A NEW MULTIPARAMETRIC MRI PROTOCOL FOR DIAGNOSIS OF
PROSTATE CANCER

by

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Abstract

In this thesis, a new magnetic resonance imaging (MRI) quantitative T₂ mapping technique, called Luminal water imaging (LWI), has been developed and used for non-invasive detection and grading of prostatic tumours.

Using this technique, we measured what we hypothesized to be the fractional amount of water content of luminal spaces in prostate, and called it Luminal Water Fraction, LWF. Based on the differences in tissue composition and fractional amount of luminal space between malignant and normal prostatic tissues and between tumors of different grades, we hypothesized that the measurements of LWF could be used for the detection and grading of prostatic tumours.

To verify these hypotheses, we performed two patient studies in which we compared MR measurements of LWI with whole-mount histology. In the first study, we evaluated the correlation between LWF and the percentage area of luminal space in the prostatic tissue. The results of this study demonstrated that LWF is significantly and strongly correlated with the percentage area of luminal space in the prostatic tissue. In the second study, we investigated the feasibility of LWI in the detection and grading of prostate cancer. The results of this study showed that LWI provides high accuracy both in the detection and grading of prostatic tumours.

After verifying our hypotheses, we performed a detailed comparison between the diagnostic accuracy of LWI and the more established MRI techniques: Dynamic Contrast-Enhanced (DCE) and Diffusion-Weighted MRI (DW-MRI). The results of this pilot study showed that
LWI alone performs better than DCE, DW-MRI, or their combination, in the detection of prostatic tumours and also in correlation with GS. Based on the results of this study, we proposed a guideline for making a more efficient, abbreviated multi-parametric MRI protocol for the diagnosis of prostate cancer.

Finally, as a side project, we explored some potential areas of improvement in DCE-MRI by investigating the impact of temporal resolution on the accuracy of DCE-MRI in detection of prostatic tumours. Our results showed that within a certain range of temporal resolutions, the diagnostic accuracy of DCE-MRI would be independent of the temporal resolution.
Lay Summary

In this thesis, a new magnetic resonance imaging (MRI) technique has been developed and used for diagnosis of prostate cancer.

Prostatic tissue is composed of water compartments of different sizes. The fractional amount of these compartments varies between normal and malignant tissue, and between tumours of different grades. Our technique has been developed for diagnosis of prostate cancer by measuring the fractional amount of water content of different compartments. We evaluated the correlation between measurements of this MRI technique and the composition of prostatic tissue. We also investigated the feasibility of this technique in the detection and grading of prostate cancer by comparing MRI measurements with histology. In addition, we compared the diagnostic accuracy of this technique with the more established MRI techniques.

Finally, as a side project, we explored some potential areas of improvement in dynamic contrast-enhanced MRI which is an established MRI technique for characterization of tumour microvasculature.
Preface

This study was approved by the University of British Columbia (UBC) Clinical Research Ethics Board (CREB) (H02-70400), and supported by the Canadian Institutes of Health Research (CIHR) (MOP-115052).

This study was performed in collaboration with multiple researchers. The idea of multi-component $T_2$ mapping in prostate was created by Dr. Piotr Kozlowski, who had the vision for this project from the very beginning and supported it all the way through with grants and providing invaluable guidance as my primary supervisor and mentor for this research. He also supported the design and development of the protocol that was developed as one of the major contribution of this study, arranged patient studies, and provided valued insight in interpretation and analysis of results. The majority of the work of this project was performed by the author. The author’s contribution to this work includes: development, performance and interpretation of the simulation and phantom studies, assistance with MRI examinations for patient studies, aid in whole-mount histology preparation, and performance of image analysis and interpretation of the entire results.

This work would have not been possible without the contributions from the following collaborators:

Dr. Silvia D Chang, an experienced radiologist and an Associate Professor at the Department of Radiology of UBC, assisted with protocol development and set up of MRI examinations for patient studies. Dr. Chang’s contribution also includes aid and confirmation of manually drawn regions of interest on the MR images.
Dr. Jing Zhang provided the pulse sequence for luminal water imaging (LWI), and assisted with the data acquisition of volunteer studies.

Dr. Edward C. Jones, an experienced pathologist at Vancouver General Hospital (VGH), performed cutting of the prostate glands using a custom made device, delineated tumours, and assigned the Gleason score (GS) on whole-mount histology slides.

Dr. Ladan Fazli, an experienced pathologist at Vancouver Prostate Center (VPC), delineated tumours, and assigned the GS on digital (high resolution) whole-mount histology slides. She also assisted with colour segmentation of the digital histology images.

Dr. S. Larry Goldenberg, a senior urology surgeon and researcher at VGH, assisted with the interpretation of the results of patient studies and with patient recruitment.

Dr. Peter Black, an experienced urology surgeon and researcher at VGH, assisted with the interpretation of the results of patient studies, and also supported this project by getting involved his fellow Dr. Richard Savdie. Dr. Savdie recruited all of the patients for this study.

The contributions from this thesis have led to the following publications, submissions and presentations:


3) Sabouri S, Chang SD, Goldenberg SL, Savdie R, Jones EC, Black PC, Kozlowski P. Comparing Diagnostic Accuracy of Luminal Water Imaging with Diffusion-Weighted and Dynamic Contrast-Enhanced MRI in Detection and Grading of Prostate Cancer (Manuscript ready to be submitted). A version of this article is also presented in Chapter 5.


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<th>Symbol</th>
<th>Description</th>
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<tr>
<td>$A_{\text{long}}$</td>
<td>Area under the peak of long $T_2$</td>
</tr>
<tr>
<td>$A_{\text{short}}$</td>
<td>Area under the peak of short $T_2$</td>
</tr>
<tr>
<td>$K^{\text{trans}}$</td>
<td>Volume transfer constant</td>
</tr>
<tr>
<td>$N_{\text{comp}}$</td>
<td>Number of peaks in the $T_2$ distribution</td>
</tr>
<tr>
<td>$T_1$</td>
<td>Spin-lattice relaxation time</td>
</tr>
<tr>
<td>$T_2$</td>
<td>Spin-spin relaxation time</td>
</tr>
<tr>
<td>$T_{2\text{-long}}$</td>
<td>Geometric mean value of long $T_2$</td>
</tr>
<tr>
<td>$T_{2\text{-short}}$</td>
<td>Geometric mean value of short $T_2$</td>
</tr>
<tr>
<td>$v_e$</td>
<td>Fractional volume of the extra-vascular extra-cellular space</td>
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<tr>
<td>$v_p$</td>
<td>Fractional volume of plasma</td>
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# List of Abbreviations

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
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<tbody>
<tr>
<td>2D</td>
<td>Two-Dimensional</td>
</tr>
<tr>
<td>3D</td>
<td>Three-Dimensional</td>
</tr>
<tr>
<td>ACR</td>
<td>American College of Radiology</td>
</tr>
<tr>
<td>ADC</td>
<td>Apparent Diffusion Coefficient</td>
</tr>
<tr>
<td>AFMS</td>
<td>Anterior Fibromuscular Stroma</td>
</tr>
<tr>
<td>AIC</td>
<td>Akaike Information Criterion</td>
</tr>
<tr>
<td>AUC</td>
<td>Area Under the Receiver Operating Characteristic (ROC) Curve</td>
</tr>
<tr>
<td>BIC</td>
<td>Bayesian Information Criterion</td>
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<tr>
<td>BPH</td>
<td>Benign Prostatic Hyperplasia</td>
</tr>
<tr>
<td>BW</td>
<td>Bandwidth</td>
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<tr>
<td>CZ</td>
<td>Central Zone</td>
</tr>
<tr>
<td>DCE</td>
<td>Dynamic Contrast-Enhanced</td>
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<tr>
<td>DCE-MRI</td>
<td>Dynamic Contrast-Enhanced MRI</td>
</tr>
<tr>
<td>DRE</td>
<td>Digital Rectal Exam</td>
</tr>
<tr>
<td>DWI</td>
<td>Diffusion Weighted Imaging</td>
</tr>
<tr>
<td>DW-MRI</td>
<td>Diffusion Weighted MRI</td>
</tr>
<tr>
<td>EES</td>
<td>Extracellular-Extravascular Space</td>
</tr>
<tr>
<td>EMF</td>
<td>Electromotive Force</td>
</tr>
<tr>
<td>ESUR</td>
<td>European Society of Urogenital Radiology</td>
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<tr>
<td>FOV</td>
<td>Field of View</td>
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<tr>
<td>Abbreviation</td>
<td>Description</td>
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<tr>
<td>GLMM</td>
<td>Generalized Linear Mixed-Effect Model</td>
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<tr>
<td>gmT₂</td>
<td>geometric mean T₂</td>
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<tr>
<td>GS</td>
<td>Gleason Score</td>
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<tr>
<td>GRASE</td>
<td>Gradient Spin Echo</td>
</tr>
<tr>
<td>H&amp;E</td>
<td>Hematoxylin and Eosin</td>
</tr>
<tr>
<td>IAE</td>
<td>Integral Approximation Error</td>
</tr>
<tr>
<td>LT2F</td>
<td>Long T₂ Fraction</td>
</tr>
<tr>
<td>MP-MRI</td>
<td>Multi-Parametric MRI</td>
</tr>
<tr>
<td>MR</td>
<td>Magnetic Resonance</td>
</tr>
<tr>
<td>MRE</td>
<td>Magnetic Resonance Elastography</td>
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<tr>
<td>MRI</td>
<td>Magnetic Resonance Imaging</td>
</tr>
<tr>
<td>MRSI</td>
<td>MR Spectroscopic Imaging</td>
</tr>
<tr>
<td>NE</td>
<td>Number of Echoes</td>
</tr>
<tr>
<td>NMR</td>
<td>Nuclear Magnetic Resonance Imaging</td>
</tr>
<tr>
<td>NNLS</td>
<td>Non-Negative Least Squares</td>
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<tr>
<td>PCa</td>
<td>Prostatic Carcinoma</td>
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<tr>
<td>PD</td>
<td>Proton Density</td>
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<tr>
<td>PFMS</td>
<td>Periurethral Fibromuscular Stroma</td>
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<tr>
<td>PI-RADS</td>
<td>Prostate Imaging Reporting and Data System</td>
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<tr>
<td>PSA</td>
<td>Prostate-Specific Antigen</td>
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<td>PZ</td>
<td>Peripheral Zone</td>
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<td>RF</td>
<td>Radio Frequency</td>
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<td>Abbreviation</td>
<td>Description</td>
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<td>--------------------------------------</td>
</tr>
<tr>
<td>ROC</td>
<td>Receiver Operating Characteristic</td>
</tr>
<tr>
<td>ROI</td>
<td>Regions of Interest</td>
</tr>
<tr>
<td>SAR</td>
<td>Specific Absorption Rate</td>
</tr>
<tr>
<td>SENSE</td>
<td>Sensitivity Encoding</td>
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<tr>
<td>SNR</td>
<td>Signal to Noise Ratio</td>
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<tr>
<td>T1W</td>
<td>T$_1$-weighted</td>
</tr>
<tr>
<td>T2W</td>
<td>T$_2$-weighted</td>
</tr>
<tr>
<td>TE</td>
<td>Echo Time</td>
</tr>
<tr>
<td>TRUS</td>
<td>Transrectal Ultrasound</td>
</tr>
<tr>
<td>TR</td>
<td>Repetition Time</td>
</tr>
<tr>
<td>TZ</td>
<td>Transition Zone</td>
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First and foremost, I would like to express my deepest gratitude to my supervisor Dr. Piotr Kozlowski for his leadership, warm encouragement, never-ending patience and unlimited support. He created many rewarding opportunities for me, and his excellent knowledge and intuition was a great asset for this research. I consider myself fortunate and privileged to have had the opportunity to perform my research under his supervision.

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Dedication

To my dearest parents, best brother, and beloved husband.
Chapter 1: Introduction

1.1 Magnetic Resonance Imaging (MRI)

MRI is a non-invasive imaging technique that has been used for over 40 years to provide anatomical, physiological, and functional information from the human body, without use of ionizing radiation.

The fundamental finding, based on which MRI is developed, was achieved in 1938 by Isidor I. Rabi (1, 2). Rabi measured the nuclear magnetic moment by using Nuclear Magnetic Resonance (NMR), and explained how the magnetic orientation of nuclei could be flipped by use of oscillating magnetic fields. In 1946, two independent teams led by Felix Bloch and Edward Purcell simultaneously discovered NMR in condensed matter (3, 4, 5). The first NMR image was produced later in 1971 by Paul Lauterbur (6), and the first MRI scan of the human body was performed in 1977 by Raymond Damadian (7). In early 1980 commercial MRI scanners were introduced and became clinically available in hospitals. Since then, there have been many revolutionary improvements in the instrumentation, computer technology, and data processing techniques in the field of MRI. Nowadays, MRI is an essential imaging tool that provides a wide range of anatomical and physiological information from the human body for diagnostic purposes.
1.1.1 Magnetization Vector

In classical mechanics, the magnetic moment of an object is a measure of its tendency to align with an external magnetic field. An object with non-zero magnetic moment that is exposed to an external magnetic field, experiences a torque, and rotates about the direction of the magnetic field. In quantum mechanics, particles possess an intrinsic form of angular momentum called spin, S. Particles with non-zero spin have a net magnetic moment and when exposed to a magnetic field will precess about the direction of the magnetic field. This motion is called Larmor precession, and the angular frequency of this precessing motion is known as Larmor frequency, \( \omega_L \), which is related to the magnitude of the applied magnetic field, B, with the following equation:

\[
\omega_L = -\gamma B
\]

Equation 1-1

where \( \gamma \) is the gyromagnetic ratio, i.e. the ratio of magnetic moment of a particle to its angular momentum.

A rotating magnetic moment can be detected using a coil placed next to it. This is because precessing magnetic moment causes a change of magnetic flux in the coil, which, based on Faraday’s law, generates an induction current in the coil.

In each atom, electrons and the nucleus can be sources of magnetic moment. Both electronic and nuclear magnetic moments precess in the presence of an external magnetic field, and therefore, can be detected using magnetic resonance techniques (Nuclear Magnetic Resonance (NMR) for the nucleus, and Electron Paramagnetic Resonance (EPR) for the electron). In MRI, the source of magnetism is the magnetic moment of nuclei. The MRI signal is generated
from Larmor precession of the magnetic moment of the hydrogen nucleus, \(^1\)H, which is a proton. \(^1\)H is used in MRI mainly due to the abundance of water in biological systems.

An MRI signal is acquired from macroscopic pieces of tissue, rather than individual nuclei. In a macroscopic piece of tissue, there are billions of \(^1\)H nuclei. Magnetic moments of such nuclei in the absence of any external magnetic field are isotropically oriented, resulting in an almost net zero magnetic moment. Right after application of an external magnetic field, magnetic moments of spins precess about the magnetic field, still with an isotropic distribution of orientations. Therefore, the nuclear magnetic moments do not contribute to the magnetism of the material immediately after being exposed to a magnetic field. However, gradually nuclear spins exchange energy with the neighboring molecules, and eventually the isotropy of the distribution of magnetic moments’ orientation breaks. As a result of this a net magnetic moment along the direction of the magnetic field appears in the sample. A simplified explanation of this phenomenon, provided by quantum mechanics, is as follows.

The proton is a spin \(\frac{1}{2}\) particle, and hence has two spin eigenstates called spin-up and spin-down (with eigenvalues of \(S=+1/2\), \(S=-1/2\), respectively). Corresponding to the two spin eigenstates, there are two energy levels, \(E(S)\), given by the equation below:

\[
E(S) = -\gamma \frac{h}{2\pi} S B
\]

Equation 1-2

where \(h\) is the Planck constant \(= 6.626 \times 10^{-34} \text{ m}^2\text{kg/s}\), and \(B\) is the magnitude of the applied magnetic field.
The state of individual spins can be expressed as a linear combination of the eigenstates. In a system at finite temperature, the population of energy states, $P(E)$, for an ensemble of spins is given by Boltzmann distribution equation, written as follows:

$$P(E) = A e^{\left(-\frac{E}{kT}\right)}$$  \hspace{1cm} \text{Equation 1-3}$$

where $A$ is a constant of the system, $k$ is the Boltzmann constant, equal to $1.381 \times 10^{-23}$ m$^2$kgs$^{-2}$K$^{-1}$, and $T$ is the absolute temperature of the system.

It can be easily observed from Equation 1-2 and Equation 1-3 that the probability for proton spins to be in state up and down is different. Therefore, in an ensemble there is a slight tendency for the spins to point in the direction of the field rather than against it. For a large number of nuclear spins, $N$, with individual magnetic moments of $m$, where $m=\gamma \frac{\hbar}{2\pi} S$, the net macroscopic magnetization, $\vec{M}$, can be calculated as below:

$$\vec{M} = \vec{m} \times N \times \left( \frac{P(E_{\text{down}}) - P(E_{\text{up}})}{P(E_{\text{down}}) + P(E_{\text{up}})} \right) = \vec{m} \times N \times \tanh \left( \frac{\Delta E}{2kT} \right) \approx \vec{m} \times N \times \left( \frac{\Delta E}{2kT} \right) = N\gamma^2 \frac{\hbar^2}{4\pi^2} \frac{\vec{B}}{4kT}$$

\hspace{1cm} \text{Equation 1-4}$$

$\vec{M}$ is called the thermal equilibrium magnetization vector. In a macroscopic piece of tissue when $\vec{M}$ is tipped away from the direction of the magnetic field, $\vec{B}$, it precesses and generates the signal that can be detected and used for image construction in MRI.
1.1.2 \( T_1, T_2, \text{ and } T_2^* \) Relaxations

As explained earlier, the first step to generate an MRI signal is having an external magnetic field to create a net magnetization vector in a macroscopic piece of matter. The second step would be to tip away the created net magnetization vector from the direction of the external magnetic field so that the net magnetization vector precesses and generates a signal. The procedure of tipping the net magnetization vectors from the direction of the magnetic field is performed by application of radio frequency (RF) excitation pulses, which is explained in more detail in Section 1.1.4. After the net magnetization vector is tipped away and the RF pulse is turned off, the precession does not last forever; eventually nuclei spins restore their equilibrium state and the net magnetization vector aligns itself with the direction of the magnetic field. The process by which the magnetization vector reaches its thermal equilibrium state is called relaxation and is divided into two independent mechanisms: spin-lattice, also known as longitudinal or \( T_1 \) relaxation, and spin-spin, also known as transverse or \( T_2 \) relaxation.

\( T_1 \) relaxation is the process by which the longitudinal component of the magnetization vector, which is along the direction of the magnetic field (here chosen to be \( \hat{z} \)), is recovered. In this process, nuclei spins exchange energy with the surrounding molecules (or lattice, in solid samples) to return to the equilibrium state. Mathematically, longitudinal relaxation is modelled by an exponential growth function with the \( T_1 \) relaxation time being defined as the time constant. This can be expressed as below:
\[ M_z(t) = M_z(0) e^{-\frac{t}{T_1}} + M_0 (1 - e^{-\frac{t}{T_1}}) \]  

Equation 1-5

where \( M_z \) is the component of magnetization vector along the direction of magnetic field (\( \hat{z} \)), and \( M_0 \) is the equilibrium magnetization.

\( T_2 \) relaxation is the process by which the transverse component of magnetization vector decays toward its equilibrium value of zero. This mechanism occurs because the local fields that spins experience is a combination of the external magnetic field and the magnetic fields of spins’ neighbours. As a result of the differences in the local fields, individual spins precess at slightly different frequencies, which dephases the spins. Mathematically, transverse relaxation is modelled by an exponential decay function with the \( T_2 \) relaxation time being the time constant. This can be expressed as below:

\[ M_T(t) = M_{T0} e^{-\frac{t}{T_2}} \]  

Equation 1-6

where \( M_T(t) \), and \( M_{T0} \) are the magnitude of the transverse component of the net magnetization vector at time \( t \), and before the beginning of relaxation, respectively.

Based on Equation 1-6, \( T_2 \) relaxation time can be described as the time at which the transverse component of \( M \) decays to approximately 37\% of its initial value.

Representative values of \( T_1 \) and \( T_2 \) relaxation times in human body tissues are presented in Table 1-1.
Table 1-1. Representative values of relaxation parameters $T_1$ and $T_2$, in milliseconds, for hydrogen components of different human body tissues at 1.5 T magnetic field and 37°C (human body temperature).

These are only approximate values. The table is reproduced from (8).

<table>
<thead>
<tr>
<th>Tissue</th>
<th>$T_1$ (ms)</th>
<th>$T_2$ (ms)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gray Matter (GM)</td>
<td>950</td>
<td>100</td>
</tr>
<tr>
<td>White Matter (WM)</td>
<td>600</td>
<td>80</td>
</tr>
<tr>
<td>Muscle</td>
<td>900</td>
<td>50</td>
</tr>
<tr>
<td>Cerebrospinal Fluid</td>
<td>4500</td>
<td>2200</td>
</tr>
<tr>
<td>Fat (CSF)</td>
<td>250</td>
<td>60</td>
</tr>
<tr>
<td>Blood</td>
<td>1200</td>
<td>100-200</td>
</tr>
</tbody>
</table>

The decay of the transverse magnetization results from any processes causing spin dephasing. In addition to the processes involved in the $T_2$ relaxation, spin dephasing can be caused by local static field inhomogeneity related to hardware imperfections and/or sample related local magnetic susceptibility effects. The general spin dephasing process is thus called $T_2^*$ relaxation, with the static component called $T_2'$ relaxation. $T_2$, $T_2^*$, and $T_2'$ relaxation times are related by the following equation:

$$\frac{1}{T_2^*} = \frac{1}{T_2} + \frac{1}{T_2'}$$  

Equation 1-7

Unlike for $T_1$ and $T_2$ relaxations, the loss of magnetization due to $T_2'$ effects can be recovered using spin-echo techniques. In these techniques, a series of refocusing RF pulses is applied, with time intervals $t$, following the initial spin excitation. By application of the refocusing pulses, the inhomogeneous static dephasing evolution reverses to form echoes at times $n \times 2t$ (where $n$ is an integer). The time between consecutive peaks of echoes, $2t$, at which the MR
signal is sampled, is called echo spacing, TE. A signal decay curve obtained from a typical spin-echo technique is shown in Figure 1-1.

![Signal decay curve](image)

**Figure 1-1.** Signal decay curve from a typical spin echo process showing the use of refocusing RF pulses to rephase the protons and to produce an echo signal.

A spin echo was first discovered by E.L. Hahn in 1950 (9). The modified spin-echo technique, known as CP, was later developed by H.Y. Carr and E.M. Purcell (10). The CP technique was further modified by S. Meiboom and D. Gill (11), and therefore was called CPMG (Carr Purcell Meiboom Gill). The CPMG sequence is nowadays widely used for T₂ mapping experiments.

In MRI, most often a complete set of data is acquired from application of a series of RF excitation and gradient pulses. The time between two consecutive excitation pulses is called the repetition time or TR. In a spin-echo experiment, where the RF excitation pulses are applied at time interval TR and signal data are sampled at time interval TE, by combining Equation

8
1-5 and Equation 1-6, the transverse component of the magnetization vector, to which the signal is proportional, can be expressed as follows:

\[ M_T(TE) = M_0 \left( 1 - e^{-TR/T_1} \right) e^{-TE/T_2} \]  

Equation 1-8

1.1.3 Bloch Equations

The equation of motion for a net magnetization vector \( \mathbf{M} \) from an ensemble of nuclei in an external magnetic field \( \mathbf{B} \), without considering the spin-lattice and spin-spin interactions, can be written as below (8):

\[ \frac{d}{dt} \mathbf{M} = \gamma \mathbf{M} \times \mathbf{B} \]  

Equation 1-9

The more accurate equation of motion, accounting for the spin-lattice and spin-spin interactions, in a coordinate system where \( \mathbf{B} \) is in z direction, can be expressed by the following equation, also known as the Bloch equation:

\[ \frac{d}{dt} \mathbf{M} = \gamma \mathbf{M} \times \mathbf{B} - \frac{M_x \hat{x} + M_y \hat{y}}{T_2} - \frac{(M_z - M_0) \hat{z}}{T_1} \]  

Equation 1-10

where \( M_x, M_y, \) and \( M_z \) are the components of the magnetization vector along the \( x, y, \) and \( z \) directions, respectively.

The Bloch equation was introduced by F. Bloch in 1946 (4) and has the following general solutions:

\[ M_x(t) = [M_x(0) \cos(\omega_L t) + M_y(0) \sin(\omega_L t)] \exp\left(-\frac{t}{T_2}\right) \]  

Equation 1-11

\[ M_y(t) = [-M_x(0) \sin(\omega_L t) + M_y(0) \cos(\omega_L t)] \exp\left(-\frac{t}{T_2}\right) \]  

Equation 1-12

\[ M_z(t) = M_0 + [M_z(0) - M_0] \exp(-\frac{t}{T_1}) \]  

Equation 1-13
In complex space, the two transverse components can be combined to generate a single transverse magnetization vector, \( M_T = M_x + iM_y \), and accordingly Equation 1-11 and Equation 1-12 can be combined and expressed as below:

\[
M_T(t) = M_T(0)\exp(i\omega_L t - \frac{t}{T_2})
\]  
Equation 1-14

As the MRI signal is generated by the transverse component of the magnetization vector, Equation 1-14 is a very practical equation and often referred to in MRI formalism.

1.1.4 RF Excitation and Signal Detection

As mentioned earlier, tilted net magnetization vectors in a magnetic field will precess and generate a detectable signal until they reach a thermal equilibrium state. This process is called free induction decay, FID, and the signal that is generated during this process is called the FID signal. Tipping away of a magnetization vector from the direction of the primary magnetic field can be performed by applying either a strong secondary magnetic field (along a different direction) or a weak oscillating magnetic field as long as it oscillates at the same frequency as the frequency of precession, known as the resonance frequency. In MRI, the magnetization vector is tilted by using the second method, i.e. application of a (secondary) oscillating magnetic field, which is much smaller and perpendicular to the direction of the primary static magnetic field. The frequency of this oscillating magnetic field is in the RF range, and so the process of tilting the magnetization vector, or excitation of spins into a non-equilibrium state, is called RF excitation. The reason why the frequency of an excitation pulse must be in the RF range can be understood based on Equation 1-1. By substituting the gyromagnetic ratio of a proton \((\gamma=2.675222\times10^8 \text{ rad.sec.T}^{-1})\) in Equation 1-1 and dividing it by \(2\pi\), the Larmor
The frequency of protons per unit magnetic field will be calculated and is equal to 42.578 MHz. T. The magnitude of the static magnetic field used in typical MRI scanners ranges from 1 to 10 T. For this range, the Larmor frequency of protons ranges between 42 and 420 MHz, which is within the RF range (3 kHz – 300 GHz).

If a coil whose surface vector is not parallel to the direction of the static magnetic field is located near the excited sample, the precession motion of the transverse component of the magnetization vector causes a change in the magnetic flux through the coil. As mentioned earlier, change of magnetic flux based on Faraday’s law induces electromotive force (EMF) in the coil. This induced EMF, generated from precessing magnetization, is the measurable signal in MRI experiments. The equation relating the detected signal to characteristics of the imaging system is derived in detail in (8). The final expression is as follows:

\[
    s(t) \propto \omega_L \int d^3r \, e^{-t/T_2(\vec{r})} M_T(\vec{r}, 0) B_T(\vec{r}) \, e^{i(\Omega - \omega_L) t + \varphi_0(\vec{r}) - \theta_B(\vec{r})}
\]

Equation 1-15

where \( s(t) \) is signal at time \( t \), \( M_T(\vec{r}, 0) \) and \( \varphi_0(\vec{r}) \) are the magnitude and phase of the complex magnetization vector at time 0 and location \( \vec{r} \) (\( \vec{r} \) is a 3D vector) determined by the initial RF pulse condition. \( B_T(\vec{r}) \) and \( \theta_B(\vec{r}) \) are the magnitude and phase of the receiver coil’s magnetic field per unit current produced at location \( \vec{r} \), and \( \Omega \) is the reference signal frequency: frequency of precession in a reference frame rotating with the Larmor frequency \( \omega_L \).

It is important to note that in Equation 1-15 the signal is related to both magnitude and phase of the magnetization vector.
1.1.5 Slice Selection

It has been mentioned earlier that excitation of spins happens only when the frequency of the applied RF pulse equates (or is close to) to the Larmor frequency of protons in the system. Larmor frequency is linearly related to the magnitude of the static magnetic field, expressed by Equation 1-1. Therefore, if an additional non-uniform magnetic field (generated by a series of coils called gradient coils), changing linearly in a desired direction is added to the existing static magnetic field during the application of the RF excitation pulse, only spins within a limited volume of the sample will be excited, and the detected signal can be spatially localized.

To clarify this matter, let us suppose that a field gradient, $\vec{G}_r$, is applied along the $\vec{r}$ direction (as demonstrated in Figure 1-2) to excite a desired slice perpendicular to $\vec{r}$. The linear perturbation of the magnetic field due to this field gradient along the $\vec{r}$ direction can be calculated by below equation:

$$\Delta B = \vec{r} \cdot \vec{G}_r = r G_r$$  \hspace{1cm} \text{Equation 1-16}

Accordingly, the total magnetic field along $\vec{r}$ can be expressed as below:

$$B(r) = B_0 + r G_r$$  \hspace{1cm} \text{Equation 1-17}

where $B_0$ is the magnitude of the static magnetic field.

By substituting Equation 1-17 in Equation 1-1 the precession frequency at position $r$ can be expressed as below:

$$\omega(r) = \omega_0 + \gamma r G_r$$  \hspace{1cm} \text{Equation 1-18}

where $\omega_0 = \gamma B_0$.

Equation 1-18 implies that under the impact of the field gradient precessing frequency is a linear function of position along the direction of the gradient. To excite a thin slice with
thickness $\Delta r$, centered at $r_0$, expanding from $r_0-\Delta r/2$ to $r_0+\Delta r/2$, the frequency of the excitation RF pulse must be tuned to a bandwidth $\Delta \omega$ ranging from $\gamma r_0 G_r - \gamma \frac{\Delta r}{2} G_r$ to $\gamma r_0 G_r + \gamma \frac{\Delta r}{2} G_r$. The thickness of the excited slice, $\Delta r$, and the bandwidth, $BW$, of the RF excitation pulse are related by the following equation:

$$BW = \Delta \omega = (\gamma r_0 G_r + \gamma \frac{\Delta r}{2} G_r) - (\gamma r_0 G_r + \gamma \frac{\Delta r}{2} G_r) = \gamma \Delta r G_r$$  

Equation 1-19

Figure 1-2. Slice selection by application of field gradient. Only spins within a plane perpendicular to field gradient, $G_r$, are excited by application of the RF pulse with a tuned bandwidth $\Delta \omega$.

In two-dimensional (2D) MRI, during each RF excitation only spins within a single slice will be excited. Following the slice excitation, signal localization in two in-plane directions is performed using frequency encoding and phase encoding, which are briefly explained in Section 1.1.6, and 1.1.7, respectively.
1.1.6 Frequency Encoding

In this localization technique, another non-uniform magnetic field, linearly varying in an in-plane direction, is applied after a slice was selected by simultaneous application of an RF excitation pulse and slice selective gradient.

Let us assume that the non-uniform magnetic field varies in the x direction. Then, during the application of a frequency encoding field gradient, spins at different locations along the x axis are affected by a different total magnetic field, and therefore they precess at different Larmor frequencies based on Equation 1-1. Through analysis of the detected signal and application of Fourier transformation it is possible to quantify the number of nuclei precessing at each frequency.

It must be noted that this technique only discriminates spins at different locations along one direction (x direction here). Therefore, the outcome of the Fourier transformation will be a projection of the 2D spin distribution onto the x axis. To complete spatial localization, another technique must be used to discriminate spins at different locations along the second dimension (y axis in here). The technique used for this purpose is phase encoding, and is explained in the following section.

1.1.7 Phase Encoding

It was explained earlier, with the aid of Equation 1-15, that the induction signal contains information from both magnitude and phase of complex magnetization vectors. The phase encoding procedure, as its name suggests, encodes spatial information across one axis (y axis here) into the phase of magnetization vector. In this technique, a non-uniform magnetic field which varies linearly across the y axis is applied for a short period of time between slice
selection and frequency encoding. Similar to what was explained for frequency encoding above, during the application of the phase encoding gradient, spins at different locations across the y axis will be exposed to different total magnetic fields and will precess at different Larmor frequencies. Therefore, when the phase encoding gradient is turned off and followed by a frequency encoding gradient, nuclei at different positions across the y axis will have different initial phases, while nuclei at different position across x axis will precess with different Larmor frequencies. A complete set of MRI data may be collected from repeated RF excitations followed by the use of different configurations of the phase encoding gradient. By applying a 2D Fourier transformation on such a data set, images of nuclei distribution in the x-y plane can be constructed.

1.1.8 Fourier Transform and Image Reconstruction

The Fourier transform is a mathematical theorem on the basis of which it is possible to transform a function into an alternative representation. In a 2D Fourier transform, a function \( f(x,y) \) can be related to its representative \( F(k_x,k_y) \), using the following equations:

\[
\rho(x, y) = \iint_{-\infty}^{\infty} S(k_x, k_y) e^{+2\pi i (k_x x + k_y y)} dk_x dk_y \quad \text{Equation 1-20}
\]

\[
S(k_x, k_y) = \iint_{-\infty}^{\infty} \rho(x, y) e^{-2\pi i (k_x x + k_y y)} dx dy \quad \text{Equation 1-21}
\]

The Fourier transform can be used to convert raw data collected as the induction signal to MR images that represent the distribution of spins or, correspondingly, the nuclei distribution. To explain this matter, let us look into the signal expression given by Equation 1-15. It can be shown (8) that in a proper rotating frame, the expression of a complex induction signal can be simplified to the following equation:
\[ s(t) \propto \int d^3r \rho(\vec{r})e^{i(\varphi(\vec{r}, t))} \]  
Equation 1-22

where \( \rho(\vec{r}) \) is nuclei density, and is proportional to the transverse magnetization, and \( \varphi(\vec{r}, t) \) is the phase of the magnetization vector in the rotating frame.

The phase can be written in terms of the precession frequency in the rotating frame, \( \omega \), using the following equation:

\[ \varphi(\vec{r}, t) = \int \omega(\vec{r}, t) \, dt \]  
Equation 1-23

For a given magnetic field gradient, \( \vec{G}(t) \), \( \omega(\vec{r}, t) \) would be equal to \( \gamma \vec{r} \cdot \vec{G}(t) \); therefore, Equation 1-23 can be re-written as below:

\[ \varphi(\vec{r}, t) = \gamma \vec{r} \cdot \int \vec{G}(t') \, dt' \]  
Equation 1-24

Now let us define a new vector, \( \vec{k} \), as below:

\[ \vec{k}(t) = \gamma \int \vec{G}(t') \, dt' \]  
Equation 1-25

By combining Equation 1-24 and Equation 1-25, the following will be obtained:

\[ \varphi(\vec{r}, t) = \vec{r} \cdot \vec{k} \]  
Equation 1-26

By substituting Equation 1-26 in Equation 1-22, the signal expression can be re-written as follows:

\[ s(\vec{k}) \propto \int d^3r \rho(\vec{r})e^{i(\vec{r} \cdot \vec{k})} \]  
Equation 1-27

Equation 1-27 is a Fourier transform relating nuclei density in an image space, \( \rho(\vec{r}) \), to a detected signal in k space \( s(\vec{k}) \). In MRI, \( s(\vec{k}) \) is sampled at a large area of k-space. By applying an inverse Fourier transform to the signal data, MR images can be reconstructed using the following equation:

\[ \rho(\vec{r}) \propto \int d^3k s(\vec{k})e^{-i(\vec{r} \cdot \vec{k})} \]  
Equation 1-28
1.2 Prostate

1.2.1 Anatomy

Prostate is an exocrine gland of the male reproductive system. The main function of the prostate is to secrete an alkaline fluid which is an important ingredient of semen. A normal human prostate is about the size and shape of a large chestnut, and weights between 17 and 19 grams (12). The lateral (left-right), vertical (superior-inferior), and sagittal (anterior-posterior) dimensions of normal prostate are about 4cm, 3cm, and 2cm, respectively. The prostate looks like a compressed inverted cone, including a base, midgland, and an apex. Its anatomical position is between the vesicle neck of the bladder and the urogenital diaphragm’s apex. It surrounds the prostatic urethra just below the bladder, and is immediately anterior to the rectum (Refer to Figure 1-3).
The prostate gland is usually described by four anatomical zones, which were first proposed by McNeal, in 1981 (13), as follows: peripheral zone, PZ, central zone, CZ, transition zone, TZ, and the anterior fibromuscular stroma, AFMS (See Figure 1-4). The key element in defining these four zones is the urethra. Posterior to the urethra, there is the glandular tissue, and anterior to the urethra there is the fibromuscular area. The transition zone is the region that surrounds the urethra proximal to the ejaculatory ducts. The central zone is the region that
surrounds the ejaculatory ducts and protrudes under the bladder base. The peripheral zone embodies the apical, the posterior and the lateral aspects of the gland. The anterior fibromuscular stroma is prolonged from the bladder neck to the striated urethral sphincter.

Figure 1-4. Zonal anatomy of the prostate as described by McNeal. The transition zone surrounds the urethra proximal to the ejaculatory ducts. The central zone surrounds the ejaculatory ducts and projects under the bladder base. The peripheral zone constitutes the bulk of the apical, posterior, and lateral aspects of the prostate. The anterior fibromuscular stroma extends from the bladder neck to the striated urethral sphincter. (Modified from Brooks JD. Anatomy of the lower urinary tract and male genitalia. In: Walsh PC, Retik AB, Vaughn ED Jr, Wein AJ (eds). Campbell's Urology, 7th Ed. Philadelphia: WB Saunders, 1998; with permission.) The image is obtained from (12), Skandalakis et al, Surgical Anatomy: The Embryologic and Anatomic Basis of Modern Surgery© 2004. Reproduced with the kind permission of McGraw-Hill Education (UK) Ltd. All rights reserved.
With aging, the prostate usually grows larger and most men develop some prostatic diseases. Common diseases of the prostate include: prostatitis (inflammation of the prostate gland), benign prostatic hyperplasia (BPH) (benign enlargement of the prostate gland) and prostate cancer.

### 1.2.2 Prostate Cancer

Prostate cancer is the most common diagnosed cancer among the Canadian men. It was estimated that in 2016, 21,600 new cases will be diagnosed, and 4,000 men will die from this disease in Canada (14). These numbers represent 21%, and 10% of all new cases of cancer and cancer deaths in men, respectively. Estimated Canadian prostate cancer statistics are presented in Table 1-2.

Table 1-2. Estimated Canadian prostate cancer statistics (2016).

Table reproduced from (14), originally obtained from (15).

<table>
<thead>
<tr>
<th>Category</th>
<th>Estimate</th>
</tr>
</thead>
<tbody>
<tr>
<td>New cases</td>
<td>21,600</td>
</tr>
<tr>
<td>Incidence rate (for every 100,000 males)*</td>
<td>114.7</td>
</tr>
<tr>
<td>Deaths</td>
<td>4,000</td>
</tr>
<tr>
<td>Death rate (for every 100,000 males)*</td>
<td>24.2</td>
</tr>
<tr>
<td>5-year net survival (estimates for 2006–2008)</td>
<td>95%</td>
</tr>
</tbody>
</table>

*Age-standardized to the 2011 Canadian Standard Population.
Over 95% of prostate cancers are adenocarcinomas originating from prostatic epithelial cells. Roughly 70-80% of prostate cancers originate from PZ, and 20-25% originate from TZ (16). The CZ of the prostate is rarely accosted with carcinoma, and only about 2.5% of carcinomas originate from this region (17). AFMS is only composed of stromal tissue; therefore, no carcinoma arises from this zone. Prostatic stromal tumours are also very rare (18).

Confirmation of prostate cancer is usually performed by pathological examination of biopsy samples. Histologic grading of prostate cancer is performed using the Gleason Grading system (19). In this system, as shown in Figure 1-5, 5 tumour patterns are defined based on the morphology of the cancerous tissue. Pathologists assign each tumour a primary grade, according to the most common pattern seen in the tumour, and a secondary grade, according to the next most common pattern with prevalence larger than 5%. The summation of two assigned grades is called the Gleason score, which is commonly used for prognostic evaluation of prostate cancer.
Figure 1-5. Histologic patterns of prostatic adenocarcinoma in Gleason grading system.

The image is copied from the original article by Gleason DF (19).

Pattern 1- Cancerous tissue is very similar to normal prostatic tissue, where glands are small, well-formed, and closely packed. Pattern 2- Glands are still well-formed but with larger separation spaces filled with more tissue. Pattern 3- Tissue still has recognizable glands while some gland tissues began to invade the surrounding tissue. Pattern 4- Very limited glands are recognizable, while many gland cells have invaded the surrounding tissue. Pattern 5- Sheets of invaded cells are seen, without recognizable glands.
1.3 Objectives and Overview of this Thesis

Early diagnosis of prostate cancer is a difficult task due to the absence of common symptoms and signs in the early stages of this disease. Meanwhile, similar to most other cancers, if it is diagnosed in the early stages, there are significantly more treatment options and the survival chances increase drastically.

Currently, most prostate cancer cases are first detected by use of Prostate-Specific Antigen (PSA) serum screening or by Digital Rectal Exam (DRE). For the cases with suspected cancer, confirmation and staging of the disease is achieved by use of transrectal ultrasound-guided (TRUS) biopsy. Although the biopsy based techniques are very reliable, they have two unavoidable and major issues: 1) being invasive and 2) not being capable of screening the entire volume of the prostate gland, as only a limited number of samples obtained from a limited area is examined. The second issue may result in discovery of inconsequential tumours in some cases, while missing significant tumours in other cases. To overcome the biopsy shortcomings, a non-invasive imaging technique that can provide pathological information through the entire prostate gland is in high demand. An accurate non-invasive imaging technique can potentially be used for: 1- detection and grading of prostate cancer, 2- monitoring treatment response, and 3- serving as a guidance for more accurate sampling of localized biopsy. Such an imaging technique will significantly decrease the probability of missing malignant tumours, which has a very high possibility when biopsies are performed.

Among the non-invasive imaging techniques, MRI is an excellent candidate for the purpose of detection and evaluation of prostate cancer. This is mainly because: 1- this technique provides high spatial resolution and high contrast in soft tissue, 2- it does not expose patients to ionizing
radiation, and 3- it is versatile and capable of quantifying a large number of tissue properties which can be used for characterization of prostatic lesions.

Using MRI, morphological properties can be obtained from T2 or T1-weighted data. Physiological properties can be investigated through a variety of techniques such as: Diffusion Weighted Imaging (DWI), which is sensitive to water diffusion processes (20, 21), Dynamic Contrast-Enhanced MRI (DCE-MRI), which can provide information about tissue structure and microvasculature properties (22), T2 mapping techniques, which can provide information about tissue composition, or Magnetic Resonance Elastography (MRE), which provides a quantitative assessment of mechanical properties of tissue (23).

While each MRI technique provides valuable information for diagnostic purposes, it has been shown that the highest accuracy is achieved when a combination of MRI techniques, known as Multi-Parametric MRI (MP-MRI) (24), is used. Over the past decade, various MP-MRI techniques have been studied and shown to improve the accuracy of prostate cancer diagnosis (25, 26, 27, 28). MP-MRI protocols typically consist of T2-weighted (T2W) imaging as an anatomical imaging technique, and a combination of at least two of the functional imaging techniques including DCE-MRI, diffusion-weighted MRI (DW-MRI), and MR Spectroscopic Imaging (MRSI).

The current guidelines for evaluation of prostate cancer based on a MP-MRI data (composed of T2W, DCE, and DW-MRI data) are called PI-RADS v2 (Prostate Imaging Reporting and Data System version 2) (29). This reporting system was developed by joint effort of the
European Society of Urogenital Radiology (ESUR), the American College of Radiology (ACR), and the AdMeTech Foundation in 2015. In the PI-RADS v2 reporting system a score on a 5-point scale is assigned to each lesion based on the likelihood that the overall findings from a combination of T2W, DCE, and DW-MRI data correlates with the presence of a clinically significant cancer. Since it was published in 2015, PI-RADS v2 has been validated and used as the main guideline for the evaluation of prostate cancer using MP-MRI in clinical practice and many research studies.

Despite the relative success of MP-MRI in diagnosis of prostatic carcinoma (PCa), similar to any other imaging techniques, it has accuracy limitations and there is still room for further improvement by implementing new technologies, sequences, and novel techniques. The PI-RADS Steering Committee also strongly supports the continued development of novel MRI methodologies for evaluation of prostate cancer by using new and/or advanced research tools that are not included in PI-RADS v2,

The main goal of this research was to develop a novel multi-parametric MRI protocol to increase the accuracy of detection and grading of prostate cancer. This goal was planned to be achieved by fulfilling the following objectives:

1. Development and assessment of a novel protocol for quantitative T₂ mapping in the prostate;
2. Investigating the relationship between the parameters measured from the developed T₂ mapping technique and the corresponding tissue composition in the prostate;
3. Comparing the diagnostic accuracy of the developed T2 mapping technique with the more established MRI techniques. Also, finding the most accurate multi-parametric MRI protocol by selecting the best combination from the investigated techniques.

Additionally, we believed that there is room for improvement in the existing DCE-MRI protocols that are used in prostate studies; therefore, we decided to explore some potential areas of progress. This supplementary goal was achieved by addressing the following objective in this thesis:

4. Investigating the effect of temporal resolution and data analysis technique in the accuracy of DCE-MRI in detection of prostatic tumours.

This rest of this thesis is organized as follows:

In Chapter 2, it is described how the new multi-exponential T2 mapping protocol, Luminal water imaging (LWI), is developed.

In Chapter 3, the correlation between measurements of LWI and the corresponding tissue composition in prostate is investigated through a direct comparison between MRI data and histology images at cellular resolution.

In Chapter 4, the relationship between measurements of LWI and the histopathology of prostatic tissue is investigated by performing a direct comparison between MRI data and
whole-mount histology, and the accuracy of this technique in the detection and grading of prostatic tumours is determined.

In Chapter 5, the diagnostic accuracy of LWI is compared with the standard MP-MRI protocols which include DCE and DW-MRI. In addition, in this chapter the significance of adding LWI into a MP-MRI protocol, consisting of DCE-MRI and DW-MRI, in detection and grading of prostatic tumours is evaluated, and the most accurate MP-MRI protocol is proposed.

In Chapter 6, the impact of temporal resolution on the accuracy of DCE-MRI in the detection of prostatic tumours is investigated.

Chapter 7 provides a summary of the key findings and contributions of this thesis, and also establishes a framework for future work.
Chapter 2: Development of a Multi-Exponential T2 Mapping Protocol for Prostate Imaging

2.1 Introduction

Magnetic resonance (MR) quantitative T2 mapping is an established imaging technique that has been used for non-invasive tracking of histopathologic changes in organs such as brain (30) and cartilage (31). In this technique, a series of T2-weighted (T2W) images is acquired by applying a spin-echo sequence, such as the CPMG sequence. From spin-echo images, a time-dependant signal decay curve can be extracted for each pixel, and consequently, the T2 relaxation time can be determined for each pixel (Figure 2-1).

If the effect of T1 relaxation can be ignored (See the Appendix for details regarding the effect of T1 relaxation on multi-echo signal), in a homogenous spin system signal decay can be described by an exponential decay function with decay constant equal to the inverse of a well-defined T2 relaxation time. This can mathematically be expressed as below:
\[ S(t) = Ae^{-\frac{t}{T_2}} \]  

Equation 2-1

where \( S(t) \) is the magnitude of the signal at a time \( t \), and \( A \) is the amplitude of the signal at time zero (which is proportional to the proton density).

However, in inhomogeneous tissues, where each voxel contains various water compartments, the signal decay curve becomes multi-exponential, with several different \( T_2 \) relaxation times. This is because hydrogen protons of water molecules experience different spin-spin interactions in different environments, and hence represent different \( T_2 \) relaxation times. Therefore, in inhomogeneous tissue one may expect multi-exponential signal decay, which can mathematically be written as follows:

\[ S(t) = \sum_{i=0}^{N} A_i e^{-\frac{t}{T_{2i}}} \]  

Equation 2-2

where \( N \) is the total number of exponential components, and is equal to the number of different water compartments, \( A_i \) is the amplitude of component \( i \), and \( T_{2i} \) is the \( T_2 \) relaxation time of component \( i \). Multiple \( T_2 \) values reflect the underlying composition of the scanned tissue, and hence their measurement can reveal important information for diagnostic purposes.

The prostate is composed of glandular and non-glandular tissue. Non-glandular tissue of prostate, which is less important for diagnostic purposes, is mainly composed of stroma. Glandular tissue of the prostate, where 99% of prostate cancers originate (32), is composed of stromal tissue fused to several ducts and acini (Figure 2-2). These ducts and acini are relatively large in size (up to 0.2 mm in diameter) (33) and consist of luminal space filled with prostatic fluid and lined with two layers of basal epithelial cells (34, 35). In this tissue arrangement, there are two distinctive water environments in a typical voxel (1-10 mm\(^3\)) of an MR image; the larger one being the lumen space, and the much smaller intra-/extra-cellular space of...
epithelial and stromal tissue. Therefore, T\(_2\) decay curves will likely be bi-exponential, with a longer T\(_2\) related to the water inside the lumen and a shorter T\(_2\) to the water within the epithelial cells and stromal tissue.

The importance of multi-exponential T\(_2\) mapping in prostate can be understood by considering the fact that the relative percentage of luminal space in glandular tissue of prostate varies between tumour and normal tissue (36), and between different pathologic grades of cancer. Therefore, measurement of multiple T\(_2\) values and their relative ratio in prostate may potentially provide useful information for diagnostic purposes.

Evidence of bi-exponential T\(_2\) decay in prostate has been shown in previous studies (37, 38, 39, 40). Reported values of T\(_2\) relaxation times of prostatic tissue from a previous study (38) are presented in Table 2-1. Potential contribution of bi-exponential T\(_2\) imaging in diagnosis of prostate cancer has also been investigated recently (41); however, no study with direct correlation between MRI and histology had been conducted by the time we started this study. This study was designed with the purpose of investigating the relationship between multi-exponential T\(_2\) mapping and the histopathology of prostatic tissue through a direct comparison between MRI and whole-mount histology. In our study, a new parameter called luminal water fraction (LWF), which represents the fractional amount of luminal space in prostatic tissue, was defined and used. Due to the differences in tissue composition and fractional amount of luminal space between normal and malignant tissues, we hypothesized that LWF could be used for the detection of prostatic tumours. Additionally, as the fractional amount of luminal space decreases with an increase in GS (See Figure 2-3), we also hypothesized that LWF could contribute to grading of prostate cancer. In summary, this study was designed to investigate
the feasibility of using multi-exponential T2 mapping in the detection and grading of prostatic tumours.

Figure 2-2. A section of normal human prostatic tissue (20×) from peripheral zone, obtained from a whole-mount histology section of a participant in this study.

Figure 2-3. A zoomed-in section (20×) of Hematoxylin and Eosin (H&E)-stained whole-mount histology slide from a) normal tissue in PZ, b) Gleason 3+4 tumour in PZ, and c) Gleason 4+4 tumour in PZ.

It can be observed that with the progression of malignancy, the fractional amount of luminal space decreases.
Table 2-1. Summary of signal fractions and T2 relaxation times and bi-exponential decays.

The table is reproduced from (42).

<table>
<thead>
<tr>
<th></th>
<th>Signal at TE=125 ms</th>
<th>Bi-exp decays: number (total)</th>
<th>Long T2 signal fraction (%) mean/median [range]</th>
<th>T2 (ms): mean/median [range]</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Long component</td>
<td>Short Component</td>
</tr>
<tr>
<td>Peripheral Zone</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Low</td>
<td>11 (14)</td>
<td>26/26 [18-34]</td>
<td>342/325 [201-516]</td>
<td>64/66 [50-80]</td>
</tr>
<tr>
<td>Central Gland</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>All</td>
<td>72 (84)</td>
<td>30/27 [3-80]</td>
<td>546/490 [161-1319]</td>
<td>64/64 [43-92]</td>
</tr>
</tbody>
</table>

In the rest of this chapter, it is described how the imaging protocol for multi-exponential T2 mapping in prostate, named Luminal Water Imaging, has been developed. In Chapter 3, the relationship between measurements of this technique and the histopathology of the prostatic tissue is investigated by performing a direct comparison between MRI data and whole-mount histology, and the accuracy of this technique in the detection and grading of prostatic tumours is determined.

2.2 Protocol Development

It has been previously shown that the sensitivity of T2 mapping techniques is highly dependent on the following scan parameters: TR, TE, number of echoes (NE), and signal to noise ratio
Without using appropriate scan parameters, there will likely be a significant uncertainty in the measurement of $T_2$ values and the relative ratio of each component. For scanning different organs, different sets of TR, TE, NE, and SNR must be determined during protocol development to maximize accuracy of $T_2$ mapping measurements. In this study, the value of TR (3073 ms) was chosen based on previous literature findings (37) such that the $T_1$ contribution was minimized while still maintaining clinically feasible acquisition times. Using this relatively long TR imposed an indirect limitation on the range of SNR (average SNR within prostate gland, on the 1st echo images of a Gradient And Spin Echo (GRASE) sequence measured later during patient study, from 17 cases: 103, 81-125 95% CI). This was because any further improvement in SNR by increasing signal averaging would require a total scan time of greater than 25 minutes. Such a long scan time was avoided as it is not practical in clinical settings. With the selected values for TR and SNR (estimated from previously acquired data), the impact of variations of NE and TE on the accuracy of $T_2$ mappings was investigated during the protocol development in this study. To find the optimum value of these parameters for $T_2$ mapping of prostate gland, we performed several simulations and phantom testing followed by a series of volunteer scans. The details about simulations, phantom scans, and volunteer study are provided in the following sub-sections.

### 2.2.1 Simulations

Simulations were performed using a fictitious two-compartment system described by two $T_2$ relaxation times: $T_{2\text{-short}}$ and $T_{2\text{-long}}$. Simulated MR images were generated for different values of $T_{2\text{-short}}$ and $T_{2\text{-long}}$ using an in-house program developed in Matlab (The Math Works Inc., Natick, MA, USA). The values of $T_{2\text{-short}}$ (25, 50, 75 ms) and $T_{2\text{-long}}$ (200, 400, 800 ms) used in
these simulations were chosen based on previously published T2 values of prostatic tissue in the literature (37, 38). Images of simulated T2 signal, $S_s(t)$, were generated in the form of simple squares with 64×64 pixels using the following equation:

$$S_s(t) = S_0(1 - LT2F) e^{-t/T_{2\text{-short}}} + (S_0 \times LT2F) e^{-t/T_{2\text{-long}}}$$  \text{Equation 2-3}

where $t$ is time, $S_0$ is the maximum signal of the short component, and LT2F is the fractional weighting of the long T2 component.

In the simulations, time was varied between TE and NE×TE with intervals of TE, $S_0$ was set at $10^4$ (in accordance with the signal intensity in our actual scan data). The LT2F was increased from 0 to 0.2 in 64 equally spaced steps throughout the field of view. This range of LT2F was selected by using the lowest ends of the ranges of long T2 fraction observed in the prostate by Storas et. al (38) to mimic the tissues with low luminal content (such as tumours) for which the long T2 component is more likely to be misidentified during the fitting process. For each set of T2-short and T2-long (9 sets in total), images of simulated T2 signal, $S_s(t)$, were generated for different combinations of TE and NE. The range of TE (7-55 ms) and NE (10-64) was chosen based on practical values for experimental settings. Gaussian noise was added to all simulated images to generate an SNR of 100 in similarity with the SNR of our previously acquired data.

Images of simulated T2 signal, $S_s(t)$, were fitted to a multi-exponential decay function using a software written in Matlab which handles the regularized Non-Negative Least Squares (NNLS) (43, 44) algorithm. From fitting, maps of LT2F were generated. More details about the software and fitting procedure are provided in Chapter 3, Data Processing (section 3.6).
From visual inspection on the series of LT2F maps obtained from simulated MR images, two criteria were identified to affect the accuracy of T2 mapping measurements. The first criterion applies a limit on the minimum value of TE×NE based on the value of T2-long. This criterion can be expressed mathematically as below:

\[ NE \times TE \geq 2 \times T_{2-long} \]  \hspace{1cm} \text{Equation 2-4}

Visual inspection on LT2F maps generated for different combinations of TE and NE showed that the relative ratio of long component is identified correctly if the last echo occurs at times bigger than 2×T2-long. To better explain this matter, two example sets of LT2F maps are presented in Figure 2-4: one is generated with identical value of NE and different values of TE, and the other is created with identical value of TE and different values of NE. It can be observed from these images that when the time of the last echo is smaller than 2×T2-long, maps of LT2F become very noisy, indicating a higher calculation error for the fractional amount of one or both T2 components.

Figure 2-4. LT2F maps generated from simulated MR images using T2-short =50 ms, T2-long =200 ms, SNR=100, with variable TE and constant NE=25 (top), and variable NE and constant TE=25 ms (bottom). For the given value of NE (25), the minimum value of TE to satisfy the first criterion is equal to (2×200)/25=16. Similarly, for the given value of TE (25 ms), the minimum value of NE to satisfy the first criterion is equal to 16.
The second criterion applies a limit only on the value of TE, based on the value of $T_{2\text{-short}}$. From visual inspection on the set of LT2F maps generated for different values of TE, and investigation of the $T_2$ distributions obtained from individual decay curves, it was concluded that the value and the relative ratio of $T_{2\text{-short}}$ and $T_{2\text{-long}}$ are identified correctly if TE is smaller than $T_{2\text{-short}}$. This criterion can be mathematically written as below:

$$TE < T_{2\text{-short}}$$  \hspace{1cm} \text{Equation 2-5}

As an example, a set of LT2F maps generated for identical value of NE and various values of TE is represented in Figure 2-5. It can be observed in these images that when the second criterion is not satisfied, the map of LT2F is close to zero for the majority of the pixels, indicating that only one component was identified.

![Figure 2-5: LT2F map generated from simulated MR images using $T_{2\text{-short}} = 50$ ms, $T_{2\text{-long}} = 200$ ms, SNR=100, NE=40. The values of TE that satisfy the second criterion are smaller than 50 ms.](image)

Based on the results of our simulations, it was concluded that the following algorithm must be followed in order to find the optimum values of TE and NE for an accurate multi-exponential $T_2$ mapping in prostate:

1- The ranges of $T_{2\text{-short}}$ and $T_{2\text{-long}}$ in prostatic tissue must be determined from literature reports;
2- The largest value of NE allowed by consideration of practical limitations of scanner and specific absorption rate (SAR) regulations must be determined;

3- Using the selected NE, the smallest value of TE which satisfies the first criterion, described earlier, must be calculated.

4- The last step would be to check whether the TE found in the third step satisfies the second criterion as well. If TE satisfies the second criterion, the set of TE and NE can be used to design a multi-exponential T$_2$ mapping protocol. If TE does not satisfy the second criterion, a reliable T$_2$ mapping protocol may not be possible.

### 2.2.2 Phantom Study

A phantom study was conducted with the purpose of verifying the findings of the simulations. The first step for this task was to find a phantom that resembles prostatic tissue in terms of T$_2$ relaxation properties. The first candidates were meat and celery. They were both scanned in fresh and formalin-fixed forms. Fixation was performed by immersing the sample in 10% formalin for a minimum of one week prior to scanning. MR signals were acquired at a 7T animal scanner (Bruker, Germany) with a quadrature birdcage coil. Axial images were obtained with a multi-echo spin echo sequence (TR=1000-5000 ms, TE=6.8-25 ms, NE=32-80, field of view (FOV)=6×6cm$^2$, voxel-size=0.47×0.47×4mm$^3$, reconstruction matrix-size=128×128, slice thickness=4mm, flip angle=90°, number of averages=2).

Similar to what was explained in the Simulations section, the MR images were fitted to a multi-exponential decay function using a software written in Matlab which handles NNLS algorithm. From fitting, a map of LT2F and also maps of T$_2$-short, T$_2$-long, were generated. More details
about the software and fitting procedure are provided in Chapter 3, the Data Processing (section 3.6).

A bi-exponential $T_2$ decay was not observed in either fresh or formalin-fixed meat. But celery showed a bi-exponential $T_2$ decay both in fresh and formalin-fixed form. The range of $T_2$ relaxation times of formalin-fixed celery was found to be overlapping with the previously reported range of $T_2$ relaxation times for prostatic tissue. In a previous quantitative $T_2$ mapping study of prostate performed by Storas et al. (38) using a 1.5T scanner, the range of short and long $T_2$ components was reported to be: 43-92 (ms) and 161-1319 (ms), respectively, and the range of fraction of long component was reported as: 0.03-0.8. In our study, using formalin-fixed celeries and a 7T scanner, the observed range of $T_{2\text{-short}}$ and $T_{2\text{-long}}$ was: 25-45 (ms) and 100-400 (ms), respectively, and the range of LT2F was: 0.1-0.2. Based on the similarity of $T_2$ relaxation properties, formalin-fixed celery was selected as our phantom in this study to investigate the findings of our simulation study.

Two sets of representative maps of $T_{2\text{-short}}, T_{2\text{-long}},$ and LT2F generated from MR images of formalin-fixed celery with different TE, and identical value of NE are presented in Figure 2-6. From these images, the necessity to satisfy the second criterion, which was described earlier in the simulation section, can be easily observed. Based on the second criterion, $T_{2\text{-short}}$ and $T_{2\text{-long}}$ are identified correctly if TE is smaller than $T_{2\text{-short}}$. In this study the range of $T_{2\text{-short}}$ was 25-45(ms), and based on the second criterion, we set TE to smaller than 45 ms in order to be able to identify both components. From Figure 2-6, it can be observed that for TE=50 ms the short component is entirely missing, which validates the second criterion. Recognition of the first criterion from the images of Figure 2-6 is not as easy as the second criterion. Comparison
between $T_{2\text{-long}}$ maps of different TE shows that as the value of $TE \times NE$ increases, $T_{2\text{-long}}$ becomes less noisy. When the first criterion ($NE \times TE \geq 2 \times T_{2\text{-long}}$) is satisfied (which happens at $TE=25$ for $NE=32$, and at $TE=12.5$ for $NE=64$, considering the maximum $T_{2\text{-long}}=400$ ms), we see that the $T_{2\text{-long}}$ is identified, i.e. it has non-zero value for most of the pixels.

Figure 2-6. $T_{2\text{-short}}$, $T_{2\text{-long}}$, and LT2F map generated from formalin fixed celery for different TE and NE=32 (left), and NE=64 (right).
In summary, results of the phantom study confirmed our simulation finding; therefore, the two criteria obtained by the simulation study were used to design the preliminary protocol for multi-exponential T2 imaging of the prostate.

The preliminary protocol was designed by following the algorithm which was proposed at the end of the Simulations section. Based on the literature values for the T2 relaxation times in prostate (T2-short [43-92 (ms)] and T2-long [161-1319 (ms)], reported by Storas et al. (38)) and taking into account the difference in the scanning system’s magnetic field (1.5 T used by Storas et al. vs. 3 T used in our study), we set the following parameters for the initial multi-exponential T2 mapping protocol: TE= 25 ms, and NE=64.

Prior to finalizing the protocol and using it for examination of participant patients, the proposed protocol was further investigated by scanning a number of healthy volunteers. Details of volunteer study are provided in the next section.

### 2.2.3 Volunteer Study

A volunteer study was performed with the purpose of testing and improving the preliminary protocol that was developed based on the results of the simulation and phantom studies. Five healthy volunteers (mean age, 35.4 years; age range 27-57 years) participated in this study, and provided informed signed consent prior to undergoing MRI examination.

MRI examinations were carried out on a 3T MRI scanner [Philips Medical Systems, Best, The Netherlands]. MR signals were acquired using a pelvic phased-array coil (Medrad, Pittsburgh, PA, USA). To shorten the total scan duration, sensitivity encoding (SENSE), which is a parallel imaging technique, was used in this study. Angled axial images were obtained in the plane...
perpendicular to the rectal wall-prostate interface, with a 3D multi-echo spin echo sequence developed by Jing Zhang at the University of British Columbia MRI Research Centre (45) (TR=3061 ms, TE=25 ms, NE=64, FOV=240×240×[24-40]mm³, voxel-size=[1×1×4 to 2×2×4]mm³, reconstruction matrix-size=240×240, slice thickness=4mm, flip angle=90°, number of averages=1, sense factor=[1.5-3]). To provide anatomical details for comparison with parametric maps of multi-exponential $T_2$ mapping, high spatial resolution T2W MR images were acquired by using a multi-slice Turbo Spin Echo (TSE) sequence (TR=1851 ms, effective TE = 80 ms, NE = 1, FOV=140×140×72 mm³, voxel-size=0.27×0.27×4 mm³, scan matrix-size=288×228, reconstruction matrix-size=512×512, slice thickness=4mm, flip angle=90°, number of averages=3, scan duration=285 sec). T2W images were acquired using the same orientation and slice location as multi-exponential $T_2$ images.

By visual inspection of multi-exponential $T_2$ images, two types of artifacts were identified: a aliasing (wraparound) artifact in the axial direction, and a Moire fringe artifact mainly in the center of images. The aliasing artifact was resolved by increasing the FOV in the axial direction, and Moire fringes were decreased (but not entirely removed) by decreasing the voxel size (increasing matrix size) in the frequency encoding (anterior-posterior) direction (See Figure 2-7).
Figure 2-7. 1st echo T2W images from 10 axial slices of a healthy prostate, with a) voxel size: 2×2×4 mm³, and b) voxel size: 1×2×4 mm³. Comparison between images of series a and b shows that Moire fringes were decreased by decreasing the voxel size in the frequency encoding (anterior-posterior) direction.

The effect of SENSE factor (a parameter in the parallel imaging technique, which is equal to the decrease of the number of phase encoding steps with respect to the full k-space sampling) on SNR was investigated by comparing SNR obtained from 1st echo images for SENSE factor ranging between 1.5 and 3. According to Figure 2-8, results showed an increase in SNR with
a decrease in SENSE factor, indicating a trade-off between scan time and SNR. For the rest of this study, SENSE=1.5 was fixed to maintain a reasonable SNR.

Figure 2-8. SNR as a function of SENSE factor for 10 slices of MR images from a healthy prostate.

Quantitative analysis was performed by fitting MR data to a multi-exponential decay function, as was explained in the Simulations section above. More details about the software and fitting procedure are provided in the sub-section 4.5: Data Processing.

From fittings, maps of $T_2$-short, $T_2$-long, and LWF were generated, and average values of $T_2$-short, $T_2$-long, and LWF were calculated within regions of interest (ROI) in the peripheral zone and central gland (which includes transition zone, central zone, and periurethral region). Representative parametric maps are shown in Figure 2-9. In the peripheral zone, two distinguishable $T_2$ components were observed in almost all pixels. The mean value (averaged over the entire peripheral zone) of $T_2$-short, $T_2$-long, and LWF were $69\pm1$ ms, $360\pm10$ ms, and $0.34\pm0.01$, respectively. In the central gland, some pixels showed two $T_2$ components and some
pixels showed only one T\textsubscript{2} component. The mean value (averaged over the entire central gland) of the T\textsubscript{2-short}, T\textsubscript{2-long}, and LWF were 73.3±0.8 ms, 340±10 ms, and 0.108±0.007, respectively. It should be noted that in the central gland, pixels with only short T\textsubscript{2} component, i.e. pixels with T\textsubscript{2-long} equal to zero, have not been accounted for in the calculation of average T\textsubscript{2-long}. Overall mean value (averaged over the entire gland) of the T\textsubscript{2-short}, T\textsubscript{2-long}, and LWF were 70.5±0.7 ms, 350±10 ms, and 0.236±0.008, respectively. Literature data (46, 47) for the percentages of lumen, epithelium, and stromal tissue, averaged over the entire normal prostate gland; are: 30%±10%, 20%±5%, and 50%±10%, respectively. Therefore, LWF measured by our technique agrees reasonably well with the lumen percentage reported in the literature.

Figure 2-9. A) Axial T2W MR image of the prostate, B) LWF map, C) T\textsubscript{2-short} map, D) T\textsubscript{2-long} map of the same slice. Scale bar of C and D images is in ‘ms’.
2.3 Summary

In summary, in this chapter we developed an initial multi-exponential T$_2$ mapping protocol, for imaging of prostate, based on a series of simulations and phantom studies. We improved and finalized the protocol by performing a volunteer study. We identified two types of artifacts and resolved them by modifying the preliminary protocol. Using the final protocol, we were able to distinguish two T$_2$ components in the T$_2$ distributions obtained from glandular regions of prostate, and the average value of LWF agreed well with the literature data for percentage of luminal space in the prostatic tissue. Based on these results, we finalized the proposed protocol for multi-exponential T2 mapping of the prostate gland, and started to recruit patients to perform the main studies, i.e. 1) investigation of the relationship between the measurements of multi-exponential T2 mapping and composition of prostatic tissue, 2) assessment of diagnostic accuracy of multi-exponential T$_2$ mapping in the detection and grading of prostate cancer, and 3) comparison between the diagnostic accuracy of this technique and the more established MRI techniques. Our patient studies are presented in the following chapters.
Chapter 3: MR measurement of Luminal Water in Prostate Gland: Quantitative Correlation between MRI and Histology

3.1 Introduction

In Chapter 2, it was described how the new multi-exponential $T_2$ mapping protocol was developed. As this technique measures LWF, which was hypothesized to be correlated with the fractional amount of luminal water in the prostatic tissue, we named this technique Luminal Water Imaging (LWI). In this chapter, the relationship between the measurements of LWI and composition of prostatic tissue will be investigated.

It has been shown in the literature that there are differences in the distribution of tissue components between normal prostate and prostate affected by diseases such as PCa and BPH (46, 48, 49). Therefore, identification of the relationships between MR parameters and tissue components, and consequently using MR parameters as a surrogate for measurement of the corresponding tissue components, could be of high importance for disease characterization. Particularly, because the fractional cross sectional area of lumen changes significantly with disease progression in PCa (49), identification of the relationship between LWF and the true fractional amount of luminal space in tissue could potentially contribute to disease assessment and grading of PCa.

This study was conducted to evaluate the correlation between LWF and the percentage area of luminal space in tissue through a direct comparison with histology. It was also aimed at evaluating the correlation between all other MR parameters measured with this technique, and
the percentage area of tissue components, including luminal space, nuclei, and cytoplasm plus stroma.

3.2 Patient Selection

This was an institutional human ethics board approved prospective study. Seventeen patients (median age, 66.4 years; age range 58-80.3 years) were recruited consecutively during January 2015 to January 2016. These were patients referred to the Vancouver General Hospital Urology Clinic and diagnosed with prostate cancer. Additional inclusion criteria included biopsy-proven prostatic adenocarcinoma, no prior treatment, no contraindications to MRI, able to tolerate the endorectal coil and scheduled for radical retropubic prostatectomy. All patients gave informed signed consent prior to undergoing MRI examination shortly before surgery (median 11 days, range 1-41 days). Clinical data for the 17 patients who participated in this study are summarized in Table 3-1.
Table 3-1. Clinical data of 17 patients participating in the study.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Median and range of patient age (y)</td>
<td>66.4 (58-80.3)</td>
</tr>
<tr>
<td>Median and range of pre-op *PSA level (ng/mL)</td>
<td>7.9 (1.4-31.6)</td>
</tr>
<tr>
<td>Pathologic tumour stage</td>
<td></td>
</tr>
<tr>
<td>pT2a</td>
<td>1</td>
</tr>
<tr>
<td>pT2c</td>
<td>4</td>
</tr>
<tr>
<td>pT3a</td>
<td>11</td>
</tr>
<tr>
<td>pT3b</td>
<td>1</td>
</tr>
<tr>
<td>Number of malignant *ROIs</td>
<td></td>
</tr>
<tr>
<td>°GS=(3+3)</td>
<td>18</td>
</tr>
<tr>
<td>°GS=(3+4)</td>
<td>24</td>
</tr>
<tr>
<td>°GS=(4+3)</td>
<td>28</td>
</tr>
<tr>
<td>°GS=(4+4)</td>
<td>11</td>
</tr>
<tr>
<td>°GS=(4+5)</td>
<td>8</td>
</tr>
<tr>
<td>°GS=(5+3)</td>
<td>3</td>
</tr>
<tr>
<td>°GS=(5+4)</td>
<td>1</td>
</tr>
<tr>
<td>°GS=(5+5)</td>
<td>3</td>
</tr>
</tbody>
</table>

*PSA = Prostate-specific antigen, *GS = Gleason score, *ROI = Region of interest

3.3 MRI Protocol

In this thesis all the MRI examinations of patient studies were carried out on a 3T MRI scanner [Philips Medical Systems, Best, The Netherlands], and MR signals were acquired with a combined endorectal/pelvic phased-array coils (Medrad, Pittsburgh, PA, USA). Angled axial images were obtained in the plane perpendicular to the rectal wall-prostate interface, with a 3D multi-echo spin echo sequence developed at the University of British Columbia MRI Research Centre (45) (TR=3073 ms, TE=25 ms, NE=64, FOV=240x240x56mm³, voxel-size=1x1x4mm³, scan matrix-size=240x119, reconstruction matrix-size=240x240, slice
thickness=4mm, flip angle=90°, number of averages=1, sense factor=1.5, scan duration=673 sec). High spatial resolution T₂-weighted MR images were acquired with the same orientation, slice thickness, and slice location, by using a multi-slice Turbo Spin Echo (TSE) sequence (TR=1851 ms, effective TE = 80 ms, NE = 1, FOV=140x140x72mm³, voxel-size=0.3x0.3x4mm³, scan matrix-size=288x228, reconstruction matrix-size=512x512, slice thickness=4mm, flip angle=90°, number of averages=3, scan duration=285 sec). The number and positioning of the slices was set to cover the entire prostate.

In addition to the multi-echo and the high resolution T₂-weighted images, we also acquired DWI and DCE-MRI data, to be used in another study for a direct comparison between the diagnostic accuracy of these techniques. The details of this study are provided in Chapter 5.

3.4 Whole-mount Histology and Registration to MR images

After the surgery, the excised specimens were immersed in 10% formalin for a minimum of 48 hours, and then dissected and examined histopathologically in a uniform and consistent manner. The seminal vesicles and vas deferens were amputated, and the remainder of the specimen was cut perpendicularly to the posterior surface of prostate, into 4 mm transverse slices by using a multi-blade cutting device, developed in-house (50). The entire prostate slices and dissected tissues were sent for histological examination and all slices were stained with hematoxylin and eosin (H & E). Areas of prostate infiltrated by tumour were manually identified and marked by an experienced genitourinary pathologist. All stained slides were then digitalized with the SL801 autoloader and a Leica SCN400 scanning system (Leica Microsystems; Concord, Ontario, Canada) at magnification equivalent to ×40 (Spatial resolution: 0.25 µm/pixel at ×40 magnification). The images were subsequently stored in the
SlidePath digital imaging hub (DIH; Leica Microsystems) of the Vancouver Prostate Centre. Using DIH software, the high resolution histology sections were examined by an experienced pathologist, who outlined the tumours and the border between peripheral zone and central gland and assigned the Gleason scores on each slide.

The digitized whole-mount histology sections were registered to MR images with a software package developed in-house (51) using Matlab. As described previously (51), the multi-step process involved registration of histology sections and high resolution T2W and multi-echo images. In the first step, histology images (as moving images) were registered to their corresponding high resolution T2W images (as fixed targets). The corresponding slices of histology and MR images were determined based on anatomical details. Prior to application of the transformations, contour of prostate was manually outlined on histology and MR images. The initial alignment of the images was performed by an affine transformation involving a 2D multi-resolution algorithm with a b-spline. The required deformation was then applied by another non-rigid b-spline transformation. The measurement of the similarity of the registration was performed by advanced mattes mutual information metric from elastix (52). The precision of registration was evaluated using the Dice Similarity Coefficient (DSC), while the exclusion criteria was set at DSC<0.85. In the second step, multi-echo images were registered to high resolution T2W images (in a similar manner to what was explained for the first step) and the inverse function of this transformation was determined. In the last step, the inverse transformation of step two was applied to the registered histology from step one, which generated the histology images that were registered to multi-echo images. This process resulted in deforming the original manually drawn ROIs, rather than MRI images, and thus the
ROI averages were calculated from the non-deformed parametric maps, ensuring their accuracy.

3.5 Image Segmentation

Identification of tissue components was performed by use of color-based segmentation using DIH software on H&E stained whole-mount histology sections. As depicted in Figure 3-1, pixels were categorized as luminal space, nuclei, or cytoplasm plus stroma, based on a manually defined set of training data for each component. Any pixel that was not categorized in any of the above classes by the software was referred to as unclassified. For each ROI, the total number of pixels identified for each component was counted, and the percentage area for each component was calculated as the ratio of the segmented pixels to all ROI pixels.

Figure 3-1. Representative example of segmentation of the whole mount histological images.

a) A magnified (20×) H&E-stained histology section; b) Tissue components are segmented into cytoplasm plus stroma (blue), lumen (yellow), and nuclei (orange).
3.6 Data Processing

All MR data were processed offline. For each pixel of the multi-echo MR images within prostate, signal decay curves were fitted to a multi-exponential decay function. Fitting was performed with a software package developed in-house which handles NNLS algorithm using Matlab. The software made no prior assumption about the number of contributing $T_2$ components in the multi-exponential decay function, and it decomposed each decay curve into multiple $T_2$ distributions, $S(T_2)$, with distinct peaks, as explained by Bjarnason and Mitchell (43) (see Figure 3-2). The number of the observed peaks for the $T_2$ distributions in this study ranged from one to three. The $T_2$ distributions with three peaks were mostly seen in discrete individual pixels. The third peak in these cases were likely an artifact, potentially due to lower SNR. Regions of continuous pixels with three peaks in $T_2$ distributions were only seen in three out of seventeen cases analyzed in this study. In these cases the 3rd peak was observed within the long $T_2$ relaxation time range. Although the reason for two distinct long $T_2$ components is not entirely clear, it may be related to distinct populations of glands with different sizes. Since the majority of $T_2$ distributions were seen to have two peaks, parameterization of the distributions was performed based on bi-exponential decay modeling with the following parameters: $N_{\text{comp}}$, $gmT_2$, $T_2$-short, $T_2$-long, $A_{\text{short}}$, $A_{\text{long}}$, and LWF. $N_{\text{comp}}$ is the number of peaks in the distribution. $gmT_2$, which is the mean $T_2$ time of the entire distribution on a log scale, was calculated as explained previously (43) by the following equation:

$$gmT_2 = \exp \left[ \frac{\sum_{T_2-\text{max}} S(T_2) \log T_2}{\sum_{T_2-\text{min}} S(T_2)} \right]$$

Equation 3-1

Where $T_2$-min and $T_2$-max were set at 20 ms and 2000 ms respectively.
T2-short and T2-long were defined as the geometric mean T2 of the short and long T2 components, and were calculated as:

\[
T_{2\text{-}\text{short}} = \exp \left[ \frac{\sum_{T_{2\text{-}min}}^{T_{2\text{-}threshold}} S(T_2) \log T_2}{\sum_{T_{2\text{-}min}}^{T_{2\text{-}threshold}} S(T_2)} \right] \quad \text{Equation 3-2}
\]

\[
T_{2\text{-}long} = \exp \left[ \frac{\sum_{T_{2\text{-}threshold}}^{T_{2\text{-}max}} S(T_2) \log T_2}{\sum_{T_{2\text{-}threshold}}^{T_{2\text{-}max}} S(T_2)} \right] \quad \text{Equation 3-3}
\]

where T2-threshold was the cut-off T2 time between the short and long component and was set at 200 ms, based on a graphical analysis of the T2 distributions as follows. The geometric T2 value of the short and long components were determined from bi-exponential decay modeling for 4700 pixels (from four slices, each obtained from a different patient) without setting a cut-off time. The distributions of the short and long T2 components were generated as the number of observation per time bin, and the corresponding histograms were plotted (See Figure 3-3). The distributions were then fitted using Matlab. The best fit curves were double Gaussian functions (f(x) = a1×exp(-((x-b1)/c1)2) + a2×exp(-((x-b2)/c2)2)) with the following parameters: a1 = 59.89, b1 = 121.8, c1= 15.21, a2 = 375.8, b2 = 122.2, c2= 43.29, and a1 = 149, b1 = 597.1, c1= 181.5, a2 = 167.2, b2 = 943.4, c2= 442.5 for T2-short and T2-long respectively. The crossing point of the fits was chosen as the cut-off value between the short and long T2 components.

A_{short} and A_{long}, were defined as areas under the short and long components (See Figure 3-2), and were calculated by summing S(T2) within the region of [T2-min - T2-threshold], and [T2-threshold -T2-max] respectively.; LWF, defined as the ratio of area under the long component over the
total area under the entire distribution, is equivalent to the area function described by Bjarnason and Mitchell (43) and was calculated as:

\[
LWF = \frac{\sum_{T_2 \geq \text{threshold}} S(T_2)}{\sum_{T_2 \geq \text{min}} S(T_2)}
\]

Equation 3-4

For \(T_2\) distributions with only one peak (\(N=1\)), depending on the \(gmT_2\) of the distribution compared to the cut-off \(T_2\), either the pair of \(T_2\)-short and \(A_{\text{short}}\) or \(T_2\)-long and \(A_{\text{long}}\) were set to zero. For cases in which the pair of \(T_2\)-short and \(A_{\text{short}}\) was set to zero, LWF was set to 1; and for cases in which the pair of \(T_2\)-long and \(A_{\text{long}}\) was set to zero, LWF was set to zero. \(T_2\) distributions with three peaks (\(N=3\)) were parameterized bi-exponentially where one peak was considered individually and the other two peaks were averaged geometrically.

Maps of the seven parameters (\(N_{\text{comp}}, gmT_2, T_2\)-short, \(T_2\)-long, \(A_{\text{short}}, A_{\text{long}}, \) and LWF) were generated for every slice. Average values of MR parameters were calculated within a total of 359 (263 non-malignant, and 96 malignant) ROIs manually outlined on the registered whole-mount histology images in malignant PZ and TZ, non-malignant PZ and TZ, normal AFMS, and Periurethral Fibromuscular Stroma (PFMS). Malignant ROIs were selected by accurately outlining the tumour boundaries delineated by pathologist. Non-malignant ROIs were selected by avoiding the tumour boundaries. Since regions of BPH were not outlined by a pathologist, non-malignant ROIs in TZ could consist of BPH.
Figure 3-2. a: 1st echo T2W image of an axial cross section from a middle level of a prostate gland. b: NNLS fitting of the multi-exponential T2 decay curve (left-hand side) generated from the pixel in non-malignant PZ (red marker in a.), and its correspondent T2 distribution (right-hand side). c: NNLS fitting of the multi-exponential T2 decay curve (left-hand side) generated from the pixel in malignant PZ (yellow marker in a.), and its correspondent T2 distribution (right-hand side). d: histology whole-mount section of the same slice.
Figure 3-3. Histogram of $T_{2\text{-short}}$ (blue) and $T_{2\text{-long}}$ (orange) for 4700 pixels. Time-axis is shown in logarithmic scale. The solid lines represent the best fit to the data, described in terms of double Gaussian functions ($f(x) = a_1 \times \exp\left(-\frac{(x-b_1)^2}{c_1^2}\right) + a_2 \times \exp\left(-\frac{(x-b_2)^2}{c_2^2}\right)$) with the following parameters: $a_1 = 59.89$, $b_1 = 121.8$, $c_1 = 15.21$, $a_2 = 375.8$, $b_2 = 122.2$, $c_2 = 43.29$, and $a_1 = 149$, $b_1 = 597.1$, $c_1 = 181.5$, $a_2 = 167.2$, $b_2 = 943.4$, $c_2 = 442.5$ for $T_{2\text{-short}}$ (dark blue) and $T_{2\text{-long}}$ (red) respectively.

3.7 Statistical Analysis

All statistical analyses were performed using MedCalc (MedCalc Software, Mariakerke, Belgium). The Kolmogrov-Smirnov test showed that none of the MR parameters and percentage areas of tissue components were normally distributed. Median and ranges of all MR parameters and percentage areas of the tissue components were calculated, with each data point corresponding to an ROI averaged value. It should be noted that as each median value was calculated from a large number of ROI averaged data points, a fraction value was obtained for $N_{\text{comp}}$, while $N_{\text{comp}}$ calculated for each individual pixel had only an integer value.
Evaluation of the correlation between MRI measurements and histology data was performed by including the whole data set from the entire prostate, including both malignant and non-malignant ROI averaged data points. The correlations between average values of MR parameters and percentage area of tissue components were evaluated with Spearman’s rank correlation test. In all analyses P<0.05 was considered statistically significant. For each of the tissue components, the MR parameter with the highest correlation was identified. Scatter plots of each histology-MRI pair (with strongest correlation) were generated and slopes of the regression lines were calculated.

3.8 Results

Median values and ranges of the percentage area of all tissue components and the MR parameters are summarized in Table 3-2 and Table 3-3, respectively.

Spearman's rank correlation coefficients between MR parameters and tissue components are presented in Table 3-4. Five MR parameters (T2_short, gmT2, A_long, LWF, and N_comp) showed significant correlation with percentage area of luminal space and percentage area of cytoplasm plus stroma. Two MR parameters (T2_short, gmT2) also showed significant correlation with percentage area of nuclei. The overall highest correlation coefficient was obtained between percentage area of luminal space and LWF (Spearman's coefficient: 0.75, P<0.001).

A sample histology section and the corresponding LWF map, presented in Figure 3-4, show good correspondence between LWF and luminal space identified with histology. The highest correlation coefficient between the percentage area of cytoplasm plus stroma and MR parameters was obtained for gmT2 (Spearman's coefficient: -0.64, P<0.001), and for the nuclei
was obtained for T₂-short (Spearman's coefficient: 0.33, P<0.001). Scattered plots of these histology-MR pairs are demonstrated in Figure 3-5.

Table 3-2. Percentage area (median values, with ranges in parentheses) of tissue components in different regions derived from histology.

<table>
<thead>
<tr>
<th>Tissue Component</th>
<th>Non-malignant *PZ</th>
<th>Non-malignant *TZ</th>
<th>Normal *AFMS</th>
<th>Normal *PFMS</th>
<th>Malignant PZ</th>
<th>Malignant TZ</th>
</tr>
</thead>
<tbody>
<tr>
<td>Luminal space</td>
<td>27.2 a, b, c, d</td>
<td>33.9 a, b, c, d</td>
<td>4.8 c, d, e, f</td>
<td>3.7 c, d, e, f</td>
<td>11.6 a, b, c, f</td>
<td>10.8 a, b, c, f</td>
</tr>
<tr>
<td></td>
<td>(13.7-65.4)</td>
<td>(7.2-64.0)</td>
<td>(1.1-21.5)</td>
<td>(0.7-15.6)</td>
<td>(2.8-31.1)</td>
<td>(2.4-26.9)</td>
</tr>
<tr>
<td>Cytoplasm plus</td>
<td>42.9 a, b</td>
<td>40.7 a, b, c</td>
<td>78.0 c, d, e, f</td>
<td>68.3 c, d, e, f</td>
<td>49.0 a, b, f</td>
<td>47.4 a, b</td>
</tr>
<tr>
<td>stroma</td>
<td>(5.9-63.2)</td>
<td>(7.7-63.6)</td>
<td>(34.3-86.3)</td>
<td>(50.8-81.6)</td>
<td>(21.6-63.2)</td>
<td>(23.1-67.2)</td>
</tr>
<tr>
<td>Nuclei</td>
<td>7.5 a, b, c, d</td>
<td>7.3 a, b, c, d</td>
<td>2.4 c, d, e, f</td>
<td>4.1 c, d, e, f</td>
<td>11.5 a, b, c, f</td>
<td>12.0 a, b, c, f</td>
</tr>
<tr>
<td></td>
<td>(2.3-28.9)</td>
<td>(2.4-18.3)</td>
<td>(1.4-7.6)</td>
<td>(1.5-20.6)</td>
<td>(3.8-38.4)</td>
<td>(6.4-38.4)</td>
</tr>
</tbody>
</table>


a Significantly different than normal AFMS (P<0.01). b Significantly different than normal PFMS (P<0.01).

b Significantly different than malignant PZ (P<0.01). d Significantly different than malignant TZ (P<0.01).

c Significantly different than non-malignant PZ (P<0.01). f Significantly different than non-malignant TZ (P<0.01).
Table 3-3. Average values (median values, with ranges in parentheses) of LWI parameters in malignant and normal tissue in different regions.

<table>
<thead>
<tr>
<th>MR Measurement</th>
<th>Non-malignant *PZ</th>
<th>Non-malignant *TZ</th>
<th>Normal *AFMS</th>
<th>Normal *PFMS</th>
<th>Malignant PZ</th>
<th>Malignant TZ</th>
</tr>
</thead>
<tbody>
<tr>
<td>T2_short (ms)</td>
<td>75 a, b, c</td>
<td>83 a, b, c</td>
<td>64 c, d, e, f</td>
<td>69 c, f</td>
<td>72 a, e, f</td>
<td>83 a</td>
</tr>
<tr>
<td></td>
<td>(50-152)</td>
<td>(49-176)</td>
<td>(32-80)</td>
<td>(40-111)</td>
<td>(25-116)</td>
<td>(36-108)</td>
</tr>
<tr>
<td>T2_long (ms)</td>
<td>503 a, b, d</td>
<td>551 b, d</td>
<td>617 c, e</td>
<td>726 c, e, f</td>
<td>507 a, b, d</td>
<td>722 c, e, f</td>
</tr>
<tr>
<td></td>
<td>(249-904)</td>
<td>(283-948)</td>
<td>(348-1074)</td>
<td>(306-1300)</td>
<td>(290-923)</td>
<td>(249-1135)</td>
</tr>
<tr>
<td>gmT2 (ms)</td>
<td>114 a, b, c, d</td>
<td>107 a, b, c, d</td>
<td>66 c, d, e, f</td>
<td>74 c, f</td>
<td>84 a, c, f</td>
<td>89 a, e, f</td>
</tr>
<tr>
<td></td>
<td>(58-295)</td>
<td>(57-237)</td>
<td>(36-86)</td>
<td>(41-120)</td>
<td>(30-139)</td>
<td>(41-120)</td>
</tr>
<tr>
<td>LWF</td>
<td>0.20 a, b, c, d, f</td>
<td>0.14 a, b, c, d, e</td>
<td>0.03 c, e, f</td>
<td>0.03 c, e, f</td>
<td>0.07 a, b, d, e, f</td>
<td>0.04 c, e, f</td>
</tr>
<tr>
<td></td>
<td>(0.03-0.50)</td>
<td>(0.02-0.40)</td>
<td>(0.02-0.11)</td>
<td>(0.00-0.13)</td>
<td>(0.01-0.18)</td>
<td>(0.00-0.12)</td>
</tr>
<tr>
<td>A_short (×10^5)</td>
<td>159 a, c</td>
<td>164 a, c</td>
<td>138 c, d, e, f</td>
<td>162 c</td>
<td>185 a, b, c, f</td>
<td>170 a</td>
</tr>
<tr>
<td></td>
<td>(98-290)</td>
<td>(109-268)</td>
<td>(84-198)</td>
<td>(111-211)</td>
<td>(64-300)</td>
<td>(96-219)</td>
</tr>
<tr>
<td>A_long (×10^5)</td>
<td>39.2 a, b, c, d, f</td>
<td>28.2 a, b, c, d, e</td>
<td>4.3 c, e, f</td>
<td>5.8 c, e, f</td>
<td>14.1 a, b, d, e, f</td>
<td>6.7 c, e, f</td>
</tr>
<tr>
<td></td>
<td>(8.0-116.0)</td>
<td>(3.5-112.7)</td>
<td>(2.4-14.6)</td>
<td>(0.2-26.1)</td>
<td>(1.9-59.8)</td>
<td>(0.1-22.0)</td>
</tr>
<tr>
<td>N_comp</td>
<td>2.00 a, b, c, d, f</td>
<td>1.90 a, b, c, d, e</td>
<td>1.55 c, e, f</td>
<td>1.44 c, e, f</td>
<td>1.82 a, b, e, f</td>
<td>1.70 c, e, f</td>
</tr>
<tr>
<td></td>
<td>(1.22-2.47)</td>
<td>(1.27-2.61)</td>
<td>(1.03-2.02)</td>
<td>(1.01-2.09)</td>
<td>(1.15-2.22)</td>
<td>(1.08-2.13)</td>
</tr>
</tbody>
</table>


a Significantly different than normal AFMS (P<0.01). b Significantly different than normal PFMS (P<0.01).

c Significantly different than malignant PZ (P<0.01). d Significantly different than malignant TZ (P<0.01).

e Significantly different than non-malignant PZ (P<0.01). f Significantly different than non-malignant TZ (P<0.01).
Table 3-4. Spearman's rank correlation coefficient (ρ), with 95% confidence intervals in parentheses, between MR measurements and the percentage area of tissue components.

<table>
<thead>
<tr>
<th>MR Measurement</th>
<th>Luminal Space</th>
<th>Cytoplasm plus stroma</th>
<th>Nuclei</th>
</tr>
</thead>
<tbody>
<tr>
<td>T2-short</td>
<td>0.42 (0.33 to 0.50)</td>
<td>-0.50 (-0.57 to -0.42)</td>
<td>0.33 (0.23 to 0.42)</td>
</tr>
<tr>
<td></td>
<td>P&lt;0.001</td>
<td>P&lt;0.001</td>
<td>P&lt;0.001</td>
</tr>
<tr>
<td>T2-long</td>
<td>-0.13 (-0.23 to -0.03)</td>
<td>0.03 (-0.07 to 0.13)</td>
<td>0.06 (-0.05 to 0.16)</td>
</tr>
<tr>
<td></td>
<td>P=0.01</td>
<td>P=0.58</td>
<td>P=0.27</td>
</tr>
<tr>
<td>gmT2</td>
<td>0.69 (0.64 to 0.74)</td>
<td>-0.64 (-0.70 to -0.58)</td>
<td>0.26 (0.16 to 0.35)</td>
</tr>
<tr>
<td></td>
<td>P&lt;0.001</td>
<td>P&lt;0.001</td>
<td>P&lt;0.001</td>
</tr>
<tr>
<td>LWF</td>
<td>0.75 (0.70 to 0.79)</td>
<td>-0.52 (-0.59 to -0.44)</td>
<td>0.02 (-0.09 to 0.12)</td>
</tr>
<tr>
<td></td>
<td>P&lt;0.001</td>
<td>P&lt;0.001</td>
<td>P=0.77</td>
</tr>
<tr>
<td>A-short</td>
<td>-0.11 (-0.26 to -0.01)</td>
<td>-0.04 (-0.14 to 0.06)</td>
<td>0.13 (0.02 to 0.23)</td>
</tr>
<tr>
<td></td>
<td>P=0.03</td>
<td>P=0.44</td>
<td>P=0.02</td>
</tr>
<tr>
<td>A-long</td>
<td>0.71 (0.65 to 0.76)</td>
<td>-0.51 (-0.58 to -0.43)</td>
<td>0.04 (-0.07 to 0.14)</td>
</tr>
<tr>
<td></td>
<td>P&lt;0.001</td>
<td>P&lt;0.001</td>
<td>P=0.47</td>
</tr>
<tr>
<td>N-comp</td>
<td>0.43 (0.34 to 0.51)</td>
<td>-0.26 (-0.35 to -0.16)</td>
<td>0.03 (-0.07 to 0.13)</td>
</tr>
<tr>
<td></td>
<td>P&lt;0.001</td>
<td>P&lt;0.001</td>
<td>P=0.58</td>
</tr>
</tbody>
</table>
Figure 3-4. Sample H&E-stained whole-mount histology sections (a,b), with the corresponding LWF map (c), and high resolution T2W image of the same slice (d). In figure (a), color-based segmentation algorithm was applied only for identification of luminal space (shown in green). Notice good correspondence between LWF values (c) and the amount of luminal space (a).
Figure 3-5. Regression results and scatter plots of: a) LWF versus percentage area of luminal space (regression line: $y = 0.45x + 0.03$, $P<0.001$), b) $gmT_2$ versus percentage area of cytoplasm plus stroma (regression line: $y = -0.2x + 0.19$, $P<0.001$), and c) $T_{2\text{-short}}$ versus percentage area of nuclei (regression line: $y = 0.1x + 0.079$, $P<0.001$). Each data point on graphs represents average value of MR parameter and corresponding percentage area value for a chosen ROI.
3.9 Discussion and Conclusion

In this study, we investigated the relationship between MR parameters of LWI and the underlying composition of prostatic tissue through a direct comparison with histology. We were particularly interested to verify and establish if there is any relationship between LWF and percentage area of lumen in prostatic tissue. The median values of $T_2$-short, $T_2$-long, and LWF (75, 503, 0.2 in PZ and 83, 551, 0.14 in TZ) measured in normal tissue in this study are comparable to the values of the corresponding parameters previously measured with bi-exponential analyses by Storås et. al (38). In their study, three median values, corresponding to high, intermediate, and low intensity, were reported for each MR parameter in peripheral zone and central gland. Reported median values in their study are as follows: short $T_2$: 73, 69, 66 (ms), long $T_2$: 837, 470, 325 (ms), long $T_2$ signal fraction (%): 54, 29, 26 in peripheral zone, and short $T_2$: 64, 61, 61 (ms), long $T_2$: 502, 450, 456 (ms), long $T_2$ signal fraction (%): 28, 21, 12 in central gland. The median and range of LWF values in the entire prostate measured in this study are in good agreement with the previously published values for percentage of lumen in the composition of normal prostatic tissue (48). The median value and range of percentage area of luminal space measured in normal and malignant tissues in PZ are also in good agreement with previously published data (48). However, median values of percentage area of cytoplasm plus stroma and nuclei measured in this study are lower than published values (48) both in normal PZ (42.9, and 7.5 vs. 54.9, and 14.5) and in malignant PZ (49.0, and 11.5 vs. 61.1, and 23.7). This underestimation could be due to inaccuracy of the segmentation algorithm, since in the calculation of percentage areas for each ROI, an average of 23% of pixels were unclassified. Because luminal space has a simpler color-based pattern (mostly plain white areas in the histology images) in comparison to the rest of the tissue, it is safe to assume
that minimum error had occurred in the calculation of luminal space percentage, and that most of the unclassified pixels belonged to cytoplasm plus stroma or nuclei.

Our correlation analyses showed that LWF is positively and significantly related to the percentage area of luminal space. With the exception of fibromuscular stroma, such strong correlation between LWF and luminal space was present in all types of tissue (data not shown).

The slope of the regression line obtained from a plot of LWF vs. fractional amount of luminal space was equal to 0.45, rather than 1. This observation implies that LWF, while proportional to the fractional volume of luminal space, is not identical to this histological measure. Several factors could have potentially contributed to this discrepancy between LWF and the fractional volume of luminal space measured with histology. Firstly, LWF measured in this study is likely to be weighted by proton density and $T_1$ values (See the Appendix for details regarding the effect of $T_1$ relaxation on the LWF measurements). In addition, we compared the 2D percentage area of lumen to the 3D fractional volume of the luminal water. Since the glands are ellipsoids, rather than elliptical cylinders aligned in the axial direction, the area of lumen is not linearly proportional to its volume. Simple comparison between the volumes of ellipsoids and elliptical cylinders suggests that the 2D percentage area of luminal space, measured with histology, overestimates the luminal volume by as much as 33%. In reality, this effect is expected to be less than 33%, taking into account that the glands are not perfect ellipsoids and are not necessarily aligned in the axial direction. Another contributing factor to be considered is morphological differences between in vivo prostate, imaged with MRI, and excised, fixed prostate, analysed with histology. However, this could make justification of the observed discrepancy even harder. That is because histological processing involves heat-induced
dehydration during which the water content is removed. Water-filled cavities are therefore more prone to shrinkage than stromal tissues. Consequently, tissue shrinkage may be non-uniform (53, 54, 55). Therefore, luminal space and the luminal water fraction may be smaller in whole-mount histology than in vivo. However, it is worth noting that the slopes of the regression lines varied very little between different types of tissue (data not shown), suggesting that the tissue composition is likely not the reason for the LWF underestimating the luminal space. Presumably a systematic error in the histology segmentation procedure resulting in an overall overestimation of percentage area of luminal space might have also been a contributing factor. Finally, water exchange between different compartments within the prostatic tissue (e.g. intra-/extra- cellular vs. luminal water) might have resulted in LWF underestimating the volume of luminal space. Determination of the true relationship between LWF and the amount of lumen in prostatic tissue is not trivial and requires further investigations.

The main limitation of this study was related to the segmentation procedure. The software that has been used in this study was not capable of differentiating loose stroma from the epithelial cytoplasm. Therefore, stroma and epithelial cytoplasm were combined into one tissue component in this study. However, according to Langer et al. (48), in cancer there is an increase in percentage area of cytoplasm, and hence in our study measuring the percentage of cytoplasm separate from stroma would be very informative and useful. In order to investigate the relationship between MR measurements of LWI and percentage area of cytoplasm more accurately, a prospective study with a more accurate segmentation algorithm, or better staining techniques, and higher number of patients is required. Another limitation to be noted here is related to the scan time. The relatively long scan time (11 min) of this technique increases the
chance of patient movement during the scan. This may result in a mismatch of data in signal decay curves and consequently the uncertainty of the data fitting procedure can be increased. Therefore, a potential future work is to reduce the total scan time of this technique.

The results of this pilot study demonstrate that LWF measured with MRI is significantly correlated with the percentage area of luminal space in the prostatic tissue. This is an important finding, considering that in PCa the amount of luminal space correlates strongly with Gleason grade, which is a key prognostic marker. With higher Gleason grade, the glands first coalesce and then disappear in solid sheets of cells (55). Therefore, LWF could potentially differentiate the aggressive forms of PCa from the more biologically benign.

In conclusion, the methodology established for measuring LWF can potentially be used as a surrogate for measurement of fractional amount of lumen in diagnosing various prostatic diseases in which the amounts of lumen differ between normal and abnormal tissues.
Chapter 4: Luminal Water Imaging: A New MRI T2 Mapping Technique for Prostate Cancer Diagnosis

4.1 Introduction

In Chapter 3, the correlation between measurements of LWI and the corresponding tissue composition in prostate was investigated, and our hypothesis that LWF represents the fractional amount of MR signal generated by water content of luminal spaces in prostate was verified. In this chapter, the relationship between the measurements of LWI and the histopathology of prostatic tissue is investigated. The diagnostic accuracy of this technique in the detection and grading of prostatic tumours is determined by direct comparisons between MRI data and whole-mount histology.

4.2 Patient Selection

The patient selection process was similar to what was explained for the previous study described in Chapter 3. Clinical data for the 18 patients who participated in this study are summarized in Table 4-1. A portion of the patient population used in this study has been previously reported in Chapter 3.
Table 4-1. Clinical data of 18 patients participating in the study.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age (y)</th>
<th>PSA level (ng/mL)</th>
<th>Prostate volume (cc)</th>
<th>Lesion characteristics</th>
<th>Pathologic tumour stage</th>
</tr>
</thead>
<tbody>
<tr>
<td>P01</td>
<td>65</td>
<td>13.1</td>
<td>32.3</td>
<td>Size (cc): 5.8</td>
<td>4+3</td>
</tr>
<tr>
<td>P02</td>
<td>65</td>
<td>4.6</td>
<td>67.5</td>
<td>0.1 3+4</td>
<td>PZ</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.1 3+3</td>
<td>PZ</td>
</tr>
<tr>
<td>P03</td>
<td>67</td>
<td>8</td>
<td>94.7</td>
<td>0.7 3+3</td>
<td>PZ</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.2 4+3</td>
<td>PZ</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.1 3+3</td>
<td>PZ</td>
</tr>
<tr>
<td>P07</td>
<td>75</td>
<td>18.2</td>
<td>41.1</td>
<td>3.3 4+3</td>
<td>PZ</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1.5 3+3</td>
<td>PZ</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.3 3+3</td>
<td>PZ</td>
</tr>
<tr>
<td>P10</td>
<td>69</td>
<td>18.4</td>
<td>39.5</td>
<td>6.2 4+5</td>
<td>PZ</td>
</tr>
<tr>
<td>P11</td>
<td>66</td>
<td>7.4</td>
<td>32.5</td>
<td>1.0 3+4</td>
<td>PZ</td>
</tr>
<tr>
<td>P12</td>
<td>67</td>
<td>6</td>
<td>17.7</td>
<td>1.3 3+4</td>
<td>PZ</td>
</tr>
<tr>
<td>P13</td>
<td>58</td>
<td>6.7</td>
<td>31.4</td>
<td>6.1 3+4</td>
<td>PZ</td>
</tr>
<tr>
<td>P14</td>
<td>58</td>
<td>12</td>
<td>20.2</td>
<td>3.9 3+4</td>
<td>PZ</td>
</tr>
<tr>
<td>P15</td>
<td>66</td>
<td>31.6</td>
<td>44.9</td>
<td>6.6 4+3</td>
<td>PZ</td>
</tr>
<tr>
<td>P16</td>
<td>75</td>
<td>8.5</td>
<td>36.6</td>
<td>3.2 4+5</td>
<td>PZ</td>
</tr>
<tr>
<td>P17</td>
<td>63</td>
<td>51</td>
<td>26.4</td>
<td>9.5 4+5</td>
<td>PZ</td>
</tr>
<tr>
<td>P18</td>
<td>62</td>
<td>13.3</td>
<td>33.3</td>
<td>2.1 4+4</td>
<td>PZ</td>
</tr>
</tbody>
</table>
4.3 MRI Protocol

Imaging protocols were similar to what was explained in the previous study. Multi-echo data were acquired using a 3D multi-echo spin echo pulse sequence and high resolution T2W images were acquired using a multi-slice TSE sequence. More details about imaging protocols can be found in Chapter 3, MRI protocol.

4.4 Histologic Examination and Registration to MR Images

Following prostatectomy, the excised specimens were immersed in 10% buffered formalin for a minimum of 48 hours. The formalin-fixed specimens were dissected and examined histopathologically in a uniform and consistent manner. The external surfaces were inked and the seminal vesicles and vas deferens amputated. The apical and bladder neck tissue were each removed as 0.5 cm thick tissue doughnuts. The complete prostate gland was then cut perpendicularly to the posterior surface of the prostate into 4 mm transverse slices with a multi-blade cutting device as explained before (section 3.4). These Hematoxylin and Eosin (H&E) stained whole-mount slides were histologically examined by a genitourinary pathologist, who outlined the tumours and assigned the Gleason scores on each slide under the light microscope. The whole-mount histology sections, digitized using a flatbed scanner, were registered to MR images with a software package developed in-house as explained in the previous Chapter (section 3.4). Tumour ROIs were manually drawn on the registered histology images, by accurately overlapping the tumour outlines drawn by the pathologist on the non-deformed histology sections. Non-malignant ROIs were manually drawn on the registered histology images while avoiding the tumour outlines.
4.5 Data Processing

All MR data were processed offline. Maps of the seven LWI parameters: $N_{comp}$, $gmT_2$, $T_{2\text{-short}}$, $T_{2\text{-long}}$, $A_{\text{short}}$, $A_{\text{long}}$, and LWF were generated for all slices across the entire prostate similarly to what was explained for the previous study described in Chapter 3. Average values of MR parameters were calculated within a total of 378 (226 non-malignant, and 152 malignant) ROIs manually outlined on the registered whole-mount histology images. ROIs were defined in six types of tissue: malignant PZ and TZ, non-malignant PZ and TZ, AFMS, and PFMS.

4.6 Statistical Analysis

Statistical analyses were performed using MedCalc and R packages (56, 57, 58). None of the MR parameters were normally distributed, as confirmed by the Kolmogorov-Smirnov test. Significant differences between malignant and non-malignant tissues were determined with paired t-test. In order to account for the correlations of MR parameters within each patient, the values of MR parameters for each type of tissue were averaged for each patient prior to the application of the paired t-test. Correlations between MR parameters and GS were evaluated by averaging Spearman’s correlation coefficients calculated for individual patients. It should be noted that the calculation of correlation coefficients for each individual patient was based on the ROIs defined in separate slices, and the ranges of Gleason scores were sufficient to calculate correlation coefficients in 17 out of 18 patients. Significance of the correlation between MR parameters and GS was evaluated with the application of a one sample t-test. $P<0.05$ was considered statistically significant. Area under the receiver operating characteristic (ROC) Curve (AUC) was calculated by ROC analysis (59) of individual and combined MR
parameters, performed in PZ, TZ, and the entire prostate. ROC analyses were performed based on logistic generalized linear mixed effect models (GLMMs) with the dependant (output) variable being malignant vs non-malignant (1 vs 0). Correlation within each patient was incorporated by having a random intercept in the model with the grouping factor for the random intercept being each patient. Multi-parametric ROC analyses were performed using logistic GLMM. In order to prevent data overfitting, only parameters that contributed significantly (P<0.05) to the model, and also minimized Akaike information criterion (AIC) and Bayesian information criterion (BIC), were included in the logistic GLMM.

### 4.7 Results

Representative maps of MR parameters are shown in Figure 4-1, and their mean values and ranges are provided in Table 4-2. A Box and Whisker plot of the average values of LWF versus different Gleason patterns in PZ is shown in Figure 4-2.

Results of the paired t-test indicate that the average values of gmT2, A\textsubscript{short}, A\textsubscript{long}, and LWF are significantly different between malignant and non-malignant tissue in the entire prostate. Results of paired t-test in PZ showed that all MRI parameters except for T2\textsubscript{long} have significantly different average values between malignant and non-malignant tissue. Paired t-test could not be conducted in TZ due to the insufficient number of patients with tumours in TZ.

Average Spearman's rank correlation coefficients between MR parameters and GS, and the results of t-test in PZ are presented in Table 4-3. T-test could not be conducted in TZ due to
the insufficient number of patients with tumours in TZ. LWF showed the strongest correlation with Spearman's coefficient values of -0.78 ± 0.01.

The values of AUCs calculated from ROC analysis are summarized in Table 4-4. LWF had the highest AUC in PZ (0.97), TZ (0.98), and the A_short had the highest AUC in the entire gland (0.82).

Multi-parametric ROC analysis in PZ showed slightly increased AUC (0.98 vs. 0.97), with only gmT2 and T2_short contributing significantly to the logistic regression model (Regression coefficients: -9±2 (P<0.0001), 6±1 (P<0.0001), respectively). Multi-parametric ROC analysis could not be performed in TZ, since the only parameter that contributed significantly to the model was LWF. In the entire prostate, multi-parametric ROC analysis again showed increased AUC (0.86 vs. 0.82), with gmT2, T2_short, A_short, and N_comp contributing significantly to the model (Regression coefficients: -5.4±0.9 (P<0.0001), 4.4±0.7 (P=0.0002), 0.7±0.2 (P=0.002), 0.9±0.2 (P=0.0001), respectively).
Figure 4-1. Representative maps of MR parameters, $T_2$-weighted image, and histology whole-mount section of the same slice. Scale bar of $gmT_2$, $T_2$-short, and $T_2$-long images are in ‘s’. Zero pixels on the $T_2$-long map indicate mono-exponential decay.
Figure 4-2. Box and Whisker plot of the average values of LWF versus different Gleason patterns in PZ.
Table 4-2. Mean value, standard deviation, and range of MR measurements in malignant and non-malignant tissues of different regions.

<table>
<thead>
<tr>
<th>Location</th>
<th>( T_2)-short (ms)</th>
<th>( T_2)-long (ms)</th>
<th>( T_2) ( a ) ( \text{gm} ) (ms)</th>
<th>( A_{\text{short}} ) ( \times 10^5 )</th>
<th>( A_{\text{long}} ) ( \times 10^5 )</th>
<th>( N_{\text{comp}} )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-malignant PZ</td>
<td>Mean</td>
<td>90±26</td>
<td>545±115</td>
<td>138±46</td>
<td>0.24±0.09</td>
<td>165±51</td>
</tr>
<tr>
<td></td>
<td>Range</td>
<td>[52-150]</td>
<td>[300-887]</td>
<td>[68-298]</td>
<td>[0.09-0.45]</td>
<td>[90-350]</td>
</tr>
<tr>
<td>Malignant PZ</td>
<td>Mean</td>
<td>81±21</td>
<td>548±188</td>
<td>94±27</td>
<td>0.10±0.05</td>
<td>188±46</td>
</tr>
<tr>
<td></td>
<td>Range</td>
<td>[44-157]</td>
<td>[207-1408]</td>
<td>[46-187]</td>
<td>[0-0.3]</td>
<td>[89-303]</td>
</tr>
<tr>
<td>Non-malignant TZ</td>
<td>Mean</td>
<td>104±38</td>
<td>548±183</td>
<td>138±49</td>
<td>0.20±0.08</td>
<td>168±40</td>
</tr>
<tr>
<td></td>
<td>Range</td>
<td>[56-202]</td>
<td>[281-914]</td>
<td>[65-276]</td>
<td>[0.06-0.43]</td>
<td>[104-250]</td>
</tr>
<tr>
<td>Malignant TZ</td>
<td>Mean</td>
<td>90±22</td>
<td>609±270</td>
<td>101±30</td>
<td>0.08±0.05</td>
<td>165±36</td>
</tr>
<tr>
<td></td>
<td>Range</td>
<td>[37-124]</td>
<td>[372-1035]</td>
<td>[39-158]</td>
<td>[0-0.15]</td>
<td>[118-292]</td>
</tr>
<tr>
<td>Normal AFMS</td>
<td>Mean</td>
<td>57±12</td>
<td>639±281</td>
<td>58±14</td>
<td>0.06±0.05</td>
<td>154±30</td>
</tr>
<tr>
<td></td>
<td>Range</td>
<td>[31-84]</td>
<td>[219-1323]</td>
<td>[28-97]</td>
<td>[0-0.21]</td>
<td>[95-262]</td>
</tr>
<tr>
<td>Normal PFMS</td>
<td>Mean</td>
<td>75±22</td>
<td>639±271</td>
<td>77±22</td>
<td>0.04±0.03</td>
<td>165±31</td>
</tr>
<tr>
<td></td>
<td>Range</td>
<td>[37-115]</td>
<td>[258-1078]</td>
<td>[38-116]</td>
<td>[0-0.11]</td>
<td>[107-228]</td>
</tr>
<tr>
<td>Entire prostate</td>
<td>Mean</td>
<td>84±28</td>
<td>569±204</td>
<td>106±45</td>
<td>0.14±0.10</td>
<td>172±46</td>
</tr>
<tr>
<td></td>
<td>Range</td>
<td>[31-202]</td>
<td>[207-1408]</td>
<td>[28-298]</td>
<td>[0-0.45]</td>
<td>[89-350]</td>
</tr>
</tbody>
</table>

\( gm T_2 \): geometric mean \( T_2 \), \( LWF \): luminal water fraction, \( PZ \): peripheral zone, \( TZ \): transition zone, \( AFMS \): anterior fibromuscular stroma, \( PFMS \): periurethral fibromuscular stroma.

Malignant PZ significantly different than non-malignant PZ (P<0.05) based on paired t-test. Paired t-test could not be conducted in TZ due to the insufficient number of patients with tumours in TZ.
Table 4-3. Statistics of correlations between MRI multi-exponential decay parameters and Gleason score in PZ; data from 17 patients were included.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>T$_2$-short</th>
<th>T$_2$-long</th>
<th>gmT$_2$</th>
<th>LWF</th>
<th>A$_{short}$</th>
<th>A$_{long}$</th>
<th>N$_{comp}$</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>PZ</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Average ρ*</td>
<td>-0.34±0.49</td>
<td>0.17±0.45</td>
<td>-0.66±0.34</td>
<td>-0.78±0.11</td>
<td>0.38±0.48</td>
<td>-0.76±0.11</td>
<td>-0.56±0.30</td>
</tr>
<tr>
<td>± standard deviation</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>95% Confidence Interval</td>
<td>-0.596 to</td>
<td>-0.065 to</td>
<td>-0.832 to</td>
<td>-0.834 to</td>
<td>0.133 to</td>
<td>-0.814 to</td>
<td>-0.716 to</td>
</tr>
<tr>
<td>for ρ</td>
<td>-0.093</td>
<td>0.399</td>
<td>-0.485</td>
<td>-0.726</td>
<td>0.628</td>
<td>-0.697</td>
<td>-0.408</td>
</tr>
<tr>
<td>Significance level</td>
<td>P = 0.010</td>
<td>P = 0.146</td>
<td>P &lt; 0.0001</td>
<td>P &lt; 0.0001</td>
<td>P = 0.005</td>
<td>P &lt; 0.0001</td>
<td>P &lt; 0.0001</td>
</tr>
<tr>
<td>Test statistic t</td>
<td>2.902</td>
<td>1.525</td>
<td>8.046</td>
<td>30.500</td>
<td>3.263</td>
<td>27.349</td>
<td>7.736</td>
</tr>
<tr>
<td>ρ*: Spearman’s coefficient of rank correlation</td>
<td></td>
<td></td>
<td></td>
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<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 4-4. ROC analysis measures for MRI parameters in PZ, TZ, and entire prostate.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>T$_2$-short</th>
<th>T$_2$-long</th>
<th>gmT$_2$</th>
<th>LWF</th>
<th>A$_{short}$</th>
<th>A$_{long}$</th>
<th>N$_{comp}$</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>PZ</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Area under the ROC curve (AUC)</td>
<td>0.84±0.05</td>
<td>0.76±0.06</td>
<td>0.96±0.03</td>
<td>0.97±0.02</td>
<td>0.85±0.05</td>
<td>0.97±0.02</td>
<td>0.88±0.05</td>
</tr>
<tr>
<td>Significance level P (Area=0.5)</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Sensitivity (95% CI)</td>
<td>0.89 (113/127)</td>
<td>0.86 (109/127)</td>
<td>0.93 (118/127)</td>
<td>0.94 (119/127)</td>
<td>0.86 (109/127)</td>
<td>0.93 (118/127)</td>
<td>0.89 (113/127)</td>
</tr>
<tr>
<td>Specificity (95% CI)</td>
<td>0.62 (53/86)</td>
<td>0.43 (37/86)</td>
<td>0.81 (70/86)</td>
<td>0.91 (78/86)</td>
<td>0.69 (59/86)</td>
<td>0.88 (76/86)</td>
<td>0.70 (60/86)</td>
</tr>
<tr>
<td><strong>TZ</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Area under the ROC curve (AUC)</td>
<td>0.97±0.03</td>
<td>0.89±0.07</td>
<td>0.98±0.02</td>
<td>0.98±0.02</td>
<td>0.94±0.05</td>
<td>0.95±0.04</td>
<td>0.92±0.05</td>
</tr>
<tr>
<td>Significance level P (Area=0.5)</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Sensitivity (95% CI)</td>
<td>0.8 (20/25)</td>
<td>0.64 (16/25)</td>
<td>0.84 (21/25)</td>
<td>0.76 (19/25)</td>
<td>0.72 (18/25)</td>
<td>0.76 (19/25)</td>
<td>0.52 (13/25)</td>
</tr>
<tr>
<td>Specificity (95% CI)</td>
<td>0.94 (61/65)</td>
<td>0.89 (58/65)</td>
<td>0.95 (62/65)</td>
<td>0.97 (63/65)</td>
<td>0.89 (58/65)</td>
<td>0.95 (62/65)</td>
<td>0.97 (63/65)</td>
</tr>
<tr>
<td><strong>Entire prostate</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Area under the ROC curve (AUC)</td>
<td>0.70±0.05</td>
<td>0.69±0.05</td>
<td>0.72±0.05</td>
<td>0.74±0.05</td>
<td>0.82±0.04</td>
<td>0.71±0.05</td>
<td>0.69±0.05</td>
</tr>
<tr>
<td>Significance level P (Area=0.5)</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Sensitivity (95% CI)</td>
<td>0.2 (30/152)</td>
<td>0.2 (30/152)</td>
<td>0.35 (53/152)</td>
<td>0.53 (81/152)</td>
<td>0.62 (95/152)</td>
<td>0.44 (67/152)</td>
<td>0.22 (33/152)</td>
</tr>
<tr>
<td>Specificity (95% CI)</td>
<td>0.89 (20/226)</td>
<td>0.93 (210/226)</td>
<td>0.81 (183/226)</td>
<td>0.76 (171/226)</td>
<td>0.88 (199/226)</td>
<td>0.74 (168/226)</td>
<td>0.92 (209/226)</td>
</tr>
</tbody>
</table>

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4.8 Discussion and Conclusion

The results of this pilot study demonstrate the feasibility of multi-exponential $T_2$ mapping for PCa diagnosis. Although multi-component nature of $T_2$ decay in prostate has been shown before (37, 38, 39, 40, 41) only one recent study investigated the application of this technique for cancer diagnosis (41); however, no direct correlation between MRI and histology has been published. In our study, we validated MR measurements with whole-mount histology, and performed statistical analysis to assess the accuracy of this technique in the detection and grading of PCa. In addition, we used a regularized NNLS algorithm, which provides an unbiased assessment of the number of $T_2$ components and the increased accuracy of $T_2$ estimation in the presence of noise (60). We also introduced a new parameter: the fractional volume of the long $T_2$ component, or LWF, which outperformed all the other MRI parameters in detecting and grading PCa. In addition, LWF represents a morphological feature of the tissue, i.e. the percentage of the luminal space, and thus lends itself to a physiological interpretation more easily than most other MRI parameters.

The mean and range of LWF values measured in non-malignant PZ in this study is in good agreement with the published values of the percentage area of lumen in normal prostatic tissue (0.24 (0.09-0.45) vs. 29.6% (15.9%-43.9%)) (48), suggesting that LWF can be a good measure of luminal space in prostate. This is strongly supported by our recent study (61), which showed a significant correlation between LWF and the histological measure of luminal space (Details of this study were provided in Chapter 3). The mean and range of gmT$_2$, $T_2$-short, and $T_2$-long also correspond well to the previously published values (38, 39).
Our results showed lower gmT₂, A_long, and LWF values in tumours and in normal dense stroma than in the non-malignant glandular tissue. In general, in PCa, some of the loose stroma that fills the area between ducts and acini is replaced by densely packed malignant epithelial cells, decreasing the percentage of lumen. Therefore it is not surprising to see a decrease in A_long and LWF in these cancers.

There was no significant difference in Ncomp, T₂-long and T₂-short between non-malignant and malignant tissues. This is likely because T₂ will depend not only on the size of the water compartment, but also on its shape, or more accurately, its surface to volume ratio (62, 63). Thus one would not necessarily expect significant differences of T₂-long between tumour and normal tissue. We observed fairly large variability in T₂-long, most likely related to the variability of individual glands’ sizes and surface to volume ratios.

Average values of Ncomp were lower in tumour and normal dense stroma than in the non-malignant glandular tissue in PZ; thus Ncomp can be considered an indicator of how glandular the tissue is. Generally, in higher grade tumours the lumen shrinks significantly and more T₂-decay curves become mono-exponential, thus decreasing the average Ncomp values. Therefore one would expect Ncomp to be significantly different between normal glandular and higher grade cancerous tissue. Since the analysis of the entire prostate included normal stroma (Ncomp ≈1) and lower grade tumours (Ncomp ≈2), it is not surprising to see no significant difference in Ncomp between non-malignant and malignant tissue.

Our results demonstrate significant correlation between LWF and GS in PZ. The decreasing LWF with increasing GS is aligned with the published values for percentage of lumen in normal tissue and in tumours of Gleason scores 6 and 7 (48). Such dependence underscores
the role of LWF as a morphologic parameter that correlates with known histologic features. It is recognized that Gleason pattern 4 carcinoma has poorly formed fused glands and is often characterized by a dense cribriform pattern. Gleason pattern 4 carcinoma has significant collapse and loss of luminal formation, contrasting with Gleason 3 carcinoma that has malignant glands with open lumina (19). Thus one may expect lowered LWF with this loss of luminal space and increased cell density.

LWF had much higher AUC in PZ and TZ than in the entire prostate. This is likely because, with regards to the luminal space, the normal dense stroma mimic the tumours in PZ and TZ. Our results show that combining $N_{\text{comp}}$ with any other MR parameter improves specificity and AUC within the entire prostate; this suggests that $N_{\text{comp}}$ can potentially be useful in tissue classification algorithms.

The main limitation of this pilot study was the relatively low number of patients. Also, the range of Gleason Score in this study was limited, with 35 lesions of GS=6, 13 GS=7, 1 GS=8, 6 GS=9 and none with GS=10. Another limitation of this study includes lack of distinction between normal gland, BPH, and prostatitis. Also, the patient selection process was potentially biased, as the recruited patients were selected from those who were scheduled for retropubic prostatectomy, indicating that the sampled lesions were weighted toward more advanced tumours. However, any study that requires validation with histology will suffer from a similar bias.

Areas of future research with this technique include expanding this study with a larger number of patients and wider range of Gleason Scores, with larger numbers in each category, to access the accuracy of this technique. Comparison of this technique with the current PI-RADS v2
protocol (29) would also be useful in determining its clinical utility. Also combining it with current PI-RADS version 2 protocol to determine if it further increases accuracy and its incremental benefit should be investigated.

In conclusion, the results of this pilot study demonstrate the feasibility of Luminal Water Imaging for diagnosis and grading of prostate cancer. Four MR parameters showed significant differences between malignant and non-malignant tissue in the prostate. LWF showed very high accuracy in detection of prostatic tumours, and a strong correlation with Gleason score. \( N_{\text{comp}} \) was found to increase the specificity of this technique in the entire prostate. Our preliminary results have shown that the proposed MRI technique can be applied for both the detection and grading of prostate cancer with high accuracy. To accurately assess the suitability of this technique for clinical application, a prospective study with much larger number of patients and broader range of tumour grades, and a comparison to the current clinical PI-RADS version 2 protocol should be done.
Chapter 5: Comparing Diagnostic Accuracy of Luminal Water Imaging with Diffusion-Weighted and Dynamic Contrast-Enhanced MRI in Detection and Grading of Prostate Cancer

5.1 Introduction
In Chapter 4, we investigated the feasibility of LWI in the detection and grading of prostate tumours, and showed that this technique provides high accuracy in the detection of tumours and high correlation with GS. Although the results of this study were very promising, it was only performed as a hypothesis-generating pilot. In order to involve LWI in clinical settings further investigation on the accuracy of this technique is required. In this prospective trial, we performed a detailed comparison of the diagnostic accuracy of LWI with the standard MP-MRI protocols, including dynamic contrast-enhanced (DCE) and DW-MRI. In addition, in this study efforts were made to evaluate the significance of adding LWI into a MP-MRI protocol, consisting of DCE-MRI and DW-MRI, for the detection and grading of the prostatic tumours.

5.2 Patient Selection
The patient selection process was similar to what was explained in Chapter 3. Clinical data for the 15 patients who participated in this study are summarized in Table 5-1. The entire patient population used in this study has been previously reported in Chapters 3 & 4.
Table 5-1. Clinical data of 15 patients participating in the study.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age (y)</th>
<th>PSA level (ng/mL)</th>
<th>Prostate volume (cc)</th>
<th>Size (cc)</th>
<th>Gleason grade</th>
<th>Location</th>
<th>Total number of ROIs per lesion (from different slices)</th>
<th>Pathologic tumour stage</th>
</tr>
</thead>
<tbody>
<tr>
<td>P01</td>
<td>65</td>
<td>13.1</td>
<td>32.3</td>
<td>5.8</td>
<td>4+3</td>
<td>PZ</td>
<td>7</td>
<td>pT3a</td>
</tr>
<tr>
<td>P02</td>
<td>67</td>
<td>8</td>
<td>94.7</td>
<td>0.7</td>
<td>3+3</td>
<td>PZ</td>
<td>2</td>
<td>pT2c</td>
</tr>
<tr>
<td>P03</td>
<td>62</td>
<td>6.5</td>
<td>31.0</td>
<td>3.2</td>
<td>4+3</td>
<td>PZ</td>
<td>6</td>
<td>pT3a</td>
</tr>
<tr>
<td>P04</td>
<td>80</td>
<td>7.2</td>
<td>128.3</td>
<td>0.1</td>
<td>3+3</td>
<td>PZ</td>
<td>2</td>
<td>pT2a</td>
</tr>
<tr>
<td>P05</td>
<td>64</td>
<td>7.6</td>
<td>32.3</td>
<td>7.5</td>
<td>4+3</td>
<td>PZ</td>
<td>8</td>
<td>pT3a</td>
</tr>
<tr>
<td>P06</td>
<td>75</td>
<td>18.2</td>
<td>41.1</td>
<td>3.3</td>
<td>4+3</td>
<td>PZ</td>
<td>5</td>
<td>pT2c</td>
</tr>
<tr>
<td>P07</td>
<td>69</td>
<td>18.4</td>
<td>39.5</td>
<td>6.2</td>
<td>4+5</td>
<td>TZ</td>
<td>5</td>
<td>pT3a</td>
</tr>
<tr>
<td>P08</td>
<td>66</td>
<td>7.4</td>
<td>32.5</td>
<td>1.0</td>
<td>3+4</td>
<td>PZ</td>
<td>1</td>
<td>pT3a</td>
</tr>
<tr>
<td>P09</td>
<td>67</td>
<td>6</td>
<td>17.7</td>
<td>1.3</td>
<td>3+4</td>
<td>PZ</td>
<td>3</td>
<td>pT3a</td>
</tr>
<tr>
<td>P10</td>
<td>58</td>
<td>6.7</td>
<td>31.4</td>
<td>6.1</td>
<td>3+4</td>
<td>PZ</td>
<td>8</td>
<td>pT3a</td>
</tr>
<tr>
<td>P11</td>
<td>58</td>
<td>12</td>
<td>20.2</td>
<td>3.9</td>
<td>3+4</td>
<td>PZ</td>
<td>4</td>
<td>pT3a</td>
</tr>
<tr>
<td>P12</td>
<td>66</td>
<td>31.6</td>
<td>44.9</td>
<td>6.6</td>
<td>4+3</td>
<td>PZ</td>
<td>8</td>
<td>pT3a</td>
</tr>
<tr>
<td>P13</td>
<td>75</td>
<td>8.5</td>
<td>36.6</td>
<td>3.2</td>
<td>4+5</td>
<td>PZ</td>
<td>4</td>
<td>pT3a</td>
</tr>
<tr>
<td>P14</td>
<td>63</td>
<td>51</td>
<td>26.4</td>
<td>9.5</td>
<td>4+5</td>
<td>PZ</td>
<td>6</td>
<td>pT3b</td>
</tr>
<tr>
<td>P15</td>
<td>62</td>
<td>13.3</td>
<td>33.3</td>
<td>2.1</td>
<td>4+4</td>
<td>PZ</td>
<td>4</td>
<td>pT3a</td>
</tr>
</tbody>
</table>
5.3 MRI Protocol

As mentioned before, MRI examinations were performed with a 3T MRI scanner, using a combined endorectal/pelvic phased-array coil. The scanning session included: T2W imaging, LWI, DCE-MRI and DW-MRI. Orientation of axial images was set perpendicular to the rectal wall-prostate interface, and identical slice thicknesses and positioning were used for all imaging techniques.

LWI protocol was similar to what was explained for the previous study described in Chapter 3. Multi-echo data were acquired using a 3D multi-echo spin echo pulse sequence developed at the University of British Columbia MRI Research Centre (8). More details about the LWI protocol can be found in Chapter 3, MRI protocol.

DW-MRI used a multi-slice diffusion weighted single shot echo-planar spin echo sequence (TR=3129-3488 ms, TE=50-64 ms, FOV=240×240×72-375×375×72mm³, voxel-size=1.4×1.4×4-1.5×1.5×4mm³, scan matrix-size=120×120-188×186, slice thickness=4mm, flip angle=90°, number of averages=4, sense factor= 2, scan duration=241-269 sec). During each DW-MRI scan, four sets of images were acquired; one without and three with diffusion gradient (b values: 0, 100, 500, and 1000 sec/mm²).

DCE-MRI data consisted of T₁-weighted (T1W) and proton density (PD) images. PD data were obtained for calculation of T1 values which consequently were used for calculation of the contrast agent concentration in the tissue (64). PD images were acquired prior to the acquisition of T1W images using a 3D ultrafast spoiled gradient echo sequence (T1-TFE) (TR=50 ms, TE=0.93 ms, FOV=295×375×72mm³, voxel-size=1.2×1.5×4mm³, scan matrix-size=188×188, slice thickness=4mm, flip angle=4°, number of averages=1, sense factor= 2, scan duration=127 sec). A series of 170 T1W images were acquired prior to and after (4, and 166 images,
respectively) the injection of contrast agent (Magnevist (Gadopentetate dimeglumine, Bayer HealthCare, NJ), 469mg/mL (0.5 mmol/mL), 0.2 mL/Kg, rate: 2mL/s, flush: 20 mL), using a 3D T1-TFE sequence (TR=2959 ms, TE=1.35 ms, temporal resolution= 3.4 ms, FOV=295x375x72mm³, voxel-size=1.2x1.5x4mm³, scan matrix-size=188x188, slice thickness=4mm, flip angle=12°, number of averages=1, sense factor= 2, scan duration=581 sec).

High spatial resolution T2W images were acquired to provide anatomical details and facilitate the process of image registration, using a multi-slice Turbo Spin Echo (TSE) sequence (TR=1851 ms, effective TE=80 ms, FOV=140x140x72mm³, voxel-size=0.3x0.3x4mm³, scan matrix-size=288x228, slice thickness=4mm, flip angle=90°, number of averages=3, scan duration=285 sec).

5.4 Histologic Examination and Registration to MR Images

Similar to what was explained in Chapters 3 & 4, following radical prostatectomy, the excised specimens were fixed by immersion in formalin and cut in to 4mm transverse slices. Whole mount histology sections were stained by H&E and submitted for histological examination. On the whole-mount slides, tumours with a minimum Gleason grade of 3 were outlined and Gleason scores were assigned.

Image registration was performed to align MR images with the corresponding histology slides. Due to the differences in the field of view, artifacts and apparent distortions between images of LWI, DCE and DW-MRI, image registration was performed separately for each technique. Digitized histology sections were generated by scanning whole-mount slides using a flatbed
scanner. Histology sections were registered to MR images with a software package developed in-house (51) using Matlab similar to what was explained before. For more information about the registration process please refer to Chapter 3, Whole-mount Histology and Registration to MR images.

5.5 Data Processing

Processing of all MR data was performed offline. From LWI data, for each voxel the signal decay curve was extracted and fitted to a multi-exponential function to generate maps of 7 parameters: $T_2\text{-short}$, $T_2\text{-long}$, geometric mean $T_2$ ($gmT_2$), LWF, $A_\text{short}$, $A_\text{long}$, and $N_\text{comp}$ as described in Chapter 3, Data Processing.

From the set of four DWI images the apparent diffusion coefficient (ADC) was calculated using the Stejskal-Tanner equation (65):

$$S_b = S_0 e^{-b*ADC}$$  

Equation 5-1

where $S_b$ is the signal intensity with non-zero $b$, and $S_0$ is the signal intensity with $b=0$.

For processing DCE-MRI data, first the concentration of contrast agent in the tissue was calculated from T1W and PD images as described in (64). The extended Tofts (also known as Kety) model (66) was used to relate the concentration of contrast in the tissue to the arterial input function (AIF). For each patient, the AIF function was generated by fitting the concentration of contrast agent in the voxels within the external iliac or femoral arteries to a double Gaussian function (67). By fitting the concentration of contrast agent in tissue to the extended Tofts model, maps of pharmacokinetic parameters: volume transfer constant ($K^{\text{trans}}$), fractional volume of the extra-vascular extra-cellular space ($v_e$), and fractional volume of
plasma \((v_p)\) were generated. These parameters, and how they were calculated, are described in greater details in Chapter 6. Fitting was performed for all the voxels within the prostate gland.

Average values of MR parameters obtained from LWI, DCE, and DW-MRI were calculated within a total of 304 (186 non-malignant, and 118 cancers) ROIs manually outlined on the registered histology sections. Tumours with dimensions of equal or higher than \(4 \times 4 \text{ mm}^2\) were included in data processing. For statistical analysis, data were divided into six categories based on the location and malignancy status of tissue as follows: malignant PZ, malignant TZ, non-malignant PZ, non-malignant TZ, normal PFMS, and normal AFMS. For selection of malignant ROIs the outlines of tumours, delineated by pathologist, were accurately replicated during the image registration process. Selection of non-malignant ROIs was performed by avoiding the tumour boundaries. Considering that in this study regions of BPH were not delineated by the pathologist, non-malignant ROIs could consist of normal or BPH tissue.

### 5.6 Statistical Analysis

Statistical analyses were performed using MedCalc and R packages (56, 57, 58). Diagnostic accuracy of individual and combined MR parameters was investigated through ROC analysis (59). Values of sensitivity, specificity, and AUC were calculated separately in PZ, TZ, and the entire prostate. ROC analyses were performed based on logistic GLMMs with the dependant variable being malignant vs non-malignant. Correlation within each patient was incorporated by having a random intercept in the model with the grouping factor for the random intercept being each patient. Multi-parametric ROC analyses were performed using logistic GLMM,
while in order to prevent data overfitting only parameters that contributed significantly to the model, and also minimized AIC and BIC, were used in the calculations.

Prior to correlation testing, normality of MR data was investigated with the Kolmogorov-Smirnov test. Since none of the MR parameters were normally distributed, correlations between MR parameters and GS were evaluated by averaging Spearman’s correlation coefficients calculated for individual patients. Significance of the correlation between MR parameters and GS was evaluated with the application of one sample t-test. In the entire study, P<0.05 was considered statistically significant.

5.7 Results

Representative MR parametric maps are shown in Figure 5-1. Values of sensitivity, specificity and AUC calculated from single and multi-parameter ROC analyses are summarized in Table 5-2 and Table 5-3, respectively.

In single parameter ROC analysis (Table 5-2), the highest value of AUC obtained from LWI was equal to or higher than the highest value of AUC obtained from DCE and DW-MRI in PZ (0.97 vs 0.9 and 0.9 respectively), TZ (0.99 vs 0.9 and 0.98), and entire prostate (0.81 vs 0.76 and 0.81). In comparing the accuracy of individual techniques, using multi-parameter ROC analysis (except for DWI which only had one parameter) (Table 5-2 and Table 5-3), the AUC value obtained from LWI was higher than the AUC value obtained from DCE and DW-MRI in PZ (0.98 vs 0.89 and 0.9), and the entire prostate (0.85 vs 0.75 and 0.81). Multi-parametric ROC analysis could not be performed for DCE in TZ since there were no combinations of DCE parameters that contributed significantly to logistic GLMM.
Comparison of the diagnostic accuracy of combinations of techniques (Table 5-3) showed that: in PZ the highest AUC value (0.98) was obtained from LWI combined with either or both DCE and DW-MRI; in TZ, the highest AUC value (0.99) was obtained from combination of LWI and DW-MRI; and in the entire prostate the highest value of AUC (0.88) was obtained from combination of all three techniques. In PZ, the differences between AUC values obtained from the combination of LWI with DCE, DW-MRI, or both were not statistically significant, but the AUC value obtained from any combination including LWI was significantly different than the AUC value obtained from the combination of DCE and DW-MRI. In TZ, none of the differences between AUC values obtained from different combinations were statistically significant. In the entire prostate, the difference between the AUC value obtained from combination of all three techniques and combination of LWI and DCE-MRI was not statistically significant. But the AUC values obtained from combination of LWI and DW-MRI or DCE and DW-MRI were significantly different than AUC values obtained from combination of all three techniques or combination of LWI and DCE-MRI.

Average Spearman's rank correlation coefficients between MR parameters and GS in PZ are presented in Table 5-4. A T-test could not be conducted in TZ due to the insufficient number of patients with tumours in TZ. The data presented in Table 5-4 indicates that the highest correlation coefficient obtained from the LWI technique was higher than the highest correlation coefficient obtained from DCE and DW-MRI (0.81 vs 0.59 and 0.53 respectively). Overall, the strongest correlation with GS was obtained from the LWF parameter with a Spearman's coefficient of -0.81±0.09.
Figure 5-1. Representative maps of MR parameters, T2-weighted image, and histology whole-mount section of the same slice.
<table>
<thead>
<tr>
<th>Location</th>
<th>Entire Prostate</th>
<th>PZ</th>
<th>TZ</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Technique</strong></td>
<td><strong>Parameter</strong></td>
<td><strong>MR</strong></td>
<td><strong>Sensitivity</strong> (95% CI)</td>
</tr>
<tr>
<td><strong>DCE</strong></td>
<td>$K_{\text{trans}}$</td>
<td>0.42 (50/118)</td>
<td>0.85 (158/186)</td>
</tr>
<tr>
<td></td>
<td>$v_c$</td>
<td>0.25 (29/118)</td>
<td>0.91 (170/186)</td>
</tr>
<tr>
<td></td>
<td>$v_p$</td>
<td>0.42 (49/118)</td>
<td>0.87 (162/186)</td>
</tr>
<tr>
<td><strong>DWI</strong></td>
<td>ADC</td>
<td>0.61 (72/118)</td>
<td>0.85 (159/186)</td>
</tr>
<tr>
<td></td>
<td>$T_2\text{-short}$</td>
<td>0.2 (24/118)</td>
<td>0.9 (168/186)</td>
</tr>
<tr>
<td></td>
<td>$T_2\text{-long}$</td>
<td>0.19 (23/118)</td>
<td>0.94 (174/186)</td>
</tr>
<tr>
<td></td>
<td>gmT$_2$</td>
<td>0.31 (37/118)</td>
<td>0.81 (150/186)</td>
</tr>
<tr>
<td></td>
<td>LWF</td>
<td>0.47 (56/118)</td>
<td>0.75 (139/186)</td>
</tr>
<tr>
<td></td>
<td>$A_{\text{short}}$</td>
<td>0.58 (68/118)</td>
<td>0.91 (170/186)</td>
</tr>
<tr>
<td></td>
<td>$A_{\text{long}}$</td>
<td>0.42 (49/118)</td>
<td>0.74 (138/186)</td>
</tr>
<tr>
<td></td>
<td>$N_{\text{comp}}$</td>
<td>0.21 (25/118)</td>
<td>0.9 (167/186)</td>
</tr>
</tbody>
</table>

*Significantly different than AUC value obtained from $K_{\text{trans}}$, based on Delong’s test. $^b$ Significantly different than AUC value obtained from $v_c$. $^c$ Significantly different than AUC value obtained from $v_p$. $^d$ Significantly different than AUC value obtained from ADC. $^e$ Significantly different than AUC value obtained from $T_2\text{-short}$. $^f$ Significantly different than AUC value obtained from $T_2\text{-long}$. $^g$ Significantly different than AUC value obtained from gmT$_2$. $^h$ Significantly different than AUC value obtained from LWF. $^i$ Significantly different than AUC value obtained from $A_{\text{short}}$. $^j$ Significantly different than AUC value obtained from $A_{\text{long}}$. $^k$ Significantly different than AUC value obtained from $N_{\text{comp}}$. 
Table 5-3. ROC analysis measures for combined MRI parameters in the entire prostate, PZ, and TZ.

<table>
<thead>
<tr>
<th>Location</th>
<th>Entire Prostate</th>
<th>PZ</th>
<th>TZ</th>
</tr>
</thead>
<tbody>
<tr>
<td>Technique</td>
<td>Contributing Parameters</td>
<td>Sensitivity</td>
<td>Specificity</td>
</tr>
<tr>
<td>LWI</td>
<td>T2_short, Ncomp, gmT2</td>
<td>0.64</td>
<td>0.82</td>
</tr>
<tr>
<td>DCE</td>
<td>T2_short, gmT2, A_short</td>
<td>0.55-0.73</td>
<td>0.76-0.87</td>
</tr>
<tr>
<td>LWI+DCE</td>
<td>T2_short, gmT2, Ncomp, A_short</td>
<td>0.73</td>
<td>0.82</td>
</tr>
<tr>
<td>LWI+DCE</td>
<td>T2_short, gmT2, ADC</td>
<td>0.62-0.79</td>
<td>0.75-0.87</td>
</tr>
<tr>
<td>DCE+DWI</td>
<td>ADC</td>
<td>0.64</td>
<td>0.88</td>
</tr>
<tr>
<td>LWI+DCE+DWI</td>
<td>T2_short, gmT2, Ncomp, ADC</td>
<td>0.75</td>
<td>0.84</td>
</tr>
</tbody>
</table>

*Significantly (P<0.05) different than value of AUC obtained from LWI, based on Delong’s test.  †Significantly different than AUC value obtained from DCE.
*Significantly different than AUC value obtained from combination of LWI and DCE.  ‡Significantly different than AUC value obtained from combination of DCE and DWI.  §Significantly different than AUC value obtained from combination of LWI, DCE, and DWI.  ‡Not significantly different than AUC value obtained from any other technique in TZ.
Table 5-4. Statistics of correlations with Gleason score in PZ.

<table>
<thead>
<tr>
<th>Technique</th>
<th>Parameter</th>
<th>Average Spearman’s coefficient of rank correlation (ρ) ±standard deviation</th>
<th>95% Confidence Interval for ρ</th>
<th>Significance level</th>
<th>Test statistic t</th>
</tr>
</thead>
<tbody>
<tr>
<td>DCE</td>
<td>$K_{\text{trans}}$</td>
<td>0.59±0.36</td>
<td>0.37 to 0.80</td>
<td>P = 0.0001</td>
<td>5.927</td>
</tr>
<tr>
<td>DCE</td>
<td>$v_e$</td>
<td>0.36±0.31</td>
<td>0.16 to 0.55</td>
<td>P = 0.0016</td>
<td>4.058</td>
</tr>
<tr>
<td>DCE</td>
<td>$v_p$</td>
<td>0.46±0.41</td>
<td>0.22 to 0.71</td>
<td>P = 0.0014</td>
<td>4.120</td>
</tr>
<tr>
<td>DWI</td>
<td>ADC</td>
<td>-0.53±0.43</td>
<td>-0.79 to -0.27</td>
<td>P = 0.0008</td>
<td>4.417</td>
</tr>
<tr>
<td>LWI</td>
<td>$T_2$-short</td>
<td>-0.46±0.40</td>
<td>-0.70 to -0.22</td>
<td>P = 0.0014</td>
<td>4.124</td>
</tr>
<tr>
<td>LWI</td>
<td>$T_2$-long</td>
<td>0.13±0.43</td>
<td>-0.13 to 0.39</td>
<td>P = 0.3021</td>
<td>1.078</td>
</tr>
<tr>
<td>LWI</td>
<td>gm$T_2$</td>
<td>-0.76±0.10</td>
<td>-0.82 to -0.70</td>
<td>P &lt; 0.0001</td>
<td>26.298</td>
</tr>
<tr>
<td>LWI</td>
<td>LWF</td>
<td>-0.81±0.09</td>
<td>-0.86 to -0.75</td>
<td>P &lt; 0.0001</td>
<td>31.663</td>
</tr>
<tr>
<td>LWI</td>
<td>$A_{\text{short}}$</td>
<td>0.38±0.50</td>
<td>0.07 to 0.68</td>
<td>P = 0.0192</td>
<td>2.702</td>
</tr>
<tr>
<td>LWI</td>
<td>$A_{\text{long}}$</td>
<td>-0.80±0.10</td>
<td>-0.86 to -0.74</td>
<td>P &lt; 0.0001</td>
<td>29.598</td>
</tr>
<tr>
<td>LWI</td>
<td>$N_{\text{comp}}$</td>
<td>-0.68±0.19</td>
<td>-0.80 to -0.57</td>
<td>P &lt; 0.0001</td>
<td>13.211</td>
</tr>
</tbody>
</table>
5.8 Discussion and Conclusion

Our results showed that LWI alone performs better than either DCE or DW-MRI, and equally well or better than the combination of DCE and DW-MRI in the detection of prostate cancer. Using LWI alone rather than a combination of DCE and DW-MRI provides advantages of being completely non-invasive as it does not require administration of contrast agent. This results in reduction of costs of contrast agent and intravenous injection materials. Also, LWI can be used as an alternative for patients who cannot undergo DCE-MRI due to potential adverse reaction to a gadolinium based contrast agent, as well as patients with metallic hip implants, for whom DWI is likely to be non-diagnostic. Finally, LWI can potentially be the basis for developing an abbreviated protocol for prostate cancer screening and diagnostics. A similar concept is becoming standard of care for breast cancer (68). However, it should be noted here that the acquisition time of LWI protocol is currently relatively long (11 minutes). Therefore, for the development of an abbreviated protocol based on this technique, further improvements of the proposed LWI technique are necessary to reduce its acquisition time. Reducing the scan time of LWI is a feasible task by for example increasing the GRASE factor and/or under-sampling the k-space in conjunction with the Compressed Sensing reconstruction.

We showed that LWI provides significantly higher correlation coefficients with Gleason score than DCE and DW-MRI. This is to be expected because LWF strongly correlates with the fractional amount of luminal space in prostate tissue (61), and the relative amount of luminal space decreases with increase in GS (19, 48). Both ADC and K\text{trans} also correlated significantly with GS in PZ, albeit with significantly lower values of the correlation coefficient. The ADC’s
significant correlation with GS is most likely related to the higher cellularity of high GS tumours (69), since it has been shown that ADC values correlate with tissue cellular density (70). The likely reason for significant correlation between $K^{\text{trans}}$ and GS is the increased tumour vascularity, as compared to normal tissue, since Borren et al. have shown a statistically significant dependence between GS and micro-vessel density (71). However, the results of our study suggest that the amount of luminal space is a much stronger factor than cellularity or vascularity in distinguishing between different Gleason scores.

In this study the highest accuracy for detecting PCa in the entire prostate was achieved when LWI was combined with both DCE and DW-MRI. However, the difference between the AUC value obtained from combination of all three techniques and combination of LWI and DCE-MRI was not statistically significant. Also, in PZ the difference between the AUC value obtained from LWI when combined with either or both DCE and DW-MRI was not statistically significant, and in TZ there was no significant difference between the AUC value obtained from any combination. Based on accuracy alone, the combination of LWI and DCE-MRI could be the preferred methodology for diagnosis of prostate cancer. But if scan time is also taken into consideration, the combination of LWI and DW-MRI would be preferred, as it still provides the highest accuracy in PZ, and TZ, with a very minor reduction in accuracy in the entire prostate.

This study has several limitations, primarily the relatively small number of patients and the limited range of GS. Also, in this study, there was no differentiation between normal gland, BPH, and prostatitis in non-malignant regions. Another limitation was related to patient selection: as the recruited patients were selected from those who were scheduled for retropubic
prostatectomy, the sampled lesions were weighted toward more advanced tumours. We had relatively long scan times (70 minutes) which increased the chance of patients’ movement, possibly resulting in data processing errors of each technique or a mismatch of images between different techniques. Another limitation was that a high b value of > 1400 was not used, which is recommended as per PI-RADS v2.

This study was a pilot study and a final decision on whether to incorporate LWI in clinical MP-MRI protocols requires further investigations. Potential future research includes expanding this study with a larger number of patients and wider range of GS, and comparison to PI-RADS version 2. Also, future work will involve further development of the LWI technique to reduce its acquisition time by increasing the Gradient Spin Echo (GRASE) factor and/or using sparse k-space sampling together with the Compressed Sensing reconstruction.

In conclusion, the results of this pilot study show that LWI alone performs better than DCE and DW-MRI in detection of PCa and in correlation with GS. LWI, when combined with either DCE or DW-MRI, provides significantly higher accuracy than either of the other two techniques individually or combined. Therefore, it can be concluded that incorporating LWI in MP-MRI protocols may result in a significant increase in accuracy of detection and grading of PCa and that a more efficient, abbreviated MP-MRI protocol for diagnosis of PCa can be achieved by combining LWI with either DCE or DW-MRI, but not both.

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Chapter 6: Optimization of Temporal Sampling of Dynamic Contrast-Enhanced MRI Protocol for Diagnosis of Prostate Cancer

6.1 Introduction

6.1.1 DCE-MRI

DCE-MRI is a MRI technique that is widely used for diagnosis of cancer and monitoring response to anticancer treatments due to its capability for characterization of tumour microvasculature (64, 72).

In this technique, a paramagnetic contrast enhancing agent is injected intravenously into the patient. Consecutive T1W MR images are acquired prior to, during, and following the administration of the contrast agent. As the contrast agent passes through the body, it leaks from the vascular system to the surrounding tissue, and decreases the T₂ and T₁ relaxation times of the adjacent tissue. The T₂ shortening effect of the contrast agent results in a decrease of signal intensity while the T₁ shortening effect of the contrast agent results in an increase of signal intensity of the T1W images. In clinical settings, depending on the tissue of interest, different DCE-MRI techniques with different contrast agents are used. For DCE-MRI of prostate, the contrast agent is usually Gadolinium-based. The dominant effect of Gadolinium on signal intensity is the decrease of T₁ relaxation time. Therefore, following the injection of Gadolinium, a signal enhancement can be obtained in the tissue in proximity to the contrast agent. The rate and level of signal enhancement is different in normal tissues and tumours. It has been shown that the signal enhancement in malignant tumours is generally faster and greater than the signal enhancement in normal tissue (73). This is mainly caused by (but not
limited to) the higher vascularity and also higher endothelial permeability of malignant tumours compared to the normal tissues. Based on these differences, by monitoring the contrast-driven enhancement, it can be determined whether a tissue is normal or not. Moreover, by knowing the amount of signal enhancement, and accordingly estimating the change in $T_1$ relaxation time of the tissue, concentration of contrast agent in the tissue can be calculated at any time. Then, by use of a pharmacokinetic model, the concentration of contrast agent in the tissue can be related to its physiological parameters such as vessel density, blood flow, endothelial permeability, and the volume of Extracellular-Extravascular Space (EES).

Among the pharmacokinetic models that are used in the literature, the Tofts (also known as Kety) (74) and extended Tofts model (75) are the most common pharmacokinetic models used for DCE-MRI. In this study, we only used the extended Tofts model which relates the concentration of the contrast agent in the tissue to three useful physiological parameters: 1) the influx volume transfer constant ($K_{\text{trans}}$), 2) the volume of EES per unit volume of tissue ($v_e$), and 3) the volume of plasma per unit volume of tissue ($v_p$).

In the extended Tofts model, the equation relating the concentration of contrast agent in the tissue ($C_t$), the concentration of contrast agent in the plasma ($C_p$), and the three physiological parameters ($K_{\text{trans}}$, $v_e$, $v_p$) is expressed as below:

$$C_t(t) = v_p C_p(t) + K_{\text{trans}} \int_0^t C_p(T) e^{-(K_{\text{trans}}/v_e)(t-T)} dT$$ \hspace{1cm} \text{Equation 6-1}

Following a DCE-MRI examination, $C_t(t)$ and $C_p(t)$ can be calculated from scan data. By using the calculated $C_t(t)$ and $C_p(t)$ and an appropriate fitting program, physiological parameters ($K_{\text{trans}}$, $v_e$, $v_p$) can be calculated (from Equation 6-1) for every pixel of a DCE image.
6.1.2 Significance of Temporal Resolution in Diagnostic Accuracy of DCE-MRI

The data fitting process based on Equation 6-1 is complicated due to the presence of the integral term. If the functional form of $C_p(t)$ is known, the integral term can be replaced by its analytical solution to simplify the fitting procedure. Otherwise, the integral term can only be estimated numerically. The difference between the result of numerical integration and the analytical solution introduces an uncertainty in the fitting procedure. Let us call the difference between the outcome of numerical integration and analytical integration, the integral approximation error, IAE. IAE can affect the accuracy of the fitting process and increase the uncertainty in calculation of physiological parameters. This error depends on the temporal resolution. It has been shown (76) that the accuracy of DCE data analysis can be maximized by using very high temporal resolution (as high as 1s). Based on this result and by considering the source of IAE, one may assume that the accuracy of DCE data analysis perhaps always increases with an increase of temporal resolution. But we will show that this is not necessarily the case for the problem at hand. In fact the IAE depends both on the temporal resolution, and the shape of $C_p(t)$. To better understand this matter, let us look at Figure 6-1. Based on this figure, as explained in its caption, it can be observed that it is possible to have a bigger IAE for a higher temporal resolution. Thus, there potentially could be an optimum value of temporal resolution minimizing the IAE. Based on the observation of this figure, one has to be cautious when choosing the temporal resolution for a DCE-MRI protocol.
Figure 6-1. Comparison between numerical integration of the integral term in Equation 6-1, from a simulated $C_p$ with temporal resolution of 12 s (left figures) and 14 s (right figures). In the top figures (a & b) the integration is performed using the rectangle method, and in the bottom figures (c & d) the integration is performed using the trapezoid method. The red curves show $C_p(T) \exp(-\left(K_{\text{trans}}/V_e\right)(t - T))$. Pink and blue colors show the areas that are overestimated and underestimated by integration, respectively. Green color shows the area that is correctly calculated by the integration. In the above figures, the total integration approximation error (IAE) can be calculated by subtraction of the pink area from the blue area. Here we can see that for both of the rectangular and trapezoidal integration techniques, in the first three steps of the integration, in the right figures ($\Delta t=14s$), the overestimated region roughly cancels the underestimated region. However, in the left figures ($\Delta t=12s$) the dominant effect is underestimation. Therefore, the cases shown at the left, while having a higher temporal resolution, have a higher IAE than the cases shown at the right.

Although many studies have investigated the effect of temporal resolution on the diagnostic accuracy of DCE-MRI (77, 78, 79, 80), to the best of our knowledge, no study has been conducted to investigate the effect of temporal resolution on the accuracy of prostate cancer diagnosis using the extended Tofts model. Therefore, the first objective of this study was to
investigate the relationship between the accuracy of calculated fit parameters ($K_{\text{trans}}, v_e, v_p$) that are used in the extended Tofts model, and the temporal resolution, by using simulation. The second objective of this study was to increase the accuracy of calculated fit parameters (obtained from simulation) for low temporal resolution data, by using an alternative data analysis technique. The third objective was to investigate the relationship between the temporal resolution and the detection accuracy of prostatic tumours, using patient data. The details of the implementation and the results of these three projects are presented in sections 6.2, 6.3, and 6.4.

6.2 Temporal Resolution and Accuracy of Fit Parameters

In this study the relationship between the accuracy of calculated fit parameters ($K_{\text{trans}}, v_e, v_p$) and temporal resolution was investigated by performing a series of simulations. In these simulations, $C_p(t)$ was generated for a series of time data points separated by $\Delta t$ by using a double-Gaussian function as described by Parker et. al (67). $C_t(t)$ was generated, with the same temporal spacing as $C_p(t)$, by substituting $C_p(t)$ and presumed values (obtained from (81)) of $K_{\text{trans}}, v_e, v_p$, in Equation 6-1. A Gaussian noise was then added to the $C_t(t)$, and $C_p(t)$ values. These $C_t(t)$ and $C_p(t)$ were then substituted in Equation 6-1, and by using a fitting program written in Matlab $K_{\text{trans}}, v_e, v_p$ were calculated for each pixel. For evaluating the accuracy of calculated fit parameters, the percentage error for each of the fit parameters was calculated by the following equation:

$$\%\text{Error} = \frac{(\text{calculated value} - \text{presumed value})}{\text{presumed value}} \times 100$$  
Equation 6-2
Maps of percentage error were generated as a function of temporal resolution and the standard deviation of the Gaussian noise that was added to $C_d(t)$ and $C_p(t)$. These maps are presented in Figure 6-2.a-c. Figure 6-2.d shows a graph of all three percentage errors as a function of temporal resolution at a single standard deviation. From Figure 6-2, it can be observed that within a certain range of temporal resolution (3-30s), the effect of standard deviation is dominated by the effect of temporal resolution. Also, from the same figure, it can be observed that the decrease in temporal resolution has a significant impact on $v_p$ values [%error: -100%-0%], while the impact on $K^{\text{trans}}$ is smaller [%error: -25%-0%], and $v_e$ remains almost unaffected [%error: -2%+2%].
6.3 An Alternative Analysis Technique for Low Temporal Resolution Data

We proposed using a different analysis technique for cases when $C_p(t)$ has to be measured with low temporal resolution. This technique is proposed based on the fact that in order to numerically calculate the integral term of Equation 6-1 with a desirably small element size, we do not necessarily need to know the $C_p(t)$ with temporal spacing equal to that element size. Instead, a lower temporal resolution $C_p(t)$ can be first fitted to an appropriate model to find its...
functional form. The functional form of \( C_p(t) \) can then be used to calculate the integral numerically with any desirably small element size.

For testing the proposed technique, a number of simulations were performed using the methodology as explained in section 6.2 with the following modification: 1) only the noiseless condition was investigated (as it was concluded in the previous section that the effect of noise is dominated by the effect of temporal resolution), and 2) regardless of temporal resolution of \( C_p(t) \), the integral term was numerically calculated with 1 s element size (while in section 6.2 the element size of the integral term was always equal to the temporal spacing of \( C_p(t) \)). Percentage errors were calculated similar to what was explained in Equation 6-2 of Section 6.2. Results are shown in Figure 6-3.
Figure 6-3. Percentage errors in calculated fit parameters obtained from simulation. The green and red graphs represent cases where the integral term is calculated with and without the proposed technique, respectively.

Figure 6-3 shows graphs of percentage error as a function of temporal resolution for the cases where the integral term is calculated with and without the proposed technique. From Figure 6-3, it can be observed that our proposed technique is successful in decreasing the error in calculation of all fit parameters, particularly $v_p$.

6.4 Temporal Resolution and Accuracy of Detection of Prostatic Tumours

In this section, we investigate the effect of temporal resolution on the diagnostic accuracy of prostate cancer detection, by using DCE-MRI data acquired from 15 patients. Scan data were
down-sampled in time by a down-sample factor, DSF, of 2, 3, 4, and 5. Parametric maps were generated for the original and the down-sampled data, and correlated to the whole-mount histology using image registration. Diagnostic accuracies were evaluated using ROC analysis. Patient selection, DCE-MRI protocol, histologic examination and registration process of this study is exactly the same as Chapter 5; therefore, they are not repeated here again. However, data processing is slightly different which is explained in the following section.

### 6.4.1 Data Processing

Data processing was similar to Chapter 5 with one modification. In addition to processing the original data, the down-sampled data were also generated and processed in this study. In an effort to mimic the lower temporal resolutions, down-sampling of data was performed by averaging every 2, 3, 4, and 5 time frames, which generated data sets with temporal resolutions of 6.8 s, 10.2 s, 13.6 s, and 17 s (original temporal resolution=3.4 s). It must be noted that averaging the signal to generate down-sampled data may not be identical to acquiring data at slower sampling rate. Particularly, averaging the data points increases the SNR of the time course of the down-sampled data to a larger degree than for the actual data acquired with lower temporal resolutions.

The original data were processed in the same manner as what was explained in Chapter 5 (Section 5.5). For processing the down-sampled data, first concentration of contrast agent in the tissue, $C_t(t)$, was calculated from down-sampled T1W images and the PD image in a similar manner as what was explained in Section 5.5. Then, by using the extended Tofts model, $C_t(t)$ was related to the concentration of contrast agent in plasma, $C_p(t)$. For each patient, the
functional form of $C_p(t)$ was determined by fitting the concentration of contrast agent in the voxels within the external iliac or femoral arteries to a double Gaussian function (67). By fitting the $C_i(t)$ to the extended Tofts model, maps of $K^{\text{trans}}$, $v_e$, and $v_p$ were generated. For each of the original and down-sampled data sets, the fitting procedure was performed twice; once with and once without using the alternative data analysis technique (which was explained in section 6.3). In both cases the functional form of $C_p(t)$ was used for numerical calculation of the integral term in Equation 6-1. Using the alternative data analysis technique, the integral term was numerically calculated with 1 s element sizes, regardless of the temporal spacing of $C_i(t)$. Without using the alternative analysis technique, the integral term was numerically calculated with the same element size as temporal spacing of $C_i(t)$.

Average values of MR parameters were calculated within a total of 304 (186 non-malignant, and 118 cancers) ROIs. Statistical analyses were performed using MedCalc. Diagnostic accuracy of individual and combined MR parameters was investigated by using ROC analysis. Multi-parametric ROC analyses were performed using logistic regression, while in order to prevent data overfitting only parameters that contributed significantly to the model were used in the calculations. Values of AUC were calculated separately in PZ, TZ, and the entire prostate.

### 6.4.2 Results

Graphs of AUC values as a function of DSF, in PZ, TZ, and the entire prostate are shown in, Figure 6-4, Figure 6-5, and Figure 6-6, respectively. Results of multi-parameter ROC analyses are summarized in Table 6-1.
Figure 6-4. Graphs of area under the (ROC) curve (AUC) as a function of down-sample factor (DSF) in the peripheral zone. The green and the red graphs represent the cases where the data were analysed with and without using the alternative technique (that was explained in section 6.3), respectively.
Figure 6-5. Graphs of AUC values as a function of DSF in the transition zone.

The green and the red graphs represent the cases where the data were analysed with and without using the alternative technique (that was explained in section 6.3), respectively. Multi-parametric ROC analyses were performed using logistic regression, while only parameters that contributed significantly to the model were used in the calculations. For DSF=3 multi-parametric ROC analysis could not be performed as no parameter was retained in the model.
Figure 6-6. Graphs AUC values as a function of DSF in the entire prostate. The green and the red graphs represent the cases where the data were analysed with and without using the alternative technique (as explained in section 6.3), respectively.
Table 6-1. Results of multi-parametric ROC analyses.

<table>
<thead>
<tr>
<th>Location</th>
<th>Fitting Technique</th>
<th>Characteristic</th>
<th>DSF=1</th>
<th>DSF=2</th>
<th>DSF=3</th>
<th>DSF=4</th>
<th>DSF=5</th>
</tr>
</thead>
<tbody>
<tr>
<td>PZ(^a)</td>
<td>Without AT(^a)</td>
<td>Contributing Parameters</td>
<td>(K_{trans})</td>
<td>(K_{trans},v_p)</td>
<td>(K_{trans})</td>
<td>(K_{trans},v_p)</td>
<td>(K_{trans},v_e,v_p)</td>
</tr>
<tr>
<td></td>
<td>With AT</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TZ(^b)</td>
<td>Without AT</td>
<td>Contributing Parameters</td>
<td>(v_e)</td>
<td>(v_e)</td>
<td>(\Delta N/A)</td>
<td>(K_{trans},v_e,v_p)</td>
<td>(v_e,v_p)</td>
</tr>
<tr>
<td></td>
<td>With AT</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Entire Prostate</td>
<td>Without AT</td>
<td>Contributing Parameters</td>
<td>(K_{trans})</td>
<td>(K_{trans},v_e)</td>
<td>(K_{trans},v_e)</td>
<td>(v_e,v_p)</td>
<td>(K_{trans},v_e,v_p)</td>
</tr>
<tr>
<td></td>
<td>With AT</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

AT\(^*\): Alternative (data analysis) technique, DSF: down-sample factor, PZ: peripheral zone, TZ: transition zone, \(\Delta \): For DSF=3 Multi-parametric ROC analysis could not be performed in TZ since no parameter was retained in the model.

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Overall, our results showed a non-linear relationship between the AUC values and temporal resolution in all locations. And, we did not observe a noticeable difference between AUC values obtained with or without using the proposed alternative data analysis technique.

We had a few interesting observations: 1) In the multi-parametric ROC analysis in PZ and the entire prostate, there is not a considerable difference between AUC values that are obtained for different temporal resolutions, ranging from 3.4s to 13.6 s. 2) for $v_p$, the impact of temporal resolution on AUC values observed here is not as dramatic as the impact of temporal resolution on the accuracy of calculation of $v_p$ which was observed in the simulation study, Section 6.2. This can be explained by hypothesizing that although a decrease in temporal resolution increases the uncertainty in the calculation of $v_p$, if the $v_p$ of both tumour and normal tissue vary in the same manner (i.e. both decrease or both increase), it is possible to have the same contrast between tumour and normal tissue in $v_p$ maps of different temporal resolutions.

6.5 Discussion and Conclusions

In this study, first we investigated the impact of temporal resolution on the accuracy of calculation of fitting parameters of Equation 6-1 by performing a simulation study. From this investigation, we concluded that a decrease in the temporal resolution has the highest impact (as decrease of accuracy) on $v_p$, less impact on $K^{\text{trans}}$, and almost no impact on $v_e$.

Second, we proposed an alternative data analysis technique to increase the accuracy of calculation of fitting parameters of Equation 6-1 for data that have been acquired with low temporal resolution. We verified the suitability of this technique against a standard data
analysis technique, using simulation. The results showed that our proposed technique successfully decreases the calculation error of all fitting parameters, particularly $v_p$.

Finally, we investigated the impact of temporal resolution on the accuracy of detecting prostatic tumours, using patient data. Our results demonstrated that when temporal resolution varies within a certain range (3.4-13.6 s), the diagnostic accuracy of DCE-MRI is not affected considerably. This result may be surprising at first, especially considering that following the bolus injection the contrast agent in arterial blood increases very fast, such that this signal may contain significant high frequency components. Thus, in order to fully capture the shape of the signal the Nyquist criterion must be satisfied by sampling the signal at greater than twice the rate of the highest frequency components. Using lower sampling rates results in a greater fitting error and inaccurate values of the fit parameters. However, our results show that cancer detection accuracy is not significantly affected by the temporal resolution. A possible reason for that is that, the calculated fit parameters for both tumour and normal tissue may be affected by a systematic error to a similar degree, resulting in a similar contrast between the tumour and normal tissue for different temporal sampling rates.

The significance of this finding can be understood by considering that if two temporal resolutions provide similar accuracy, the lower temporal resolution may be preferred to allow for improvement of spatial resolution or SNR (while maintaining the same scan time) to achieve better results. An increase in spatial resolution of DCE-MRI decreases false-negative diagnoses and improves specificity as demonstrated by Furman-Haran et al. (82).
Chapter 7: Conclusions

7.1 Summary

In this work, LWI, a new quantitative $T_2$ mapping technique, has been developed for diagnosis and grading of prostate cancer. This technique was developed to measure the fractional amount of water content in luminal space of prostate tissue. As the fractional amount of luminal space is different between normal and malignant tissue, and between tumours of different grades, it was hypothesized that this technique can be used for both detection and grading of prostatic tumours.

We developed the LWI protocol based on a series of simulations and phantom studies. The preliminary protocol was then improved and validated by acquiring data from a number of healthy volunteers.

We investigated the correlation between parameters measured with LWI, particularly LWF, and the corresponding tissue composition in prostate. The results demonstrated that LWF is significantly and strongly correlated with the percentage area of luminal space in the prostatic tissue. Hence, our hypothesis that LWF represents the fractional amount of MR signal generated by water content of luminal spaces in prostate was verified.

We evaluated the diagnostic accuracy of the proposed MRI technique by performing a patient study. In this pilot study, we demonstrated the feasibility of LWI for diagnosis of prostate cancer. Our results showed that LWI measurements provide high accuracy in detection of tumours, and strong correlation with Gleason score.
Additionally, we compared the diagnostic accuracy of LWI with the more established MP-MRI protocols: DCE and DW-MRI. We also investigated the significance of combining LWI with DCE and/or DW-MRI, in the accuracy of detection and grading of prostatic tumours. The results showed that LWI alone performs better than DCE and DW-MRI in the detection of tumours as well as in correlation with GS. LWI, when combined with either DCE or DW-MRI, provided significantly higher accuracy than either of the other two techniques individually or combined. Combination of all three techniques did not provide significant improvement on the accuracy that is obtained from combination of LWI with either DCE or DW-MRI. Therefore, it was concluded that a more efficient, abbreviated MP-MRI protocol for the diagnosis of prostate cancer could be achieved by combining LWI with either DCE or DW-MRI, but not necessarily with both.

Finally, as a side project, we investigated the impact of temporal resolution on the accuracy of DCE-MRI in detection of prostatic tumours. Our results showed that for a temporal resolution ranging between 3.4s to 13.6 s, the diagnostic accuracy of DCE-MRI in prostate cancer is independent of the temporal resolution.

7.2 Contributions

The contributions of this thesis can be summarized as follows:

- Development of a novel MRI technique for measurement of luminal water content in prostate.
A clinically applicable protocol was developed for non-invasive measurement of luminal water content in prostate.

- Verification of the hypothesized relationship between MR measurement of luminal water and the fractional amount of luminal space in prostatic tissue.

Correlation between LWF and the fractional amount of luminal space in prostatic tissue was evaluated. Establishment of the relationship between LWF and fractional amount of luminal space allows for potential application of LWI for the diagnosis and evaluation of prostatic diseases (including but not limited to prostate cancer) in which the extent of luminal space differs between normal and abnormal tissues.

- Clinical evaluation of diagnostic accuracy of LWI.

Preliminary clinical assessment of LWI for the diagnosis of prostate cancer was performed, and the feasibility of this technique in detection and grading of prostatic tumours was demonstrated. Incorporation of LWI in clinical settings could potentially result in a significant increase in the accuracy of detection and grading of prostate cancer.

- Evaluation of the diagnostic accuracy of LWI compared to DCE and DW-MRI.

A detailed comparison of the diagnostic accuracy of LWI with the standard MP-MRI protocols was performed. It was demonstrated that LWI performs better than DCE and DW-MRI in diagnosis of prostate cancer; therefore, this technique could become clinically preferred to DCE-MRI as it does not require administration of any contrast agent. Substitution of DCE-
MRI by LWI saves the cost of contrast agent and intravenous injection materials; moreover, LWI could be the alternative option for patients who cannot undergo DCE-MRI at all due to the potential adverse reaction to a gadolinium-based contrast agent. Finally, substitution of DW-MRI by LWI could be the alternative option for patients with metallic hip implants, for whom DWI is likely to be non-diagnostic.

- Proposing a potential area of improvement in accuracy of DCE-MRI for diagnosis of prostate cancer.

The impact of temporal resolution on the accuracy of DCE-MRI in the detection of prostatic tumours was investigated. It was demonstrating that when temporal resolution varies within a certain range, the diagnostic accuracy of DCE-MRI is not affected considerably; hence a lower temporal resolution can be used, allowing for improvement of spatial resolution or SNR compared to a higher temporal resolutions, to achieve better results.

### 7.3 Future Work

The main limitation of the LWI pilot studies in this thesis is having access to the data from a relatively low number of patients, and also, a limited range of Gleason Scores. Another limitation of LWI studies is the lack of distinction between normal gland, and BPH. Another important area of improvement for LWI is reduction in its scan time. Based on above limitations, areas of future research with our developed LWI technique are as follows:
- Expanding the study of diagnostic accuracy of LWI with a larger number of patients and wider range of Gleason Scores with larger numbers in each category to better access the accuracy of this technique.

- Comparing LWI with the current PI-RADS (version 2) protocol to further assess the clinical utility of LWI.

- Further development of our proposed LWI technique to reduce its acquisition time by increasing the GRASE factor and/or under-sampling the k-space in conjunction with the Compressed Sensing reconstruction.

- Investigating the accuracy of LWI in distinction between normal tissue and BPH.

For the DCE-MRI study, the future research direction could include:

- Performing a simulation study to investigate the impact of temporal resolution on the contrast between the calculated physiological parameters of a presumed normal tissue and a presumed malignant tissue. This result can be compared to our clinical result to better characterize the relationship between the temporal resolution and the diagnostic accuracy of DCE-MRI.
References


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Appendix

Effect of the T1 relaxation on the measurement of Luminal Water Fraction

In Chapter 2, for the development of a mathematical model to describe the T2 decay in prostatic tissue, the effect of T1 relaxation was ignored. In this appendix, we will demonstrate the effect of including T1 relaxation in the modeling of the T2 decay.

By including the effect of T1 relaxation in the decay model the signal decay which was previously described by Equation 2-2, can be expressed as below:

\[ S(t) = \sum_{i=0}^{N} A_i e^{-\frac{t}{T_{2i}}}(1 - e^{-\frac{t}{T_{1i}}}) \]  

Equation A-1

where \( S(t) \) is the signal magnitude at a time t, \( A \) is the amplitude of the signal at time zero, and \( T_{2i} \) and \( T_{1i} \) are respectively the T2, and T1 relaxation times of water compartment i.

As described in chapters 2 and 3, in the absence of T1 relaxation effects, LWF is defined as:

\[ LWF = \frac{A_{long}}{A_{short} + A_{long}} \]  

Equation A-2

where \( A_{short} \) and \( A_{long} \) are the maximum amplitudes of the short and long T2 components, respectively.

By considering the effect of T1 relaxation, the \( A_{short} \) and \( A_{long} \) terms of Equation A-2 can be expressed as below:
\[ A_{\text{short}} = A'_{\text{short}} \left(1 - e^{-\frac{TR}{T_{1\text{-short}}}}\right) \]  
Equation A-3

\[ A_{\text{long}} = A'_{\text{long}} \left(1 - e^{-\frac{TR}{T_{1\text{-long}}}}\right) \]  
Equation A-4

where \( T_{1\text{-short}} \) and \( T_{1\text{-long}} \) are the \( T_1 \) relaxation times of the small and the large water compartments, respectively, and \( A'_{\text{short}} \) and \( A'_{\text{long}} \) are the maximum amplitudes of the short and long \( T_2 \) components when the effect of \( T_1 \) relaxation is taken into account.

By substituting Equation A-3 and Equation A-4 in Equation A-2 LWF can be expressed as below:

\[
LWF = \frac{A_{T_{1\text{-long}}}(1 - e^{-\frac{TR}{T_{1\text{-long}}}})}{A_{T_{1\text{-short}}}(1 - e^{-\frac{TR}{T_{1\text{-short}}}}) + A_{T_{1\text{-long}}}(1 - e^{-\frac{TR}{T_{1\text{-long}}}})}.
\]  
Equation A-5

From Equation A-5 we can see that LWF no longer represents the fractional amount of water within the luminal space, unless \( T_{1\text{-short}} \) and \( T_{1\text{-long}} \) are equal in value. Therefore we can define the true luminal water fraction as:

\[
LWF' = \frac{A_{T_{1\text{-long}}}}{A_{T_{1\text{-short}}} + A_{T_{1\text{-long}}}}.
\]  
Equation A-6

By comparing Equation A-5 and A-6 it can be realized that the measurements of luminal water fraction that were reported in Chapters 2 - 5 were in fact \( T_1 \)-weighted measures of the true fractional amount of luminal water content.
To better understand the effect of $T_1$ relaxation on the measurements of luminal water fraction, let us perform a very simplistic analysis. In this analysis we calculate the values of LWF and LWF’ for some assumed values of $A'_\text{short}$, $A'_\text{long}$, $T_1$-short, $T_1$-long and TR. $A'_\text{short}$ and $A'_\text{long}$ are assumed to be equal to $A_{\text{short}}$ and $A_{\text{long}}$ measured in normal PZ ($165\times10^5$, and $52\times10^5$ respectively, see Table 4.2). $T_1$-short is assumed to be equal to the $T_1$ relaxation time in human muscle (1250 ms) at 3T, and $T_1$-long is assumed to be equal to the $T_1$ relaxation time of free water (4000 ms). TR = 1473 ms (which is the effective TR of our LWI protocol = 3073 ms – (64×25 ms)). By substituting these values in Equations A-5 and A-6, LWF and LWF’ values will be calculated as: 0.12, and 0.24, respectively. In this example, the effect of ignoring $T_1$ relaxation appears as underestimation of the true luminal water fraction.

Now, let us perform another simple analysis and calculate the ratio of LWF/LWF’ for two more general cases where; 1) $T_1$-long is presumed to be equal to 4000 ms but $T_1$-short varies between 800ms and 1600ms, and 2) $T_1$-short is presumed to be equal to 1250 ms but $T_1$-long varies between 2500ms and 5000ms. The results of these analyses are shown in figure A-1 and Figure A-2.
Figure A-1. The ratio of LWF to LWF’ for $T_{1\text{-long}}=4000$ ms, and $800\text{ms}<T_{1\text{-short}}<1600\text{ms}$.

Figure A-2. The ratio of LWF to LWF’ for $T_{1\text{-short}}=1250$ ms, and $2500\text{ms}<T_{1\text{-long}}<5000\text{ms}$.

From Figure A-1 and Figure A-2, we can see that for the interested ranges of $T_{1\text{-short}}$ and $T_{1\text{-short}}$
LWF always underestimates LWF’.
In conclusion, $T_1$ relaxation affects measurement of luminal water fraction in prostatic tissue. Ignoring the effect of $T_1$ relaxation in the modeling of $T_2$ decay results in underestimation of the true fractional amount of luminal water. This is the most likely reason for the slope of the correlation between LWF measured with MRI vs. fractional volume of the luminal space measured with histology to be about 0.45 (See the Results section of Chapter 3).

It should be noted that the simplistic analyses presented in this appendix was only performed by using a single set of values for $A'_{short}$, and $A'_{long}$. Determination of the $T_1$ relaxation effect on measurements of luminal water under general conditions requires further investigation in form of an extensive simulation study.