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Automated brain tissue segmentation of Magnetic Resonance Images in Multiple Sclerosis

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- **Sergi Valverde**, Arnau Oliver, Yago Díez, Mariano Cabezas, Joan Carles Vilanova, Lluís Ramió-Torrentà, Àlex Rovira, and Xavier Lladó. Evaluating the effects of white matter multiple sclerosis lesions on the volume estimation of six brain tissue segmentation methods. *American Journal of Neuroradiology*. Vol. 36(6), pp. 1109-1115. 2015. American Society of Neuroradiology. Quality index: [JCR RNMMI IF 3.589, Q1(19/125)].
- **Sergi Valverde**, Arnau Oliver, Mariano Cabezas, Eloy Roura and Xavier Lladó. Comparison of ten brain tissue segmentation methods using revisited IBSR annotations. *Journal of Magnetic Resonance Imaging*. Vol. 41, Issue 1, pp. 93-101. January 2015. Wiley. Quality index: [JCR RNMMI IF: 3.210 Q1(23/125)].
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Journals

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- Eloy Roura, Arnau Oliver, Mariano Cabezas, **Sergi Valverde**, Deborah Pareto, Joan Carles Vilanova, Lluís Ramió-Torrentà, Àlex Rovira and Xavier Lladó. A toolbox for multiple sclerosis lesion segmentation. *Neuroradiology*. Vol. 57(10), pp. 1031-1043. 2015. Springer. Quality index: [JCR RNMMI IF 2.485, Q2(41/125)].
- Mariano Cabezas, Arnau Oliver, **Sergi Valverde**, Brigitte Beltrán, Joan Carles Vilanova, Lluís Ramió-Torrentà, Àlex Rovira and Xavier Lladó. BOOST: a supervised approach for multiple sclerosis lesion segmentation. *Journal of Neuroscience Methods*. Vol. 237, pp 108-117. 2014. Elsevier. Quality index: [JCR N IF 2.025, Q3(174/252)].
- Yago Díez, Arnau Oliver, Mariano Cabezas, **Sergi Valverde**, Robert Martí, Joan Carles Vilanova, Lluís Ramió-Torrentà, Àlex Rovira and Xavier Lladó. Intensity based methods for brain MRI longitudinal registration. A study on multiple sclerosis patients. *Neuroinformatics*. Vol 12(3), pp 365-379. 2014. Springer. Quality index: [JCR CSTM IF 2.825, Q1(13/102)]

Conferences

- Eloy Roura, Arnau Oliver, Mariano Cabezas, **Sergi Valverde**, Deborah Pareto, Joan Carles Vilanova, Lluís Ramió-Torrentà, Àlex Rovira and Xavier Lladó. An SPM12 extension for multiple sclerosis lesion segmentation. *SPIE Medical Imaging 2016*. February 2016, San Diego, USA.
- **Sergi Valverde**, Arnau Oliver, Eloy Roura, Deborah Pareto, Joan Carles Vilanova, Lluís Ramió-Torrentà, Jaume Sastre-Garriga, Xavier Montalban, Àlex Rovira and Xavier Lladó. Evaluation of two automated lesion segmentation and filling pipelines for brain tissue segmentation of multiple sclerosis patients.

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- Eloy Roura, Arnau Oliver, Mariano Cabezas, **Sergi Valverde**, Deborah Pareto, Joan Carles Vilanova, Lluís Ramió-Torrentà, Àlex Rovira and Xavier Lladó. A toolbox for segmenting multiple sclerosis lesions using T1w and FLAIR images. *ECTRIMS 2015. Multiple Sclerosis.* October 2015, Barcelona, Spain. Quality index: [JCR CN IF:4.472 Q1(25/191)].
- **Sergi Valverde**, Arnau Oliver, Deborah Pareto, Joan Carles Vilanova, Àlex Rovira , Lluís Ramió-Torrentà and Xavier Lladó. SLF: a MS white matter lesion filling toolbox for the SPM software. *ECTRIMS 2014. Multiple Sclerosis.* September 2014, Boston, USA. Quality index: [JCR CN IF:4.822 Q1(22/192)].
- Ester Quintana, Brigitte Beltrán, **Sergi Valverde**, René Robles-Cedeno, Héctor Perkal, Xavier Lladó, José Manuel Fernández-Real and Lluís Ramió-Torrentà. Expression of miRNAs in multiple sclerosis cerebrospinal fluid and their relation to MR activity. *ECTRIMS 2014. Multiple Sclerosis.* September 2014, Boston, USA. Quality index: [JCR CN IF:4.822 Q1(22/192)].
- Ester Quintana, Brigitte Beltrán, **Sergi Valverde**, René Robles-Cedeno, Héctor Perkal, Xavier Lladó, José Manuel Fernández-Real and Lluís Ramió-Torrentà. Relación entre la expresión de mirnas en LCR y variables de RM. *Neurología*, vol 29, pp 66-67. 2014. Quality index: [JCR CN IF:1.322 Q3(142/191)].
- **Sergi Valverde**, Arnau Oliver, Mariano Cabezas, Yago Díez, Jordi Freixenet, Xavier Lladó, Joan Carles Vilanova, Àlex Rovira and Lluís Ramió-Torrentà. A quantitative study of the effects of White Matter MS lesions on tissue segmentation methods. *ECTRIMS 2013. Multiple Sclerosis.* October 2013, Copenhagen, Denmark. Quality index: [JCR CN IF:4.472 Q1(25/191)].

Acronyms

- ANN** Artificial Neural Network
BET Brain Extraction Tool
BSE Brain Surface Extractor
CNS Central Nervous System
CSF Cerebrospinal Fluid
CIS Clinically Isolated Syndrome
EDSS Extended Disability Status Scale
FANTASM Fuzzy and Noise Tolerant Adaptive Segmentation Method
FAST FMRIB's Automated Segmentation Tool
FCM Fuzzy-C Means
FLAIR Fluid Attenuated Inversion Recovery
FMRIB Oxford Centre for Functional MRI of the Brain
FSL FMRIB Software Library
GAMIXTURE Image segmentation toolbox based on genetic algorithm and mixture model optimization
GM Gray Matter
IBSR Internet Brain Segmentation Repository
LST Lesion Segmentation Toolbox
MARGA Multispectral Adaptive Region Growing Algorithm
MRI Magnetic Resonance Image
MRBrainS13 Magnetic Resonance Brain Segmentation Challenge 2013
MS Multiple Sclerosis
KNN K-Nearest Neighbor
PD Proton Density
PVC Partial Volume Classifier
SLS SALEM Lesion Segmentation
SPASEG Image segmentation toolbox based on local Markov random fields
SPM Statistical Parametric Mapping
T1-w T1-weighted
T2-w T2-weighted
WM White Matter

Abstract

Resum

Resumen

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Chapter 1

Introduction

In this first chapter, we first introduce the reader to the research context and background of the thesis, situating this work within the research line of our group. Afterwards, we describe the proposed objectives and the respective stages to cover. Finally, we summarize the main structure of this thesis, highlighting the conceptual thread between each of the articles that compose its main core.

1.1 Research context

1.1.1 Multiple Sclerosis

The human nervous system can be divided into the central nervous system (CNS) consisting on the brain and the spinal chord, and the peripheral nervous system, which connects the CNS with the sense organs [7]. CNS is mainly constituted by two tissue components: gray matter (GM), which consists of neuronal cell bodies; and white matter tissue (WM), which is mainly composed of myelinated axon tracts [63]. In the case of the brain, it is mostly composed by GM and WM, both evolved by the Cerebro-spinal fluid (CSF), which provides basic mechanical and immunological protection to the brain inside the skull [63].

Multiple sclerosis (MS) is the most common chronic immune-mediated disabling neurological disease of the CNS [64], in which the insulating covers of the nerve cells in the spinal chord and brain are damaged [18]. Nowadays, MS is the most frequent non-traumatic neurological disease that causes more disability in young adults. It follows a similar behavior also seen in other putative autoimmune diseases, and affects twice as many women as it does in men [19]. It has a low incidence in childhood, but it increases rapidly in adulthood reaching a peak between 25 and 35

years, and then slowly declines, becoming rare at 50 and older [8]. So far, the world estimate for the disease is between 1.3 to 2.5 million cases, being relatively common in Europe, the United States, Canada, New Zealand, and parts of Australia, but rare in Asia, and in the tropics and subtropics [8].

MS is characterized by areas of inflammation, demyelination, axonal loss, and gliosis scattered throughout the CNS, often causing motor, sensorial, vision, coordination, deambulation, and cognitive impairment [17]. Demyelination is the process of progressive damage to the protective covering (myelin sheath) around the axon of the neurons. Demyelinated axons conduct impulses at reduced or spontaneous velocity causing impairment in sensation, movement and cognition [18]. The different clinical courses of the disease are generally grouped into four subtype forms [43]. The *Relapsing/Remitting* (RRMS) form of the disease is characterized by exacerbation times where symptoms are present. These periods are followed by periods of remission, where the patient recovers partial or totally from the disease symptoms. The *Secondary Progressive* (SPMS) form is characterized by a gradual intensification of symptoms between affection relapses. The *Progressive remitting* (PRMS) form is typified by an increase in the relapsing times with significant recovery but with worsening symptoms in new relapsing intervals. Lastly, the *Primary Progressive* (PPMS) form is characterized by a severe decrease of remitting times with special localization in the brain. In general, 50% of RRMS patients after 10 years develop the SPMS form of the disease. After 25 years, the 90% of RRMS patients would develop the SPMS form [43].

1.1.2 Magnetic Resonance Imaging in MS

Magnetic Resonance Imaging (MRI) is a noninvasive medical imaging technique that is used in radiology to generate image representations of different internal anatomical organs and physiological processes of the body. Over the last 40 years, MRI have evolved as a clinical modality [33], and in particular as an essential tool for the diagnosis and evaluation of central nervous system disorders such as MS [24]. On MRI, MS plaques are well-delimited regions with hypointense signal intensity with respect to GM on T1-weighted (T1-w), while hyperintense with respect to GM on T2-weighted (T2-w), Proton Density-weighted (PD-w) and Fluid Attenuated Inversion Recovery (FLAIR) modalities (see Figure 1.1).

In this aspect, new criteria for MS diagnosis and monitoring has been revised in the last years [48], due to the MRI sensitivity to reveal focal white matter (WM) lesions and disease activity in time and space [26]. Additionally, various studies have also analyzed the correlation between MRI brain tissue atrophy measures and MS disability status, showing that tissue loss is an important component of the disease progression [14, 25, 27, 56]. Tissue loss seems to increase through the course of MS

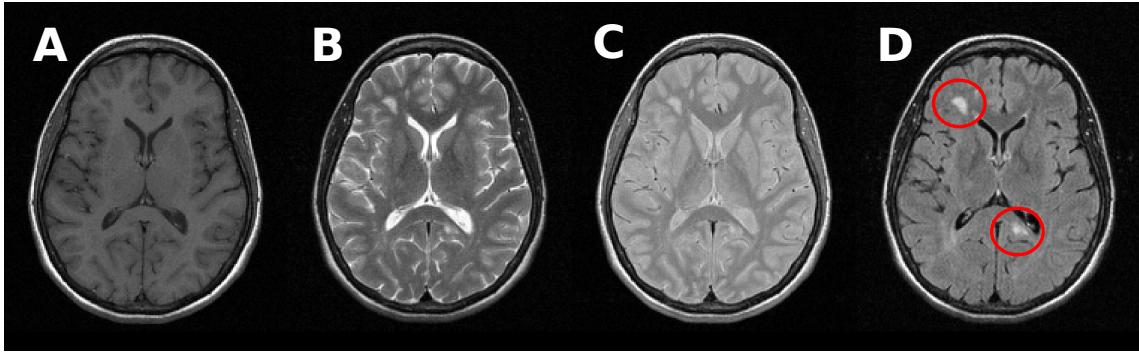


Figure 1.1: MRI image modalities. A) T1-weighted (T1-w) image sequence. B) T2-weighted (T2-w) image sequence. C) Proton Density-weighted (PD-w) image sequence. D) Fluid Attenuated Inversion Recovery (FLAIR) sequence. MS plaques are shown inside red circles on the FLAIR modality. MS plaques are hypointense with respect to GM and WM in T2-w, PD-w and FLAIR sequences, while hypointense with respect to WM on the T1-w modality.

with a similar rate between 0.3% and 0.5% per year, and independently of the MS subtype [21, 56]. In general, GM atrophy is more associated with disability changes than WM atrophy [28], and not only in the RRMS and SPMS MS subtypes [27, 56], but also in Clinically Isolated Syndrome (CIS) patients where several studies have shown a significantly greater ventricular cavities and an associated GM loss on MRI scans of CIS patients that will develop MS compared to those who not [13, 25].

1.1.3 Image analysis in MS

Manual analysis of brain images is unfeasible in practice, given the large number of two-dimensional slices of each three-dimensional MRI patient image and the possible intra/inter observer variability between experts [8]. This has led to the development since the early nineties of a wide number of lesion and tissue segmentation methods, with the aim to reduce the time of manual interaction and the inherent variability of manual annotations [16, 32, 39].

Pre-processing of MRI images

Acquired brain MRI volumes incorporate non-brain tissue parts of the head such as eyes, fat, spinal cord or brain skull. Brain tissue extraction from non-brain tissue is commonly referred in the literature as skull-stripping (see Figure 1.2 B and C). Skull-stripping has a direct effect on the performance of automated methods, as differences

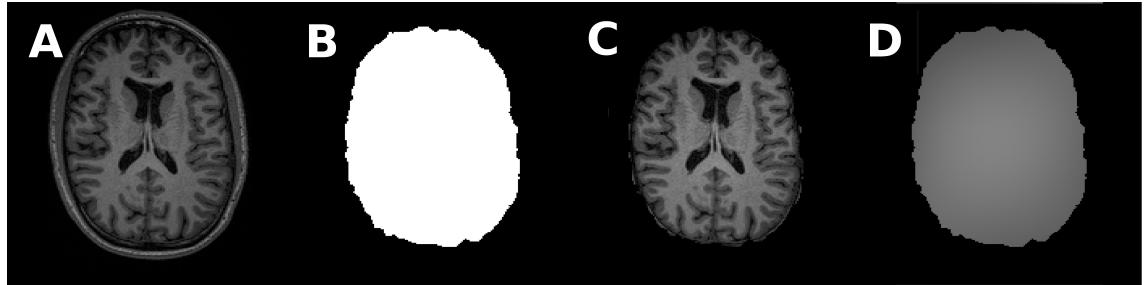


Figure 1.2: MRI pre-processing steps. A) T1-weighted (T1-w) image sequence. B) Computed brain mask using the BET approach [62] and C) skull stripped T1-w sequence . D) Estimated T1-w bias-field using the N3 method proposed by [61].

in skull stripping would lead into unexpected results in the tissue classification if skull or eyes are included as brain tissue [1, 49]. Among the different proposed methods for skull-stripping [1, 40, 54], the Brain Extraction Tool (BET) [62] and the Brain Surface Extractor (BSE) [59] are the most commonly used methods by the neuroimaging community.

Furthermore, inherent characteristics of the MRI acquisition process such as differences in the magnetic field, bandwidth filtering of the data or eddy currents driven by field gradients usually derive in image artifacts that may also have a negative impact on the performance of methods [60]. In these cases, intensity correction of MRI images is either performed before lesion/tissue segmentation, or as an integrated part of the tissue segmentation pipeline (see Figure 1.2 D). Among the former available strategies proposed [3, 37], the N3 [61] and N4 [68] methods are currently the most widely used tools used for intensity correction.

Automated lesion segmentation

MRI based diagnostic criteria for MS has led to an increasing need to analyze quantitatively focal MS lesions in individual and temporal studies [9, 48]. Different sequences such as T2-w, PD-w and FLAIR are often used in lesion detection and segmentation, as MS lesions appear brighter than GM and WM on them. However, WM lesions often present a similar signal intensity profile to CSF on T2-w. In contrast, FLAIR sequences suppress fluids from the image, restraining the CSF tissue effects on the acquired image, although some severe T2-w hyperintense lesions appear similar to CSF in FLAIR [36].

A wide number of automated lesion segmentation techniques have been proposed during the last years [30, 42]. In these methods, lesion segmentation is based either in supervised or unsupervised strategies. Supervised methods employ a training

set of correctly-identified observations that are used as prior information to learn the lesion characteristics. Newer proposed strategies integrate spatial decision forest [31], statistical methods [65], patch-based models [35] or adaptive dictionary learning strategies [22]. In contrast, unsupervised learning methods do not use any prior information in the segmentation task, which involves grouping data into categories based on some measure of inherent similarity or distance characteristic of the input images. Among these, most recent methods include probabilistic models which separate WM lesions from normal-appearing tissue by considering lesions as an outlier class [36, 38, 67], or techniques that make use of the signal intensity of lesions on FLAIR to apply several thresholding methods with post-processing steps to automatically segment lesions [53, 57].

Automated brain tissue segmentation in MS

The existent correlation between brain tissue atrophy measures and MS disability status [25, 27], has increased the necessity to develop robust automated brain tissue segmentation methods capable to perform accurate brain tissue volume measurements [34]. However, automated segmentation of brain tissue is still a challenging problem due to the complexity of the images, existence of lesions, differences in tissue intensities, noise, intensity inhomogeneities and the absence of models of the anatomy that fully capture the possible deformations in each structure [8, 39].

A wide number of brain tissue segmentation methods have been proposed so far. General purpose intensity based methods usually perform tissue segmentation on T1-w sequences, as this modality clearly separates gray matter from white matter. These include probabilistic strategies based on Bayesian inference [4, 46, 55, 59], Markov Random Fields models [6, 66, 72], or unsupervised clustering methods [11, 47]. In contrast, supervised learning approaches also combine T1-w sequences with other modalities such as T2-w and PD-w using *K-Nearest-Neighbor* classifiers [20, 70], *Support Vector Machines* [2, 69], *Random Forests* [71, 45], or trained *Gaussian mixture models* [51].

However, different studies have shown that tissue abnormalities found in MS image patients such as WM lesions reduce the accuracy of tissue segmentation methods [5, 15]. Effectively, WM lesions on T1-w are hypointense with respect to normal-appearing WM, and therefore, lesion voxels that are classified as GM are distorting the overall GM volume. However, lesion voxels may also have an effect in the observed differences in normal-appearing tissue. WM lesions which are actually classified as WM decrease the mean overall signal intensity of the WM, causing that GM voxels with signal intensities similar to WM lesions may be also miss-classified as WM. In contrast, if WM lesions are classified as GM, normal-appearing WM voxels with signal intensities similar to lesions may be miss-classified as GM.

Lesion filling

In MS, hypointense WM lesions have to be pre-processed before tissue segmentation in order to reduce the effects of WM lesions on tissue segmentation. Historically, WM lesions have been masked-out of the T1-w before segmentation, and their volume have been added to WM afterwards [14]. Although this method effectively reduces the error in tissue volume, it has been shown in several studies that this approach is not optimal [5, 15].

In this aspect, several strategies have proposed to in-paint lesions on the T1-w with signal intensities of the normal-appearing WM before tissue segmentation [5, 15, 44, 58], a process which is usually known in the literature as lesion filling. However, most of the available lesion filling methods require manual delineations of lesions, which may be tedious, challenging and time-consuming task depending of the characteristics of the image [42]. When available, lesion filling has demonstrated not only a significant reduction in the associated errors of WM lesions in tissue volume measurements [50], but also in image registration [12, 23, 58] and cortical thickness measurements [44].

1.2 Research background

This thesis is located within the framework of different research projects associated to the Computer Vision and Robotics Institute (VICOROB) of the University of Girona¹. VICOROB has been working on numerous medical image analysis projects since 1996, mainly in segmentation and registration of mammography images. Lately in 2009, the research group started a fruitful collaboration with several medical MS research teams with the aim to develop new automated techniques capable to segment MS lesions and to perform atrophy measurements that can be transferred to experts for clinical use. In particular, our research in the MS field has been carried out within the following research projects:

1. [2009 – 2012] PI09/91918 “SALEM: Segmentación Automática de Lesiones de Esclerosis Múltiple en imágenes de resonancia magnética” awarded by the Instituto Carlos III.
2. [2009 – 2012] VALTEC09-1-0025 “SALEM: Eines per a la segmentació automàtica de lesions d’Esclerosi Múltiple en ressonància magnètica” awarded in 2009 by the Generalitat de Catalunya within the “Projectes de valorització VALTEC”.

¹<http://vicorob.udg.edu>

3. [2015–2017] TIN2014-55710-R: NICOLE: “Herramientas de neuroimagen para mejorar el diagnóstico y el seguimiento clínico de los pacientes con Esclerosis Múltiple” awarded in 2014 by the spanish call Retos de investigación 2014.
4. [2015 – 2019] BiomarkEM.cat: “New technologies applied to clinical practice for obtaining biomarkers of atrophy and lesions in magnetic resonance images of patients with multiple sclerosis”. Awarded in 2015 by the Fundació la Marató de TV3.

Since then, the research group has published original contributions in different fields such as image pre-processing [54], MS lesion segmentation [9, 10, 42, 53], temporal analysis [29, 41], image registration [23, 52], and tissue segmentation [8]. All the works have been done in collaboration with different medical MS teams from:

- The Hospital Vall d’Hebron: Dr. Rovira, who is the director of the “Unitat de Ressonància Magnètica-Centre Vall d’Hebron” (URMVH) and has participated in numerous research projects funded by public and private institutions in the last few years, as well as Dr. Pareto and technicians Huerga and Corral. This group is part of the MAGNIMS network, a European network of centers that share an interest in the MS study through MRI.
- The Clínica Girona / Hospital Santa Caterina: Dr. Vilanova and Dr. Barceló are the codirectors of the “Unitat de Ressonància Magnètica” at the Clínica Girona and are members of several national and international radiology societies.
- The Hospital Josep Trueta: Dr. Ramió-Torrentà, who is the current coordinator of the ”Unitat de Neuroimmunología i Esclerosis Múltiple”, as well as Drs. Robles and Beltrán, who work for the radiology unit.

1.3 Objectives

As part of the SALEM, NICOLE and BiomarkEM.cat research project frameworks, the main goal of this thesis is:

to develop a novel fully automated brain tissue segmentation method capable of computing accurate tissue volume measurements on images of MS patients.

Different stages have to be covered first in order to fulfill the main proposed goal. All them can be considered as sub-objectives that allow us to gain a better knowledge of the different parts that compose a fully automated tissue segmentation method for MS images containing lesions. In what follows, we detail these proposed sub-goals:

- **to analyze and evaluate the state-of-the-art of tissue segmentation methods.** This stage aims to quantitative review and evaluate the different proposed tissue segmentation techniques in order to understand their advantages and drawbacks. In order to fulfill this goal, we plan to perform different experiments using public databases that incorporate manual tissue annotations, which will allow to perform a quantitative evaluation of the accuracy of the methods.
- **to study and evaluate the effect of WM lesions on tissue segmentation of MS patient images.** Although it is known that the inclusion of WM lesions on tissue segmentation distort the brain volume measurements, this effect has not been studied and compared across different tissue segmentation methods. In this aspect, the second stage is focused on the analysis of the effects of WM lesions on the tissue distributions of a set of tissue segmentation approaches. Our hypothesis here is that a better knowledge of the correlation between lesion attributes, such as signal intensity and lesion size, and the observed differences in tissue volume of the analyzed algorithms may be beneficial to design a tissue segmentation method for MS. Hence, we aim to perform several experiments using multi-center 1.5T MS data from different scanners in order to analyze the effects of WM lesions on tissue segmentation.
- **to reduce the effect of WM lesions on tissue segmentation of MS patient images designing and implementing a new lesion filling algorithm.** As said in section 1.1.3, WM lesions have to be pre-processed before tissue segmentation in order to reduce the effects of those lesions on tissue segmentation. In this regard, the third sub-goal is two-fold: firstly, to compare the accuracy of different lesion filling techniques proposed in the literature, analyzing their accuracy on both 1.5T and 3T databases. Secondly, after analyzing the benefits and drawbacks of each proposed method, we aim to propose a new lesion filling algorithm in order to overcome the possible limitations of existent methods.
- **to analyze and evaluate the effect of automated algorithms that perform WM lesion segmentation and filling on the tissue segmentation.** Although lesion filling techniques have already been successfully applied to reduce the effect of WM lesions on tissue segmentation, usually WM lesions

are annotated manually before tissue segmentation. In contrast, the effect of both automated lesion segmentation and filling on tissue segmentation is still unclear. The fourth stage of the thesis aims to understand the effects of the inherent errors in automated lesion segmentation on the posterior lesion filling and tissue segmentation. Thus, we plan to perform several experiments with different pipelines that incorporate automated lesion segmentation, lesion filling and tissue segmentation. Using these experimental data, we aim to evaluate the accuracy of these pipelines on MS data, analyzing and evaluating the extend of the effect of remaining WM lesions on the differences in tissue segmentation, which may be beneficial to update the knowledge gained in previous stages.

- **to propose a new fully automated tissue segmentation method for MS patient images.** Finally, we aim to benefit from the stages to propose a novel **fully automated tissue segmentation method** able to deal with images of MS patients with different level of brain atrophy and lesion load. In this last stage, we aim to validate the accuracy of the proposed method by comparing it with the state-of-the-art in tissue segmentation in MS.

This objective refers to the brain tissue segmentation of MS patient images into GM, WM, CSF in transversal studies. We do not concentrate in differences in tissue volume at different stages, but in the effect of WM lesions in the final tissue segmentation. All these stages will be carried out using not only public databases but also different 1.5T and 3T databases of MS patients from the collaborating hospital centers. **Furthermore, as part of the goals of the research frameworks from which this thesis is located, implementations of all the proposed methods will be publicly available for the research community.**

1.3.1 Document structure

A graphical description of the structure of the thesis linking all the chapters presented is shown in Figure 1.3. Connections between the chapters depict the conceptual link between them. The rest of the document is organized as follows:

- **Chapter 2. Comparison of 10 brain tissue segmentation methods using revisited IBSR annotations.** We present here comprehensive comparison of the accuracy of 10 brain tissue segmentation methods on two public MRI databases. This chapter is based on the paper published in the *Journal of Magnetic Resonance Imaging* in 2015.

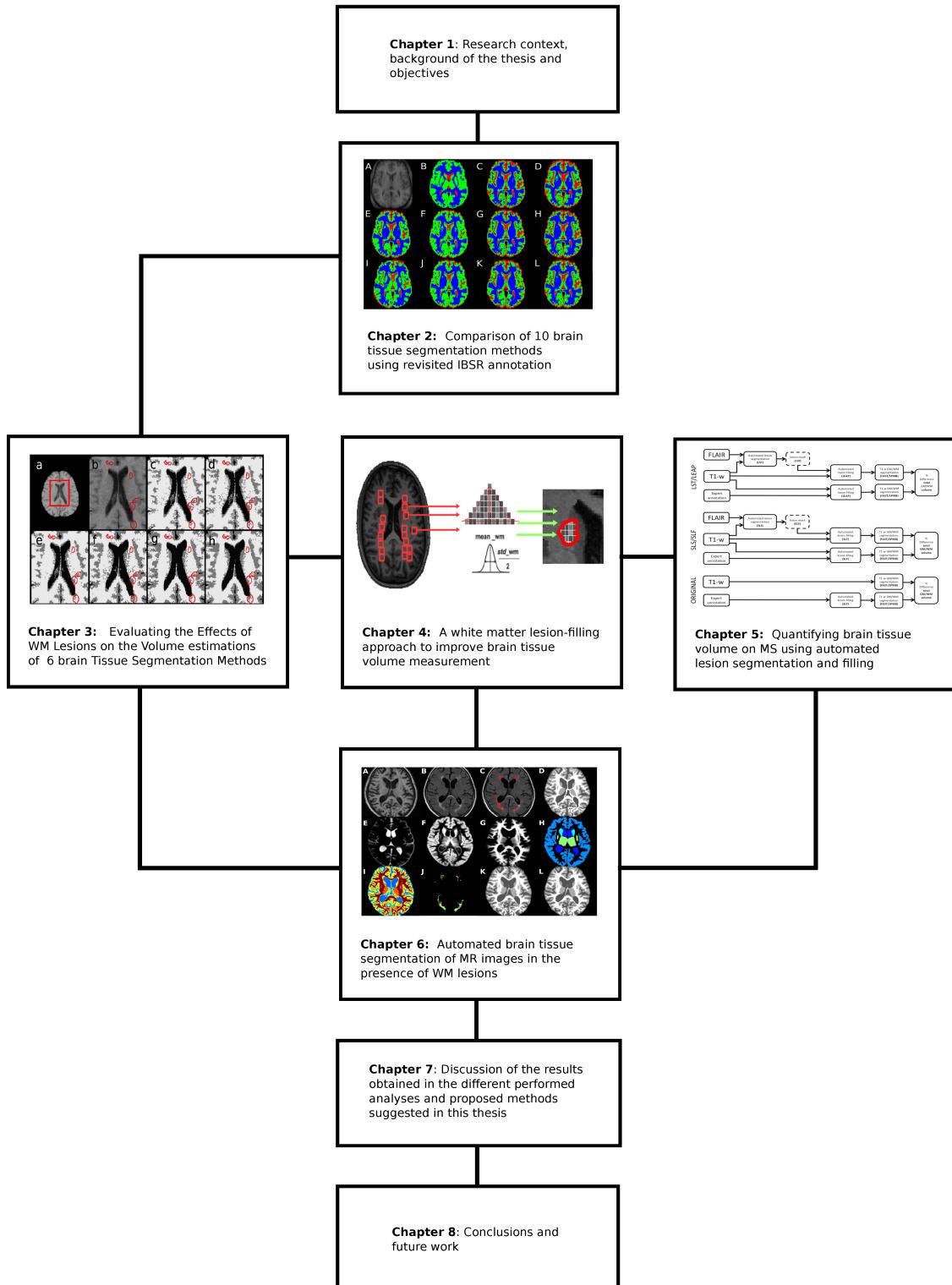


Figure 1.3: Organization of the document. Preliminary chapter 1 describes the research context and the main objectives of this thesis. Chapters 2 to 6 introduce the main contributions of this work based on the different works submitted or published in research journals. Chapter 7 presents a general discussion of the results obtained from Chapters 2 to 6. Finally, the main conclusions and the proposed future work are presented in Chapter 8. Connections between chapters depict a conceptual link between them.

- **Chapter 3. Evaluating the effects of white matter multiple sclerosis lesions on the volume estimation of 6 brain tissue segmentation methods.** After reviewing different tissue segmentation techniques using public data, we perform a detailed analysis of the effects of WM lesions on the brain tissue volume measurements of six of these tissue segmentation methods using MS data from different hospital centers collaborating in the research projects. This chapter is based on the paper published in the *American Journal of Neuroradiology* in 2015.
- **Chapter 4. A white matter lesion-filling approach to improve brain tissue volume measurements.** In this chapter, we propose a new technique to fill WM lesions on 1.5T and 3T data, validating its accuracy with respect to other methods in the literature. This chapter is based on the paper published in the *NeuroImage: Clinical* journal in 2014.
- **Chapter 5. Quantifying brain tissue volume in multiple sclerosis with automated lesion segmentation and filling.** In this chapter we present a detailed evaluation of the performance of different automated pipelines that incorporate lesion segmentation, lesion filling and tissue segmentation on MS data. This analysis is novel in the sense that this is the first work that evaluates two automated pipelines on MS data. This chapter is based on the paper published in the *NeuroImage: Clinical* journal in 2015.
- **Chapter 6. Automated brain tissue segmentation of MR images in the presence of white matter lesions.** We propose here a novel fully automated tissue segmentation pipeline designed to deal with MS patient images containing lesions. We validate the accuracy of the proposed method comparing the performance with other state-of-the-art techniques. Data from the MRBrainS13 challenge as well as data from our hospital collaborators is used to perform the evaluation. This chapter is based on the paper submitted to the *Medical Image Analysis* journal in 2016.
- **Chapter 7. Results and discussion.** This chapter provides a comprehensive discussion of the results obtained in this thesis.
- **Chapter 8. Conclusions and future work.** Finally, the main conclusions based on the contributions of this thesis are defined. Based on these conclusions, we also point out different future works to improve and extend the work carried out in this thesis.

Chapter 2

Comparison of 10 brain tissue segmentation methods using revisited IBSR annotations.

In this chapter, we perform a quantitative evaluation of the accuracy of 10 automated brain tissue segmentation methods. Methods are compared based using the Internet Brain Segmentation Repository (IBSR) databases IBSR20 and IBSR18¹. The performance of the methods is evaluated by ranking their accuracy based on the significant differences with respect the rest of methods. This proposed evaluation has been published in the following paper:

Paper published in **Journal of Magnetic Resonance in Medicine (JMRI)**
Volume: 41, Issue: 1, Pages: 93-101, Published: January 2015
DOI: 10.1002/jmri.24517
Quality Index: 3.21 (Quartile 1)

¹<https://www.nitrc.org/projects/ibsr/>

Chapter 3

Evaluating the Effects of White Matter Multiple Sclerosis Lesions on the Volume Estimation of 6 Brain Tissue Segmentation Methods.

In this chapter, we present an study of the impact of MS white matter lesions on the brain tissue measurements of six well-known segmentation techniques. These include straightforward techniques such as Artificial Neural Network (ANN) and fuzzy C-means (FCM) as well as more advanced techniques such as the Fuzzy And Noise Tolerant Adaptive Segmentation Method (FANTASM), FMRIB's Automated Segmentation Tool (FAST), and Statistical Parametric Mapping (SPM) with versions SPM5 and SPM8. This proposed evaluation has been published in the following paper:

Paper published in the **American Journal of Neuroradiology (AJNR)**
Volume: 36, Pages: 1109-1115, Published: February 2015
DOI: 10.3174/ajnr.A4262
Quality Index: 3.59 (Quartile 1)

Chapter 4

A white matter lesion-filling approach to improve brain tissue volume measurements.

In this chapter, we propose a new technique to fill WM lesions before tissue segmentation. The proposed approach is evaluated in both 1.5T and 3T data. We validate our method comparing its accuracy with other proposed automated lesion filling methods on the same data. Furthermore, the proposed technique has been released for public use both as a standalone program or as SPM8/SPM12 library. This work has been published in the following paper:

Paper published in the **NeuroImage: Clinical** journal (NICL)
Volume: 6, Pages: 86-92, Published: August 2014
DOI: [10.1016/j.nicl.2014.08.016](https://doi.org/10.1016/j.nicl.2014.08.016)
Quality Index: 2.53 (Quartile 2)

Chapter 5

Quantifying brain tissue volume in multiple sclerosis with automated lesion segmentation and filling.

In this chapter we present a detailed evaluation of the performance of different pipelines that incorporate fully automated processes such as lesion segmentation, lesion filling and tissue segmentation on MS data. For each automated pipeline, we analyze the percentage of error in tissue segmentation between a set of 70 MS images where WM lesions have been refilled before segmentation and the same images processed different levels of automation from manually masking lesion to fully automated lesion segmentation and filling. This analysis has been published in the following paper:

Paper published in the **NeuroImage: Clinical** journal (NICL)

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DOI: doi:10.1016/j.nicl.2015.10.012

Quality Index: 2.53 (Quartile 2)

Chapter 6

Automated tissue segmentation of MR brain images in the presence of white matter lesions.

In this chapter, we propose a novel automated pipeline for tissue segmentation of MS patient images containing lesions. The accuracy of the method is evaluated using both the challenge MRBrainS13 database¹ and a 3T MS database of MS patient images. We validate the accuracy of the proposed method with other state-of-the-art techniques. A public version of the method has been released for public use. The proposed pipeline has been described in detail in the next paper and submitted to the Medical Imaging Journal:

Paper submitted to the **Medical Image Analysis** (MIA) journal
Currently in revision.
Quality Index: 3.68 (Quartile 1)

¹<http://mrbrains13.isi.uu.nl/>

Chapter 7

Main results and discussion

MRI tissue segmentation techniques are increasingly being used as the standard tools to assess brain tissue volume. However, automated tissue segmentation is still a challenging task in MS, due to tissue abnormalities found in MS image patients such as WM lesions that are known to reduce the accuracy of tissue segmentation methods. When expert manual annotations of WM lesions are available, lesion filling has been shown to be an effective method to reduce the effects of those lesions on tissue segmentation. However, manual annotations are time-consuming and prone to variability between experts, which in combination of the need to analyze quantitatively focal MS lesions in individual and temporal studies has led to the development of a wide number of automated lesion segmentation of MS lesions. Therefore, a solid understanding of the effects of MS lesions on automated pipelines that concatenate processes such as lesion segmentation, lesion filling and tissue segmentation is important.

As said in section 1.3, each of these processes cover part of the necessary knowledge needed to tackle the problem of automated brain tissue segmentation of images containing lesions. This chapter provides a comprehensive discussion of the main results obtained on previous chapters, analyzing each of these necessary processes until the development of a fully automated tissue segmentation method for MS images.

7.1 Effect of WM lesions on tissue segmentation

A wide number of automated tissue segmentation methods have been proposed in the literature so far. In Chapter 2, we evaluated the accuracy of ten approaches using the two public available databases of healthy subjects IBSR18 and IBSR20. With the aim to include a wide set of different segmentation approaches and available tools, the analysis included well-known intensity based algorithms such as ANN, FCM,

and KNN, and also public available toolboxes such as FAST, SPM5, SPM8, PVC, GAMIXTURE, SVPASEG, and FANTASM. Results were presented before and after correcting CSF masks, as it was shown that available annotations ignored sulcal CSF tissue on original masks. When sulcal CSF was corrected, SVPASEG, SPM8 and FAST yielded the highest accuracy on both databases. However, most of the methods were variant to changes in acquisition sequences, intensity inhomogeneities or special attributes of the different databases, which highlighted the fact that brain tissue segmentation problem was still open, because there was not a single method that achieved a significant higher accuracy on all tissues.

Afterwards, six of these methods (ANN, FCM, FANTASM, FAST, SPM5 and SPM8) were evaluated on MS data from different hospital centers and scanners in order to analyze the extent to which tissue volume estimations were affected by changes in WM lesion volume and intensity. Our results showed that SPM8 was the method with the lowest differences in total volume, while FANTASM and again SPM8 were the methods where the incidence of WM lesions was the lowest on normal-appearing tissue. In general, differences in tissue volume were lower on methods combining morphological prior information such as SPM5 and SPM8, or spatial constraints such as FANTASM and FAST. In contrast, these differences were higher on simpler intensity based algorithms that lacked of spatial correction such as FCM and ANN. This fact and the higher performance on healthy data of former methods, stressed the necessity of adding morphological prior information and/or spatial constraints in automated brain tissue segmentation, not only to overcome inherent MRI artifacts but also as an important component to deal with WM lesions.

The main factor in the observed differences in tissue volume across methods was caused by lesion volume. Furthermore, WM lesion voxels tended to be classified as GM on images where the variation between lesion signal intensity and the mean signal intensity of normal-appearing WM was higher, which indicates a direct relationship between the differences in brain tissue volume and changes in lesion load and WM lesion intensity. However, lesion voxels had also a direct effect on the observed differences in GM and WM outside lesion regions. As already commented in Chapter 3, these differences are especially important because they highlight the bias introduced by WM lesions on the estimation of tissue volume that is not pathologically affected. Our analysis showed that if lesion voxels were not considered to compute brain volume, still methods tended to overestimate GM, specially on images with higher lesion load. Observed differences in normal-appearing tissue volume were important, because although lesion voxels could be reassigned to WM after segmentation, if these lesions were present in image segmentation, part of the bias was still present. Furthermore, differences in total tissue volume may be canceled between the errors produced in the same lesion regions and the effect of these voxels in normal-appearing tissue. This fact clearly shows the necessity to process

WM lesion before segmentation.

7.2 Effect of lesion filling in tissue segmentation

In the last years, different techniques have been proposed to reduce the bias introduced by WM lesions on brain tissue volume measurements of MS images, mostly by in-painting WM lesions on T1-w with signal intensities similar to normal-appearing WM. After reviewing the related available literature in Chapter 4, we classified existing methods by those that filled WM lesions using the *local* intensities from the surrounding neighboring voxels of lesions, and those that used *global* WM intensities from the whole brain to fill WM lesions. Although all these methods had been already validated separately, we performed a general comparison of all the available techniques in order to analyze their accuracy on the same 1.5T and 3T data and also to investigate its performance with different tissue segmentation techniques such as FAST and SPM8.

This analysis served as a basis to propose a new technique to refill WM lesions which was a compromise between *global* and *local* methods. In contrast to other existing techniques, the proposed method filled lesion voxels intensities with random values of a normal distribution generated from the mean WM signal intensity of each two-dimensional slice. Our results showed that when compared to other methods, our approach yielded the lowest deviation in GM and WM volume on 1.5T and 3T data when FAST tissue segmentation was used. When SPM8 tissue segmentation method was used, the performance of our lesion filling method was also very competitive, yielding the lowest differences or similar to the best method in GM and WM. In contrast to the rest of pipelines, differences in tissue volume between the same images filled with our method and afterwards segmented with either FAST or SPM8 were very low ($< 0.1\%$), which indicates that the proposed strategy was equally efficient independently of the tissue segmentation chosen.

The proposed algorithm performed significantly better than local methods on images with higher lesion load. In contrast to *global* methods, *local* methods may be limited by the range of similar intensities coming from the neighboring voxels, which on images with a large lesions may be introducing a bias on GM and WM tissue distributions by the addition of a considerable number of voxels with similar intensities. Furthermore, the performance our approach was also better on images with high lesion load when compared with *local* methods, specially on images with lower resolution such as 1.5T data, most probably because our method estimated the mean global NAWM intensity for each slice independently, being more sensible to reproduce possible changes in the intensities between slices.

7.3 Effect of automating lesion segmentation and filling on tissue segmentation

As already said earlier, lesion filling has been shown to be an effective method to reduce the effects of these lesions on tissue segmentation. However, in all the lesion-filling approaches including ours, MS lesions have to be known *a priori*, which requires to delineate lesions manually. This was a clear limitation in terms of fully automatizing brain tissue on the presence of MS lesions, which motivated the evaluation of the effect of automated lesion segmentation on tissue segmentation. Although different automated tissue segmentation methods have been proposed, most of them are based on supervised learning, which require to explicitly train them usually with a large amount of labeled data. Labeled data may be not available, which had pointed out the interest of the community in unsupervised methods that can operate without prior data. As shown in Chapter 5, we compared two fully unsupervised pipelines that combined automated lesion segmentation and filling as a first step to understand the effect of fully processed images in tissue segmentation.

Given the performance shown in our previous studies and its widely use in clinical studies, SPM8 was used as a reference tissue segmentation method to measure tissue volume on a set of 70 3T images of CIS patients. On these images, available manual expert annotations were employed to refill WM lesions before segmentation using the filling method of the pipeline, and were considered as ground-truth. Afterwards, we evaluated the differences in GM and WM volume between the set of filled images using manual annotations and the same images processed using different variations of the SLS and LST toolkits that differed in the level of manual intervention. Evaluating different pipelines with distinct levels of automation permitted us to analyze the effect of each of the automated process involved in the obtained differences in total and normal-appearing tissue volume.

As already suggested in Chapter 3, this new analysis showed that the effect of lesions in total tissue volume was limited due to a canceling effect between the errors produced in the same lesion regions, and the effect of these voxels in normal-appearing tissue. In all the pipelines that incorporated automated lesion segmentation, most of the observed differences in normal-appearing tissue were produced by the effect of false positive lesion voxels that were already segmented without refilling them. In contrast, there was not a relevant correlation between the number of false positive lesion voxels and the observed differences in normal-appearing GM and WM, which suggested that most of these miss-classified voxels were actually WM before refilling them. The relationship between errors in automated lesion and tissue segmentation suggest also the importance of not only to keep reducing the number of missed lesions, but also stress the necessity of contextual spatial informa-

tion of lesion regions in order to confine them in WM and hence reduce the effect of miss-classified voxels on tissue segmentation.

As shown in the results presented in Chapter 4, masking-out lesion voxels before tissue segmentation might not be optimal, as leaving lesion voxels out of the tissue distributions appears to increase the differences in tissue volume with respect to lesion filled images, even if these voxels are re-assigned to WM afterwards. However, although not optimal, masking lesion before segmentation has been found a valid alternative to reduce the effects of WM lesions in research and clinical settings, and only in the recent years, lesion filling techniques are been already applied on research and clinical studies. Regarding to this, our results show that at least with the evaluated data, the differences in tissue volume between images where expert lesion masks have been masked-out and the same images where lesions have been automatically segmented and filled, are similar on images with low lesion load ($< 10ml$). In contrast, from our experiments we observed that differences in tissue volume tend to increase with lesion load on masked-out images, while the increase of the error is more moderated on the fully-automated images. However, given the available data and maximum lesion load considered in our analysis ($< 20ml$), these findings should be considered with care.

In any case, our analysis points out the fact that automated lesion segmentation and filling methods reduced significantly the impact of WM lesions on tissue segmentation, and with a similar performance to the pipelines that required manual expert intervention. These results are relevant and validate that each of these automated processes can be useful not only in terms of time and economic costs, but also as active processes in fully automated tissue segmentation in the presence of WM lesions.

7.4 Fully automated tissue segmentation of images containing WM lesions

Previous sections have stressed the necessity to deal with MS lesions before tissue segmentation, showing several general insights that can be useful for automated tissue segmentation of images containing lesions. The obtained results of the different evaluated methods in Chapter 2 and 3 have pointed out the superiority of methods that were benefited by morphological prior information or spatial constraints in automated brain tissue segmentation. More importantly, the results obtained in Chapter 3 have evidenced the effect of WM lesion on tissue segmentation and the necessity to deal with MS lesions in order to reduce not only the bias produced by the same lesions but also the effect of these lesion voxels in normal-appearing tis-

sue. In this scenario, we have proposed a new lesion filling technique that was very competitive with different databases and tissue segmentation methods, as shown in Chapter 4. Finally, we have shown in Chapter 5 that the addition of unsupervised lesion segmentation and filling into already existing tissue segmentation pipelines reduced significantly the error in tissue volume when compared with previous pipelines where lesions were segmented as normal tissue.

Following these insights, we have developed a novel multi-channel method designed to segment brain tissues in MRI images of MS patients. As explained in Chapter 6, this approach makes use of a combination of intensity, anatomical and morphological prior maps to guide the tissue segmentation. Tissue segmentation has been tackled based on a robust partial volume segmentation where WM outliers have been estimated and refilled before segmentation using a multi-channel post-processing algorithm also integrating partial volume segmentation, spatial context, and prior anatomical and morphological atlases. Furthermore, the proposed method takes advantage of new affordable processors such as GPUs that reduce up to four times the execution time to register and segment tissue when compared to general purpose CPUs. This property makes this method useful for studies containing a large number of subjects to analyze.

The proposed method has been quantitatively and qualitatively evaluated using different databases of images containing WM lesions. In order to analyze the extent to which T1-w and FLAIR modalities intervened in the obtained accuracy, the proposed method was run in all experiment using only T1-w or using both T1-w and FLAIR image sequences. As shown by the presented results, the proposed technique yielded competitive and consistent results in both general and MS specific databases without parameter tweaking. In the MRBrainS tissue segmentation challenge¹, our method combining both T1-w and FLAIR was the best non-supervised technique of the challenge, being ranked in the 7th position out of 31 participant methods. When only the T1-w modality was used, still the accuracy of the proposed method was clearly superior to general purpose methods such as FAST (ranked 21th) and SPM12 (best ranked 17th), even if those used both image modalities. In MS data, the performance of our method combining T1-w and FLAIR sequences was similar or better to the best evaluated pipeline incorporating lesion segmentation and filling. Obtained differences in tissue volume between images processed with the proposed algorithm and the same images where lesions were filled using expert lesion annotations, were lower than 0.15% on all tissues, validating the overall capability of the proposed method to reduce the effects of WM lesions on tissue segmentation.

In general, our results showed that the percentages of error in tissue volume of our proposed approach were low and similar in both databases. The percentages of error

¹<http://mrbrains13.isi.uu.nl/>

were the lowest when the FLAIR modality was used, which evidences that this image sequence has a direct effect on the efficiency of the algorithm, and consequently it should be used when available. However, the accuracy of the method using only the T1-w modality was also superior to other general purpose strategies, which also evidences that the improvement in tissue segmentation was not only generated by the addition of the FLAIR modality, but also by the combination of intensity, anatomical and morphological priors, and the use of an specific outlier algorithm with integrated lesion filling.

Chapter 8

Conclusions

This thesis synthesizes our work done during the last three years. Following the same objectives defined in the Introduction chapter, we summarize in what follows the main conclusions and contributions of this thesis:

- We analyzed and evaluated the state-of-the-art of brain tissue segmentation methods. This first stage aimed to quantitative review and evaluate different proposed tissue segmentation techniques in order to understand their advantages and drawbacks. As part of the resulting analysis published in the *Journal of Magnetic Resonance Imaging* in January of 2015, **our results showed a higher accuracy on several methods that incorporated morphological prior information and/or spatial constraints such as FAST, SP-VASEG and SPM8. These methods were also less prone to changes in acquisition sequences and intensity inhomogeneities.**
- We studied the effect of WM lesions on tissue segmentation of MS patient images. The second stage to cover was focused on the analysis of the effects of WM lesions on the tissue distributions. Six of the analyzed methods on Chapter 2 were evaluated on multi-center 1.5T MS data from different scanners. Related to the previous stage, our results stressed **the necessity of adding morphological prior information and/or spatial constraints in automated brain tissue segmentation, not only to overcome inherent MRI artifacts but also as an important component to deal with WM lesions.** Furthermore, our analysis of the effects of WM lesions on tissue volume showed that **the inclusion of WM lesions on tissue segmentation not only biased the total tissue volume measurements by the addition of miss-classified lesion voxels, but also by the effect of these lesions in observed differences in normal-appearing tissue volume.**

The entire analysis was published in the *American Journal of Neuroradiology* in February 2015.

- We proposed a new technique to reduce the effects of WM lesions on tissue segmentation of MS patient images. The third stage required first to compare the accuracy of different proposed lesion filling techniques in the literature with the aim to propose then a new technique to reduce the effects of WM lesions on tissue segmentation. **The lesion filling method proposed in this thesis, was shown effective in different data and independently of the tissue segmentation method used afterwards.** The proposed approach outperformed the rest of methods on both 1.5T and 3T data when FAST was used, while its performance was similar or lower to the best available strategy when SPM8 was used. The proposed lesion filling method was published in the *NeuroImage: Clinical journal* in August of 2014. **Furthermore, we released a public version on the proposed method that can be freely downloaded from our research team web page¹.** This software is already been used in the collaborating hospitals.
- We analyzed and evaluated the effect of automated WM lesion segmentation and filling on the tissue segmentation. During the fourth stage proposed, we quantitatively evaluated the accuracy of two state-of-the-art automated pipelines that incorporate unsupervised lesion segmentation, lesion filling and tissue segmentation on MS data. As shown in the published paper in the *NeuroImage: Clinical journal* in October of 2015, our analysis showed that **pipelines that incorporated automated lesion segmentation and filling were capable to reduce significantly the impact of WM lesions on tissue segmentation, performing similarly to the pipelines that required manual expert intervention.**
- Finally, we proposed a new fully automated tissue segmentation method for MS patient images containing lesions. The main goal of the thesis was to propose a fully automated tissue segmentation method capable to deal with images of MS patients. As shown in Chapter 6, the proposed method incorporated all the major insights obtained from previous stages with the aim of provide a robust fully automated tissue approach for accurate brain volume measurements. Our results showed that **when compared with existent tissue segmentation methods, the presented approach yielded a higher accuracy in tissue segmentation while the influence of MS lesions on tissue segmentation was lower or similar to the best state-of-the-art pipeline incorporating automated lesion segmentation and filling.**

¹The latest version on the proposed lesion filling method can be download from <http://atc.udg.edu/nic/slftoolbox/index.html>

This work has been submitted for publication in the *Medical Image Analysis journal* in January 2016. **As part of this work, we also released a public version on the proposed method that can be freely downloaded from our research team web page².**

During this PhD thesis, different collaborations have been done with other researchers of the VICOROB group. First, we evaluated the effect of MS lesions on longitudinal registration in the published study of Diez et al. [23], where we contributed with several processing steps such as lesion filling. Lately, we were also involved in the development of several automated lesion segmentation pipelines that allowed us to gain knowledge about this topic. In this regard, we helped to implement two different lesion segmentation pipelines for MS such as those published in the papers of Cabezas et al. [10] and Roura et al. [53], respectively. Furthermore, we also collaborated on a new pipeline for automated lesion segmentation of Lupus lesions proposed by Roura et al., which has been submitted for publication recently.

8.1 Future work

Unfortunately, there are several aspects that have been not investigated during this thesis. One of the main limitations on several stages has been the lack of 3T images with high lesion load. As pointed out in Chapters 5 and 6, the low mean lesion load of the cohorts analyzed, which indeed has the major interest for the medical experts, has not allowed to investigate better the performance of the analyzed pipelines in the presence of images with higher lesion load. In the case of our tissue segmentation method, we believe that an additional analysis of the performance with images with higher lesion load would be helpful not only to analyze the robustness of the proposed algorithm, but also to investigate the benefits of adding other image sequences such as T2-w or PD-w.

Although the proposed tissue segmentation method has been designed for cross-sectional data, there is an increasingly clinical interest in the measurements of longitudinal changes in tissue volume. We believe that the proposed method could be extended to longitudinal changes by re-adapting the pipeline with prior registering of time point images before tissue segmentation. This is in fact one of the goals that our team has in mind to tackle first within the research framework of the BiomarkEM.cat project, in order to release suitable tools that can be used in clinical settings.

²A public version of the method can be download from <http://atc.udg.edu/nic/msseg/index.html>

The ultimate goal should be to provide state-of-the-art tools for the collaborating hospitals involved in these research projects that may be useful not only to diagnose and monitorize the progression of disease, but also to evaluate new treatments in MS patients. Related to that, the tools developed in this thesis should be integrated with other tools developed in our group in order to implement this complete system capable to provide robust useful biomarkers in MS such as the number of lesions, lesion volume, brain tissue volume or brain atrophy.

Bibliography

- [1] Julio Acosta-Cabronero, Guy B. Williams, João M S Pereira, George Pengas, and Peter J. Nestor. The impact of skull-stripping and radio-frequency bias correction on grey-matter segmentation for voxel-based morphometry. *NeuroImage*, 39(4):1654–1665, 2008.
- [2] Ayelet Akselrod-Ballin, Meirav Galun, Moshe John Gomori, Ronen Basri, and Achi Brandt. Atlas guided identification of brain structures by combining 3D segmentation and SVM classification. *Medical image computing and computer-assisted intervention : MICCAI ... International Conference on Medical Image Computing and Computer-Assisted Intervention*, 9(Pt 2):209–16, 2006.
- [3] J B Arnold, J S Liow, K a Schaper, J J Stern, J G Sled, D W Shattuck, a J Worth, M S Cohen, R M Leahy, J C Mazziotta, and D a Rottenberg. Qualitative and quantitative evaluation of six algorithms for correcting intensity nonuniformity effects. *NeuroImage*, 13(5):931–943, 2001.
- [4] John Ashburner and Karl J. Friston. Unified segmentation. *NeuroImage*, 26:839–851, 2005.
- [5] Marco Battaglini, Mark Jenkinson, Nicola De Stefano, and Nicola De Stefano. Evaluating and reducing the impact of white matter lesions on brain volume measurements. *Human Brain Mapping*, 33(9):2062–71, 2012.
- [6] S. Bricq, Ch. Collet, and J.P. Armspach. Unifying framework for multimodal brain mri segmentation based on hidden markov chains. *Medical Image Analysis*, 12(6):639 – 652, 2008. *[ce:title]Special issue on information processing in medical imaging 2007[ce:title]*.
- [7] Per Brodal. *The central nervous system*. 2010.
- [8] Mariano Cabezas, Arnau Oliver, Xavier Lladó, Jordi Freixenet, and Meritxell Bach Cuadra. A review of atlas-based segmentation for magnetic resonance brain images. *Computer Methods and Programs in Biomedicine*, 104(3):e158–e177, 2011.

- [9] Mariano Cabezas, Arnau Oliver, Eloy Roura, Jordi Freixenet, Joan C. Vilanova, Llu??s Rami??-Torrent??, ??lex Rovira, and Xavier Llad?? Automatic multiple sclerosis lesion detection in brain MRI by FLAIR thresholding. *Computer Methods and Programs in Biomedicine*, 115(3):147–161, 2014.
- [10] Mariano Cabezas, Arnau Oliver, Sergi Valverde, Brigitte Beltran, Jordi Freixenet, Joan C. Vilanova, Llu??s Rami??-Torrent??, ??lex Rovira, and Xavier Llad?? BOOST: A supervised approach for multiple sclerosis lesion segmentation. *Journal of Neuroscience Methods*, 237:108–117, 2014.
- [11] Beno??t Caldairou, Nicolas Passat, Piotr A Habas, Colin Studholme, and Fran??ois Rousseau. A non-local fuzzy segmentation method: application to brain mri. *Pattern Recognition*, 44(9):1916–1927, 2011.
- [12] A Ceccarelli, J S Jackson, S Tauhid, A Arora, J Gorky, E Dell’Oglio, A Bakshi, T Chitnis, S J Khouri, H L Weiner, C R G Guttmann, R Bakshi, and M Neema. The impact of lesion in-painting and registration methods on voxel-based morphometry in detecting regional cerebral gray matter atrophy in multiple sclerosis. *American Journal of Neuroradiology*, 33(8):1579–85, September 2012.
- [13] Antonia Ceccarelli, Maria a Rocca, Mohit Neema, Vittorio Martinelli, Ashish Arora, Shahamat Tauhid, Angelo Ghezzi, Giancarlo Comi, Rohit Bakshi, and Massimo Filippi. Deep gray matter T2 hypointensity is present in patients with clinically isolated syndromes suggestive of multiple sclerosis. *Multiple sclerosis (Hounds Mills, Basingstoke, England)*, 16(1):39–44, 2010.
- [14] D T Chard, G J M Parker, R Kapoor, A J Thompson, and D H Miller. Brain atrophy in clinically early relapsing remitting multiple sclerosis. 2002.
- [15] Declan T. Chard, Jonathan S. Jackson, David H. Miller, and Claudia A M Wheeler-Kingshott. Reducing the impact of white matter lesions on automated measures of brain gray and white matter volumes. *Journal of Magnetic Resonance Imaging*, 32:223–228, 2010.
- [16] Harvey E. Cline, William E. Lorensen, Ron Kikinis, and Ferenc Jolesz. Three-Dimensional Segmentation of MR Images of the Head Using Probability and Connectivity. *Journal of Computer Assisted Tomography*, 14(6):1037–1045, 1990.
- [17] Alastair Compston and Alasdair Coles. Multiple sclerosis. *Lancet*, 359(9313):1221–31, 2002.
- [18] Alastair Compston and Alasdair Coles. Multiple sclerosis. *Lancet*, 372(9648):1502–17, 2008.

- [19] Devic M Confavreux C, Aimard G. Course and prognosis of multiple sclerosis assessed by the computerized data processing of 349 patients. *Brain*, 103(2):281–300, 1980.
- [20] Renske De Boer, Henri A Vrooman, Fedde Van Der Lijn, Meike W Vernooij, M Arfan Ikram, Aad Van Der Lugt, Monique M B Breteler, and Wiro Niessen. White matter lesion extension to automatic brain tissue segmentation on mri. *NeuroImage*, 45(4):1151–1161, 2009.
- [21] N. De Stefano, A. Giorgio, M. Battaglini, M. Rovaris, M. P. Sormani, F. Barkhof, T. Korteweg, C. Enzinger, F. Fazekas, M. Calabrese, D. Dinacci, G. Tedeschi, A. Gass, X. Montalban, A. Rovira, A. Thompson, G. Comi, D. H. Miller, and M. Filippi. Assessing brain atrophy rates in a large population of untreated multiple sclerosis subtypes. *Neurology*, 74(23):1868–1876, 2010.
- [22] Hrishikesh Deshpande, Pierre Maurel, and Christian Barillot. Classification of Multiple Sclerosis Lesions using Adaptive Dictionary Learning. *Computerized Medical Imaging and Graphics*, 2015.
- [23] Yago Diez, Arnau Oliver, Mariano Cabezas, Sergi Valverde, Robert Martí, Joan Carles Vilanova, Lluís Ramió-Torrentà, Àlex Rovira, and Xavier Lladó. Intensity based methods for brain MRI longitudinal registration. A study on multiple sclerosis patients. *Neuroinformatics*, 12(3):365–379, 2014.
- [24] Robert R Edelman and Steven Warach. Magnetic Resonance Imaging. *New England Journal of Medicine*, 328(10):708–716, 1993.
- [25] Massimo Filippi, Paolo Preziosa, Massimiliano Copetti, Gianna Riccitelli, Mark a Horsfield, Vittorio Martinelli, Giancarlo Comi, and Maria a Rocca. Gray matter damage predicts the accumulation of disability 13 years later in MS. *Neurology*, 81(20):1759–67, nov 2013.
- [26] Massimo Filippi and Maria A Rocca. MR imaging of multiple sclerosis. *Radiology*, 259(3):659–81, 2011.
- [27] Elizabeth Fisher, Jar-Chi Lee, Kunio Nakamura, and Richard A Rudick. Gray matter atrophy in multiple sclerosis: A longitudinal study. *Annals of Neurology*, 64(3):255–265, sep 2008.
- [28] Leonora K Fisniku, Declan T Chard, Jonathan S Jackson, Valerie M Anderson, Daniel R Altmann, Katherine A Miszkiel, Alan J Thompson, and David H Miller. Gray Matter Atrophy Is Related to Long- Term Disability in Multiple Sclerosis. *Ann Neurol*, 64:247–254, 2008.

- [29] Onur Ganiler, Arnau Oliver, Yago Diez, Jordi Freixenet, Joan C. Vilanova, Brigitte Beltran, Llu??s Rami??-Torrent??, ??lex Rovira, and Xavier Llad?? A subtraction pipeline for automatic detection of new appearing multiple sclerosis lesions in longitudinal studies. *Neuroradiology*, 56(5):363–374, 2014.
- [30] Daniel Garc??a-Lorenzo, Simon Francis, Sridar Narayanan, Douglas L Arnold, and D Louis Collins. Review of automatic segmentation methods of multiple sclerosis white matter lesions on conventional magnetic resonance imaging. *Medical Image Analysis*, 17(1):1–18, January 2013.
- [31] Ezequiel Geremia, Olivier Clatz, Bjoern H. Menze, Ender Konukoglu, Antonio Criminisi, and Nicholas Ayache. Spatial decision forests for MS lesion segmentation in multi-channel magnetic resonance images. *NeuroImage*, 57(2):378–390, 2011.
- [32] Guido Gerig, John Martin, Ron Kikinis, Olaf Kubler, Martha Shenton, and Ferenc A. Jolesz. Unsupervised tissue type segmentation of 3D dual-echo MR head data. *Image and Vision Computing*, 10(6):349–360, 1992.
- [33] Tal Geva. Magnetic resonance imaging: historical perspective. *Journal of Cardiovascular Magnetic Resonance*, 8(4):573–580, 2006.
- [34] Antonio Giorgio and Nicola De Stefano. Clinical use of brain volumetry. *Journal of Magnetic Resonance Imaging*, 37(1):1–14, 2013.
- [35] Nicolas Guizard, Pierrick Coup??, Vladimir S. Fonov, Jose V. Manj??n, Douglas L. Arnold, and D. Louis Collins. Rotation-invariant multi-contrast non-local means for MS lesion segmentation. *NeuroImage: Clinical*, 8:376–389, 2015.
- [36] Rola Harmouche, Nagesh K Subbanna, D Louis Collins, Douglas L Arnold, and Tal Arbel. Based on Modeling Regional Intensity Variability and Local Neighborhood Information. 62(5):1281–1292, 2015.
- [37] Zujun Hou. A review on MR image intensity inhomogeneity correction, 2006.
- [38] Saurabh Jain, Diana M. Sima, Annemie Ribbens, Melissa Cambron, Anke Maertens, Wim Van Hecke, Johan De Mey, Frederik Barkhof, Martijn D. Steenwijk, Marita Daams, Frederik Maes, Sabine Van Huffel, Hugo Vrenken, and Dirk Smeets. Automatic segmentation and volumetry of multiple sclerosis brain lesions from MR images. *NeuroImage: Clinical*, 8:367–375, 2015.
- [39] T Kapur, W E Grimson, W M Wells, and R Kikinis. Segmentation of brain tissue from magnetic resonance images. *Medical image analysis*, 1(2):109–27, 1996.

- [40] Jong Min Lee, Uicheul Yoon, Sang Hee Nam, Jung Hyun Kim, In Young Kim, and Sun I. Kim. Evaluation of automated and semi-automated skull-stripping algorithms using similarity index and segmentation error. *Computers in Biology and Medicine*, 33(6):495–507, 2003.
- [41] Xavier Lladó, Onur Ganiler, Arnau Oliver, Robert Martí, Jordi Freixenet, Laia Valls, Joan C. Vilanova, Lluís Ramió-Torrentà, and Àlex Rovira. Automated detection of multiple sclerosis lesions in serial brain MRI. *Neuroradiology*, 54:787–807, 2012.
- [42] Xavier Lladó, Arnau Oliver, Mariano Cabezas, Jordi Freixenet, Joan C. Vilanova, Ana Quiles, Laia Valls, Lluís Ramió-Torrentà, and Àlex Rovira. Segmentation of multiple sclerosis lesions in brain MRI: A review of automated approaches. *Information Sciences*, 186(1):164–185, March 2012.
- [43] F. D. Lublin and S. C. Reingold. Defining the clinical course of multiple sclerosis: results of an international survey. National Multiple Sclerosis Society (USA) Advisory Committee on Clinical Trials of New Agents in Multiple Sclerosis. *Neurology*, 46(4):907–911, April 1996.
- [44] Stefano Magon, Laura Gaetano, M Mallar Chakravarty, Jason P Lerch, Yvonne Naegelin, Christoph Stippich, Ludwig Kappos, Ernst-Wilhelm Radue, and Till Sprenger. White matter lesion filling improves the accuracy of cortical thickness measurements in multiple sclerosis patients: a longitudinal study. *BMC Neuroscience*, 15(1):106, January 2014.
- [45] D Mahapatra. Analyzing Training Information From Random Forests for Improved Image Segmentation. *Image Processing, IEEE Transactions on*, 23(4):1504–1512, 2014.
- [46] J.L. Marroquin, B.C. Vemuri, S. Botello, E. Calderon, and A. Fernandez-Bouzas. An accurate and efficient bayesian method for automatic segmentation of brain mri. *Medical Imaging, IEEE Transactions on*, 21(8):934 –945, aug. 2002.
- [47] Dzung L Pham. Spatial Models for Fuzzy Clustering. *Computer Vision and Image Understanding*, 297:285–297, 2001.
- [48] Chris H Polman, Stephen C Reingold, Brenda Banwell, Michel Clanet, Jeffrey a Cohen, Massimo Filippi, Kazuo Fujihara, Eva Havrdova, Michael Hutchinson, Ludwig Kappos, Fred D Lublin, Xavier Montalban, Paul O’Connor, Magnhild Sandberg-Wollheim, Alan J Thompson, Emmanuelle Waubant, Brian Weinshenker, and Jerry S Wolinsky. Diagnostic criteria for multiple sclerosis: 2010 revisions to the McDonald criteria. *Annals of neurology*, 69(2):292–302, 2011.

- [49] V. Popescu, M. Battaglini, W. S. Hoogstrate, S. C J Verfaillie, I. C. Sluimer, R. A. van Schijndel, B. W. van Dijk, K. S. Cover, D. L. Knol, M. Jenkins, F. Barkhof, N. de Stefano, H. Vrenken, F. Barkhof, X. Montalban, F. Fazekas, M. Filippi, J. Frederiksen, L. Kappos, D. Miller, J. Palace, C. Polman, M. Rocca, A. Rovira, and T. Yousry. Optimizing parameter choice for FSL-Brain Extraction Tool (BET) on 3D T1 images in multiple sclerosis. *NeuroImage*, 61(4):1484–1494, 2012.
- [50] V. Popescu, N. C G Ran, F. Barkhof, D. T. Chard, C. A. Wheeler-Kingshott, and H. Vrenken. Accurate GM atrophy quantification in MS using lesion-filling with co-registered 2D lesion masks. *NeuroImage: Clinical*, 4:366–373, 2014.
- [51] Martin Rajchl, John S.H. Baxter, a. Jonathan McLeod, Jing Yuan, Wu Qiu, Terry M. Peters, and Ali R. Khan. Hierarchical max-flow segmentation framework for multi-atlas segmentation with Kohonen self-organizing map based Gaussian mixture modeling. *Medical Image Analysis*, 000:1–12, 2015.
- [52] E. Roura, T. Schneider, M. Modat, P. Daga, N. Muhlert, D. Chard, S. Ourselin, X. Lladó, and C. A. M. Wheeler-Kingshott. Multi-channel registration of fa and t1w images in the presence of atrophy: application to multiple sclerosis. *Functional Neurology*, 30(4), 2015.
- [53] Eloy Roura, Arnau Oliver, Mariano Cabezas, Sergi Valverde, Deborah Pareto, Joan C Vilanova, Lluís Ramió-Torrentà, Àlex Rovira, and Xavier Lladó. A toolbox for multiple sclerosis lesion segmentation. *Neuroradiology*, 57(10):1031–1043, 2015.
- [54] Eloy Roura, Arnau Oliver, Mariano Cabezas, Joan C. Vilanova, ??lex Rovira, Lluís Ramió-Torrentà, and Xavier Lladó. MARGA: Multispectral Adaptive Region Growing Algorithm for brain extraction on axial MRI. *Computer Methods and Programs in Biomedicine*, 113(2):655–673, 2014.
- [55] Snehashis Roy, Aaron Carass, Pierre-Louis Bazin, Susan Resnick, and Jerry L. Prince. Consistent segmentation using a rician classifier. *Medical Image Analysis*, 16(2):524 – 535, 2012.
- [56] Richard a Rudick, Jar-Chi Lee, Kunio Nakamura, and Elizabeth Fisher. Gray matter atrophy correlates with MS disability progression measured with MSFC but not EDSS. *Journal of the neurological sciences*, 282(1-2):106–11, jul 2009.
- [57] Paul Schmidt, Christian Gaser, Milan Arsic, Dorothea Buck, Annette Förtschler, Achim Berthele, Muna Hoshi, Rüdiger Ilg, Volker J Schmid, Claus Zimmer, Bernhard Hemmer, and Mark Mühlau. An automated tool for detection of

- FLAIR-hyperintense white-matter lesions in Multiple Sclerosis. *NeuroImage*, 59(4):3774–3783, February 2012.
- [58] Michaël Sdika and Daniel Pelletier. Nonrigid registration of multiple sclerosis brain images using lesion inpainting for morphometry or lesion mapping. *Human Brain Mapping*, 30(4):1060–1067, 2009.
- [59] D W Shattuck, S R Sandor-Leahy, K a Schaper, D a Rottenberg, and R M Leahy. Magnetic resonance image tissue classification using a partial volume model. *NeuroImage*, 13(5):856–876, 2001.
- [60] A. Simmons, P. S. Tofts, G. J. Barker, and S. R. Arridge. Sources of intensity nonuniformity in spin echo images at 1.5 T. *Magnetic Resonance in Medicine*, 32(1):121–128, 1994.
- [61] J G Sled, A P Zijdenbos, and A C Evans. A nonparametric method for automatic correction of intensity nonuniformity in MRI data. *IEEE transactions on medical imaging*, 17(1):87–97, 1998.
- [62] Stephen M. Smith. Fast robust automated brain extraction. *Human Brain Mapping*, 17(3):143–155, 2002.
- [63] G H Sperber. Clinically Oriented Anatomy. *Journal of anatomy*, 208(3):393, 2006.
- [64] Lawrence Steinman, M.D. Multiple Sclerosis: A Coordinated Immunological Attack against Myelin in the Central Nervous System. *Cell*, 85(3):299–302, may 1996.
- [65] Elizabeth M Sweeney, Russell T Shinohara, Navid Shiee, Farrah J Mateen, Avni a Chudgar, Jennifer L Cuzzocreo, Peter a Calabresi, Dzung L Pham, Daniel S Reich, and Ciprian M Crainiceanu. OASIS is Automated Statistical Inference for Segmentation, with applications to multiple sclerosis lesion segmentation in MRI. *NeuroImage: Clinical*, 2:402–13, jan 2013.
- [66] Jussi Tohka, Ivo D. Dinov, David W. Shattuck, and Arthur W. Toga. Brain mri tissue classification based on local markov random fields. *Magnetic Resonance Imaging*, 28(4):557 – 573, 2010.
- [67] Xavier Tomas-Fernandez and Simon Keith Warfield. A Model of Population and Subject (MOPS) Intensities with Application to Multiple Sclerosis Lesion Segmentation. *IEEE transactions on medical imaging*, 0062(c):1–15, jan 2015.

- [68] Nicholas J. Tustison, Brian B. Avants, Philip A. Cook, Yuanjie Zheng, Alexander Egan, Paul A. Yushkevich, and James C. Gee. N4ITK: Improved N3 bias correction. *IEEE Transactions on Medical Imaging*, 29(6):1310–1320, 2010.
- [69] Annegreet van Opbroek, Fedde van der Lijn, and Marleen de Bruijne. Automated brain-tissue segmentation by multi-feature svm classification. In *Proceedings of the MICCAI Workshopsâ€The MICCAI Grand Challenge on MR Brain Image Segmentation (MRBrainSâ€13)*, 2013.
- [70] H Vrooman, Fedde van der Lijn, and W Niessen. Auto-knn: brain tissue segmentation using automatically trained knearest-neighbor classification. In *Proceedings of the MICCAI Workshopsâ€The MICCAI Grand Challenge on MR Brain Image Segmentation (MRBrainSâ€13)*, 2013.
- [71] Zhao Yi, Antonio Criminisi, Jamie Shotton, and Andrew Blake. Discriminative, semantic segmentation of brain tissue in mr images. In Guang-Zhong Yang, David Hawkes, Daniel Rueckert, Alison Noble, and Chris Taylor, editors, *Medical Image Computing and Computer-Assisted Intervention – MICCAI 2009*, volume 5762 of *Lecture Notes in Computer Science*, pages 558–565. Springer Berlin / Heidelberg, 2009.
- [72] Y Zhang, M Brady, and S Smith. Segmentation of brain MR images through a hidden Markov random field model and the expectation-maximization algorithm. *IEEE Transactions on Medical Imaging*, 20:45–57, 2001.