a 'tortuosity' model consisting of a geometry of impenetrable 'blocks', a 2-D model incorporating a flow channel within the lattice, and finally, a 2-D model which tracks the phase of the particle (see Section 5.1.7) and computes its contribution to the NMR signal.

The lattice in all programs is defined by choosing boxes of dimension $B_x \times B_y$ placed on a grid with units of size $A_x \times A_y$. This basic unit is then repeated in all directions on the grid. For the 1-D lattice in Fig. 2.9, it is only repeated along the $\hat{x}$ -direction. In the 2-D models, shown in Figs. 3.14 and 3.15, the box $B_x \times B_y$ represents 'barriers' to the simulated particle. If the particle attempts to cross a barrier, it will do so with a probability, p , which depends on the permeability of the barriers, P , and v , the velocity of the particle [7]:

$$p = \frac{2P}{v}. \quad (3.9)$$

If it fails to permeate the barrier, the simulation will 'bounce' the particle off the barrier. After a time equal to the diffusion time has elapsed, the net displacement, $\langle x \rangle$, of the particle is computed and the apparent diffusion coefficient is given by

$$D_a = \frac{\langle x \rangle^2}{2T_d}. \quad (3.10)$$

The 2-D models in Figs. 3.14 and 3.15 are similar to the two compartment

model described by Szafer *et al.* [7]. The compartment $B_x \times B_y$ in Fig. 3.14 represents the intracellular space of the neuron while the remaining space within $A_x \times A_y$ represents the extracellular space. The barriers represent the effect of the cell membrane and the myelin sheath of the axon. For the white matter fibre simulations B_x was set equal to B_y and was varied from $5 \mu\text{m}$ to $20 \mu\text{m}$.

Diffusion sensitization for the leg muscle experiments is along the z-axis, parallel to the tibia. Relative to the tibia, the longitudinal axis of the soleus muscle fibres make an angle of about 20° [107]. The average length of a sarcomere is $1.5 - 2.5 \mu\text{m}$ [112], depending on the state of the muscle fibre. The 20° inclination of the fibres increases the 'effective' sarcomere length by a small factor of $(\cos 20^\circ)^{-1}$. See Fig. 5.12 for a schematic of the sarcomere and myofibril within a muscle fibre bundle. The myofilaments that make up the sarcomere are also shown. For our experiments, we will assume that all of the sarcomeres are in the resting state, since the volunteer is resting in a supine position with his/her leg inserted into the gradient coil. There are no forces on the leg that would put the soleus muscle in a state of contraction.

In Fig. 3.15A(i), the larger compartment, with dimensions B_x, B_y , represents the length of the sarcomere and the width of the myofibril, respectively. The second compartment with dimensions $B_x, A_y - B_y$, represents the sarcoplasmic reticulum and the sarcoplasm outside of the myofibril. For these simulations, A_x, A_y , and B_y were set equal to $3 \mu\text{m}$, $1.27 \mu\text{m}$, and $1.20 \mu\text{m}$ respectively. The intrinsic diffusion coefficient in each of these compartments may be different, however, we have kept them equal in our simulations. The ends of the sarcomere are assumed to have a permeability, P .

The above model assumes that the myofilaments do not hinder diffusion along

Figure 3.14: 2-D model of human white matter. The bold structure shown is repeated in all directions. $B_x \times B_y$ represents the intracellular space of the neuron while the remaining space within $A_x \times A_y$ represents the extracellular space.

Figure 3.15: Several models of the muscle cell myofibrils used in computer simulations. **A.** 2-D Model. The bold structure (i.e. $A_x \times A_y$) is repeated in all directions. Two 2-D models were simulated in (i) to represent the intra and extra myofibril spaces and in (ii) to represent only the sarcomeres. **B.** Tortuosity Model. The particles must take a tortuous path around the impenetrable blocks $B_x \times B_y$.

the y-direction. This may not be the case: a second valid set of parameters may be to use only one compartment with dimensions B_x, B_y , as in Fig. 3.15A(ii), representing the length of the sarcomere and the spacing of the thin myofilaments respectively. Both of these parameters have been used to simulate the muscle cell geometry.

We have also tried to model the diffusion using a ‘tortuosity’ model [113], as in Fig. 3.15B. The idea behind this model is that the myofilaments and large organic molecules hinder the diffusion by forcing the molecule to take a tortuous pathway around them. The tortuous pathway is modelled by placing impermeable ‘blocks’ within the diffusing medium.

Another model geometry includes a ‘flow channel’, which can simulate flow within the extracellular environment of the white matter, or it can simulate the preferential transport of water along the sarcomere. This is described in more detail in Section 5.1.8 and Section 5.2. The model is shown in Fig. 3.16.

Figure 3.16: The 2-D Monte Carlo model of the myofibril. The periodic structure is $A_x \times A_y$, where the length of the sarcomere is represented by $A_x - B_x$, the width of the myofibril is A_y , the bold lines are impermeable barriers, and the flow channel is the area $B_x \times (A_y - B_y)$. The flow channel area represents the effect of the sarcoplasmic reticulum, which we believe may direct the water motion pathway preferentially along the myofibril.

Table 3.1: A list of all the diffusion sequences and the parameters used. More parameters for the same sequences are also given in Table 3.2.

Sequence name	Type	G_{READ} (mT/m)	G_D dir.	ADC period (μ s)	τ_{du} (μ s)	Filter (kHz)	Column resolution (mm)
YAP	PGSE	$3.0 \hat{y}$	$\hat{y}$	8192	8	30	1.0
YRL	PGSE	$3.0 \hat{y}$	$\hat{x}$	8192	8	30	1.0
YAPL	PGSE	$3.0 \hat{y}$	$\hat{y}$	8192	8	30	1.0
YRLL	PGSE	$3.0 \hat{y}$	$\hat{x}$	8192	8	30	1.0
STEAMYAP	STEAM	$1.0 \hat{y}$	$\hat{y}$	16384	16	31.5	1.4
STEAMYRL	STEAM	$1.0 \hat{y}$	$\hat{x}$	16384	16	31.5	1.4
STEAMYAP_20	STEAM	$1.0 \hat{y}$	$\hat{y}$	16384	16	31.5	1.4
STEAMYRL_20	STEAM	$1.0 \hat{y}$	$\hat{x}$	16384	16	31.5	1.4
STEAMYAP_15	STEAM	$1.0 \hat{y}$	$\hat{y}$	16384	16	31.5	1.4
STEAMYRL_15	STEAM	$1.0 \hat{y}$	$\hat{x}$	16384	16	31.5	1.4
YAP_BIP50	AGPS	$3.0 \hat{y}$	$\hat{y}$	8192	8	30	1.0
YRL_BIP50	AGPS	$3.0 \hat{y}$	$\hat{x}$	8192	8	30	1.0
YAP_BIP37	AGPS	$3.0 \hat{y}$	$\hat{y}$	8192	8	30	1.0
YRL_BIP37	AGPS	$3.0 \hat{y}$	$\hat{x}$	8192	8	30	1.0
YAP_BIP24	AGPS	$1.5 \hat{y}$	$\hat{y}$	8192	8	30	2.0
YRL_BIP24	AGPS	$1.5 \hat{y}$	$\hat{x}$	8192	8	30	2.0
YAP_BIP20	AGPS	$1.5 \hat{y}$	$\hat{y}$	8192	8	30	2.0
YRL_BIP20	AGPS	$1.5 \hat{y}$	$\hat{x}$	8192	8	30	2.0
YAP_BIP14	AGPS	$1.5 \hat{y}$	$\hat{y}$	8192	8	30	2.0
YRL_BIP14	AGPS	$1.5 \hat{y}$	$\hat{x}$	8192	8	30	2.0
COIL_TD50	PGSE	$2.0 \hat{z}$	$\hat{z}$	8192	8	20	1.4
COIL_TD19	PGSE	$2.0 \hat{z}$	$\hat{z}$	8192	8	20	1.4
COIL_TD14	PGSE	$2.0 \hat{z}$	$\hat{z}$	8192	8	20	1.4
COIL_TD9	PGSE	$2.0 \hat{z}$	$\hat{z}$	8192	8	20	1.4
COIL_BITE41	AGPS	$2.0 \hat{z}$	$\hat{z}$	8192	8	20	1.4
COIL_BITE39	AGPS	$2.0 \hat{z}$	$\hat{z}$	8192	8	20	1.4
COIL_BITE77	AGPS	$2.0 \hat{z}$	$\hat{z}$	8192	8	20	1.4

Table 3.2: A list of all the diffusion sequences and the parameters used. More parameters for the same sequences are also given in Table 3.1.

Sequence name	# Acqu.	Col. Area (mm ²)	TE (ms)	τ_2 (ms)	δ (ms)	Δ (ms)	T_d (ms)
YAP	10	10	111	-	36	62.6	50.6
YRL	10	10	111	-	36	62.6	50.6
YAPL	10	10	159	-	40	106.6	93.3
YRLL	10	10	159	-	40	106.6	93.3
STEAMYAP	10	10	89	140.240.355	30	$\tau_2 + 40$	$\tau_2 + 30$
STEAMYRL	10	10	89	140.240.355	30	$\tau_2 + 40$	$\tau_2 + 30$
STEAMYAP ₂₀	10	10	69	490.640	20	$\tau_2 + 30$	$\tau_2 + 23$
STEAMYRL ₂₀	10	10	69	490.640	20	$\tau_2 + 30$	$\tau_2 + 23$
STEAMYAP ₁₅	14	10	59	800.1000	15	$\tau_2 + 25$	$\tau_2 + 20$
STEAMYRL ₁₅	14	10	59	800.1000	15	$\tau_2 + 25$	$\tau_2 + 20$
YAP_BIP50	20	10	182	-	24	58.6	50.6
YRL_BIP50	20	10	182	-	24	58.6	50.6
YAP_BIP37	20	10	165	-	28	46.3	37.0
YRL_BIP37	20	10	165	-	28	46.3	37.0
YAP_BIP24	20	10	161	-	35	36	24.3
YRL_BIP24	20	10	161	-	35	36	24.3
YAP_BIP20	20	10	133	-	28	29	19.7
YRL_BIP20	20	10	133	-	28	29	19.7
YAP_BIP14	20	10	177	-	19	20	13.7
YRL_BIP14	20	10	177	-	19	20	13.7
COIL_TD50	10	20	69	-	3	51	50
COIL_TD19	10	20	41	-	5	20.7	19
COIL_TD14	10	20	36	-	5	15.7	14
COIL_TD9	10	20	28	-	5	10.7	9
COIL_BITE41	10	20	41	-	4.5	7.5	6
COIL_BITE39	10	20	39	-	4.5	6.5	5
COIL_BITE77	10	20	77	-	3	5	4

Chapter 4

Results and Analysis

4.1 White Matter Data

A total of 12 volunteers were studied for diffusion of water in the white matter of the brain. Data for long diffusion times, $50.6 \text{ ms} \leq T_d \leq 1020 \text{ ms}$, were obtained on six of these volunteers while data for short diffusion times, $13.7 \text{ ms} \leq T_d \leq 50.6 \text{ ms}$, were obtained on the remaining 6 volunteers.

For a diffusion time of $T_d = 170 \text{ ms}$, we show in Fig. 4.1(a) the frequency domain signal profiles obtained from each G_D value used in the sequence. From these signal profiles, a diffusion profile is obtained by fitting each point from the *same* location to Eq. (2.78) using a least squares fitting algorithm. An example is shown in Fig. 4.1(b), where data from a location $Y \approx 32 \text{ mm}$, shown by the arrow in Fig. 4.1(a), are plotted vs. the b factor and a line representing a linear least squares fit to the points is shown. No systematic deviation from exponentiality was observed for any of these decays. This issue will be addressed again in Section 5.1.5.

Fig. 4.1 demonstrates the noise level and typical scatter of the points for these diffusion measurements. The noise level is high here (relative to measurements on the leg muscle tissue in Section 4.5) because of the long TE required to accomodate

(a)

(b)

Figure 4.1: (a) Diffusion weighted signal profiles for each value of G_D obtained from the sequence STEAMYRL ($T_d = 170$ ms) for one volunteer. The right side of the profiles correspond to positions at the back of the head while the left side of the profiles corresponds to the front. A diffusion profile is obtained from the curves shown by fitting each point from the *same* location to a least squares fitting algorithm. An example is shown in (b), where the attenuation of the NMR signal, $\ln M(b)$ vs. b , in human white matter in vivo is plotted. A semi-log plot is used to show the exponential dependence. The data shown here are taken from the position shown by the arrow in (a) ($Y \approx 32$ mm), where the diffusion sensitization is perpendicular to the white matter fibres.

the long duration of the diffusion gradients. Although our acquisition technique has good immunity to bulk tissue motion due to its lack of phase encoding and the averaging of several signals, part of the scatter may also be associated with relative motion of the brain caused by CSF pulsations. Involuntary head movements from the volunteer will also cause errors in spatial encoding, which could contribute to the scatter. This contribution, however, is considered to be less important. The effect of CSF pulsations will be analysed in more detail in Section 5.1.7.

For each volunteer, diffusion profiles were obtained for a set of T_d values corresponding to either $T_d \geq 50.6$ ms or $T_d \leq 50.6$ ms. A complete set of diffusion profiles is shown in Fig. 4.2. The profiles for the short diffusion times are shown for one volunteer and the profiles for the long diffusion times are shown for a different volunteer. Both short and long diffusion time protocols measured diffusion for $T_d = 50.6$ ms, so as to have some overlap.

Anisotropy can clearly be seen in the diffusion profiles for $T_d \geq 50.6$ ms by comparing the right and left hand columns in Fig. 4.2. These results are consistent with previously published work [28]. In the central region $D_a^{RL} > D_a^{AP}$, where D_a^{RL} is the apparent diffusion coefficient measured with G_D oriented RL, and D_a^{AP} is the apparent diffusion coefficient measured with G_D oriented AP. However, this behaviour is reversed at anterior and posterior locations. This is not surprising since the orientation of the white matter fibres, the structure that is primarily responsible for the observed anisotropy, is predominantly RL in the central region whereas in anterior and posterior locations it is predominantly AP [28, 107]. We believe the extrema in Fig. 4.2 correspond to locations where the observed behaviour is dominated by diffusion in fibres oriented nearly parallel or perpendicular to G_D . For this reason we have decided to concentrate our attention on these locations. At

all positions along the COI there will be white matter fibres with various orientations and there will be other cell types as well as extracellular fluid that affect the observed behaviour. However, the relative contribution from these sources should be substantially reduced at positions corresponding to the extrema in the diffusion profiles, shown by the arrows in Fig. 4.2.

From the values of the central extrema ($Y \approx -16$ mm) in Fig. 4.2, we have plotted D_a^{RL} and D_a^{AP} as a function of $T_d^{1/2}$ in Fig. 4.3(a). Similar plots for D_a^{RL} and D_a^{AP} for the extrema in the posterior location ($Y \approx 32$ mm) are given in Fig. 4.3(b). The error on the data, σ_{D_a} , in Fig. 4.2 is obtained from Eq. (3.8), as described in Section 3.6. However, σ_{D_a} is not shown in the figures because the graphs would look too busy and also because σ_{D_a} is usually less than half the root mean square variation in the profile at the given location. The error bars in Fig. 4.3 represent our careful estimate of the root mean square variation in the profile at the locations given by the arrows in Fig. 4.2. The error from the linear least squares fit, σ_{D_a} , is negligible compared to the error obtained from the signal variation.

The diffusion plots for the other volunteers are completely consistent with the plots in Fig. 4.3. The diffusion profiles for all volunteers were averaged and are shown in Fig. 4.4. The error on the data is the standard deviation of the mean.

The very close agreement between the central and posterior data strongly supports our position that the D_a values at these two locations are dominated by fibres that are perpendicular and parallel to the COI, respectively.

The profiles for D_a^{AP} in Fig. 4.4(a) (solid circles) and D_a^{RL} in Fig. 4.4(b) (open circles) decrease significantly as T_d increases and asymptotically approach a value close to $0.60 \pm 0.05 \times 10^{-3} \text{ mm}^2/\text{s}$. This is the type of behaviour expected for diffusion which is impeded by a semi-permeable barrier. The profiles for D_a^{RL} in Fig. 4.4(a)

Figure 4.2: Diffusion profiles for all of the T_d values studied. Diffusion sensitization for both AP and RL directions are shown.

(i)

(j)

(k)

(l)

(m)

(n)

(o)

(p)

Figure 4.3: D_a vs. $T_d^{1/2}$ for water in human white matter at (a) a central location in the column ($Y \approx -16 \text{ mm}$), and in (b) a posterior location in the column ($Y \approx 32 \text{ mm}$), in the corona radiata for 1 volunteer. Due to the large acquisition time, the data for $T_d \leq 50.6 \text{ ms}$ ($T_d^{1/2} \leq 0.225 \text{ s}^{1/2}$) were acquired in a different session (a different day) than the data for $T_d \geq 50.6 \text{ ms}$. Profiles for the diffusion sensitization gradient oriented AP and RL are shown. The above profiles are typical of all of the volunteers studied.

Figure 4.4: D_a vs. $T_d^{1/2}$ for water in human white matter averaged over all volunteers. Profiles for the diffusion sensitization gradient oriented AP and RL are shown for (a) a central location of the column, and in (b) a posterior location of the column, in the corona radiata.

(open squares) and D_a^{AP} in Fig. 4.4(b) (solid squares) show a more T_d independent behaviour. This is consistent with diffusion in the absence of restricting barriers and is the expected behaviour when G_D is applied parallel to the fibres. The slight decrease observed in the profiles is probably due to a small percentage of fibres that are not oriented parallel to G_D . Small amounts of grey matter may be contributing to the signal in the posterior region of the column, explaining the overall smaller values of D_a in Fig. 4.4(b).

We believe our work is the first to show, unambiguously [31, 58, 64], the asymptote for diffusion across the fibres, and the expected behaviour of restricted diffusion both across and along the fibre direction. We have also covered a very large range of diffusion times in our protocol, which is necessary in order to fully observe the time dependence of the diffusion coefficient. As far as we know, our data cover the widest range of T_d in the literature for a human study [31, 58, 64]. We also report D_a for the shortest diffusion time (13.7 ms) yet measured in human white matter in vivo [31, 37, 58, 64], yet the sequence for this T_d has a b_{max} of 122 s/mm^2 , which is larger than other published measurements of D_a for short T_d values. (i.e. $b_{max} = 95 \text{ s/mm}^2$ [58], $b_{max} = 44 \text{ s/mm}^2$ [45]). At the low values of T_d , we observe that the anisotropy is removed in Fig. 4.4(a) and close to being removed in Fig. 4.4(b). This may be useful for DWI. Diffusion weighted images free from signal intensity variations from anisotropy have been shown [29] to be more effective in visualizing tumours.

4.2 Phantom Results

To demonstrate the accuracy of the experimental protocol, and to ascertain that neither the anisotropy nor the time dependent behaviour of D_a observed in our

measurements were due to systematic artifacts or sequence variability, we have repeated the full experiment on a water and acetone phantom. Fig. 4.5 shows D vs. $T_d^{1/2}$ for water in the lower curves and the upper curves are D vs. $T_d^{1/2}$ for acetone. As expected, the anisotropy in D does not exceed $\pm 0.1 \times 10^{-3} \text{mm}^2/\text{s}$ for water or acetone. The errors in the data are obtained using the same method described for Fig. 4.3 in Section 4.1, or it is σ_{D_a} from Eq. (3.8) in Section 3.6, whichever is greater. Also, the profiles are constant with respect to $T_d^{1/2}$ within $\pm 0.1 \times 10^{-3} \text{mm}^2/\text{s}$. All values of D show excellent agreement with literature values of the diffusion coefficient of water [51, 52] ($2.05 \times 10^{-3} \text{mm}^2/\text{s}$ at 22°C) and acetone [114, 67] ($4.0 - 4.6 \times 10^{-3} \text{mm}^2/\text{s}$). Representative signal decay plots are shown in Fig. 4.6.

We have also repeated our experimental protocol on a ‘fibre’ phantom (see Section 3.3). This known anisotropic system further confirms the reliability of our experimental results. It was also performed to confirm the reliability of the Monte Carlo simulations (see Section 4.3). Fig. 4.7 shows D_a vs. $T_d^{1/2}$ for water within the fibre phantom, both parallel and perpendicular to the fibres. Parallel to the fibres, D_a shows the expected time independent behaviour for free, unhindered diffusion, while there is a time dependence for D_a perpendicular to the fibres, indicating that the fibre walls are restricting the water mobility. The diffusion profiles showed no dependence on position for any of the T_d values studied. The signal decay plots were observed to be exponential to within experimental error. See Fig. 4.16 for the results of the Monte Carlo simulation for this phantom. The errors in Fig. 4.7 are obtained using the same method described for Fig. 4.5.

Figure 4.5: Self Diffusion coefficient of water and acetone at 22°C . The measurements were obtained using the same protocol as for the human data. As expected, there is no significant dependence on the diffusion time or the direction of the diffusion sensitizing gradient (G_D).

Figure 4.6: Attenuation of the NMR signal, $\ln M(b)$ vs. b , for water (circles) and acetone (squares) at 22°C for a diffusion time of $T_d = 37$ ms. The error in the data is $\approx 0.7\%$, less than the size of the symbols. A linear least squares fit of the data gives a slope of $-2.17 \pm 0.01 \times 10^{-3} \text{ mm}^2/\text{s}$ for water and $-4.34 \pm 0.01 \times 10^{-3} \text{ mm}^2/\text{s}$ for acetone.

Figure 4.7: D_a vs. $T_d^{1/2}$ for water within a Baxter Cellulose Acetate Hollow Fiber Dialyzer, model C'A-170. The water was inside the hollow fibres, which had a diameter of $200 \mu\text{m}$. Diffusion sensitization was parallel to the fibres for the square symbols and perpendicular to the fibres for the circles. The temperature of the water was 20°C .

4.3 Monte Carlo Results

For the Monte Carlo algorithms described in Section 3.7, we must be certain that the number of iterations and the diffusing step size are set at points equal to or beyond the point of convergence. That is, we must be sure that our results do not change due to a lack of iterations. We used 20000 iterations (the number of particles in the system) for all simulations. In Fig. 4.8, we show the diffusion coefficient vs. number of iterations for the 2-D Monte Carlo simulation, keeping all other simulation parameters constant. The results indicate that 20000 iterations are well past the point of convergence.

We used a step size of $5 \mu\text{s}/\text{step}$ for simulating our head data (lowest diffusion time is 13.7 ms) and $2 \mu\text{s}/\text{step}$ for simulating our leg data (lowest diffusion time is 4 ms). Fig. 4.9 shows the diffusion coefficient vs. diffusing step size for $T_d = 13.7$ ms and $T_d = 4$ ms, keeping all other simulation parameters constant. Fig. 4.9 shows that convergence is reached at about $50 \mu\text{s}/\text{step}$ for both of these T_d 's.

We have run the 1-D and 2-D Monte Carlo simulations to model our head data shown in Fig. 4.4. It was found that both the 1-D and 2-D two compartment models predict that the asymptote is reached well before it is reached in our experimental results. Simulations using a larger barrier spacing than $a = 5 \mu\text{m}$ show better agreement with our data. Fig. 4.10 shows the 1-D and 2-D Monte Carlo results for barrier spacings of $a = 5 \mu\text{m}$ and $a = 20 \mu\text{m}$. Fig. 4.11 shows the best fit obtained from the 2-D Monte Carlo simulation to the head data. The barrier spacings in this simulation are $a = 20 \mu\text{m}$ for the APC data and $a = 14 \mu\text{m}$ for the RLP data.

The barrier spacings of $a = 20 \mu\text{m}$ and $a = 14 \mu\text{m}$ are close to the known average axon diameter of $5 \mu\text{m}$ in human white matter [36, 115], but unfortunately,

Figure 4.8: D_x vs. number of iterations, N . The 2-D Monte Carlo code is tested for convergence by changing the number of iterations. The other parameters used were $a = 5 \mu\text{m}$, $f = 0.7$, $T_d = 13.7 \text{ ms}$, $D_I = D_E = 3.0 \times 10^{-3} \text{ mm}^2/\text{s}$, $P = 0.005 \text{ cm/s}$, and a diffusion step time of $5 \mu\text{s}/\text{step}$. The data show convergence at 3000 iterations, however, 20000 iterations were used in the simulations.

Figure 4.9: Da vs. step size. The 2-D Monte Carlo code is tested for convergence by changing the duration of the diffusing step for $T_d = 13.7 \text{ ms}$ in (a) and $T_d = 4 \text{ ms}$ in (b). The other parameters used were $a = 5 \mu\text{m}$, $f = 0.7$, $D_I = D_E = 3.0 \times 10^{-3} \text{ mm}^2/\text{s}$, $P = 0.005 \text{ cm/s}$, and $N = 20000$ iterations. The data show convergence at a step size of about $50 \mu\text{s}/\text{step}$ in both (a) and (b).

Figure 4.10: Monte Carlo simulation of the apparent diffusion coefficient D_a vs. $T_d^{1/2}$ for a 1-D model (bold symbols) and 2-D model (open symbols). A small barrier spacing of $5 \mu\text{m}$ was used (squares) as well as a large barrier spacing of $20 \mu\text{m}$ (circles).

(a)

(b)

Figure 4.11: Comparison of D_a vs. $T_d^{1/2}$ for water in human white matter in vivo for (a) the Central AP location, as in Fig. 4.4(a), and the Monte Carlo simulation for $a = 20 \mu\text{m}$, $P = 0.003 \text{ cm/s}$, as in Fig. 4.10. In (b), the Posterior RL location is shown, as in Fig. 4.4(b), and the Monte Carlo simulation for $a = 14 \mu\text{m}$, $P = 0.004 \text{ cm/s}$, is shown, as in Fig. 5.4. Both (a) and (b) used a cellular volume fraction of $f = 0.8$ with $D_I = D_E = 3.0 \times 10^{-3} \text{ mm}^2/\text{s}$. Good agreement is obtained between the experimental and simulated curves.

when the simulation uses a barrier spacing of $a = 5 \mu\text{m}$, the data for low T_d 's are not in agreement. The barrier spacings of $a = 20 \mu\text{m}$ and $a = 14 \mu\text{m}$ are, however, in complete agreement with the results of others [116, 117] who have obtained MR data in human white matter in vivo. Even though the average axon diameter in white matter is $5 \mu\text{m}$, there exists a range of axon diameters in humans from $1 - 30 \mu\text{m}$ [36, 115]. Despite this, it is unclear at this point why the 'best fit' to the experimental data is obtained with a simulated barrier size of $a = 20 \mu\text{m}$ and $a = 14 \mu\text{m}$. We will discuss this further in Chapter 5.

We have confirmed that the 2-D Monte Carlo program is operating correctly by comparing its results to several other works, both theoretical and experimental. We also compare the simulation with the 'fibre' phantom results in Fig. 4.7. We now describe each of these comparisons.

The 2-D Monte Carlo algorithm uses a geometry, explained in Section 3.7, identical to the geometry of the Monte Carlo program of Szafer *et al.* [7]. As a first test, then, we compare our data with their published results. Fig. 4.12 shows two of the figures presented in Szafer's paper and we also show our Monte Carlo results for the same parameters. Excellent agreement is obtained, as expected.

To show that the algorithm itself is valid, we compare our results with a complex theoretical equation derived by Tanner [54] (see Section 2.7). The equation predicts the diffusion coefficient as a function of time in a system of semi-permeable regularly spaced planar barriers. We can simulate this geometry in our program by making the intracellular space take up 100% of the available space, that is, by setting $B_x = A_x$ and $B_y = A_y$ (see Section 3.7). We have run our program for a barrier spacing of $5 \mu\text{m}$, and several values of permeability as shown in Fig. 4.13. The data in Fig. 4.13 are presented as reduced parameters. That is, 'reduced time'

Figure 4.12: D_a vs. T_d from Monte Carlo simulations in the literature [7] is shown in (a) and (c) and data from our 2-D Monte Carlo program, using identical simulation parameters, are shown in (b) and (d). (a) and (b) show the change in the diffusion profile for several values of barrier permeability. (c) and (d) show the change in the diffusion profile for several values of barrier spacing, a . The parameters used were $N=10000$ iterations, step size = $5 \mu\text{s}/\text{step}$, $D_I = 1.0 \times 10^{-3} \text{ mm}^2/\text{s}$ and $D_E = 2.5 \times 10^{-3} \text{ mm}^2/\text{s}$. Our data show excellent agreement with the literature Monte Carlo results.

is defined as $T_r = D_I(0)T_d/a^2$, and reduced permeability is $P_r = aP/D_I(0)$. The diffusion time, T_d , ranged from 0 to 83.3 ms ($T_d^{1/2} = 0.289 \text{ ms}^{1/2}$) which corresponds to only a small part of the range of T_d values investigated experimentally. For larger values of T_d it became increasingly difficult and time consuming to get Tanner's equations to converge. However, it should be clear from Fig. 4.13 that our data are in very close agreement with Tanner's theoretical curves.

We have repeated the simulations for the 3 profiles shown in Fig. 4.13, using instead a $15 \mu\text{m}$ barrier spacing, to see how sensitive the profile is to this parameter. Fig. 4.14 shows the deviation of the profiles when this is done, for $P_r = 2.0$ and $P_r = 0.5$. The bold symbols are the new profiles with $a = 15 \mu\text{m}$ and the open symbols are the original profiles in Fig. 4.13. The sensitivity of the result to the parameter a shows that the agreement in Fig. 4.13 represents a good test of the Monte Carlo code's simulation of the barrier spacing. The $P_r = 0.001$ curve in Fig. 4.13 does not change appreciably because of the very low value of permeability.

Tanner and Stejskal [118, 119] have also performed experiments of restricted diffusion of water in artificial systems of thin liquid layers. One of their systems consisted of a stack of mica leaves separated by thin aluminum foil spacers, all held in place by string. Water is taken up into the space between the mica leaves, which had an average separation of $14.5 \pm 1.3 \mu\text{m}$. They measured the diffusion attenuation of the water within the stack with a pulsed gradient spin echo (PGSE) technique (see Section 2.3), for several diffusion times. This system is ideally represented by our Monte Carlo geometry (see Section 3.7). We have modelled their experimental system and we show the results of the simulation in Fig. 4.15. There are no adjustable input parameters to the simulation. The permeability, P , was set to $P=0$. The 'free' diffusion coefficient of the water, $D_I = D_E$, was

Figure 4.13: $D_a(T_d)/D_I(0)$ vs. T_r for several values of reduced permeability. $P_r = aP/D_I(0)$. The bold lines are from a theoretical equation derived by Tanner [54], which predicts the apparent diffusion coefficients as a function of diffusion time for a system of planar barriers. The normalized diffusion coefficients are plotted vs. the reduced time, defined as $T_r = T_d D_I(0)/a^2$. The square symbols are the results from our 2-D Monte Carlo program, using similar parameters, with $a = 5 \mu\text{m}$ and $D_I(0) = 3.0 \times 10^{-3} \text{mm}^2/\text{s}$.

Figure 4.14: $D_a(T_d)/D_I(0)$ vs. T_r for several values of reduced permeability, $P_r = \alpha P/D_I(0)$. The open symbols are data from Fig. 4.13 for comparison and the bold symbols are the profiles when the barrier spacing is changed from $5 \mu\text{m}$ to $15 \mu\text{m}$, with the other parameters being unchanged for the same symbol type.

set according to their reported water temperature of $26 \pm 1^\circ\text{C}$. The barrier spacings were set according to the average separation of the mica leaves.

We have set the A_y parameter slightly larger to allow for a 3% unrestricted volume. This was done because of Tanner's [118] comment,

"...3% of the water volume was unrestricted over the strings and edges of the stack."

We obtain excellent agreement with Tanner's data in Fig. 4.15. The agreement with the experimental data, without any adjustable input parameters to the simulation, clearly demonstrates that our 2-D Monte Carlo simulation is operating correctly.

As a final test, we have used a dialysis filter as a phantom for restricted diffusion. The phantom is described in Section 3.3. The experimental data from the phantom were shown earlier in Fig. 4.7.

Fig. 4.16 shows the results from the 2-D Monte Carlo simulation of the phantom and the experimental results are shown for comparison. Once again, there are no adjustable input parameters to the simulation. The inner diameter of the fibres [104] is known to be $200\ \mu\text{m}$. (the error on the diameter is unknown.) The temperature of the water bath in the experiment was $20.9 \pm 0.4^\circ\text{C}$, and the permeability of the fibres to water is zero. Only the interior of the fibres was filled, so there is no 'extracellular' fraction.

The good agreement of the simulated data with the experimental data in Fig. 4.16, shows that our Monte Carlo program is working as it was designed to function. It correctly predicts the restricted diffusion coefficients in simple systems.

The success of our simulation in Figs. 4.13, 4.14, 4.15 and 4.16 shows that the algorithm is correct, and our Monte Carlo data are correct, for the geometry that

Figure 4.15: D_a vs. T_d for water in the mica stack phantom described in the text. The experimental data points are from Tanner [119] and the upper and lower curves are from the 2-D Monte Carlo simulation. The error on the experimental values for barrier spacing and temperature give upper and lower values to be used as input parameters for the simulation. For the upper curve, we used $A_x = B_x = B_y = 15.8 \mu\text{m}$, $A_y = 16.29 \mu\text{m}$, and $D_I = D_E = 2.33 \times 10^{-3} \text{mm}^2/\text{s}$. For the lower curve, we used $A_x = B_x = B_y = 13.2 \mu\text{m}$, $A_y = 13.61 \mu\text{m}$, and $D_I = D_E = 2.22 \times 10^{-3} \text{mm}^2/\text{s}$. For both simulations, we used $P=0$.

Figure 4.16: Da vs. $T_d^{1/2}$ for water in a fibre phantom. The experimental data points are the bold circles and the open circles are from the 2-D Monte Carlo simulation. For the simulation, we used $A_x = A_y = B_x = B_y = 200 \mu\text{m}$, $D_I = D_E = 1.99 \times 10^{-3} \text{mm}^2/\text{s}$, and $P=0$. The error bars on the simulated data represent the range of values obtained when the simulation was repeated using $D_I = D_E = 2.01 \times 10^{-3} \text{mm}^2/\text{s}$ and $1.97 \times 10^{-3} \text{mm}^2/\text{s}$, in accordance with our experimental error on the temperature reading.

the program was designed to simulate. The discrepancy of the simulation with the experimental data *in vivo*, (see Figs. 4.10 and 4.11), is probably because the Monte Carlo geometry does not take into account the many complex physiological processes occurring in the neurons (e.g. extracellular flow from pressure applied from CSF pulsations). Neurons are also cylindrical in shape, not square, as in the simulation. This approximation, however, should only result in a minor variation in D_a . Furthermore, the fibres in the dialysis phantom are cylindrical, yet we see only a minor variation in the data in Fig. 4.16. Another possibility is that compartmentalization with slow exchange may exist in the white matter, and that each compartment may have very different $D_a(0)$'s. These possibilities are explored further in Section 5.1.

4.4 Gradient Coil Calibration

The isocentre of the gradient coil was calibrated using a 2 cm³ water phantom as described in Section 3.4.1. The coil's gradient amplitude G_z vs. Current is shown in Fig. 4.17. It is from this important calibration line that we obtain the b -values for the diffusion sequences on the leg. The gradient pulse profile, obtained from an oscilloscope trace, is converted from a 'current vs. time' profile to a 'gradient(mT/m) vs. time' profile using the calibration constant, $7.38 \pm 0.05 \frac{mT}{m-A}$, from Fig. 4.17. Then, the profile is used by DPRO (see Section 3.6) to compute the b -values for a specific sequence.

The error in the data points are obtained by standard propagation of error techniques [110], using Eq. (3.2). The error increases as G_z increases because, from Eq. (3.2), the phantom profile is wider (in kHz) at high G_z , resulting in a lower S/N. This is one reason why gradient coil positions off centre were not

calibrated using this technique. Although S/N is the same for any position, the profile is offset from 0 kHz by $\frac{\gamma}{2\pi}G_z \times (\text{position offset}, z_o)$. This effectively means that, for positions off centre by ≥ 1 cm, some parts of the profile will have frequency components exceeding the bandwidth of the ADC, so the profile will be cut off, and its width is impossible to determine.

The calibration profile for the entire coil volume is shown in Fig. 4.18. This 3-D plot is essentially an error map of the gradient amplitude for $I=12$ Amps. An ideal gradient coil would have a flat plane profile, or a constant gradient value everywhere. Fig. 4.18 shows that our coil's gradient is linear to within $\pm 1.5\%$ within a 5.5 cm radius sphere. The agreement of the calibration profile with the theoretical profile is excellent. The theoretical profile is shown in Fig. 4.19, and was obtained by a computer model of the wire loop positions, and Eqs. (2.99) to (2.101).

4.5 Leg Muscle Data

In this study, we measured the diffusion coefficient of water in the soleus muscle (see Fig. 3.12 in Section 3.5.) of the leg from 7 normal volunteers. A typical signal decay curve is shown in Fig. 4.20(b). No systematic deviation from exponentiality was observed for these decay curves. The scatter for these decay curves is lower than for the measurements in the brain since, unlike in the brain where complications such as CSF motions may cause some scatter, the muscle tissue does not move. The error in the data points is σ_{M_j} , as described in Section 3.6. The decay curve was obtained from the data in Fig. 4.20(a) at a position $z=-10$ mm (shown by the arrow). Fig. 4.20(a) shows each of the signal profiles obtained, (corresponding to each G_D amplitude) similar to Fig. 4.1 in Section 4.1. For this particular volunteer,

Figure 4.17: G_z vs. current for a 2 cm^3 water phantom positioned in the coil's isocentre. The line shown is a linear least squares fit to the data, and has a slope of $7.38 \pm 0.05 \frac{\text{mT}}{\text{m-A}}$. This value was used as the calibration constant to determine the b -values for the diffusion sequences.

Figure 4.18: 3-D plot of the coil's gradient amplitude vs. axial position within the coil, z and vs. the radial position within the coil R , for a current of $I=12$ Amps. Ideally the profile should be a flat plane. The profile shows that our coil's gradient is linear to within $\pm 1.5\%$ within a 5.5 cm radius sphere.

Figure 4.19: Theoretical 3-D profile of the gradient coil's amplitude vs. axial and radial position, z and R . The plot was generated by computer modelling as described in the text. This plot shows that we should expect a $\pm 1.5\%$ linearity from our coil for positions within a 5.5 cm diameter sphere. The experimental profile is shown in Fig. 4.18.

the $20 \times 20 \text{ mm}^2$ column was placed at a location $R = 10 \text{ mm}$ off axis. From Fig. 4.18, this gives an axial range of -55 mm to $+55 \text{ mm}$ where the magnetic field gradient inhomogeneity is $\leq \pm 1.5\%$. For the higher values of G_D in Fig. 4.20(a) (the lower amplitude curves), peaks in the curve can be seen at $\pm 120 \text{ mm}$. These peaks are due to the gradient coil's large inhomogeneity at this location. The same peaks are seen in a similar plot for the diffusion weighted profiles of a water phantom, shown in Fig. 4.21. Bold, vertical lines are placed on the graph to show the $\pm 1.5\%$ cut off position in the magnetic field gradient inhomogeneity. The spatial variations in the profiles in Fig. 4.20(a), for $-55 \text{ mm} \leq Y \leq 55 \text{ mm}$, are likely indicative of a distribution of water density or T_2 relaxation times in the muscle tissue. However, since the variation occurs even for $b \sim 0$, it is not a diffusion effect. This can also be seen from Fig. 4.22(f) (the corresponding diffusion profile from the data in Fig. 4.20(a)) since D_a is quite constant over this region.

As explained in Section 4.1, from Fig. 4.20, we can obtain a diffusion coefficient profile. We show typical diffusion profiles in Fig. 4.22 for each of the 7 diffusion times investigated. For our analysis of the time dependence of the diffusion coefficient, we choose a location on the profiles where the MRI images show that our column is encapsulating only tissue from the soleus muscle. Fig. 3.12 shows a typical example of the placement of the column and the position where mostly soleus muscle is included in the COI. For the volunteer in Fig. 4.22, this position is $z = -10 \text{ mm}$, and is shown by the arrows. From the diffusion coefficients, D_a , obtained at these locations, we have plotted D_a vs. T_d in Fig. 4.23(a). The error on the data, σ_{D_a} , in Fig. 4.22 is obtained from Eq. (3.8), as described in Section 3.6. However, σ_{D_a} is not shown in the figures because the graphs would look too busy and also because σ_{D_a} is usually less than half the root mean square variation in the profile

Figure 4.20: (a) Diffusion weighted profiles for each value of G_D obtained from the sequence COIL-TD19 ($T_d = 19$ ms) for one volunteer in human soleus leg muscle. A diffusion profile is obtained from the curves shown by fitting each point from the *same* location to a least squares fitting algorithm. An example is shown in (b), where the attenuation of the NMR signal, $\ln M(b)$ vs. b , for water in human soleus leg muscle in vivo is plotted. This example is typical of the decay curves obtained in the leg. From the slope of the line, we obtain $D_a = 1.47 \pm 0.03 \times 10^{-3} \text{ mm}^2/\text{s}$. For this particular volunteer, the column was placed at a location $R = 10$ mm off axis. From Fig. 4.18, this gives a range of -55 mm to $+55$ mm where the magnetic field gradient inhomogeneity is $\leq \pm 1.5\%$.

Figure 4.21: Diffusion weighted profiles for each value of G_D obtained from the sequence COIL-TD14 ($T_d = 14$ ms) on a water phantom. The solid, vertical black lines on the graph show the $\pm 1.5\%$ cut off positions in the magnetic field gradient inhomogeneity. The magnetic field gradient profile is shown in Fig. 4.18.

at the given location. The error bars in Fig. 4.23(a) represent our careful estimate of the root mean square variation in the profile at the locations given by the arrows in Fig. 4.22. The error from the linear least squares fit, σ_{D_a} , is negligible compared to the error obtained from the signal variation.

We see a small decline in D_a with T_d in Fig. 4.23(a), suggesting that restricted diffusion effects are occurring along the long axis of the muscle fibres in the soleus muscle. In order to increase accuracy in our measurements, and to establish reproducibility of the experiment, we have repeated the measurements on 6 other normal volunteers. All of the diffusion plots showed similar declines. The average of the 7 volunteers' diffusion profiles is shown in Fig. 4.23(b). The error on the data is the standard deviation of the mean.

To demonstrate that the variations in the diffusion profile are from restriction in the leg muscle, the experiment was also performed on water and acetone phantoms. The resulting diffusion coefficients, shown in Fig. 4.24, are independent of diffusion time within the experimental error, and agree with literature values of $2.05 \times 10^{-3} \text{mm}^2/\text{s}$ and $4.0 - 4.6 \times 10^{-3} \text{mm}^2/\text{s}$, for water and acetone respectively [51, 52, 67]. The errors in the data are obtained using the same method described for Fig. 4.3 in Section 4.1, or it is σ_{D_a} from Eq. (3.8) in Section 3.6, whichever is greater.

The diffusion data for the human leg in Fig. 4.23 show a slight decline for increasing diffusion time. Although the magnitude of the drop is only 22%, the decline is large enough that it cannot be explained by experimental error fluctuations, since the water and acetone data do not vary by more than 5%. Also, the profiles for each volunteer show the same decline in D_a , all by a magnitude greater than 15%.

Figure 4.22: Typical diffusion profiles from one volunteer for human soleus muscle in vivo. Diffusion profiles for $T_d = 4, 5, 6, 9, 14, 19$, and 50 ms are shown in (a), (b), (c), (d), (e), (f), and (g), respectively.

Figure 4.23: (a) The apparent diffusion coefficient, D_a vs. T_d in human soleus muscle is shown for one volunteer. As described in the text, each data point is taken from the region shown by the arrows in Fig. 4.22. (b) D_a vs. T_d in human soleus muscle, averaged over 7 normal volunteers. The slight decline seen in the diffusion profile suggests restricted diffusion is occurring along the z-direction within soleus muscle.

Figure 4.24: The diffusion coefficient, D vs. T_d for water at $T = 22^\circ\text{C}$ and acetone at $T = 23^\circ\text{C}$. As expected, there is little variation in the diffusion coefficient for the different sequences.

Chapter 5

Discussion and Further Results and Analysis

5.1 White Matter Experiments

There are many structures and processes that could, and most likely do, have an effect on the mobility of water molecules in the brain. Structures at the molecular level (e.g. large proteins) will affect D_a only for extremely short diffusion times, well below the range of diffusion times that we can study on the MR imager at the Ottawa General Hospital. For the smallest diffusion times that we can measure, the average diffusion distances will be of the order of a few microns which is about the size of the diameter of a typical axon. Thus, it is reasonable to expect that the main properties affecting D_a , for the range of diffusion times that we have measured, will be associated with structures, or compartments, that are greater than 1 micron in size. Intracellular and extracellular water should make up $\sim 95\%$ of our NMR signal [64, 120]. Each of these compartments may have different $D_a(0)$'s, and may experience different levels of restriction. Other compartments to be considered are the blood capillaries, and the bodies of the neurons and the neuroglia. Let us consider these compartments in view of our experimental data.

In the human cerebrum, there are two main types of cells, the neurons and the neuroglia. The neuroglia outnumber the neurons by more than 10 to 1 [121]. There are in turn, several types of neuroglia. There are the astrocytes which, in the white matter, form a web of fibrous, connective tissue among the neurons [122]. They also surround the blood capillaries and form part of the blood brain barrier [107]. Based on anatomical considerations [122] of the relative size and number of astrocytes in the white matter we believe that the astrocytes do not contribute significantly to the NMR signal. Thus they should not play a major role in the restriction effect seen in white matter [36, 112]. The random orientation of the astrocytes [122] means that they would not lead to the anisotropic diffusion results seen in white matter. We will categorize the water signal from the astrocytes along with the extracellular water. The extracellular water should therefore be considered to be the water 'outside' the neurons in the white matter.

The oligodendrocyte is another kind of neuroglia found in the white matter. The myelin sheath of the axon is one of the 'branches', or processes of the oligodendrocyte. The white matter is made up mainly of axons surrounded by such a myelin sheath. Numerous axons are connected to the oligodendrocyte in this way. Based on anatomical considerations [107] of the relative size and number of oligodendrocytes, we believe that, like the astrocytes, the oligodendrocytes do not contribute significantly to the NMR signal, or the observed directional dependence of D_a .

The neurons are cells which consist of a cell body, an axon, and one or more dendrites [107]. The cell body of the neuron is found in the cerebral cortex, or the subcortical gray matter [112]. The axon of the cortical neuron originates at the cell body and extends to other parts of the cerebral cortex, or to the spinal neurons

along well defined routes or tracts. Each tract in the brain consists of a bundle of myelinated axons which connect one area of the brain with another [36]. The white matter is made up mainly of axons that have been myelinated by the processes of the oligodendrocytes [107]. These bundles of fibre tracts have a clear organization, and their directional orientation [36] gives rise to the diffusional anisotropy seen in our experiments. The bulk of our NMR signal comes from axonal water in the myelinated fibre tracts [120], water in the myelin, and extracellular water [64].

Finally, there is the vasculature. The water molecules inside the major blood vessels of the brain are pushed along by the pulsatile flow of the blood. The flow rates in these vessels are high enough, however, that this motion is not detected in MRI as a diffusion effect. In the capillaries, the flow rate is much lower and can sometimes mimic a diffusion process. The amount of MR signal that comes from water in the capillaries of the brain is generally considered to be about 4% of the total signal [28, 67]. However, capillary density varies considerably from one region of the brain to another.

Based on the preceding discussion, it should be reasonable to approximate the brain white matter in terms of a two compartment model where one of the compartments corresponds to the intracellular space and the other is the extracellular space. For the purposes of modelling, we will consider the cells to be axons of uniform diameter and orientation. Thus, for diffusion parallel to the axon fibre orientation, the water molecules will diffuse freely while for diffusion perpendicular to the fibre orientation, the water molecules will encounter a series of barriers to diffusion. The barrier corresponds to the cell membrane and/or the myelin sheath. See Section 3.7 for a complete description of the model.

5.1.1 The 1-Compartment Model of Tanner

Before presenting the analysis based on the 2-compartment model discussed above, it is useful to discuss Tanner's 1-compartment model, introduced in Section 2.7. This study was for one dimensional diffusion of water in a system of equidistant semi-permeable parallel planes. The following equation was presented [54] (shown also in Eq. (2.86)).

$$D_a(\infty) = \left[\frac{1}{D(0)} + \frac{1}{aP} \right]^{-1}. \quad (5.1)$$

where, for this model, $D(0)$ is the 'free' diffusion coefficient, unhindered by the barriers. As $T_d \rightarrow 0$, we expect that $D_a \rightarrow D(0)$, since the water molecule does not have time to interact with the barriers. $D_a(\infty)$ is the asymptotic value that is expected to be reached for long T_d 's, a is the barrier separation and P is the permeability of the barriers. Eq. (5.1) does not describe the behaviour of $D_a(T_d)$ for intermediate values of T_d , however, it is quite useful since if three of the four parameters $D(0)$, $D_a(\infty)$, a , and P are known, then the remaining parameter can be calculated using this equation.

In our case, we can use Eq. (5.1) to get an estimate of P . From the data in Fig. 4.4, we can extrapolate to $T_d = 0$ to get $D_a(0) = 1.5 \pm 0.2 \times 10^{-3} \text{ mm}^2/\text{s}$ and as $T_d \rightarrow \infty$ we obtain $D_a(\infty) = 0.60 \pm 0.05 \times 10^{-3} \text{ mm}^2/\text{s}$. From the literature [36, 107] it is expected that a will be about $5 \mu\text{m}$. If we insert these values into Eq. (5.1), we find that $P = 0.020 \pm 0.002 \text{ cm/s}$. The permeabilities of cell membranes range from [7] 0.001 cm/s to [70] 0.020 cm/s . Most cell membrane permeability studies focus on red blood cells, which typically [32] have a permeability of 0.005 cm/s . The value we obtain with this simplistic model is on the high side of accepted

membrane permeabilities, but it is reasonable nonetheless. It is in agreement with Le Bihan's value of $P=0.016$ cm/s, which was obtained with the same equation [64].

5.1.2 The 2-Compartment Model of Szafer

The two compartment model presented by Szafer *et al.* [7], although still rather simplistic, should be a better representation of reality. The model, discussed in more detail in Section 3.7, has two compartments which can be associated with the intra and extra cellular spaces. They have not presented an analytical expression for D_a as a function T_d , however, Szafer *et al.* [7] have given two expressions (see Eqs. (2.87) and (2.88)) to relate $D_a(\infty)$ with a , P , D_I , D_E and f , where D_I is the 'free', unhindered diffusion coefficient for water in the intracellular environment, D_E is the 'free', unhindered diffusion coefficient for water in the extracellular environment, and f is the cellular volume fraction, defined by

$$f = \frac{B_x B_y}{A_x A_y}. \quad (5.2)$$

A_x , A_y , B_x and B_y were defined in Section 3.7 (see also Fig. 3.14). Unfortunately, the use of these equations is limited by the fact that the values of several of these parameters are not well known. However, we will estimate these values so that we can make use of these equations to get an approximate value for P . We will use Szafer's [7] values of $D_E = 2.5 \times 10^{-3}$ mm²/s, and $f = 0.8$ and use $B_x = B_y = a = 5$ μ m as the average axon diameter. From Eq. (5.2) we find that this implies a spacing between cells of 0.5 μ m. As in Section 5.1.1, we will use $D_a(\infty) = 0.60 \pm 0.05 \times 10^{-3}$ mm²/s and $D_a(0) = 1.5 \pm 0.2 \times 10^{-3}$ mm²/s. Our extrapolated value of $D_a(0)$, in this model, corresponds to a volume average of D_I and D_E , given by,

$$D_a(0) = fD_I + (1 - f)D_E \quad (5.3)$$

from which we compute $D_I = 1.3 \pm 0.2 \times 10^{-3} \text{ mm}^2/\text{s}$. Then, the permeabilities from Eqs. (2.87) and (2.88) are found to be $P^{PS} = 0.019 \pm 0.004 \text{ cm/s}$ and $P^{SP} = 0.017 \pm 0.004 \text{ cm/s}$, respectively. It is interesting to note that these values are not significantly different from that obtained using Tanner's equation in Section 5.1.1.

5.1.3 Monte Carlo Models

The equations discussed above are useful for getting estimates of P , however, they do not help us to understand the behaviour of D_a as a function T_d . To do this we resort to computer modelling. We have developed a Monte Carlo simulation program based on the 2-compartment model of Szafer *et al.* [7], described in Section 3.7. Using the same five parameters as in Section 5.1.2 in this simulation, we obtain the curve shown in Fig. 5.1. We have also included the results of a 1-D simulation (described in Sections 2.7 and 3.7) using the same values for $D_a(0)$, a , and P , as were used in Section 5.1.1.

Clearly, there is poor agreement at small T_d values for both the 1-D and 2-D simulations, when compared with the experimental data in Fig. 4.4 (circle symbols). This was not completely unexpected, since accurate values for some of the simulation's input parameters are not available. For example, the literature value [7] for D_E is not very well known, and f may range from 0.7 [33] to 0.95, depending on the location in the brain. Our extrapolated value of $D_a(0)$ may also be incorrect. It is very possible that both D_I and D_E could be as high as $3.0 \times 10^{-3} \text{ mm}^2/\text{s}$, the value for pure water at 37°C .

In an attempt to obtain agreement with our experimental data, and also to

Figure 5.1: Monte Carlo simulation of water diffusion using 1-D (dotted) and 2-D geometries. The 2-D simulation used $D_I = 1.3 \times 10^{-3} \text{ mm}^2/\text{s}$, $D_E = 2.5 \times 10^{-3} \text{ mm}^2/\text{s}$, $a = 5 \mu\text{m}$, $f = 0.8$, and $P = 0.018 \text{ cm/s}$. The 1-D simulation used the same values for a and P , but used $D(0) = 1.55 \times 10^{-3} \text{ mm}^2/\text{s}$ since its geometry does not allow for a D_I , D_E , or f : see text for details.

illustrate how the Monte Carlo curves depend on the input parameters, we have run many simulations using $D_I = D_E = 3.0 \times 10^{-3} \text{mm}^2/\text{s}$ (the diffusion coefficient of free water at 37°C), and altering only one of the parameters a , f , and P . In Fig. 5.2, we vary the permeability, P , while using $a = 14 \mu\text{m}$, and $f = 0.8$. We see that the permeability has a large effect on $D_a(\infty)$, but less so for lower T_d values. Varying only f has a similar effect, as seen in Fig. 5.3 where we hold $P = 0.004 \text{cm/s}$ constant.

Since we are interested in obtaining agreement with the experimental data, it is more instructive to see how the barrier spacing, a , affects the curves while we force $D_a(\infty)$ of the simulation to be around $0.6 \times 10^{-3} \text{mm}^2/\text{s}$, the value of the asymptote for the data in Fig. 4.4 (circle symbols). To accomplish this, we hold $f = 0.8$ constant, and force the asymptote to remain close to $0.6 \times 10^{-3} \text{mm}^2/\text{s}$ by varying P . The results are shown in Fig. 5.4, where a is varied from $5 \mu\text{m}$ to $20 \mu\text{m}$. P ranged from 0.003cm/s to 0.012cm/s . We see from Fig. 5.4 that a has a large impact on the *shape* of the curve at low T_d values, in contrast to the effect of the parameters f and P .

In Fig. 5.4, good agreement with the experimental data is shown. The profile for $a = 14 \mu\text{m}$ best fits the posterior RL data in Fig. 4.4(b), while the curve for $a = 20 \mu\text{m}$ best fits the central AP data in Fig. 4.4(a). This is illustrated more clearly in Fig. 4.11.

Unfortunately, such large values for the barrier spacing are not supported by the diameter of white matter axons reported in the literature for human brain, which is typically $5 \mu\text{m}$. Although it is known that there is a distribution of cell diameters ranging from [36, 115] $1 \mu\text{m}$ to $20 \mu\text{m}$, the literature is unclear on what the distribution is for particular areas of the brain. Hajnal *et al.* [36] state that

Figure 5.2: Monte Carlo simulation of water diffusion for several values of permeability, P . All curves used the parameters $D_I = 3.0 \times 10^{-3} \text{mm}^2/\text{s}$, $D_E = 3.0 \times 10^{-3} \text{mm}^2/\text{s}$, $a = 14 \mu\text{m}$ and $f = 0.8$. The curve for $P=0.004 \text{ cm/s}$ best fits the experimental data in Fig. 4.4 (circle symbols) for the high T_d values.

Figure 5.3: Monte Carlo simulation of water diffusion for several values of cellular volume fraction, f . All curves used the parameters $D_I = 3.0 \times 10^{-3} \text{ mm}^2/\text{s}$, $D_E = 3.0 \times 10^{-3} \text{ mm}^2/\text{s}$, $a = 14 \mu\text{m}$ and $P = 0.004 \text{ cm/s}$. The curve for $f = 0.8$ best fits the experimental data in Fig. 4.4 (circle symbols) for the high T_d values.

Figure 5.4: Monte Carlo simulation of water diffusion for several values of barrier spacing, a . All curves used the parameters $D_I = 3.0 \times 10^{-3} \text{mm}^2/\text{s}$, $D_E = 3.0 \times 10^{-3} \text{mm}^2/\text{s}$, $f = 0.8$ and the values of a are shown in the inset. P varied from 0.003 cm/s for the largest a value, up to $P = 0.012$ cm/s for the lowest a value. The curve for $a = 14 \mu\text{m}$ ($P = 0.004$ cm/s) best fits the posterior RL experimental data in Fig. 4.4(b), while the curve for $a = 20 \mu\text{m}$ ($P = 0.003$ cm/s) best fits the central AP data in Fig. 4.4(a).

fibres ranging from 1 μm to 10 μm make up 94% of the myelinated fibres in each corticospinal tract. It is unclear whether this ‘average’ diameter can be applied to the area of the brain that we are studying. However, in the absence of more specific data, we will assume that it does. As mentioned previously in Section 4.3, it is interesting to note that some other workers [116, 117] who have obtained diffusion data from human white matter using MR, compute an effective barrier spacing of about 20 μm , in agreement with our observations.

5.1.4 Consideration of the White Matter Fibre Orientation

We will address later some possibilities which may explain why our experimental data deviate from the simulated data. Before this, we will discuss another observation that cannot be explained by the two compartment model. The $D_{a\parallel}$ values in Fig. 4.4 do not stay constant as a function of T_d as would be expected based on our model. Instead, they decrease as T_d increases. In fact, the behaviour of $D_{a\parallel}$ is qualitatively very similar to that of $D_{a\perp}$ except that the variation with T_d is less pronounced. This can be explained if the predominant fibre orientation is not the exclusive orientation.

One of the limitations of the model that we have proposed is the assumption that all of the axonal fibres are oriented parallel to each other. Although this is the ideal condition, in reality, there is likely to be a certain fraction of the fibres that are at some non-zero angle to the rest. If this is the case, then we would expect to see a reduction in $D_{a\parallel}$ as T_d increases since some of the water molecules are restricted. Similarly, the $D_{a\perp}$ component will likely be too high because some of the water molecules are not restricted. Thus, our data appear to be consistent

with a distribution of orientations.

A proper analysis of this effect requires a complete knowledge of the orientation of all of the fibres in the region of interest. Since we do not have this information, we will use a more simplistic approach. We will consider that there is a certain fraction, ε , of the fibres that are parallel to the G_D and to each other, while the rest are perpendicular to this direction. In this case we should be able to write, as a first order approximation:

$$\begin{aligned} D_{a\perp}^{\text{measured}} &= \varepsilon D_{a\perp}^{\text{actual}} + (1 - \varepsilon) D_{a\parallel}^{\text{actual}} \\ D_{a\parallel}^{\text{measured}} &= \varepsilon D_{a\parallel}^{\text{actual}} + (1 - \varepsilon) D_{a\perp}^{\text{actual}}. \end{aligned} \quad (5.4)$$

Based on our knowledge of anatomy [28, 107], we know that ε should be fairly large, since we have purposely chosen areas of the brain where there is a predominant fibre orientation (see Section 3.2). If ε were 0.5, then $D_{a\perp}^{\text{measured}}$ would equal $D_{a\parallel}^{\text{measured}}$ for all T_d . This is clearly not the case. We can see from our data that ε must be considerably greater than 0.5. It should be reasonable then, to consider values for ε in the range $0.75 \leq \varepsilon \leq 1.0$.

We can recompute the data in Fig. 4.4, using Eq. (5.4) rearranged to solve for $D_{a\parallel}^{\text{actual}}$ and $D_{a\perp}^{\text{actual}}$. (Again, we repeat for clarity that $D_{a\parallel}^{\text{measured}}$ represents the data with square symbols in Fig. 4.4 and similarly, $D_{a\perp}^{\text{measured}}$ represents the data with circle symbols.) If we use these equations with several values for ε in the range $0.75 \leq \varepsilon \leq 1.0$, we get the family of curves shown in Fig. 5.5. If this analysis is correct, then the true behaviour for $D_{a\perp}$ and $D_{a\parallel}$ should be similar to one of these curves. None of these corrected curves for $D_{a\parallel}$ is strictly flat but the curve for $\varepsilon = 0.75$ is the closest.

Figure 5.5: D_a vs. $T_d^{1/2}$ for water in human white matter fibres, adjusted using Eq. (5.4). In (a) we show data from a central location, and in (b) from a posterior location, as in Fig. 4.4. The lines labelled ϵ_1 , ϵ_2 , and ϵ_3 are the adjusted values of D_a for the fraction of fibre components parallel to the predominant fibre orientation of $\epsilon_1 = 0.75$, $\epsilon_2 = 0.80$ and $\epsilon_3 = 0.90$. The corresponding permeabilities, computed using Eq. (2.87), are $P=0.002\pm0.003$, 0.007 ± 0.004 , and 0.014 ± 0.004 cm/s respectively for (a), and $P=0.004\pm0.003$, 0.008 ± 0.003 , and 0.014 ± 0.003 cm/s respectively for (b).

This proposal seems to offer at least a partial explanation for why $D_{a||}$ is not independent of T_d . However, if we attempt to fit these corrected curves using the Monte Carlo simulation, we still must use a barrier spacing of $a = 20 \mu\text{m}$ and $a = 14 \mu\text{m}$ in order to get a good fit to the central and posterior data, respectively. The simulated data and the corrected $D_{a||}$ data for $\varepsilon = 0.75$, are shown in Fig. 5.6. The error in the data points are obtained by standard propagation of error techniques [110], using Eq. (5.4). The only input parameter in the simulation that has changed appreciably from the simulation in Fig. 4.11 is P , which is now lower, but still close to the ‘typical’ cell membrane permeability [7] of 0.001 cm/s . Although this result may be valid, it is not used in the rest of the thesis in order to avoid making the analyses in the following sections unnecessarily complicated.

5.1.5 Analysis of the Signal Decay in Human White Matter

The previous section suggests that the white matter fibres can be viewed as two types: one type which is oriented parallel to the predominant fibre orientation (the majority, by a percentage of $\varepsilon \times 100$), and the other type which is oriented perpendicular to the predominant fibre orientation. This interpretation leads to an important observation concerning the decay of the NMR signal. If the diffusion of water in the two parts of the volume being studied is different and the water molecules are in one or the other but they cannot move from one to the other in the time it takes to measure D_a , then we would expect the signal decay to be described by an equation of the form

$$S(b) = S_1 e^{-bD_1} + S_2 e^{-bD_2}, \quad (5.5)$$

Figure 5.6: Comparison of D_a vs. $T_d^{1/2}$ for water in human white matter in vivo for the (a) central AP location (as in Fig. 4.4(a)), adjusted using Eq. (5.4), and the Monte Carlo simulation for $a = 20 \mu\text{m}$, $P = 0.0001 \text{ cm/s}$, $D_I = D_E = 3.0 \times 10^{-3} \text{ mm}^2/\text{s}$, and $f = 0.8$. In (b), the posterior RL location is shown (as in Fig. 4.4(b)), adjusted using Eq. (5.4), and the Monte Carlo simulation for $a = 14 \mu\text{m}$, $P = 0.0005 \text{ cm/s}$, $D_I = D_E = 3.0 \times 10^{-3} \text{ mm}^2/\text{s}$, and $f = 0.8$ is shown. Good agreement is obtained between the experimental and simulated curves.

where S_1 is the signal contribution from the spins in one part of the volume and the other part contributes a signal S_2 when there are no diffusion gradients used (i.e. $b=0$). However, we have observed that the signal decay at $T_d = 513$ ms is exponential, not bi-exponential (see Fig. 5.7).

For a system such as this we would expect an exponential decay if the regions are close enough together that the water molecules, on average, spend an equal amount of time in each of these environments. In this case the signal decay should be described by an equation of the form

$$S(b) = S(0)e^{-bD_{av}}, \quad (5.6)$$

where $S(0) = S_1 + S_2$ and D_{av} is the weighted average value of the diffusion coefficients D_1 and D_2 . For the largest T_d values used in our experiments, a free water molecule will travel a mean displacement of about $80 \mu\text{m}$ (using $D_f = 3.0 \times 10^{-3} \text{mm}^2/\text{s}$). In brain tissue, where $D_a \leq 0.5D_f$, the diffusion of a water molecule will actually be considerably less. This means that if there is a mixture of orientations in our region of interest, then this mixture must occur on a very small scale. Otherwise the water molecules would not have a chance to diffuse in the two environments and the decay would not be exponential.

An exponential decay could also be obtained if $D_1 \approx D_2$. However, this would not be consistent with results in the literature [28, 57, 31, 64, 70] or our results shown in Fig. 5.5.

From this analysis we can conclude that our assumption that most of the fibres in the volume of interest are parallel, is a good one. Otherwise, we would have a bi-exponential decay with the same fractions for the two components for all T_d .

It was mentioned above that the signal decay has been observed to be expo-

Figure 5.7: The NMR signal $\ln S(b)$ vs. b . for 33 points, a large range of b -values, and $T_d = 513$ ms. We show volunteer 1 in (a) and volunteer 2 in (b). The attenuation in (a) and (b) appears to follow a single exponential decay. The error in the data points is σ_M , as described in Section 3.6.

nential for $T_d = 513$ ms. However, at $T_d = 50$ ms the decay is observed to be slightly multi-exponential, see Fig. 5.8. In fact, it appears that the point at which the Monte Carlo simulation result (in Fig. 5.1 and for $a = 5 \mu\text{m}$ in Fig. 5.4) begins to deviate from the experimental results is where the multi-exponentiality begins. This is to be expected since the Monte Carlo calculation implicitly assumes that the signal decay will be a single exponential. If we have a multi compartment system, then this would be equivalent to explicitly assuming, in the Monte Carlo simulation, a ‘fast exchange’ of the water among the various environments so that the signal decay should be given by Eq. (5.6). This issue will be discussed again in Section 5.1.7.

Another possible explanation for the elevated $D_{a\perp}$ values and the multi-exponential signal decay for this region of T_d values is that the multi-exponentiality is caused by blood perfusion in the capillaries within the volume of interest. However, the fraction of spins corresponding to the ‘fast diffusion process’, assumed here to be perfusion, is about 6%. This is slightly higher than expected for this region of the brain. The average blood perfusion fraction for the brain is about 4% [28, 67], as mentioned in Section 5.1. Furthermore, if the two compartments are intracellular water and intravascular water, then the experimental observation of an exponential decay at 513 ms means that the blood vessels must be close enough to the axons to allow complete mixing of the two compartments. This implies that the capillaries are densely distributed throughout the tissue. However, this is not consistent with a blood perfusion fraction of only 4%. Therefore, this does not appear to be the correct interpretation.

A third possible explanation for the observations is that there is a broad distribution of axonal diameters. However, judging from the extent of multi-

Figure 5.8: The NMR signal $\ln S(b)$ vs. b , for 33 points, a large range of b -values and $T_d = 50$ ms. We show volunteer 1 in (a) and volunteer 2 in (b). The error in the data points is σ_{M_j} , as described in Section 3.6. The signal decays appear to deviate from a straight line at the lower b -values, indicating a bi-exponential (or multi-exponential) behaviour. The bi-exponential behaviour could be caused by a division of the volume of water into two compartments. See text for details.

exponentiality in Fig. 5.8, this distribution would either have to be very broad or else the D_a values are strongly and preferentially affected by the larger axons. Since 94% of the myelinated axons fall within 1 μm to 10 μm [36], it is unlikely that this is the explanation.

Based on the discussion above, it appears that the most likely explanation for our data is that we are seeing the effects of a two compartment system where the water from the two compartments has not had enough time to visit both compartments for long enough to average out the effects of diffusion in the two compartments until T_d increases to ≈ 100 ms. So far in this chapter we have discussed several possible compartments that may be responsible for the multi-exponentiality in Fig. 5.8 and the single-exponential in Fig. 5.7. We believe that the most likely compartments responsible for this effect in the white matter are the intracellular and extracellular spaces. These compartments are close enough together that we might expect a multi-exponential decay at short diffusion times to evolve into a single exponential decay as the diffusion time is increased.

5.1.6 T_2 Effects on the Apparent Diffusion Coefficient

Signal attenuation from T_2 relaxation does not affect the values of D_a in a single diffusion experiment because the attenuation due to T_2 is constant for each G_D value, and is cancelled out when the slope is computed from the attenuation curve. In a single compartment system the values of D_a will be independent of TE, the measurement parameter that determines how much T_2 relaxation has occurred when the signal is measured. In this case, there is no need to keep TE fixed when D_a is measured as a function of T_d . However, for diffusion in a multicompartment system with slow exchange, (see Section 5.1.5) T_2 relaxation may have an effect

on D_a if, in each compartment, T_2 is significantly different. The overall NMR signal will be preferentially weighted to the compartment with the highest T_2 . If the diffusion in each compartment is also significantly different, then D_a measured for different values of TE would differ. This possibility may be significant for our experimental protocol because we were not able to hold the echo time, TE, constant for all sequences (see Table 3.2). Since T_2 attenuation varies with TE, each sequence may be measuring signal with a different weighting of the intracellular and extracellular compartments. This could affect the value of D_a that we obtain. In 1994, Mackay *et al.* [123] reported a multiexponential T_2 behaviour in human white matter. The decay curve was decomposed into three different T_2 values, which were attributed to i) water in the intracellular and extracellular space (the main peak at $T_2 = 68$ ms, making up 85% of the signal), ii) the myelin membranes (a small peak at $T_2 = 10$ ms, making up 8% of the signal), and iii) the cerebrospinal fluid (a small peak with $T_2 = 200 - 1000$ ms, making up 7% of the signal).

To address this issue, we have measured D_a vs. TE on two volunteers, to see if there is a dependence. We do not observe a TE dependence on D_a (see Fig. 5.9). This is what we expected, since there is no experimental evidence that T_2 in human white matter is different for the intracellular and extracellular environments. We would not expect our measurements to be affected by the two small peaks in Mackay's [123] results mentioned above because the TE in our sequences varied from 59-182 ms (see Table 3.2), well above the peak attributed to myelin water, and we do not expect cerebrospinal fluid to be contaminating our results like Mackay reported. Whittall and Mackay *et al.* later reported a study [124] with similar peaks, but in that work, the cerebrospinal fluid peak was always at $T_2 = 2000$ ms.

a more reasonable T_2 value for cerebrospinal fluid. This later study [124] confirms that in our experiments, we are measuring signal mainly from spins undergoing relaxation from a single T_2 value.

5.1.7 Monte Carlo Simulation of the Signal Decay

It may be that the poor agreement between the Monte Carlo simulation (in Fig. 5.1 and for $a = 5 \mu\text{m}$ in Fig. 5.4) and the experimental data is due to the fact that the experimental signal decay is multi-exponential at low T_d values, and the Monte Carlo assumes a single exponential behaviour (mentioned previously in Section 5.1.5). To address this possibility, we have revised the Monte Carlo model to generate an attenuation similar to that which would be measured for the real system. The new Monte Carlo algorithm computes the phase of the spin for each diffusing step. We will call this algorithm the 'phase Monte Carlo'. The algorithm for the phase Monte Carlo is described in more detail in Appendix B. From the simulated attenuation curve, we then calculate D_a by a best straight line fit to the $\ln[S(b)]$ vs. b graph. The same b -values as in the experiment are used in the fit. The values of D_a determined with the phase Monte Carlo should be much more consistent with the experimental data.

To get reasonable values for the input parameters to the Monte Carlo simulation, we fit our experimental attenuation curve in Fig. 5.8(a) with the bi-exponential function in Eq. (5.5) and we obtain the values $S_1 = 41.5 \pm 0.7$, $S_2 = 2.8 \pm 0.7$, $D_1 = (0.589 \pm 0.022) \times 10^{-3} \text{mm}^2/\text{s}$, and $D_2 = (9.5 \pm 1.3) \times 10^{-3} \text{mm}^2/\text{s}$. D_1 and D_2 correspond to the intracellular and extracellular diffusion coefficients, D_I and D_E , respectively. If we enter these values into the phase Monte Carlo simulation along with $a = 5 \mu\text{m}$, $f = \frac{S_1}{S_1 + S_2} = 0.936$ and $P = 0.05 \text{ cm/s}$, we get very

(a)

(b)

Figure 5.9: D_a vs. TE in human white matter in vivo for $T_d = 110$ ms. For a central location along the column, in (a), we do not see a dependence on TE for either G_D AP or RL. Similarly for a posterior location in (b), there is no TE dependence observed. The errors in the data are obtained using the same method described for Fig. 4.3 in Section 4.1.

good agreement with the measured signal decay. Consequently, we also get the same value of D_a with the simulation as we got experimentally.

All of the parameters used in this simulation are in good agreement with values reported in the literature with the possible exception of $D_E = 9.5 \times 10^{-3} \text{mm}^2/\text{s}$. No reliable values for D_E are available in the literature [7], however, since we are modelling it as a diffusion coefficient we would expect $D_E \leq 3.0 \times 10^{-3} \text{mm}^2/\text{s}$ which is the diffusion coefficient for free water at 37°C . The fact that D_E is this high suggests that there must be some active mechanism contributing to the motion of the spins.

This is likely caused by hydrostatic pressure applied to the brain tissue by the pulsations of the cerebrospinal fluid in the ventricles which are known to swell and relax with the cardiac cycle [125]. As the ventricles swell they apply hydrostatic pressure to the brain parenchyma. The main effect of this pressure will be to ‘squeeze out’ the extracellular fluid from between the axons. Note that the displacement of the extracellular fluid could be in any direction and will not necessarily depend on the direction of the pressure applied by the CSF. Although this cannot be called a diffusion process it will cause attenuation of the MRI signal in a similar way.

The value of $D_E = 9.5 \times 10^{-3} \text{mm}^2/\text{s}$ was obtained from the experimental decay measured for $T_d = 50 \text{ ms}$. This is long enough for the intra and extra cellular compartments to have undergone a significant amount of exchange although not yet enough to yield an exponential signal decay. This implies, however, that $D_E = 9.5 \times 10^{-3} \text{mm}^2/\text{s}$ is lower than the D_E value that we would obtain for very short diffusion times where no exchange is occurring. Unfortunately, it was not possible to measure this ‘short T_d ’ D_E value reliably.

Our success with explaining our results at $T_d = 50$ ms encouraged us to use the same input parameters for different diffusion times. Unfortunately, the values of D_a obtained as a function of T_d did not agree well with experiment. In order to get better agreement with our data for low T_d values we must incorporate the dynamics of the extracellular fluid into our model as flow rather than as a diffusion process. However, to do this rigorously would require significant changes to the framework of the existing code and would probably increase computation time considerably. Consequently, we decided to use the more feasible approach of incorporating extracellular flow into a simplified Monte Carlo diffusion model. This is discussed in Section 5.1.8.

5.1.8 2-D Monte Carlo With a Flow Channel

In this section we will show that by modelling the motion in the extracellular space with a flow mechanism rather than diffusion we can get good agreement with our experimental data. The model has been presented earlier in Section 3.7. The geometry that we will use has $A_x = 10 \mu\text{m}$, $A_y = 6 \mu\text{m}$, $B_x = 5 \mu\text{m}$ and $B_y = 4 \mu\text{m}$. The area of size $A_y \times (A_x - B_x)$ (i.e. the white part in Fig. 5.10) can be associated with the intracellular space. The area of size $A_y \times B_x$ (i.e. the shaded part of Fig. 5.10) can be thought of as extracellular space.

To reduce the computation time for the Monte Carlo simulation we have replaced the membrane of permeability P by a membrane with $P=0$ (shown as bold lines in Fig. 5.10) and a ‘flow channel’ where the water can flow freely into the extracellular space. This means that the membrane permeability, which, in the simulation, amounts to a probability of exiting the compartment, has been replaced with the chance encounter of entering the flow channel. The number of

water molecules leaving the cell per unit time should be the same for these two models. Inside the cell the molecule will diffuse as before. However, once the molecule leaves the cell it will flow with a constant velocity through the extracellular space and into an adjacent cell where its motion is again diffusional. Although we describe the flow channel as the area $B_x \times (A_y - B_y)$, the probability of the molecule entering the flow channel is directly related to the ratio of $(A_y - B_y)$ to the perimeter of the 'cell', or the intracellular space. There is no diffusional motion during the time spent in the flow channel, thus, the parameter B_x is simply an estimation of the distance travelled while the molecule is caught in the 'flow' effect of the simulation.

Using the parameters $D_I \rightarrow D_a(0) = 0.9 \times 10^{-3} \text{mm}^2/\text{s}$ (there are no D_I or D_E parameters in this simulation), a flow velocity in the channel of 0.05 cm/s , and a barrier size of $5 \mu\text{m}$ (see Fig. 5.10), we obtain very good agreement with the experimental $D_{a\perp}$ for the central location. See Fig. 5.11(a) for a comparison of the Monte Carlo simulation and these experimental data.

For a comparison with the experimental $D_{a\perp}$ in the posterior region of our column, in Fig. 4.4(b), we have used the same parameters as above except $A_x = 9 \mu\text{m}$ and $B_x = 5 \mu\text{m}$, for an effective axon diameter of $a = A_x - B_x = 4 \mu\text{m}$. This simulation also gives good agreement and is shown with the data for $D_{a\perp}$ from Fig. 4.4(b) in Fig. 5.11(b).

From the good agreement in Fig. 5.11, we conclude that some sort of facilitated water motion is occurring in the brain white matter. The source of this mechanism is probably the pulsations of the CSF fluid. It is known from the literature [125] that the pressure on the CSF in the ventricles follows the cardiac cycle. These pressure changes are likely transmitted through the ventricular membranes and

Figure 5.10: The geometry for the 2-D Monte Carlo with a flow channel incorporated to model the effect of high extracellular motions. The periodic structure is $A_x \times A_y$, where the diameter of the axon is represented by $A_x - B_x = 5 \mu\text{m}$, and the flow channel is the area $B_x \times (A_y - B_y)$. The values of the parameters used in the simulation are $A_x = 10 \mu\text{m}$, $A_y = 6 \mu\text{m}$, $B_x = 5 \mu\text{m}$ and $B_y = 4 \mu\text{m}$.

(a)

(b)

Figure 5.11: Monte Carlo simulation of human brain white matter using a model which incorporates a flow channel in the geometry. The experimental data are $D_{a\perp}$ from (a) a central location along the COI, from Fig. 4.4(a), and from (b) a posterior location along the COI, from Fig. 4.4(b).

are felt by the surrounding brain tissue. Since the axons are surrounded by several layers of myelin we will consider them to be relatively incompressible. Thus, we believe the predominant effect of the pressure exerted on the brain tissue by the CSF will be to cause displacement of the extracellular fluid.

We have assumed in the simulation that the resultant displacement of the extracellular water, or the ‘flow’ direction, is perpendicular to the white matter fibres. The tortuous environment of the extracellular space probably results in flow for any direction. For flow parallel to the fibres, there should not be as much of a restriction effect. Thus, although we expect that flow in the extracellular environment will affect the magnitude of $D_{a||}$, we should not expect to see $D_{a||}$ change with T_d as a result of flow parallel to the fibres. It may also be true, due to the relatively low restriction in this direction, that the flow of water is fast enough for its NMR signal to be completely attenuated and not be seen when G_D is parallel to the fibres.

Bulk motion effects, CSF and vascular pulsations all contribute to the difficulty in carrying out diffusion measurements of brain tissue [44, 51]. Care must be taken when placing the COI that the ventricles are not included in the selected volume, otherwise severe artifacts and/or CSF signal contamination will result. In this study, our column is close to the ventricles because we wanted to capture the highly RL-oriented fibre emanations from the corpus callosum. Multiple acquisition averaging and at least six points on the decay curve help to average out artifacts related to bulk tissue motion from blood or CSF pulsations. A better way to eliminate bulk motion effects may be to implement navigator spin echoes [2, 126] along with the diffusion echo. Echo planar and other fast imaging sequences show promise as a method to obtain diffusion images free of motion artifacts [51]. These

sequences acquire the entire image data set in a single shot (≈ 100 ms), thus ‘freezing’ the effects of motion between lines in Fourier space. Bulk motion during the diffusing interval (i.e. between G_D ’s), however, will affect these sequences as much as it affects the STEAM, PGSE, and BIPOLAR sequences used in this work.

5.2 Leg Muscle Experiments

In Section 4.5, the diffusion profiles suggest that restricted diffusion is occurring along the long axis of the muscle fibres in the soleus muscle. In order for restricted diffusion to affect the bulk sample, there must be a repetitive tissue structure within the sample. Muscle tissue consists of bundles of fibres, or cells which generally extend the length of the muscle, with a diameter of $10 - 100 \mu\text{m}$. Each fibre contains bundles of very fine myofibrils, a cellular structure which extends the length of the muscle fibre and takes up most of the sarcoplasm. The myofibrils, in turn, are made up of myofilaments, as shown in Fig. 5.12. The myofilaments have a repetitive structure along the length of the cell called a sarcomere. The sarcomere is the active unit of the myofibrils that provides contraction. The contraction of the sarcomere, based on the sliding filament theory [112], occurs when the thick myofilaments exert a force on the thin myofilaments such that they slide past each other, causing a net contraction ($\sim 0.5 \mu\text{m}$) of the muscle cell. We propose that the Z-line of the sarcomere is an obstacle to water diffusion and is responsible for the restriction we observe in human soleus leg muscle.

On this assumption, we can proceed to model the periodic structure of the sarcomeres in a computer simulation. If the water does indeed move principally via diffusion and the Z-lines of the sarcomere are a source of restriction, then the computer simulation of diffusion should agree relatively well with our experimental

B.

Figure 5.12: Taken from Thibodeau *et al.* [107]. **A.** A single muscle cell, or muscle fibre, from which skeletal muscles are composed. Each muscle fibre contains many myofibrils. Also notice the intricate network of the sarcoplasmic reticulum, which forms tubular channels surrounding the myofibrils and extends the length of the fibre. **B.** Fine structure of the myofibril, showing the components of the sarcomere. The Z-line anchors the thin myofilaments from adjacent sarcomeres. This structure may also act as a barrier to water diffusion.

data. One and two dimensional models of diffusion within a periodic lattice have been presented and discussed in Section 3.7. The results of simulations using these models are shown in Fig. 5.13.

With the parameters shown in Fig. 5.13, the results of the 1-D, 2-D, and tortuosity simulations show no correlation with the experimental data. The results of the simulations are not surprising however, since similar results by other groups [7, 54] show that, for a barrier spacing of $3\text{ }\mu\text{m}$, the bulk of the drop in the diffusion coefficient occurs in under 4 ms. The size of the barrier seems to be the only parameter in the model that determines the time at which the asymptotic behaviour is reached. We infer from this poor agreement that the 1-D, 2-D, and tortuosity models are not sufficiently realistic models of water mobility in leg muscle tissue.

These models perhaps can accurately represent the effect of the Z-lines on a diffusing molecule, but considering the complexity of cell tissues, it is doubtful that the effect of the Z-line is the main one governing the behaviour of water motion in live tissue.

Cell processes such as active and passive transport of molecules like Ca^{++} must also affect the net water motion. The high energy requirements of muscle cells demand that their ability to distribute adenosine triphosphate (ATP) throughout the sarcomeres be quick and efficient. The mitochondria supply the ATP to the myofibrils and probably can move themselves to sites of utilization by transport mechanisms involving microtubules [107]. In brain white matter, the axon lengths are large enough that diffusion alone cannot supply important molecules to the entire cell fast enough. A mechanism of intracellular axonal transport involving microtubules [34] and other filaments of the cytoskeleton is in place to accom-

Figure 5.13: 1-D and 2-D Monte Carlo simulation of water diffusion. The experimental data from Fig. 4.23(b) are shown for comparison as solid black circles. The values of the parameters A_x , A_y , B_x and B_y (in μm) used in the simulation (see Section 3.7) are shown in the figure. None of the simulated curves show a decline as seen in the experimental data. This indicates that the 1-D, 2-D and tortuosity models are an insufficient representation of water motion within tissue in vivo.

plish this. The muscle cells, although abundant with mitochondria, also need to have a system of facilitated transport to distribute the ATP and other important molecules.

A system of active transport crucial for contraction in muscle cells is the pumping of Ca^{++} by the sarcoplasmic reticulum. The sarcoplasmic reticulum is an interconnected system of tubular canals and sacs. In muscle cells, it surrounds each myofibril as shown in Fig. 5.12A. When the motor neuron fires, its action potential stimulates the sarcoplasmic reticulum to release its store of Ca^{++} , which is a trigger for ATP uptake by the myofilaments which leads to contraction of the sarcomere. After the stimulation, the sarcoplasmic reticulum pumps the Ca^{++} ions back into its sacs and channels. The subsequent decrease in Ca^{++} concentration in the myofibril ceases contraction. In the relaxed state of the muscle, then, the sarcoplasmic reticulum is continually pumping Ca^{++} ions up a concentration gradient. The entire process of Ca^{++} release and reuptake is complete in about 15 ms [127]. Water molecules may follow this 'flow' of Ca^{++} ions into and out of the reticulum. Once in the reticulum, the water can diffuse or may be preferentially directed along its longitudinally oriented membrane channels.

We propose an addition to the 2-D computer model described above so as to model the effect of directed motion. We believe the sarcoplasmic reticulum is the predominant channel for directed longitudinal transport of water. In our model we simulate the motion of the water molecules inside the myofibril using a diffusion process. The flow is introduced using a 'flow channel' which acts in the same way as did the flow channel introduced in Section 5.1.8. If a diffusing molecule enters the 'flow' area, it will be directed along $\hat{x}$ with a constant velocity until it exits the channel. The model is shown in Fig. 3.16. The flow channel is defined by the area

$B_x \times (A_y - B_y)$. The direction of flow in the channel is determined by the point of entry: entry from the left side of the channel results in flow along $+\hat{x}$, while $-\hat{x}$ flow results from a right side entry.

Fig. 5.14 shows the results of the simulation. We used 60000 iterations, a diffusion step time of $1 \mu\text{s}$, a free diffusion coefficient $D(0) = 1.24 \times 10^{-3} \text{mm}^2/\text{s}$ and a constant velocity in the flow channel of 0.03 cm/s . Note the increase in the apparent diffusion coefficient over the free diffusion coefficient, due to the presence of the ‘flow’ channel. We have modelled the Z-lines as being impermeable to water.

The agreement of the simulation with experiment indicates that some mechanism for directed transport, or flow, exists within the muscle tissue. The fact that the 1-D, 2-D, and tortuosity models show virtually no time dependence is a strong indication that these models are lacking a crucial element which is important to the understanding of water mobility in muscle: directed transport.

The flow mechanism modelled in our simulation, however, cannot simulate blood perfusion or coherent capillary flow. The motion of blood is continuous throughout the experiment, whereas in our model, the flow is simply a jump from one sarcomere to the next. Also, blood perfusion usually results in a pseudo-diffusion coefficient, D^* , 8 to 10 times larger [52] than that of free water. If a significant majority of the NMR signal is from blood, then we would expect to see bi-exponential behaviour [52] in the attenuation curves. We do not observe bi-exponential behaviour, and an example attenuation plot for a volunteer is shown in Fig. 4.20(b). The absence of the bi-exponential behaviour indicates that the signal contribution from blood is negligible, or that D^* of blood perfusion in the leg is comparable to the diffusion coefficient of water in the muscle fibres. Neither should we expect our hypothesized ‘flow’ mechanism to result in a bi-exponential decay.

Figure 5.14: Monte Carlo simulation of the leg muscle using a model which incorporates a 'flow' channel. The agreement of the simulation with experimental data suggests that some sort of 'flow', or directed transport of water is occurring in the muscle cells. The parameters used from Fig. 3.16 are $A_x = 3 \mu\text{m}$, $A_y = 9 \mu\text{m}$, $B_x = 2 \mu\text{m}$ and $B_y = 5 \mu\text{m}$.

This is because 95% of the particles in the simulation reach the flow channel at least once for a 4 ms simulation. For a 14 ms and above simulation, all the particles have reached a flow channel at least once. This means there is a fast exchange situation (see Section 5.1.5) for all the T_d 's simulated.

It could be argued that our model does not accurately represent the situation in muscle tissue since the size and geometry of the flow channels in the model are very different from the size and shape of the sarcoplasmic reticulum. Furthermore, the Z-lines have been assumed to be impermeable in this model whereas in reality water likely passes through the Z-line relatively easily. These are valid objections, however, our objective in modelling the leg data was not to develop a model that describes all aspects of diffusion in muscle tissue but rather to understand the main effects. This we have done. We believe that our results demonstrate that directed transport of water in the sarcoplasmic reticulum is the dominant process affecting our measurements but it is not likely the only significant contribution.

Chapter 6

Conclusions

6.1 Human White Matter

We have obtained D_a values in human white matter over a large range of T_d (14-1000 ms). We believe our work is the first to show, unambiguously [31, 58, 64], the asymptotic behaviour of D_a for large values of T_d in white matter with diffusion parallel and perpendicular to the white matter fibres. Our data also show behaviour consistent with restricted diffusion models where diffusion across the fibre is restricted but parallel to the fibre the diffusion is unhindered. This feature, to our knowledge, has not been previously experimentally demonstrated in humans in vivo.

The anisotropy in human white matter has been well established in other works [28, 34, 64], and our data confirm this behaviour at the longer T_d 's. Our data demonstrate, for the first time, that the anisotropy is removed at lower values of T_d . This may be useful for DWI. Diffusion weighted images free from signal intensity variations from anisotropy have been shown to be more effective in visualizing tumours [29] and the region of brain tissue affected by stroke [46, 73, 128]. The method currently used to remove anisotropy is diffusion tensor imaging [29, 30, 59].

which is costly in acquisition time [29, 74]. By developing short T_d diffusion weighted imaging sequences, the anisotropy could be removed without the long acquisition times.

Spanning such a large range of T_d with reasonable accuracy was accomplished by using three different MR sequences optimized for a specific window of T_d : AGPS for the lower diffusion times, PGSE for intermediate values and STEAM for the long diffusion times. As far as we know, no other groups have attempted this approach. Usually, only one sequence is used for restricted diffusion experiments [31, 64] because this minimizes the number of experimental variables. However, it also restricts the practical range of T_d that can be probed. One of our goals was to characterize the time dependence of D_a in human white matter. This could not have been achieved with just a single sequence. Our data represent the largest range of T_d investigated. We also report D_a for the shortest T_d measured [31, 37, 57, 64] (to the best of our knowledge) in human white matter in vivo, yet the sequence for this T_d has a comparatively large b_{max} of 122 s/mm². Other works [31, 58, 129] have published diffusion coefficients (at higher T_d) with a b_{max} close to or less than this.

Our initial attempt to explain our D_a vs. T_d data using Monte Carlo simulations (see Fig. 4.11) resulted in very good agreement between the simulation results and the experimental data. Furthermore, the parameter values used in the simulations were, for the most part, quite realistic. However, the axonal diameter that we were forced to use may have been too large.

Axon diameters in human white matter range from 1 μm to 20 μm , but most are generally considered to be about 5 μm [36, 64, 107, 115, 116, 117]. If the results of our simulations are to be believed, then the axons in the region studied

are 14 μm to 20 μm in diameter. We extensively searched the literature for axonal diameters in the region of the brain we investigated, however, this information is sparse and we were unable to find more specific data on the distribution of axonal diameters. However, based upon the information that we were able to obtain [36, 64, 115, 116, 117], we concluded that, while we cannot rule out the possibility of having such large axon diameters, this is more likely an over estimation.

This over estimation suggests that the Monte Carlo model is not comprehensive enough to realistically reflect the transport properties of water in brain cells. A model incorporating a 'flow channel' similar to the model used successfully for leg muscle measurements has provided better agreement (see Fig. 5.11). This demonstrates that a non-diffusion mechanism is affecting some of the water in the white matter. We believe a likely source for this mechanism is the CSF pulsations applying pressure to the white matter, and in turn, forcing the extracellular water to flow haphazardly in a tortuous manner. The model with the flow channel is still a simplistic representation of the dynamics of water transport in the brain. Nevertheless, we believe it accounts for a crucial element contributing to the observed behaviour of the NMR signal.

Some interesting future work would be to combine the flow channel with the same intra/extra cellular environment of the original 2-D Monte Carlo model. The flow channel would probably be better modelled as periodic 'bursts' of flow in random directions in the extracellular space, to better model the periodic CSF pulsations. This model could also be combined with the phase Monte Carlo method described in Section 5.1.7. Such an algorithm, however, is non-trivial, and would undoubtedly require enormous computation time.

If the model described above were efficiently implemented, the parameters f , P ,

and D_E would re-enter the model. With P obtained from the simulation, generating ‘permeability weighted’ images may be feasible. The computation time required will likely be too long to make the Monte Carlo fitted ‘permeability image’ clinically viable in the near future. However, if the permeability values obtained from the Monte Carlo fit are reasonably close to the values of P obtained from Eq. (2.87), then a quick, ‘real time’ calculation of P using Eq. (2.87) could be used to generate a ‘permeability’ image. Our method accurately determines $D_a(\infty)$, and so would give credible upper and lower limits for the true value of P , as we have demonstrated in Sections 5.1.2 and 5.1.4, where we obtain the lower and upper limits of $P=0.002$ cm/s and $P=0.019$ cm/s. It is likely that the ‘flow’ effect will alter the value of $D_a(\infty)$ by a small amount from what it would otherwise be, and therefore, the value of P obtained from Eq. (2.87) will be slightly elevated. However, this should not diminish the usefulness of ‘permeability weighted’ images since, for clinical applications, it is more important to have reliable spatial *variations* of P than to have precise values of P at a given location. Such information could be useful for diagnosing and monitoring demyelinating diseases such as MS [129]. A permeability map may be able to distinguish demyelination from edema or gliosis, something current T_1 or T_2 images cannot do [12]. It may also be possible to detect regions in the early stages of demyelination, making early diagnosis of MS and early intervention a possibility.

6.2 Human Leg Muscle

We have demonstrated a practical method of obtaining diffusion measurements for very short T_d ’s on a clinical MR imager. We have implemented a double Maxwell gradient coil in a non-standard way which maximizes gradient amplitudes and in

turn, allows very short T_d diffusion sequences to be used. We have developed a unique circuit which isolates our gradient coil from rf pick-up, even though it is placed inside the rf coil. The circuit allows us to use an exterior rf coil, which means that the gradient coil's amplitude can be made larger than one where an rf coil insertion must be accounted for (see Section 3.4).

Diffusion coefficients of water in human soleus leg muscle *in vivo* have been obtained for several diffusion times. To the best of our knowledge, it is the first study of the time dependence of the diffusion coefficient in human leg muscle [67, 130]. Included in our data are coefficients for the shortest T_d values measured. The short T_d measurements have given us a view of water diffusion on the scale of a few microns. These data may be important as reference values for future investigations of peripheral vascular disease and the effects of hydration due to diabetes [65]. The Z-line of the sarcomere likely has an effect on the diffusion of water in muscle cells, however, it does not appear to be the dominant mechanism. In order to explain our experimental results it was necessary to also include a flow channel.

Simulations using other models fail to reproduce the structure in our experimental data. This indicates that some mechanism for directed transport of water within muscle cells could exist and we speculate that the sarcoplasmic reticulum could be instrumental in this process. It also clearly demonstrates that simplistic 1-D and 2-D diffusion models are not sufficient to explain the complexity of water mobility in human muscle tissue. They cannot represent the effect of active transport mechanisms within the cell, or motion caused by cellular organelles and the active pumping of Ca^{++} ions by the sarcoplasmic reticulum. Our model does not incorporate all of the complexities of water motion, however we believe it does

include a prominent mechanism.

Chapter 7

Future Directions

7.1 Clinical Studies for Multiple Sclerosis

In this thesis, we have established that the asymptote of the D_a vs. $T_d^{1/2}$ profile is reached at about 500 ms. Therefore, any future clinical protocols (and computer simulations) need not investigate much beyond this T_d value.

One such clinical study which should be feasible, based on this thesis, is to obtain ‘permeability weighted’ images of the brains of patients with multiple sclerosis. In order to obtain ‘weighting’ of the permeability in such an image, our flow Monte Carlo model in Section 5.1.7 must be modified to re-include the effect of a permeable barrier with permeability P , as well as the extracellular environment of the original 2-D model. As mentioned in Chapter 6, the ‘flow’ effect should be replaced with a model that gives periodic (close to the period of CSF pulsations, ~ 1 s) ‘bursts’ of flow in random directions in the extracellular space. This model should also be combined with the phase Monte Carlo method described in Section 5.1.7 so that ‘slow exchange’ effects are taken into account.

This clinical study need only obtain a few T_d values of D_a in order to ‘fit’ it to the above simulation: one at a large T_d to obtain $D_a(\infty)$, and a few at the

lowest T_d 's. The pixel intensity in the image would be weighted to P , the output parameter from the simulation.

The accuracy of the permeability, P , would depend of course, on the accuracy of the simulation's input parameters. It remains unclear what the correct values of f , D_E , and D_I are for healthy human brain, let alone what they would be in MS plaques. It would be more plausible to consider P as 'proportional to' the true permeability, rather than as a quantitative measurement. With this caveat, it is likely that such permeability weighted images should give good contrast between healthy and diseased white matter. This information could be useful for diagnosing and monitoring MS.

7.2 Linescan Imaging and Anisotropy Removal

Larsson *et al.* [35] have studied D_a in the human brain of MS patients. In that study, involuntary head movements of the patients (due to the disease) caused image artifacts severe enough to obliterate reasonable measurements of D_a . Thus, we recommend using a fast linescan imaging technique, similar to that by Poorter *et al.* [41], which avoids image artifacts from motion which most 2D-FFT imaging techniques are very susceptible to.

Not only could linescan imaging be used for the permeability weighted project mentioned above, it could also be used to acquire diffusion images at low T_d . This thesis has demonstrated that the effects of diffusion anisotropy are removed at low T_d (see Fig. 4.4 and Section 4.1). As mentioned in Section 6.1, diffusion images free from anisotropy effects have been shown [29] to be more effective in visualizing tumours. Research is underway in other laboratories [29, 59] to remove the anisotropy effects from white matter by using diffusion tensor imaging.

Diffusion tensor imaging, however, is extremely costly in acquisition time, a critical factor for clinical studies. Our proposed method is fast and should be effective at removing the anisotropy.

A lot of excitement in stroke research [6, 7, 73, 128] has emerged from observations of a large drop [75] in D_a in areas of the brain affected by the acute infarct [77]. This effect may occur within minutes of the insult [75], which is not seen on T_2 weighted images [73]. Once again, the effect of diffusion anisotropy may obscure the delineation between ischemic and normal brain, which would lead to errors in defining the size and location of the lesion [77]. Our proposed method should be faster and as effective in removing the anisotropy as diffusion tensor imaging.

In this thesis, we have demonstrated that D_a values as low as $T_d = 4$ ms can be obtained with a custom made gradient coil using modest electrical power. The next logical step would be to increase the size of our gradient coil to accommodate a human head and also include gradients along $\hat{x}$ and $\hat{y}$. With this, clinical diffusion images free of anisotropy effects could quickly be obtained.

Appendix A

Calculation of Field Derivatives

In Chapter 2, it was mentioned that it is convenient to compute $\frac{dB_z}{dz}$ of Eq. (2.99) for gradient coil optimization. Here, we present the derivation of $\frac{dB_z}{dz}$.

Showing Eqs. (2.99) to (2.101) from Chapter 2, we have

$$B_z(y, z) = \frac{\mu_o I k}{4\pi (ay)^{1/2}} \left[J_1 + \frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} J_2 \right]. \quad (\text{A.1})$$

where

$$k = \left(\frac{4ay}{(a + y)^2 + z^2} \right)^{1/2}. \quad (\text{A.2})$$

and J_1 and J_2 are the elliptical integrals of Legendre: [102]

$$\begin{aligned} J_1 &= \int_0^{\pi/2} \frac{d\Upsilon}{(1 - k^2 \sin^2 \Upsilon)^{1/2}} \\ J_2 &= \int_0^{\pi/2} (1 - k^2 \sin^2 \Upsilon)^{1/2} d\Upsilon. \end{aligned} \quad (\text{A.3})$$

Taking the derivative of Eq. (A.1), we get

$$\begin{aligned} \frac{dB_z}{dz} &= \frac{\mu_o I}{4\pi (ay)^{1/2}} \left[\frac{dk}{dz} \left[J_1 + \frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} J_2 \right] + \right. \\ &\quad \left. k \left[\frac{dJ_1}{dz} + \frac{d}{dz} \left[\left(\frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} \right) J_2 \right] \right] \right]. \end{aligned} \quad (\text{A.4})$$

Evaluating some of the terms in Eq. (A.4), we have

$$\frac{dk}{dz} = \frac{-zk^3}{4ay}, \quad (\text{A.5})$$

and

$$\frac{dJ_1}{dz} = \frac{-zk^4}{4ay} \int_0^{\pi/2} \frac{\sin^2 \Upsilon d\Upsilon}{(1 - k^2 \sin^2 \Upsilon)^{3/2}}. \quad (\text{A.6})$$

and

$$\begin{aligned} \frac{d}{dz} \left(\frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} \right) J_2 &= \left(\frac{-2z((a - y)^2 + z^2) - 2z(a^2 - y^2 - z^2)}{((a - y)^2 + z^2)^2} \right) J_2 + \\ &\quad \frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} \frac{dJ_2}{dz} \\ &= \left(\frac{-4za^2 + 4zay}{((a - y)^2 + z^2)^2} \right) J_2 - \frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} \frac{zk^2}{4ay} (J_2 - J_1). \end{aligned} \quad (\text{A.7})$$

We then define J_3 as

$$J_3 \equiv \int_0^{\pi/2} \frac{\sin^2 \Upsilon d\Upsilon}{(1 - k^2 \sin^2 \Upsilon)^{3/2}}. \quad (\text{A.8})$$

and substitute it into Eq. (A.6). Eq. (A.4) finally becomes, after substituting in Eqs. (A.5) to (A.8),

$$\begin{aligned} \frac{dB_z}{dz} &= \frac{\mu_o I}{4\pi(ay)^{1/2}} \left[\frac{-zk^3}{4ay} \left[J_1 + \frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} J_2 \right] + k \left(\frac{-zk^4}{4ay} J_3 - \right. \right. \\ &\quad \left. \left. \frac{4az(a - y)}{((a - y)^2 + z^2)^2} J_2 - \frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} \frac{zk^2}{4ay} (J_2 - J_1) \right) \right] \\ &= \frac{\mu_o I}{4\pi(ay)^{1/2}} \left[J_1 \left(\frac{-zk^3}{4ay} + \frac{zk^3}{4ay} \frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} \right) + J_2 \left(\frac{-zk^3}{2ay} \frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} - \right. \right. \\ &\quad \left. \left. \frac{4azk(a - y)}{((a - y)^2 + z^2)^2} \right) - \frac{zk^5}{4ay} J_3 \right] \\ &= \frac{\mu_o I}{4\pi(ay)^{1/2}} \left[\frac{zk^3}{4ay} J_1 \left(\frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} - 1 \right) - J_2 \left(\frac{zk^3}{2ay} \frac{a^2 - y^2 - z^2}{(a - y)^2 + z^2} + \right. \right. \\ &\quad \left. \left. \frac{4azk(a - y)}{((a - y)^2 + z^2)^2} \right) - \frac{zk^5}{4ay} J_3 \right]. \end{aligned} \quad (\text{A.9})$$

Eq. (A.9) can be used to compute the field gradient. J_1 , J_2 and J_3 are easily evaluated by standard computer integration algorithms [111].

Appendix B

Details of the ‘phase Monte Carlo’ Algorithm

In Section 5.1.7, we introduced a new Monte Carlo model, the ‘phase Monte Carlo’, which computes the phase of the spin for each diffusing step. This is different from the previous models, described in Section 3.7, which do not use the phase to compute D_a , but rather, D_a is computed from the spin’s net displacement, $\langle x \rangle$, using Eq. (3.10).

In the phase Monte Carlo, the phase change of the spin is computed for each diffusion step, using a formula similar to Eq. (2.56).

$$\phi = \gamma \int_0^{\Delta+\delta} Gz \, dt. \quad (\text{B.1})$$

Of course, in the computer calculation, Eq. (B.1) is a sum over all diffusing steps j , written as

$$\phi_k = \gamma \sum_{j=1}^{\text{int}(\frac{\Delta+\delta}{T_{step}})} G_j z_j T_{step}, \quad (\text{B.2})$$

where ϕ_k is the net phase change for spin k (corresponding to Eq. (2.58)), G_j is the gradient amplitude during spin k ’s j^{th} step, z_j is the position of spin k after

the j^{th} step, and T_{step} , the ‘step size’, is the duration of a diffusing step, as in Section 4.3 and Fig. 4.9.

Clearly (see Fig. 2.5), G_j is a function of time during the simulation, and its time dependent ‘profile’, for each b value, is an input parameter to the simulation.

The resulting signal attenuation, $S(b)$, for a particular b value is then computed using (corresponding to Eq. (2.59)).

$$S(b) = \sum_{k=1}^N \cos(\phi_k), \quad (\text{B.3})$$

where N is the number of iterations, or number of spins.

At the end of the simulation, we have the signal attenuation for the same b values that were used in the experiment. The simulated attenuation can be plotted and analyzed just like the experimental data. The simulated D_a values are obtained by the same method described for the experimental data in Section 3.6.

Bibliography

- [1] P.A. Bandettini, E.C. Wong, R.S. Hinks, R.S. Tikofsky, J.S. Hyde. Time course EPI of human brain function during task activation. *Magn. Reson. Med.* **25**, 390–397 (1992).
- [2] R.J. Ordidge, J.A. Helpert, Z.X. Qing, R.A. Knight, V. Nagesh. Correction of motional artifacts in diffusion-weighted MR images using navigator echoes. *Magn. Reson. Imag.* **12**, 455–460 (1994).
- [3] D. Le Bihan, J. Delannoy, R.L. Levin. Temperature Mapping with MR Imaging of Molecular Diffusion: Application to Hyperthermia. *Radiology* **171**, 853–857 (1989).
- [4] G.E. Santyr, E.J. Fairbanks, F. Kelcz, J.A. Sorenson. Off-Resonance Spin Locking for MR Imaging. *Magn. Reson. Med.* **32**, 43–51 (1994).
- [5] G.O. Cron, G. Santyr, F. Kelcz. Correction of Errors due to Slice Profile Imperfections in Quantitative Dynamic Contrast-Enhanced Breast MRI. *Proc., SMRM, 5th Meeting, Vancouver, Canada*, 1050 (1997).
- [6] M.E. Moseley, Y. Cohen, J. Mintorovitch, L. Chileuitt, H. Shimizu, J. Kucharczyk, M.F. Wendland, P.R. Weinstein. Early Detection of Regional

- Cerebral Ischemia in Cats: Comparison of Diffusion and T2 Weighted MRI and Spectroscopy. *Magn. Reson. Med.* **14**, 330-346 (1990).
- [7] A. Szafer, J. Zhong, J.C. Gore. Theoretical Model for Water Diffusion in Tissues. *Magn. Reson. Med.* **33**, 697-712 (1995).
- [8] K.L. Malisza, U.I. Tuor, P. Kozlowski, J.K. Saunders. Prevention of both T2 and Diffusion Weighted Increases in Image Intensity during Cerebral Hypoxia-Ischemia Correlates with Lack of Damage in Neonates. *Proc., SMRM, 5th Meeting*, Vancouver, Canada, 594 (1997).
- [9] Rabi II. S. Millman, P. Kusch, J.R. Zacharias. The Molecular Beam Resonance Method for Measuring Nuclear Magnetic Moments. *Phys. Rev.* **55**, 526-535 (1939).
- [10] F. Bloch. Nuclear Induction. *Phys. Rev.* **70**, 460-474 (1946).
- [11] E.M. Purcell, H.C. Torrey, R.V. Pound. Resonance Absorption by Nuclear Magnetic Moments in a Solid. *Phys. Rev.* **69**, 37-38 (1946).
- [12] R.R. Edelman, J.R. Hesselink. *Clinical Magnetic Resonance Imaging*. W.B. Saunders Company, Philadelphia, 1990, pp. 3-16.
- [13] E.L. Hahn. Spin Echoes. *Phys. Rev.* **80**, 580-594 (1950).
- [14] H.Y. Carr, E.M. Purcell. Effects of Diffusion on Free Precession in NMR Experiments. *Phys. Rev.* **94**, 630-638 (1954).
- [15] D.W. McCall, D.C. Douglas, E.W. Anderson. Self-Diffusion in Liquids: Paraffin Hydrocarbons. *Phys. Fluids.* **2**, 87-91 (1959).

- [16] E.O. Stejskal and J.E. Tanner. Spin Diffusion Measurements: Spin Echoes in the Presence of a Time-Dependent Field Gradient. *J. Chem. Phys.* **42**, 288-292 (1965).
- [17] P. Mansfield, P.G. Morris. NMR Imaging in Biomedicine. In: J.S. Waugh, ed. *Advances in Magnetic Resonance Imaging*. New York: Academic Press: 1982:[Suppl 2].
- [18] D. Le Bihan, E. Breton, A. Syrota. In-vivo self-diffusion magnetic resonance imaging. *Proc.,SMRM, 4th Meeting*, Berkely, California, 1238-39 (1985).
- [19] D.G. Taylor, M.C. Bushell. The spatial mapping of translational diffusion coefficients by the NMR imaging technique. *Phys. Med. Biol.* **30**, 345-349 (1985).
- [20] K. Merboldt, W. Hanicke, J. Frahm. Self-Diffusion NMR Imaging Using Stimulated Echoes. *J. Magn. Reson.* **64**, 479-486 (1985).
- [21] A.M. Cormack. Representation of a Function by Its Line Integrals, with some Radiological Applications. *J. Appl. Phys.* **34**, 2722-2727 (1963).
- [22] P.C. Lauterbur. Image Formation by Induced Local Interactions: Examples Employing Nuclear Magnetic Resonance. *Nature*, **242**, 190-191 (1973).
- [23] P. Mansfield, P.K. Grannell. NMR 'diffraction' in solids? *J. Phys. C.*, **6**, L422 (1973).
- [24] J.F. Shen, J.K. Saunders. Ultra-Fast Imaging with Orthogonal Phase Encoding. *Proc., SMR, 2nd Meeting*, San Franscisco, 470 (1994).

- [25] R. Turner, D. Le Bihan, J. Maier, R. Vavrek, L.K. Hedges, J. Pekar. Echo-Planar Imaging of Intravoxel Incoherent Motion. *Radiology* **177**, 407–414 (1990).
- [26] G.E. Wesbey, M.E. Moseley, R.L. Ehman. Translational molecular self-diffusion in magnetic resonance imaging: effects and applications. In: T.L. James, A.R. Margulis, eds. *Biomedical magnetic resonance*. San Francisco: Radiology Research and Education Foundation, 63–78 (1984).
- [27] C. Thomsen, O. Henriksen, P. Ring. In vivo measurement of water self diffusion in the human brain by magnetic resonance imaging. *Acta Radiol.* **28**, 353–361 (1987).
- [28] T.L. Chenevert, J.A. Brunberg, J.G. Pipe. Anisotropic Diffusion in Human White Matter: Demonstration with MR Techniques in Vivo. *Radiology* **177**, 401–405 (5)(1990).
- [29] P.J. Basser, J. Mattiello, D. Le Bihan. MR Diffusion Tensor Spectroscopy and Imaging. *Biophys. J.* **66**, 259–267 (1994).
- [30] C. Pierpaoli, P. Jezzard, P.J. Basser. High-resolution diffusion tensor imaging of the human brain. *Proc., SMR, 3rd Meeting*, Nice, France, 899 (1995).
- [31] M.A. Horsfield, G.J. Barker, W.I. McDonald. Self Diffusion in CNS Tissue by Volume Selective Proton NMR. *Magn. Reson. Med.* **31**, 637–644 (1994).
- [32] L.L. Latour, K. Svoboda, P. Mitra, C.H. Sotak. Time-Dependent Diffusion of Water in a Biological Model System. *Proc. Natl. Acad. Sci. USA* **91**, 1229–1233 (1994).

- [33] G.J. Stanisz, A. Szafer, G.A. Wright, R.M. Henkelman. An Analytical Approximation to a Monte Carlo Model of Diffusion in Tissues. Proc., SMR, 3rd Meeting, Nice, France, 917 (1995).
- [34] C. Beaulieu, P.S. Allen. Determinants of Anisotropic Water Diffusion in Nerves. *Magn. Reson. Med.* **31**, 394-400 (1994).
- [35] H.B.W. Larsson, C. Thomsen, J. Frederiksen, M. Stubgaard, O. Henriksen. In vivo Magnetic Resonance Diffusion Measurements in the Brain of Patients with Multiple Sclerosis. *Magn. Reson. Imag.* **10**, 7-12 (1992).
- [36] J.V. Hajnal, M. Doran, A.S. Hall, A.G. Collins, A. Oatridge, J.M. Pennock, I.R. Young, G.M. Bydder. MR Imaging of Anisotropically Restricted Diffusion of Water in the Nervous System: Technical, Anatomic, and Pathologic Considerations. *J. Comput. Assist. Tomogr.* **15**, 1-18 (1991).
- [37] D. Le Bihan. Anisotropic diffusion in brain white matter revisited: Restriction, Permeability and Tortuosity. Proc., ISMRM, 4th Meeting, New York, New York, 1324 (1996).
- [38] G. Liu, P. van Gelderen, C.T.W. Moonen. Single-Shot Diffusion MRI on a Conventional Clinical Instrument. Proc., SMR, 2nd Meeting, San Francisco, 1034 (1994).
- [39] A.W. Anderson, J.C. Gore. Using Navigator Echoes to Correct for Rigid Body Motion in Diffusion Weighted Imaging. Proc., SMR, 2nd Meeting, San Francisco, 354 (1994).

- [40] C. MacGillivray. Diffusion-Weighted MR Imaging of Moving Structures Using A Three Echo Navigator Imaging Technique. M.Sc. Thesis. Carleton University. Physics Dept. (1996).
- [41] J. De Poorter, Y. De Deene, R. Van de Walle, C. De Wagter, C. Thomsen, S. Brockstedt, F. Stahlberg, E. Achten. An Improved Line-Scanning Technique for In Vivo Diffusion Imaging without Motion Artifacts. Proc., SMR, 3rd Meeting, Nice, France, 907 (1995).
- [42] H. Gudbjartsson, S.E. Maier, R.V. Mulkern, I.A. Morocz, F.A. Jolesz. Diffusion Weighted Linescan Imaging. Proc., SMR, 3rd Meeting, Nice, France, 908 (1995).
- [43] K. Harada, N. Fujita, K. Sakurai, Y. Akai, S. Kim, K. Nakanishi, T. Kozuka. Diffusion Imaging of the Brain Using a Sequence Employing Gradient Sensitization in Three Orthogonal Directions Proc., SMRM, 9th Meeting, New York, New York USA, 375 (1990).
- [44] D. Le Bihan, E. Breton, D. Lallemand, P. Grenier, E. Cabanis, M. Laval-Jeantet. MR Imaging of Intravoxel Incoherent Motions: Application to Diffusion and Perfusion in Neurological Disorders. Radiology **161**, 401–407 (1986).
- [45] R.B. Buxton. The Diffusion Sensitivity of Fast Steady-State Free Precession Imaging. Magn. Reson. Med. **29**, 235–243 (1993).
- [46] G. Liu, P. van Gelderen, J. Duyn, C.T.W. Moonen. Single-Shot Diffusion MRI of Human Brain on a Conventional Clinical Instrument. Magn. Reson. Med., **35**, 671–677 (1996).

- [47] Y. Zur, E. Bosak, N. Kaplan. A New Diffusion SSFP Imaging Technique. Proc., SMR, 3rd Meeting, Nice, France, 906 (1995).
- [48] H. Gudbjartsson, S.E. Maier, S. Patz, F.A. Jolesz. Diffusion Imaging on Low-Field Systems using the LSDI Technique. Proc., ISMRM, 4th Meeting, New York, New York, 185 (1996).
- [49] R.C. Mckinstry, E. Akbudak, A.Z. Snyder, J.A. Aronovitz, T.E. Conturo. Echo Planar Human Diffusion Tensor Imaging with Tetrahedral Encoding. Proc., ISMRM, 4th Meeting, New York, New York, 192 (1996).
- [50] A.G. Sorenson, R.G. Gonzalez, R.M. Weisskoff, T.L. Davis, T.G. Reese, W.A. Copen, J.L. Boxerman, J.P. Sasson, B.R. Rosen. In Vivo Full Diffusion Tensor Measurement in Normal and Pathologic Human Brain. Proc., ISMRM, 4th Meeting, New York, New York, 1327 (1996).
- [51] D. Le Bihan, R. Turner, C.T.W. Moonen, J. Pekar. Imaging of Diffusion and Microcirculation with Gradient Sensitization: Design, Strategy, and Significance. JMRI 1, 7-28 (1991).
- [52] D. Le Bihan. Molecular Diffusion Nuclear Magnetic Resonance Imaging. Magn. Res. Quarterly, 7(1), 1-30 (1991).
- [53] S. Brockstedt, C. Thomsen, R. Wirestam, J. De Poorter, M. Olsson, B.R.R. Persson, F. Stahlberg. Development of a Diffusion-Sensitive Pulse Sequence using an Enhanced Whole-Body Gradient System. Proc., SMR, 2nd Meeting, San Francisco, 1035 (1994).

- [54] J.E. Tanner. Transient diffusion in a system partitioned by permeable barriers. Application to NMR measurements with a pulsed field gradient. *J. Chem. Phys.* **69**, 1748-1754 (1978).
- [55] L.L. Latour, P.P. Mitra, R.L. Kleinberg, C.H. Sotak. Time-Dependent Diffusion of Fluids in Porous Media as a Probe of Surface-to-Volume Ratio. *J. Magn. Reson. A* **101**, 342-346 (1993).
- [56] M.A. Horsfield, W.I. McDonald. Restricted Self-Diffusion in CNS Tissue by Volume Selective Proton NMR. Proc., SMRM, 12th Meeting, New York, New York USA, 291 (1993).
- [57] L. Gates, I. Cameron. Restricted Diffusion in Human White Matter For Long Diffusion Times. Proc., SMR, 2nd Meeting, San Francisco, 1037 (1994).
- [58] M.A. Horsfield, M. Lai, G.J. Barker, R. Turner, S.L. Webb, P.S. Tofts, D.H. Miller, G.R. Cherryman, W.I. McDonald. Time-Dependent Diffusion Tensors by Volume-Selective NMR. Proc., SMR, 2nd Meeting, San Francisco, 1039 (1994).
- [59] A.M. Ulug, P.B. Barker, R.N. Bryan, P.C.M. van Zijl. Diffusion Tensor Imaging of the Human Brain. Proc., SMR, 3rd Meeting, Nice, France, 901 (1995).
- [60] E. Fukushima, S.B.W. Roeder. Experimental Pulse NMR. A Nuts and Bolts Approach. Addison-Wesley Publishing Company, Massachusetts, USA, 1981.
- [61] W.M. Cowan. The Use of Axonal Transport for Studies of Neuronal Connectivity. Elsevier Scientific Publishing Company, New York, 1975.

- [62] M.D. Kamen. Radioactive Tracers in Biology. An Introduction to Tracer Methodology. Academic Press Inc., New York, 1955.
- [63] M.K. Ahn, S.J.K Jenson, D. Kivelson. Molecular theory of diffusion constants in liquids. J.Chem. Phys., **57**, 2940–2951 (1972).
- [64] D. Le Bihan, R. Turner, P. Douek. Is water diffusion restricted in human brain white matter? An echo planar NMR imaging study. Neuroreport **4**, 887–890 (1993).
- [65] R.A. Damion, W. Vennart, I.R. Summers, R.E. Ellis. Water Diffusion Coefficient Measurements in the Finger by Magnetic Resonance Imaging. Magn. Reson. Imag. **12**, 873–879 (1994).
- [66] R.R. Edelman, J. Gaa, V.J. Wedeen, E. Loh, J.M. Hare, P. Prasad, W. Li. In Vivo Measurement of Water Diffusion in the Human Heart. Magn. Reson. Med., **32**, 423–428 (1994).
- [67] T.L. Chenevert, J.G. Pipe, D.M. Williams, J.A. Brunberg. Quantitative Measurement of Tissue Perfusion and Diffusion in Vivo. Magn. Reson. Med. **17**, 197–212 (1991).
- [68] A. Szafer, J. Zhong, J.C. Gore. Understanding MR Measurements of Water Diffusion in Tissues. Proc., SMR, 3rd Meeting, Nice, France, 918 (1995).
- [69] L. Gates, I. Cameron. Time Dependence of Water Diffusion In Human White Matter. Proc., SMR, 3rd Meeting, Nice, France, 355 (1995).

- [70] A. Finkelstein. Water Movement Through Lipid Bilayers, Pores, and Plasma Membranes: Theory and Reality. John Wiley & Sons, New York, 1987. pp. 155–165.
- [71] P. Douek, R. Turner, J. Pekar, N. Patronas, D. Le Bihan. MR Color Mapping of Myelin Fiber Orientation. *J. Comput. Assist. Tomogr.* **15**, 923–929 (1991).
- [72] J. Coremans, R. Luypaert, F. Verhelle, T. Stadnik, M. Osteaux. A Method for Myelin Fiber Orientation Mapping Using Diffusion-Weighted MR Images. *Magn. Res. Imag.* **12**, 443–454 (1994).
- [73] S. Mori, P.C.M. van Zijl. Diffusion Weighting by the Trace of the Diffusion Tensor within a Single Scan. *Magn. Reson. Med.*, **33**, 41–52 (1995).
- [74] E.W. Hsu. A Simplified Method for Determining Fiber Orientation Using New Interpretations of NMR Anisotropic Diffusion Measurements. *Proc., SMRM, 12th Meeting*, New York, USA, 583 (1993).
- [75] D. Davis, J. Ulatowski, S. Eleff, M. Izuta, S. Mori, D. Shungu, P.C.M. van Zijl. Monitoring of Changes in Water Diffusion Coefficients During Reversible Ischemia in Cat and Rat Brain. *Magn. Reson. Med.*, **31**, 454–460 (1994).
- [76] T. Niendorf, U. Flogel, D.G. Norris, D. Leibfritz. Monitoring of cell volume changes during osmotic stress and transient ischemia: The implications for diffusion weighted imaging. *Proc., SMR, 2nd Meeting*, San Fransisco, 1012 (1994).

- [77] S. Warrach, D. Chien, W. Li, M. Ronthal, R.R. Edelman. Fast magnetic resonance diffusion-weighted imaging of acute human stroke. *Neurology*, **42**, 1717-1723 (1992).
- [78] L. Gates, I. Cameron. Diffusion Measurements at Very Short Diffusion Times in Human Muscle Using a Double Maxwell Gradient Coil. Proc., SMR, 3rd Meeting, Nice, France, 912 (1995).
- [79] D. Morvan, A. Leroy-Willig. Simultaneous Measurements of Translational Diffusion and Transverse Relaxation in Normal Skeletal Muscle at Rest and During Recovery After an Exercise. Proc., SMRM, 12th Meeting, New York, USA, 606 (1993).
- [80] G.G. Cleveland, D.C. Chang, C.F. Hazelwood, H.E. Rorschach. Nuclear Magnetic Resonance Measurement of Skeletal Muscle. *Biophys. J.* **16**, 1043-1053 (1976).
- [81] D.C. Chang, H.E. Rorschach, B.L. Nichols, C.F. Hazelwood. Implications of Diffusion Coefficient Measurements for the Structure of Cellular Water. *Ann. NY. Acad. Sci.* **204**, 434-443 (1973).
- [82] L.A. Abetsedarskaya, F.G. Miftakhutdinova, V.D. Fedotov. State of Water in Live Tissues (results of investigations by the NMR-spin echo method) *Biophys.* **13**, 750-758 (1968).
- [83] J.E. Tanner. Self Diffusion of Water in Frog Muscle. *Biophys. J.* **28**, 107-116 (1979).

- [84] P.G. Morris. Nuclear Magnetic Resonance Imaging In Medicine and Biology. Oxford University Press. New York. 1986.
- [85] F.W. Wehrli. D. Shaw. J.B. Kneeland. Biomedical Magnetic Resonance Imaging. Principles. Methodology. and Applications. VCH Publishers. Inc.. New York. New York. 1988.
- [86] P.T. Callaghan. Principles of Nuclear Magnetic Resonance Microscopy. Oxford University Press. New York. 1991.
- [87] C.P. Slichter. Principles of Magnetic Resonance. Springer-Verlag Berlin Heidelberg 1978.
- [88] A.P. French. E.F. Taylor. An Introduction to Quantum Physics. W.W. Norton & Company. Inc. New York. N.Y. 1978.
- [89] G. Baym. Lectures on Quantum Mechanics. Addison-Wesley Publishing Company. Massachusetts. USA. 1973.
- [90] J.J. Sakurai. Modern Quantum Mechanics. Addison-Wesley Publishing Company. Massachusetts. USA. 1985.
- [91] C. Kittel and H. Kroemer. Thermal Physics. W.H. Freeman and Company. New York. 1980. pp. 398-399.
- [92] F. Reif. Fundamentals of statistical and thermal physics. McGraw-Hill Book Company. Toronto. 1965. pp. 251-488.
- [93] R.K. Pathria. Statistical Mechanics. Pergamon Press. New York. N.Y.. 1972.

- [94] D. Le Bihan. Diffusion and Perfusion Magnetic Resonance Imaging: Applications to Functional MRI. Raven Press. New York. 1995. pp. 7-9.
- [95] A. Abragam. The Principles of Nuclear Magnetism. Oxford University Press. London. 1961.
- [96] D. D. Stark, W.G. Bradley. Magnetic Resonance Imaging. Mosby Year Book. Inc. Toronto. 1992. pp. 130-131.
- [97] H. Saint-Jalmes, J. Tacquin, Y. Barjhoux. Design Data for Efficient Axial Gradient Coils: Application to NMR Imaging. *Magn. Reson. Med.* **2**, 245-252 (1985).
- [98] V.D. Barger, M.G. Olsson. Classical Electricity and Magnetism. A Contemporary Perspective. Allyn and Bacon. Inc., Boston. 1987. pp. 197-199.
- [99] P.A. Tipler. Physics. Worth Publishers. Inc. 2nd edition. New York. 1982. pp. 746-747.
- [100] F. Romeo, D.I. Hoult. Magnetic Field Profiling: Analysis and Correcting Coil Design. *Magn. Reson. Med.* **1**, 44-65 (1984).
- [101] R. Turner. Gradient Coil Design: A Review of Methods. *Magn. Reson. Imag.* **11**, 903-920 (1993).
- [102] H. Saint-Jalmes, J. Tacquin, Y. Barjhoux. Optimization of homogeneous electromagnetic coil systems: Application to whole-body NMR imaging magnets. *Rev. Sci. Instrum.* **52**(10), 1501-1508 (1981).
- [103] M. Durand. Magnetostatique (Masson. Paris. 1968).

- [104] Baxter Inc., Chicago USA. Private communication.
- [105] C.N. Chen, D.I. Hoult. Biomedical Magnetic Resonance Technology. Adam Hilger, IOP Publishing Ltd. 1989.
- [106] P.A. Bottomley. A versatile magnetic field gradient control system for NMR imaging. *J. Phys. E: Sci. Instrum.* **14**, 1081-1087 (1981).
- [107] G.A. Thibodeau, K.T. Patton. Anatomy and Physiology. Mosby Year Book, Inc. St. Louis, USA. 1993. pp. 227-273.
- [108] A.F. Gmitro, A.L. Alexander. Use of a Projection Reconstruction Method to Decrease Motion Sensitivity in Diffusion-Weighted MRI. *Magn. Reson. Med.*, **29**, 835-838 (1993).
- [109] J.W. Hugg, E.J. Butterworth. Diffusion Mapping with Modest Gradients Using a Double PGSE Preparation. *Proc..ISMRM*, 4th Meeting, New York, New York. 186 (1996).
- [110] P.R. Bevington. Data Reduction and Error Analysis for the Physical Sciences. McGraw-Hill, New York (1969).
- [111] W.H. Press, S.A. Teukolsky, W.T. Vetterling, B.P. Flannery. Numerical Recipes in FORTRAN. Cambridge University Press, New York, N.Y., 1986.
- [112] D.W. Fawcett. A Textbook of Histology. Chapman and Hall, New York. 1994. pp. 279-283.
- [113] G.E. Bell, J. Crank. Influence of Imbedded Particles on Steady-state Diffusion. *J. Chem. Soc. Faraday Trans. II*, **70**, 1259-1273 (1974).

- [114] T.J. Norwood, S.C.R. Williams. A new method for diffusion weighted imaging. *Magn. Reson. Imag.* **11**, 367-373 (1993).
- [115] M.B. Carpenter, J. Sutin. *Human Neuroanatomy*. 8th edition. Williams & Wilkins, Baltimore, 1983, pp. 228-238.
- [116] J.V. Hajnal, M. King, A. Oatridge. Measurement of Diffusion Coefficients and Barrier Spacings in the Corpus Callosum. *Proc., SMRM, 11th Meeting*, Berlin, Germany, 1204 (1992).
- [117] K. Merboldt, W. Hanicke, H. Bruhn, M.L. Gyngell, J. Frahm. Diffusion Imaging of the Human Brain in Vivo Using High-Speed STEAM MRI. *Magn. Reson. Med.* **23**, 179-192 (1992).
- [118] J.E. Tanner. Use of a Pulsed Magnetic Field Gradient for Measurements of Self-Diffusion by Spin-Echo Nuclear Magnetic Resonance with Applications to Restricted Diffusion in Several Tissues and Emulsions. Ph.D. Thesis, University of Wisconsin, Chemistry (1966).
- [119] J.E. Tanner and E.O. Stejskal. Restricted Self-Diffusion of Protons in Colloidal Systems by the Pulsed-Gradient, Spin-Echo Method. *J. Chem. Phys.* **49**, 1768-1777 (1968).
- [120] S.H. Koenig, R.D. Brown, M. Spiller, N. Lundbom. Relaxometry of the Brain: Why White Matter Appears Bright in MRI. *Magn. Reson. Med.*, **14**, 482-495 (1990).
- [121] C. Romero-Sierra. *Neuroanatomy. A Conceptual Approach*. Churchill Livingstone Inc. New York, New York, 1986.

- [122] H.C. Elliot. Textbook of Neuroanatomy. J.B. Lippincott Company, Philadelphia. 1963.
- [123] A. Mackay, K. Whittall, J. Adler, D. Li, D. Paty, D. Graeb. In Vivo Visualization of Myelin Water in Brain by Magnetic Resonance. *Magn. Reson. Med.*, **31**, 673–677 (1994).
- [124] K. Whittall, A. Mackay, D. Graeb, R.A. Nugent, D. Li, D. Paty. In Vivo Measurement of T_2 Distributions and Water Contents in Normal Human Brain. *Magn. Reson. Med.*, **37**, 34–43 (1997).
- [125] S. Ohara, H. Nagai, T. Matsumoto, T. Banno. MR imaging of CSF pulsatory flow and its relation to intracranial pressure. *J. Neurosurg.* **69**, 675–682 (1988).
- [126] A. de Crespigny, M. Yenari, D. Enzmann, M. Marks, M. Mosely. Navigated Spin-echo Diffusion Imaging of Human Stroke. *Proc., SMR, 2nd Meeting*, San Francisco, 137 (1994).
- [127] Berne and Levy. *Physiol. Rev.* **64**, 1240–1269 (1984).
- [128] P. van Gelderen, M.H.M. de Vleeschouwer, D. DesPres, J. Pekar, P.C.M. van Zijl, C.T.W. Moonen. Water Diffusion and Acute Stroke *Magn. Reson. Med.* **31**, 154–163 (1994).
- [129] P. Gideon, C. Thomsen, O. Henriksen. Increased Self-Diffusion of Brain Water in Normal Aging. *J. Magn. Reson. Imag.*, **4**, 185–188 (1994).
- [130] D. Morvan, A. Leroy-Willig, P. Jehenson, C.A. Cuenod, A. Syrota. Temperature Changes Induced by ^1H -Decoupling in Human Muscle Measured by

a MRI Diffusion Technique. Proc., SMRM, 11th Meeting, Berlin, Germany, 590 (1992).

IMAGE EVALUATION TEST TARGET (QA-3)

APPLIED IMAGE, Inc.
1653 East Main Street
Rochester, NY 14609 USA
Phone: 716/482-0300
Fax: 716/288-5989

© 1993, Applied Image, Inc., All Rights Reserved