UNIVERSITY OF CALGARY

Relationship between Blood Pressures and Brain Volumes: Systematic Review and Analysis of A Canadian Population-Based Study

by

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A THESIS

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES

IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE

DEGREE OF MASTER OF SCIENCE

DEPARTMENT OF MEDICAL SCIENCES

CALGARY, ALBERTA

DECEMBER 2013

Abstract

Long-standing hypertension has been associated with global and regional brain atrophy. Little is known regarding the timing of atrophy onset and its relationship with duration and severity of hypertension. A systematic literature review was performed to identify the relationships between hypertension and brain atrophy measured using neuroimaging, to summarize the existing knowledge and to identify areas for future investigation. Most studies identified that higher systolic blood pressure was associated with late-life brain atrophy; however, these studies did not include a concurrent assessment of brain imaging in mid-life, and participants in these studies were middle-aged in the 1980s when definitions of blood pressure (BP) were more permissive and population BP control was poorer. To investigate the relationship between BP and brain atrophy in mid-life in a contemporary cohort, brain magnetic resonance (MR) images were analyzed in 778 individuals participating in the population-based PURE-MIND study, a sub-study of the international Prospective Urban Rural Epidemiologic (PURE) Study designed to identify the risk factors for common non-communicable diseases. MR data were processed for global and regional brain volumes and cortical thickness. No significant association was seen between brain volume and hypertension, after controlling for age and gender. However, few participants had markedly elevated BP, probably due to improving population-wide control of hypertension due to lower thresholds for successful treatment than in past decades. Analysis showed that brain atrophy is not a common consequence of mid-life hypertension in today's Canadians. It

is possible that aggressive screening and control of mid-life BP might prevent atrophic changes.

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List of Abbreviations and Symbols

Abbreviation or Symbol	Definition
ABP	Ambulatory blood pressure
AM	Automated method
B1	RF rotating magnetic field
BP	Blood pressure
C	Clinic recruitment
CA	Community advertisement
CSF	Cerebrospinal fluid
CT	Computed tomography
D	Dementia
DBP	Diastolic blood pressure
DTI	Diffusion tensor imaging
fMRI	Functional magnetic resonance imaging
FSL	FMRIB software library
GM	Gray matter
GMV	Gray matter volume
HTN	Hypertension
HU	Hounsfield unit
MeSH	Medical subject heading
MPRAGE	Magnetization-prepared rapid acquisition with gradient echo
MR	Magnetic resonance
N	No of participants
n/a	Not applicable
ND	No dementia
NR	Not reported
P	Population based
PD	Proton density
MoCA	Montreal cognitive assessment test
PP	Pulse pressure
PURE	Prospective urban rural epidemiological
RCT	Randomized control trial
SBP	Systolic blood pressure
SD	Standard deviation
SPM	Statistical parametric mapping
STRIVE	Standards for reporting vascular changes on neuroimaging

Abbreviation or Symbol	Definition
SVD	Small vessel disease
T1	Spin-lattice relaxation time
T1W	T1-weighted
T2	Spin-spin relaxation time
T2*	Spin-spin relaxation time accounting for magnetic field inhomogeneities and susceptibility effects
TBM	Total brain matter
TBV	Total brain volume
TE	Echo time
TIA	Transient ischemic attack
TR	Repetition time
WMH	White matter hyperintensities

Acknowledgements

I would like to thank Dr. Eric E Smith, my supervisor, and Dr. Richard Frayne, my co-supervisor, first for making me part of their research group and more so for their support, skilled guidance and feedback throughout the course of this project. I also want to say heart felt thank to Dr Nathalie Jette, Dr Cheryl McCreary my committee members for their valuable guidance through out my graduate school.

I would also like to acknowledge the individuals who were integral to the completion of this work: Dr. Cheryl McCreary for her expert knowledge of FreeSurfer software and for her time and guidance, and Vanessa Hill for her help during quality control check process of the data.

A heartfelt thank you is also due to Mari Boesen for her help in formatting the thesis and for more general support throughout graduate school. Thanks also to my family for their support throughout the course of my degree.

Operating grants for the PURE-MIND study, provided by the Canadian Institutes of Health Research, Canadian Stroke Network and the Heart and Stroke Foundation of Canada, supported my graduate student stipend. I also want to thank NSERC CREATE I3T program for providing me scholarship during the second year of my masters.

Chapter One: Introduction

Hypertension is clinically defined based on measured elevation in blood pressure levels. High blood pressure leads to secondary changes in the vascular system, most prominently in the heart, kidney and brain. Persistent elevated blood pressure (BP) can cause coronary artery disease leading to an increased risk of myocardial infarction, damage to the microvasculature of the kidney resulting in renal failure, and cerebrovascular disease causing stroke and dementia. In the kidney and brain, hypertension has been associated with progressive atrophy of the organ. Brain atrophy is shrinkage of brain volume due to loss of brain cells *i.e.,* neurons, glial cells and axons. The public health impact of hypertension is large, because it is so common. According to a report from the Canadian Chronic Disease Surveillance System in 2006/07 of the nearly 6 million Canadians aged 20 years and older, more than 1 in 5 had diagnosed hypertension.[1] This introductory chapter will review definitions and causes of hypertension, effects of hypertension on the vasculature, effects of hypertension on key organs, and will present the scope of and hypotheses to be tested in this thesis.

1.1 Definition and Causes of Hypertension

The Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure (JNC 7) [2] provides definitions of hypertension based on systolic and diastolic blood pressures. Systolic blood pressure (SBP) is the maximum pressure generated by contraction of left ventricle that pushes the blood out into aorta. The diastolic blood pressure (DBP) is defined as lowest pressure when the left ventricle relaxes between contractions. According to JNC 7, individuals are considered pre-hypertensive if systolic blood pressure is 120 mmHg to 139 mmHg or diastolic blood pressure is 80 mm Hg to 89 mmHg. They are classified as hypertensive if either systolic blood pressure is ≥140 mmHg or diastolic blood pressure is ≥90 mmHg or increase of both SBP and DBP.[2] Stage I hypertension is >140-159 mmHg/90-99 mmHg and stage II hypertension is >160 mmHg/100 mmHg. (The SI unit of pressure is Pascal where 1 mmHg = 133.3 Pascal, but conventionally mmHg is used for measurement of blood pressure for clinical purposes.[3] Therefore, mmHg will be used as unit for blood pressure measurement throughout the thesis.)

The Canadian Hypertension Education Program (CHEP) also provides recommendations for the management of blood pressure. [4] According to CHEP, hypertension is defined as persistently elevated BP of ≥140 mmHg/90 mmHg. “High normal” blood pressure is defined as SBP >120-130 mmHg and/or DBP 85-89 mmHg, and warrants annual follow-up. Antihypertensive therapy is recommended for patients with macrovascular target organ damage when the SBP is higher than 140 mmHg or

DBP is higher than 90 mmHg, and for patients without macrovascular target organ damage when the SBP is higher than 160 mmHg or DBP is higher than 100 mmHg.

Hypertension may have multiple causes, and can be described as either primary (without an identified specific cause) or secondary (associated with another identified medical condition). The majority of adults with hypertension (90% to 95%) have primary (or essential) hypertension.[2] Genetic and environmental factors likely play an important role in essential hypertension. Hypertension due to known medical conditions is called secondary hypertension.[5] The most common causes of secondary hypertension are anatomical abnormalities (*e.g.*, coarctation of aorta, or aortic stenosis due to atherosclerosis); renal disease (*e.g.*, chronic renal failure), renal artery stenosis; or endocrine abnormalities (*e.g.*, pheochromocytoma, hyperaldosteronism, or Cushing's syndrome). Pharmacologic agents like hormonal contraceptives and alcohol can also induce hypertension.

1.2 Measurement of Hypertension

1.2.1 Sphygmomanometry

Several methods are used for the measurement of BP including physician office-based, home-based and 24-hour ambulatory BP measurements. Both mercury and automatic sphygmomanometers can be used for recording BP. The most commonly used equipment for recording BP is the mercury sphygmomanometer. The bladder cuff of the sphygmomanometer is placed on the upper arm and is inflated until the pressure, as measured by an attached pressure gauge, exceeds the systolic blood pressure. While simultaneously auscultating over the brachial artery with a stethoscope, a valve is

opened allowing air to escape from the bladder cuff. Subsequently, the cuff pressure falls. When the cuff pressure falls below the systolic pressure Korotkoff sounds can be heard, and this pressure is recorded as the systolic blood pressure. When the cuff pressure falls below the diastolic blood pressure the sounds disappear; the pressure at the time of disappearance is recorded as the diastolic blood pressure. The CHEP [6] recommends obtaining three readings from the right arm taken five minutes apart with the patient seated, and recording blood pressure as the average of the last two readings, discarding the first. In clinical practice, it is common to take only one or two readings, however. In case of suspicion of postural hypertension, standing blood pressure should be also measured.

Automated blood pressure measurement devices offer a convenient alternative to the manual measurement described in the preceding paragraph. Home-based automated blood pressure measurement devices are widely available, allowing patients to monitor their own blood pressure at home. However, automatic sphygmomanometers used mostly for recording of BP in home can be prone to errors and should be checked for accuracy regularly by taking sequential measurements; variable readings on sequential measurements indicate faulty equipment and need for replacement of batteries or equipment itself.[7]

1.2.2 Ambulatory Blood Pressure Recordings

Ambulatory blood pressure (ABP) monitoring refers to serial automated measurement of BP at regular intervals throughout an extended time period, *e.g.*, over 24 hours, using a portable device. ABP provides a more comprehensive assessment of average blood

pressure and may better estimate risk of hypertension-induced end organ damage.[8] ABP may also provide more representative readings if office BP measurement are confounded by elevated blood pressure as a consequence of patient anxiety – the so-called “white coat hypertension” syndrome. A typical representative ABP monitoring scheme might include 12 BP readings during sleep and 16 readings while awake, recorded over a 24 h period.[9]

ABP also allows for comparisons between waking blood pressure and nocturnal sleeping blood pressures. Normally the BP drops during sleep, which is called “dipping”. The lack of the normal dipping has been associated with hypertensive end-organ clinical changes such as cardiovascular events, chronic renal failure and stroke.[10] ABP is considered as abnormal if awake BP is greater than 140 mmHg/90 mmHg; sleep BP is higher than 125 mmHg/80 mmHg; or the 24-hour average is more than 135 mmHg/85 mmHg.[11]

1.3 Effects of Hypertension on the Microvasculature

High BP exerts a force on the walls of arteries, leading to functional and structural changes in the endothelium and the vascular wall. These changes include thickening of the arterial wall through proliferation of the vascular media smooth muscle cells and collagen deposition (fibrosis) and dilation and lengthening of the vessels (dolichoectasia). In the small brain arterioles this process of thickening of the arteriolar walls is called arteriolosclerosis. The thicker walls promote vascular integrity, but potentially at the expense of vascular reactivity and elasticity. Hypertensive vessels become stiffer (*i.e.*, less elastic). A consequence of this loss of elasticity in the aorta can

be an increase in the pulse pressure (PP). PP is defined as the difference between systolic and diastolic blood pressure. The normal pulse pressure range is 30 mmHg to 40 mmHg. Pulse pressure measurements of >40 mmHg suggest reduced vascular compliance and are seen in isolated systolic hypertension, a common pattern of hypertension in the elderly in which systolic pressure is elevated (*i.e.*, 140 mmHg) but diastolic blood pressure is normal or even reduced.[12]

More severe vascular changes due to hypertension include microscopic arterial wall trauma leading to aneurysmal dilations. Pathological wall changes include necrosis of segments of the vessel wall with loss of smooth muscle cells, in conjunction with collagen deposition (*i.e.*, fibrinoid necrosis). Loss of wall integrity can cause microscopic leakage of red blood cells or small hemorrhages (which can be detected on MR imaging, as microbleeds). Hypertension is a risk factor for atherosclerosis and likely accelerates atherosclerotic changes. In small arteries (*i.e.*, typically <200 μm diameter), atherosclerotic changes are not seen but pathology studies can identify elevated lipid content in the degenerated walls, a finding called lipohyalinosis.

These hypertensive changes can cause narrowing of the vessel lumen and predispose the vessel to thrombosis, resulting in some cases in thrombotic occlusion of the vessel. Conversely, the loss of vascular wall integrity can predispose to haemorrhaging. Either thrombosis or haemorrhage can result in clinical diseases linked to hypertension (see subsequent section).[13] Vessel occlusion, stenosis and altered vascular reactivity may lead to decreased blood flow. End organ atrophy, defined as loss of cellular elements of the tissue, may result from decreased blood volume,

microscopic infarctions, cellular atrophy because of energy failure leading to lack of protein synthesis, or autophagy.[14] In animal models, predominantly using spontaneously hypertensive rats, hypertension leads to brain atrophy. [15] This above described mechanism of brain atrophy was also proven in animal studies.[15]

1.4 Management of Hypertension

According to CHEP [4] pre-hypertensive individuals and patients with established hypertension should be treated with lifestyle modification. Lifestyle modifications include daily exercise for 30-60 minutes, weight reduction, reduce sodium intake, and modifications of dietary habits with greater intakes of fruit and vegetables than dairy products, grain and high cholesterol food. Stress management can also play an important role in the reduction of blood pressure.

Pharmaceutical agents should be prescribed in those who have blood pressure >160 mmHg/100 mmHg. The first line therapy is diuretics and beta-blocker in patients under 60 year of age; angiotensin converting enzyme (ACE) inhibitor or long acting calcium channel blockers are recommended in older patients. Combinations of these drugs can be used in cases of uncontrolled hypertension.

1.5 Clinical Consequences of Hypertension

The mechanism of microvasculature damage by hypertension was described in section 1.3 and is responsible for initiation of the clinical complications. Hypertension has been called a “silent killer” because it can damage essential body organs – such as the brain, heart, kidney and eyes – without symptoms until devastating clinical events, such as

stroke or myocardial infarction, occur. The major clinical consequences are summarized below.

In the heart, hypertension causes arteriosclerosis and a subsequent narrowing of coronary arteries, eventually leading to cardiac ischemia manifest as angina, myocardial infarction or arrhythmia. Because hypertension opposes the force of contraction of the left ventricle, the workload of the left ventricle is increased which leads to compensatory ventricular hypertrophy and, when compensation is no longer possible, to heart failure.

Hypertensive vascular changes in the kidney result in renal atrophy and renal failure. After diabetes, hypertension is the second most common cause of end stage renal damage.[16] Kidneys have autoregulatory mechanisms that allow the organ to withstand the increases in BP. One of the responses of kidney to increase arterial pressure is natriuresis and diuresis. Increased arterial pressure stimulates the release of endothelial nitric oxide, which decreases the tubular sodium reabsorption and also induces vasodilatation. Renal afferent arterioles constrict in response to elevated BP, which reduces the blood flow to kidney and reduces glomerular filtration as a compensatory mechanism to protect the nephron from excessive pressure and flow. However persistently elevated BP may overwhelm the renal auto-regulatory mechanisms and cause necrosis of afferent arterioles, leading to malignant nephrosclerosis. The kidney also has an important role in regulating blood pressure. When hypertension-induced renal injury severely reduces the glomerular filtration rate, intra-renal angiotensin II level increases because of low sodium, increase sympathetic vascular tone and stress induced stimuli, which activates renin and increases the

conversion of angiotensinogen into angiotensin, which cause retention of sodium,[17] and leads to even more hypertension. Thus, a vicious circle can ensue where hypertension causes renal injury, which begets more severe hypertension.[18]

The most dramatic cerebral consequence of hypertension is stroke. Hypertension is the strongest modifiable risk factor both ischemic stroke and intracerebral hemorrhage. In the brain, hypertension has also been linked to more subtle lesions that probably impair neurological function. High blood pressure, high pulsatile pressure and high blood flow cause damage to small vessels of brain [19] that result in microbleeds, silent brain infarcts, and disruption of connecting pathways of the brain [20] which cause changes in the structure and function of the brain. These hypertensive brain lesions can be detected on MR images. The symptoms depend on the size, localization, and number of ischemic insults,[21] but are often subtle and not clinically recognized. Even when asymptomatic, silent brain infarcts associated with hypertension have been linked to poor performance on cognitive tests and future risk of dementia.[22] Because these clinically unrecognized infarcts may be asymptomatic but are not without consequence on more sensitive testing, they have also been termed “covert” strokes.

The brain normally reduces in size with advancing age;[23] this phenomenon is called senile atrophy. After the age of 40, brain volume is on average 5% lower per additional age decade.[24] The mechanism of senile atrophy is still unclear. Gray matter atrophy is related to death of neurons.[25] White matter volume also reduces with advancing age; part of this reduction may be related to increasing burden of white

matter hyperintensities of presumed vascular origin, possibly caused by ischemic demyelination.[26]

Existing data associate hypertension or higher blood pressure with excessive brain atrophy for a given age. However, the previously published literature is not always consistent and it is often old, reflecting dated management strategies for hypertension that did not include the more aggressive treatment advocated by CHEP and JNC 7. For example, some older studies found that higher BP in midlife was associated with brain atrophy, while others found that higher BP in later life was not, and was actually associated with lower BP.[27] Atrophy in brain sub-regions was evaluated in some studies, but sub-regions were not consistently defined and evaluated across studies. I am not aware of any systematic reviews that have attempted to synthesize this somewhat disparate prior literature.

Knowledge of the presence and rate of brain atrophy in treated and untreated hypertensives would facilitate research into the consequence of hypertension and its treatment. Because atrophy can precede clinical consequences such as cognitive impairment or stroke but can be identified with great accuracy using MR imaging, the identification of hypertension-related atrophy on MR could identify patients that are at higher risk for other forms of end-organ damage such as cognitive impairment or stroke, before the patient is symptomatic. Knowing the circumstances under which atrophy occurs, for example at what blood pressure level or for how long, could help stage the severity of hypertension based on end-organ damage, which might correlate better with future risk of hypertension-related disabilities than simple pressure measurements.

Because atrophy may precede clinical impairments but can be measured, it could be an attractive surrogate marker for treatment effects. That is, the effectiveness of antihypertensive treatment strategies could be based, in part, on the ability of the treatment to reduce the rate of brain atrophy. Finally, the degree of hypertension-associated brain atrophy in a population could potentially be used as a marker of the success of population hypertension control.

1.5 Motivation of the Project

Brain atrophy can be measured reliably using neuroimaging, particularly MR imaging, and may be an important manifestation of hypertension that identifies persons at risk for subsequent hypertension-induced cerebral consequences such as stroke and cognitive impairment. However, the following gaps in current knowledge should be addressed to better understand the relationship between hypertension and brain atrophy. These gaps are:

1. The relationship between duration of hypertension and time of onset of brain atrophy is not known,
2. The relationship between hypertension and specific brain regional atrophy is poorly understood,
3. The relationship between pulse pressure and brain atrophy is not known,
4. The relationship between hypertension-related atrophy and cognitive function is not known, and
5. The relationship between the effect of antihypertensive treatment and brain atrophy is not known.

To address some of these limitations two projects were carried out. In the first project (Chapter 2), a systematic review was performed of all existing literature on blood pressure, hypertension and radiological measures of brain atrophy. In the second project (Chapters 3 and 4), brain MR data were analyzed from 778 persons age 40-70 years participating in the Canadian portion of an international population-based study, PURE-MIND, to determine whether blood pressure or hypertension were associated with global and regional brain atrophy, and thinning of the cerebral cortex. The overall objective of this thesis was to establish a relationship between hypertension and its effect on brain volume.

1.6 Hypotheses

It is hypothesized that higher blood pressure is associated with brain atrophy. This hypothesis is based on the following understanding:

1. Increases in blood pressure and changes in flow velocity cause trauma to small arteries resulting in arteriosclerosis, which in turn causes narrowing and occlusion of small vessels leading to reduced blood flow in brain, and
2. Reduced perfusion of the brain causes deficiency of oxygen and essential nutrients leading to cell death in the brain.

To generate new knowledge that would support or refute general hypothesis, the following specific hypotheses were tested:

1. That the prior literature would provide supporting evidence for a relationship between BP and brain atrophy.

2. In analyses of the PURE-MIND data, that higher systolic BP, diastolic BP and history of hypertension would be associated with greater brain atrophy. In addition the following secondary hypotheses were tested:
    a. That systolic and diastolic BP would be associated with atrophy of certain brain sub-regions including total cortical gray matter and white matter. Based on literature review, it is hypothesized that these sub-regions would be most likely to atrophy: the prefrontal cortex, basal ganglia and cerebellum. It is also hypothesized that ventricular volume would be higher in persons with higher blood pressure, reflecting ex vacuo dilatation due to atrophy in the brain parenchyma.
    b. That the relationship between BP and brain atrophy would depend on whether the person blood pressure was controlled on antihypertensive medications or not. It is hypothesized that patients with poorly controlled blood pressure whether taking antihypertensive medications or not would exhibit the greatest atrophy.

1.7 Thesis Outline

This thesis consists of five chapters. This first chapter provides a broad overview of the motivation, objectives, and hypothesis for this research. Chapter 2 presents a systematic literature review on the relationship between hypertension and brain atrophy. Chapter 3 reviews the most commonly used methods for the analysis of brain volume and describes the methods and advantages of using the particular approach, FreeSurfer, [28] that I used in my research. Chapter 4 presents the methodology and

results of my study of the relationship between blood pressure and brain atrophy in the PURE-MIND study. In the last chapter, the work presented in this thesis is summarized and future directions of the project are proposed.

1.8 Scientific Contributions and Publication Record

I wrote and am responsible for the content of this thesis. Together with my supervisor Dr. Smith, I conceived the projects goals and designed the study analyses. For the systematic review (Chapter 2), I designed the literature search strategy, carried out the searches in PubMed and Web of Science databases, selected the eligible studies, abstracted the study data into our own database, and analyzed and interpreted the findings. In accordance with good practice for systematic review methods, Dr. Smith served as a second reviewer for study eligibility and independently abstracted the study findings; final study eligibility and results were determined by consensus between Dr. Smith and I.

For the MR analysis of BP and brain atrophy (Chapters 3 and 4), I analyzed the PURE-MIND dataset. The PURE-MIND study was designed and coordinated by Dr. Eric E Smith. PURE-MIND study investigators recruited patients at four Canadian sites. Dr. McCreary designed and implemented acquisition protocol across four sites using ADNI protocol, collected all MR data and maintained the local database for consistency with central site. She also exported all T1 data from database and processed it through the FreeSurfer pipeline. Dr. Smith and a radiology fellow (Dr. Jayesh Modi) reviewed study MR images for silent brain infarcts and microbleeds. A summer student, Karla Sanchez, analyzed the study MR for white matter hyperintensity volume. I analyzed the PURE-

MIND FreeSurfer output to determine brain volumes and cortical thickness. A summer student (Vanessa Hill) assisted me for two months and worked under my supervision. In conjunction with Dr. Smith, I designed the statistical analysis plan for determining the relationship between BP and brain atrophy. Dr. Smith acted as my study statistician and conducted study analyses. I interpreted the study findings.

Some components of this project required input of other scientists working at the Seaman Family Magnetic Resonance Research Centre. Dr. Smith, as my thesis supervisor, provided mentorship and support throughout my project. Dr. Richard Frayne, my co-supervisor, provided guidance and feedback on multiple occasions. Both assisted with study design and helped to frame the study conclusions. Dr. McCreary helped me in understanding the FreeSurfer software and guided me on multiple occasions. Dr. Jette provided valuable feedbacks during my committee meetings for improvement of my work.

Portions of this work have been presented as posters at the Alberta Imaging Symposium in Calgary in 2012 and the Campus Alberta Neuroscience Symposium in Edmonton in 2012. Chapters 2 and 4 are being prepared for publication in the journals *Hypertension* and *Neurology*, respectively.

Chapter Two: Association Between Hypertension and Brain Atrophy – A Systematic Literature Review

2.1 Introduction

With advancing age, brain volume decreases.[23] This change is evident by sulcal and ventricular widening. The exact pathogenesis of senile atrophy is still under debate, however cerebrovascular risk factors are thought to be important candidate contributors to brain atrophy. Among those factors, hypertension is a very common and modifiable risk factor, whose prevalence is increasing as the population ages. Previous studies disagree with respect to the relationship between blood pressure and atrophy, with some studies associating hypertension with increased brain atrophy, while others identify lower blood pressure as also being associated with increased atrophy. Across these studies, there has been wide variation in the populations studied, measurement methodology for blood pressure, study design (cross-sectional *vs*. longitudinal) and methods for measuring brain atrophy. All of these factors may have contributed to discordant findings.

It was hypothesized that hypertension or higher blood pressure would be associated with increased brain atrophy independent of age. To test this hypothesis, we

systematically reviewed existing published studies on the relationship between hypertension or high blood pressure and brain atrophy, including both observational and interventional studies. The included studies that were population-based, clinic-based or disease-based (including those focussing on Alzheimer's disease or dementia), but excluded studies focusing specifically on post-stroke brain volume changes due to the difficulty in separating stroke-related atrophy from more general brain atrophy.

2.2 Methods

2.2.1 Search Strategy

Search terms were determined from the consensus of two investigators (EES, a neurologist, and SB, a radiologist), and were designed to have high sensitivity for identification of articles relating hypertension or blood pressure to brain atrophy. These keywords were used to search Pubmed *via* Ovid Medline and Web of Science on 26 March 2013, in order to find articles relevant to hypertension or blood pressure and brain atrophy that were published in English from January 1978 to March 2013. The medical subject headings (MeSH) and search strategy used are given in Table 2-1. Manually searching of reference lists of relevant articles identified using Medline and Web of Science was employed as a strategy to identify additional articles; however, no additional relevant articles were identified.

Table 2.1 Key words used in Medline and Web of Science for systematic literature review.

Search	Terms
Pubmed	**1.** exp Hypertension/ **2.** exp Blood Pressure/ **3.** hypertens*.tw. **4.** blood pressur*.tw. **5.** 1 or 2 or 3 or 4 **6.** exp Brain/ **7.** (brain or cerebr* or cort*).tw. **8.** (atrophy or volum* or Thickness).tw. **9.** 5 and 6 and 7 and 8 **10.** limit 9 to english language **11.** limit 10 to yr="1978 -Current" **12.** limit 11 to humans
Web of Science	**1.** hypertens* or blood pressure* **2.** brain or cerebr* or cort* **3.** atroph* or volume or thickness **4.** #1 and #2 and #3 **5.** Limit to yr="1978 –Current **6.** Excluding] Research Areas=(life sciences biomedicine other topics, allergy, cardiovascular system cardiology, toxicology, mechanics, surgery, nursing, immunology, dentistry oral surgery medicine, acoustics, environmental sciences ecology, rheumatology, endocrinology metabolism, nutrition dietetics, oncology, microscopy, urology nephrology, audiology speech language pathology, paediatrics, medical laboratory technology, food science technology, anaesthesiology, chemistry, spectroscopy, haematology, genetics heredity, research experimental medicine, evolutionary biology, zoology, engineering, substance abuse, marine freshwater biology, ophthalmology, microbiology, sport sciences , materials science, plant sciences, fisheries, geriatrics gerontology, developmental biology, instruments instrumentation, obstetrics gynaecology, otorhinolaryngology, respiratory system, imaging science photographic technology, parasitology, science technology, optics, thermodynamics, transplantation, virology, cell biology, biotechnology applied microbiology anthropology, automation control systems, education educational research, pathology, agriculture, electrochemistry, infectious diseases, oceanography, veterinary sciences, orthopaedics, robotics, biophysics, dermatology, social issues, gastroenterology hepatology, tropical medicine, emergency medicine, general internal medicine, biochemistry molecular biology)

exp=exploded; tw=text word

2.2.2 Eligibility Criteria

Studies were considered eligible if they reported an association, and effect estimate of the degree of association such as an odds ratio or regression model beta coefficient, between either hypertension or blood pressure values and the degree of brain atrophy, measured radiologically, in a defined population. Radiological measures could include qualitative or quantitative assessments of total or regional brain volumes on either CT or MR scanning. Articles reporting either adjusted or unadjusted effects were included. The details of the exclusion criteria are given in Table 2-2. Articles were excluded that did not report a measure of association or quantify the degree of association, were not published in English, involved animal studies only, involved only patients in the acute and post-acute phases of stroke (because it would be difficult to distinguish stroke-related atrophy from general atrophy), included only pathological measures of atrophy without radiological measures, or were published in abstract form only. Studies published prior to 1978 were excluded because CT and MR imaging were not routinely available for clinical use or research prior to then. Authors were not contacted for unpublished data. In cases where multiple studies reported on the same study population, the single best study was selected for inclusion to avoid reporting duplicate studies. The selected articles were screened and reviewed by two individuals (SB, EES).

Table 2.2 Exclusion criteria for Medline and Web of Science of studies in systematic literature review

Medline and Web of Science
1. Not in English Language ***2.*** Animal Studies ***3.*** Effect of hypertension on other systems ***4.*** Effects of metabolic disorders in brain **5.** Studies did not use MR/CT for measurement of brain volume

2.2.3 Data Extraction

Each of the selected articles was evaluated using a pre-specified article report form (see Appendix I) by two individuals (SB, EES) to extract the appropriate data. Extracted data included study author and year of publication and sample size. The study design was categorized as cohort, case control, randomized controlled trial or other, and as a cross-sectional or longitudinal design. Study setting was characterized as either clinic-based, community via advertising or other non-population based recruitment strategy, or population-based. The methodology for measuring atrophy including the radiological modality used (either CT or MR), the method for atrophy assessment, and the brain regions measured, was recorded. Other study data abstracted were mean age, mean BP systolic, diastolic BP, the proportion with hypertension, and the strength of the association of BP changes with brain atrophy, the strength of association was

determined by description of relationship between blood pressure and brain atrophy after controlling for cofounders and by odd ratios.

2.2.4 Quality Assessment of Primary Studies

All the included studies were assessed for their quality by using Newcastle-Ottawa Scale [29] and Standard for Reporting Vascular Changes on neuroimaging (STRIVE) methods [30]. The Newcastle-Ottawa Scale assessed the quality using an eight-point scale for the selection and exposure categories of case control and cohort studies separately. Case control studies received one point for each of the following eight features:

1. The case definition is adequate with independent validation,
2. Representation of the case is appropriate, (*i.e.,* defined time period, catchment area, hospital or clinic or random sample of the cases),
3. Selection of controls from the same community,
4. Controls have no history of disease,
5. Cases and controls are comparable for design and analysis,
6. Exposure is determined by secure medical record or interview from the hospital and not self reported,
7. Same methods for cases and controls, and
8. Non-response rate is similar for both groups.

For cohort studies one point is given for each of the following categories:

1. Sample is representative of the exposed community,
2. Non-exposed cohort is derived from same community,

3. Exposure is determined from secure record or structured interview,
4. The outcome was not present at the start of study,
5. Cohorts are comparable for design and analysis,
6. Blind assessment of outcome,
7. Follow up is long enough for outcome, and
8. Sufficient number of subjects came for follow up.

The recently published STRIVE standards [30] were used to develop a six-point scale that assess the quality of the imaging methods. One point for each of the following features:

1. The image acquisition parameters were described in detail,
2. The qualitative or quantitative method of measuring brain atrophy was described in detail or a URL was provided,
3. The reproducibility of the method was given,
4. The qualification of the raters was reported, and
5. The inter-rater reliability was reported using an appropriate metric, such as a kappa statistic or intra-class correlation coefficient.
6. Total brain volume adjusted for intracranial head size.

For studies that used completely automated methods for brain analysis without user input, the category of 'inter-rater reliability reported' was considered not applicable.

2.2.5 Statistical analysis

Because of wide variation in the categorization of BP, radiological methods used and brain regions measured, there were few studies that reported the association between

BP and brain atrophy in the same way. Therefore, it was not considered appropriate to attempt a meta-analysis of the data. Instead, the findings of the systematic review were tabulated according to setting:

1. Population-based studies,
2. Non-population based studies, and
3. Studies in specific clinical settings, such as diabetes or Alzheimer's disease.

2.3 Results

A flow diagram of the systematic literature search following Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) standards [31] is given in Fig 2-1. The search yielded 672 articles from Medline and 2,222 articles from Web of Science. After eliminating duplicate articles, a total of 2,697 unique articles were identified. Both reviewers screened all 2,697-article titles independently and identified 489 potentially relevant articles. Only non-relevant studies that did not establish relationship between BP and brain atrophy or animal studies were excluded at this stage. Next, both reviewers independently reviewed the article abstracts of these 489 articles and by consensus identified 90 articles for full article review. To increase the sensitivity of our review, the second reviewer (EES) re-reviewed all 2,697 abstracts and identified another 18 articles for full article review. A total of 108 articles underwent full article review. Of these 108 articles, 8 were not accessible (*i.e.*, they could not be retrieved electronically or through inter-library loan at our institution). After independent full article review of the 100 remaining articles by both reviewers, there were 55 articles identified that met all study inclusion and exclusion criteria.

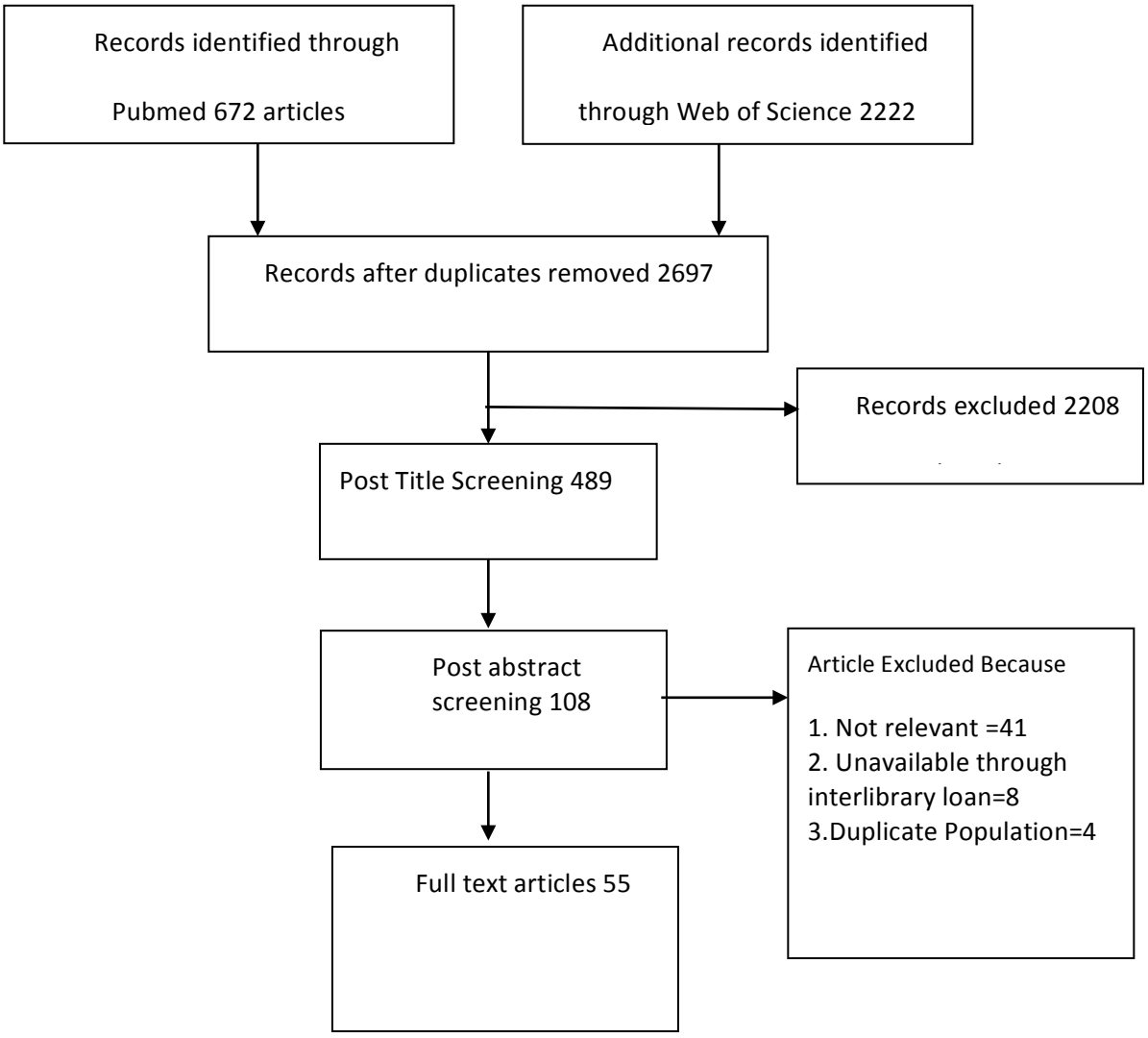


Figure 2.1 Preferred reporting items for systematic reviews and meta-analyses (PRISMA) 2009 flow diagram [31] of our systematic literature review

Among these 55 relevant articles there were 14 case control studies (12 were prospective and 2 were retrospective case studies), 40 cohort studies (36 were prospective, and 4 were retrospective cohort studies), and 1 was a prospective

randomized control trial. The details of recruitment criteria, number of participants, mean SBP and DBP, and method of brain volume measurement are given in Table 2-3

Table 2.3 Data extracted from the 55 reviewed studies that underwent full article review.

Study Author	Year of Publication	N	Type of Study	Settings[1]	Mean Age	Mean SBP/DBP	Atrophy Method[2]	Global/ Regional Volume	Longitudinal
Akiyama [32]	1997	75	Cohort	CA	57.4	NR	Manual Tracing	Y/Y	Y
Brundel [33]	2010	56	Case control	C	70	NR	FreeSurfer	N/Y	N
Burgmans [34]	2010	93	Cohort	CA	62	126/76	Manual Tracing	N/Y	Y
Celle [35]	2012	183	Cohort	P	65	123/78	SPM	N/Y	N
Chen [36]	2006	337	Cohort	P	62.6	NR	SPM	N/Y	N
Chowdhury [37]	2011	1108	Cohort	C	61.8	123/71	4 point scale	N/Y	N
Colloby [38]	2011	68	Case control	C, CA (controls)	74.2	Cases 137/80, Controls 131/75	FIRST	N/Y	N

Davis [39]	2012	79	Cohort	RCT	69	139/NR	SIENAX	Y/Y	N
De Leeuw [40]	2004	152	Cohort	C	68.4	No SVD 147/85 SVD 151/87	Visual Rating	Y/Y	N
Debette [41]	2011	1352	Cohort	P	54	123/NR	Semi automated Methods	Y/N	Y
DeCarli [42]	1999	414	Cohort	C	72.5	138/75	Automated method	Y/N	N
den Heijer [27]	2003	1077	Cohort	P	72	Initial 131/81, Follow-up 147/70	Visual Rating	Y/N	Y
Du [43]	2006	42	Cohort	CA	73.6	NR	Semi automated Method	N/Y	Y
Enzinger [44]	2005	201	Cohort	P	59.8	MR Follow-up139/86, No Follow-up 140/85	SIENAX	Y/N	Y
Firbank [45]	2010	97	Cohort	C	79	144/77	FSL	N/Y	Y
Firbanks [46]	2007	95	Secondary analysis of RCT	RCT	76.4	Cases 158/84 Controls 133/73	MIDAS	Y/N	N
Gattringer [47]	2012	287	Cohort	C	66.6	142/85	SIENAX	N/Y	Y

Gianaros [48]	2006	134	Cohort	CA	60.7	131/78	SPM	N/Y	N
Glodzik [49]	2012	115	Cohort	CA	62.6	134/82	Voxel Based Morphometry	N/Y	N
Godin [50]	2009	1792	Cohort	P	72.4	NR	SPM	Y/Y	N
Goldstein [51]	2002	155	Cohort	C	71	Older 127/73 Younger 125/75	Manual Tracing	Y/Y	N
Goldstein [52]	2005	121	Cohort	CA	66.2	Time1; 119/72 Time2; 126/74	Manual Tracing	Y/Y	Y
Harris [53]	2008	28	Cohort	P	77.2	132/71	Manual Tracing	N/Y	N
Hatazawa [54]	1984	123	Case control	C	50.2	NR	CT pixel based on HU	Y/N	N
Havlik [55]	2002	575	Cohort	P	82	132/83	Visual Rating	Y/Y	N
Hoogendam [56]	2012	3922	Cohort	P	60	135/82	FreeSurfer	N/Y	N
Ikram [57]	2008	490	Cohort	P	73.4	NR	Semi automated Algorithm	Y/Y	N

Jack [58]	1998	48	Case control	C	80.4	NR	Hippocampus Manual Tracing Temporal Lobe, AM	N/Y	Y
Leritz [59]	2011	115	Cohort	C, CA	68.3	133/84	FreeSurfer	N/Y	N
Maillard [60]	2012	579	Cohort	P	39.2	117/76	FSL	Y/Y	N
Manolio [61]	1994	303	Nested case control	P	70.5	NR	Visual Rating	Y/Y	N
Meyer [22]	1999	224	Cohort	C	59.5	NR	Manual Tracing	Y/Y	Y
Muller [62]	2010	965	Cohort	C	58	141/82	Automated Method	N/Y	N
Nagai [63]	2008	55	Cohort	C	72.7	148/82	Manual Tracing Intensity contour Mapping	Y/N	N
Nagai [64]	2009	55	Cohort	C	72.7	Atrophy 162/89, Non-atrophy 143/80	Manual Tracing & pixel intensity threshold	N/Y	N
Nagai [65]	2009	331	Cohort	C	59	Cases 147/84, Controls 123/74	Automated Method	Y/N	Y
Pirtilla [66]	1992	416	Cohort	C	NR	NR	Visual Rating	Y/N	N

Raz [67]	2003	80	Case control	P	61	Cases 138/85 126/80 controls	Manual Tracing	Y/Y	Y
Raz [23]	2005	72	Cohort	NC	52.6	NR	Manual Tracing	Y/Y	Y
Sakurai [68]	2006	67	Case control	C	69.45	Cases 135/74, Control 110/65	Manual Tracing	N/Y	N
Salerno [69]	1992	35	Case control	C	68.5	Cases 145/89, Controls 131/79	Manual Tracing	Y/Y	N
Savazzi [70]	1985	80	Case control	C	NR	NR	Visual Rating	Y/N	N
Schmidt [71]	2004	1252	Cohort	P	68.9	NR	Visual Rating	Y/Y	N
Seo [72]	2012	385	Cohort	C	72.1	NR	Semi automated Method	Y/Y	N
Skoog [73]	1998	484	Cohort	P	85	162/79 ND, 148/77 D	4-pt ordinal scale for each lobe	N/Y	N
Strassburger [74]	1997	47	Case control	C, NR (control)	67.4	NR	Manual Tracing segmentation of CSF by signal intensity	Y/N	N
Taki [75]	2006	405	Cohort	CA	47	131/80	SPM segmentation of GM divided By ICV ('GM ratio')	N/Y	N

Vlek [76]	2009	331	Cohort	C	58	138/80	Automated Probabilistic Segmentation	Y/Y	Y
Waldstein [77]	2012	113	Cohort	p	66	133/75	Visual Rating	Y/N	N
Walters [78]	2003	86	Case control	C	71.7	Cases 152/86, controls 146/83	Brain boundary shift integral	Y/N	Y
Ward [79]	2005	114	Cohort	C, CA	54.2	132/79	SIENAX	Y/N	N
Widya [80]	2011	471	RCT	C	74.8	Cases 158/87 Controls 155/84	SIENAX	Y/Y	N
Wiseman [81]	2004	154	Case control	C	76.88	Cases 166/88, controls 132/74	ANALYZE	Y/Y	N
Yamano [82]	1999	78	Case control	C	63	Cases149/85 Controls 128/76	Evans ratio	N/Y	Y
Ylikoski [83]	2000	113	Cohort	P	NR	NR	4 pt scale	Y/Y	N

RCT = randomized controlled trial; NR = not reported.

[1]C = clinic-based recruitment, CA = community advertising, indicating recruitment from the community via advertising or another non-population based strategy, P =population-based.

[2]ANALYZE = Software to calculate brain volume; FSL = FMRIB software library; FIRST = FSL program ; SIENAX = FSL program ; SPM = Statistical parametric mapping ; AM=Automated method; HU=Hounsfield unit

Study quality, according to Newcastle-Ottawa scale, is given for case-control studies in Table 2-4 and for cohort studies in Table 2-5. The overall mean of study quality scores for case control studies was 4.5 and for cohort studies were 4.5 (out of a possible 8 points). The most common reasons for low study quality were non-representativeness of the cases (12/14) 85.7%, controls (12/14) 85.7% or cohort participants (31/41) 75.6%, and lack of longitudinal follow-up for cohort studies (10/41) 24.3%. The quality assessment for image acquisition and atrophy measurement according to STRIVE is given Table 2-6. Almost all studies described image acquisition parameters according to STRIVE standards while only (11/42) 26.2% studies reported details about the qualifications of study raters, and (12/39) 30.7% studies reported inter-rater reliability for atrophy measurements. None of the studies were excluded on the basis of the quality assessments.

Table 2.4 Quality of the case control studies according to Newcastle-Ottawa scale

Study Author and Year	Is the case definition adequate?	Case Representativeness	Selection of Control	Definition of Controls	Ascertainment Of exposure	Comparability Of case and Control	Same method of exposure ascertainment for cases and controls	Non-response rate for exposure	Total score (out of 8)
Hatazawa 1984	no	no	no	no	yes	no	yes	no	2
Savazzi 1985	yes	no	no	no	yes	yes	yes	no	4
Salerno 1992	yes	no	no	yes	yes	yes	yes	yes	6
Strassburger 1997	no	no	no	yes	yes	n/a	yes	yes	4
Jack 1998	yes	no	no	yes	yes	yes	yes	no	5
Yamano 1999	yes	no	no	yes	yes	no	no	no	3
Raz 2003	no	no	no	yes	yes	yes	yes	yes	5
Walters 2003	yes	no	no	yes	yes	yes	yes	yes	6
Wiseman 2004	yes	no	no	yes	yes	yes	yes	yes	6
Sakurai 2006	yes	no	no	no	yes	n/a	yes	yes	4
Brundel 2010	no	no	no	yes	yes	yes	yes	no	4
Colloby 2011	yes	no	yes	yes	yes	yes	yes	yes	7
Seo 2012	yes	yes	no	no	yes	yes	no	no	4
Waldstein 2012	yes	yes	yes	n/a	n/a	n/a	n/a	no	3

n/a = not applicable

Table 2.5 Quality of selected cohort studies according to Newcastle-Ottawa scale

Study Author Year	Representativeness of the exposed cohort	Selection of the non exposed cohort	Ascertainment of exposure	Demonstration that outcome of interest was not present at start of study	Comparability of cohorts on the basis of the design or analysis	Assessment of out come	Was follow-up long enough for outcomes to occur	Adequacy of follow up of cohorts	Total Score (out of 8)
Pirtilla 1992	no	no	no	n/a	no	yes	n/a	n/a	1
Manolio 1994	no	yes	yes	n/a	yes	yes	n/a	n/a	4
Akiyama 1997	no	yes	no	yes	yes	no	yes	no	4
Skoog 1998	yes	yes	yes	n/a	yes	yes	n/a	n/a	5
DeCarli 1999	no	no	yes	no	yes	yes	yes	no	4
Meyer 1999	no	no	no	yes	yes	yes	yes	no	4
Ylikoski 2000	no	yes	yes	no	yes	yes	n/a	n/a	4
Goldstein 2002	no	yes	yes	no	yes	yes	n/a	n/a	4
Havlik 2002	yes	yes	yes	no	yes	yes	yes	no	6
den Heijer 2003	no	yes	yes	no	yes	yes	yes	no	5
De Leeuw 2004	no	yes	yes	no	no	yes	n/a	n/a	3
Schmidt 2004	yes	yes	yes	no	no	yes	n/a	n/a	3
Enzinger 2005	no	yes	yes	yes	yes	yes	yes	no	4
Goldstein 2005	no	yes	yes	yes	yes	yes	yes	yes	7
Raz 2005	no	yes	no	yes	yes	yes	yes	no	5
Ward 2005	no	no	yes	no	yes	yes	n/a	n/a	3
Chen 2006	yes	yes	yes	yes	yes	yes	n/a	n/a	6
Du 2006	no	yes	no	yes	yes	yes	no	no	4
Gianaros 2006	no	yes	yes	no	yes	yes	n/a	n/a	4
Taki 2006	no	yes	yes	no	yes	yes	n/a	n/a	4
Firbanks 2007	no	yes	yes	yes	yes	yes	yes	no	6
Harris	no	yes	yes	no	yes	yes	n/a	n/a	4

2008									
Ikram 2008	no	yes	yes	no	yes	yes	n/a	n/a	4
Nagai 2008	no	yes	yes	no	yes	yes	n/a	n/a	4
Godin 2009	yes	yes	yes	no	yes	yes	n/a	n/a	5
Nagai 2009	no	yes	yes	no	yes	yes	n/a	n/a	4
Vlek 2009	no	yes	yes	yes	yes	yes	yes	no	6
Burgmans 2010	no	yes	yes	no	yes	yes	n/a	n/a	4
Muller 2010	no	yes	yes	no	yes	yes	n/a	n/a	4
Chowdhury 2011	no	yes	yes	no	yes	yes	n/a	n/a	4
Debette 2011	yes	yes	yes	yes	yes	yes	yes	no	7
Leritz 2011	no	yes	yes	no	no	yes	n/a	n/a	4
Celle 2012	no	yes	yes	no	yes	yes	n/a	n/a	4
Davis 2012	no	yes	yes	no	yes	yes	n/a	n/a	4
Glodzik 2012	no	yes	yes	no	yes	yes	n/a	n/a	5
Hoogend-am 2012	yes	yes	yes	no	yes	yes	n/a	n/a	5
Maillard 2012	no	yes	yes	no	yes	yes	n/a	n/a	4
Gattringer 2012	yes	yes	yes	no	no	yes	n/a	n/a	4
Nagai 2009	no	yes	yes	yes	no	no	yes	yes	5
Firbank 2010	yes	yes	yes	no	yes	yes	yes	yes	7
Widya 2011	yes	yes	yes	no	yes	yes	n/a	n/a	5

n/a = not applicable

Table 2.6 Assessment of study neuroimaging quality according to STRIVE criteria

Study Author and Year	Image acqusition	Whole brain adjusted for intracranial volume	Details of qualitative visual rating and quantitative computational methods, including the URL if available for download or an appendix describing the method in detail.	For studies using computational image analysis programmes: training of the analysts, any expert supervision, and the background of the expert; repeatability.	qualification of rater	inter rater reliability	Total score (out of 6)
Hatazawa 1984	yes	yes	yes	no	n/a	n/a	3
Savazzi 1985	yes	no	yes	n/a	no	no	2
Pirtilla 1992	yes	n/a	no	n/a	yes	yes	3
Salerno 1992	yes	yes	yes	yes	yes	no	5
Manolio 1994	yes	n/a	yes	n/a	yes	no	3
Akiyama 1997	yes	yes	yes	n/a	no	no	3
Strassburger 1997	yes	yes	yes	n/a	no	no	3
Jack 1998	yes	no	n/a	yes	yes	no	3
Skoog 1998	yes	no	yes	n/a	yes	no	3
DeCarli 1999	yes	yes	yes	yes	no	no	4
Meyer 1999	yes	yes	yes	no	no	no	3
Yamano 1999	yes	yes	yes	n/a	no	no	3
Ylikoski 2000	yes	n/a	yes	n/a	no	no	2
Goldstein 2002	yes	yes	yes	n/a	no	no	3
Havlik 2002	yes	n/a	yes	n/a	no	no	2
Den Heijer 2003	yes	n/a	yes	n/a	no	yes	3
Raz 2003	yes	yes	yes	yes	no	yes	5
Walters 2003	no	n/a	yes	no	n/a	n/a	1
De Leeuw 2004	yes	yes	no	n/a	no	no	2
Schmidt 2004	yes	n/a	yes	n/a	no	no	2
Wiseman 2004	yes	yes	yes	no	yes	no	4

Enzinger 2005	yes	yes	yes	no	no	n/a	3
Goldstein 2005	yes	yes	yes	n/a	no	no	3
Raz 2005	yes	yes	yes	n/a	no	yes	4
Ward 2005	yes	yes	yes	no	n/a	n/a	3
Chen 2006	yes	yes	yes	no	n/a	n/a	3
Du 2006	yes	yes	yes	no	no	yes	4
Sakurai 2006	yes	no	yes	n/a	no	no	2
Gianaro 2006	yes	no	yes	no	n/a	n/a	2
Taki 2006	yes	yes	yes	no	n/a	n/a	3
Firbanks 2007	yes	no	yes	yes	yes	yes	5
Harris 2008	yes	yes	yes	no	no	yes	4
Ikram 2008	yes	no	yes	yes	no	yes	4
Nagai 2008	yes	no	yes	yes	yes	no	4
Godin 2009	yes	yes	yes	no	n/a	N/A	3
Nagai 2009	yes	no	yes	yes	yes	yes	5
Vlek 2009	yes	n/a	yes	yes	n/a	n/a	3
Nagai 2009	yes	no	yes	no	no	no	2
Firbank 2010	yes	no	yes	no	no	yes	3
Widya 2011	yes	no	yes	no	no	no	2
Brundel 2010	yes	yes	yes	no	n/a	n/a	3
Burgmans 2010	yes	yes	yes	no	no	no	3
Muller 2010	yes	yes	yes	no	n/a	n/a	3
Chowdhury 2011	yes	n/a	yes	n/a	yes	no	3
Colloby 2011	yes	yes	yes	yes	no	n/a	4
Debette 2011	yes	no	yes	no	no	no	2
Leritz 2011	no	no	yes	no	n/a	n/a	1
Celle 2012	no	n/a	yes	no	n/a	n/a	1
Davis 2012	yes	yes	yes	no	n/a	n/a	3

Glodzik 2012	yes	yes	yes	no	n/a	n/a	3
Hoogendam 2012	yes	yes	yes	yes	no	n/a	4
Maillard 2012	no	yes	no	no	no	no	1
Gattringer 2012	yes	yes	yes	no	yes	yes	5
Seo 2012	yes	no	yes	no	no	no	2
Waldstein 2012	yes	no	yes	no	no	no	2

n/a = not applicable

The results of the 14 population-based studies are summarized in Table 2-7. There were 12 population-based studies involving 9,483 participants that showed an association between hypertension and brain atrophy. In 11 out of 12 studies, higher BP was associated with more brain atrophy. However, one study (that involved the oldest participants) showed an association between lower BP and brain atrophy. There were two population-based studies including 1,905 participants that did not show an association between BP and brain atrophy.

Table 2.7 Summary of 14 population-based studies

Study author and year	N	Mean age	Mean BP	Prevalence of Hypertension	Longitudinal	Findings
Studies reporting an association between hypertension or blood pressure and brain atrophy						
Manolio 1994	303	70.5	NR	35%	no	HTN is associated with global atrophy
Skoog 1998	484	85	162/79 ND, 148/77 D	NR	no	Low SBP, DBP is associated with frontal and parietal cortical atrophy
Havlik 2002	575	82	132/83	22.6%	no	SBP is associated with ventricular atrophy
den Heijer 2003	1077	72	Initial 131/81, Follow-up 147/70	35%	yes	High DBP is associated with cortical atrophy
Chen 2006	337	62.7	NR	32.7%	no	In men HTN is associated with GM atrophy in frontal & Temporal lobe
Harris 2008	28	77.2	132/71	46.4%	no	High SBP is associated with atrophy of posterior corpus callosum
Ikram 2008	490	73.4	NR	51%	no	DBP is associated with brain atrophy
Debette 2011	1352	54	123/NR	26.4%	yes	Midlife HTN or SBP is not associated with longitudinal change in brain volumes
Celle 2012	183	65	123/78	33.3%	no	HTN is associated with GM atrophy of frontal region
Hoogendam 2012	3962	60	135/82	53.9%	no	DBP is associated with brain atrophy
Maillard 2012	579	39.2	117/76	NR	no	SBP is associated with GMV loss
Waldstein 2012	113	66	133/75	29%	no	Higher BP is associated with brain atrophy
Studies that failed to find an association between hypertension or blood pressure and brain atrophy						
Ylikoski 2000	113	NR	NR	27.4%	no	HTN is not associated with brain atrophy
Godin 2009	1792	72.4	NR	77%	no	HTN is not associated with GM, Hippocampus or CSF volumes

NR = not reported

The results of 25 studies recruiting participants via clinics, community advertising or other non-population-based strategies are shown in Table 2.8. There were 17 studies with 3,217 participants that found an association between hypertension or higher BP and more brain atrophy. Another 8 studies involving 2,385 participants did not find an association between BP and atrophy.

Table 2.8 Results of studies recruiting from clinics or via community advertising.

Study author and year	N	Mean age	Mean BP	Prevalence of hypertension	Longitudinal	Findings
Studies reporting an association between hypertension, blood pressure and brain atrophy						
Hatazawa 1984	123	50.2	NR	35.2%	no	HTN is associated with lower brain volume index
Salerno 1992	35	68.5	Cases 145/89, Control, 131/79	51.4%	no	HTN is associated with ventricular dilatation
Akiyama 1997	75	57.4	NR	70.2%	yes	HTN is associated with brain atrophy
Strassburger 1997	47	67.4	NR	57.4%	no	HTN causes atrophy of temporal and occipital lobes
DeCarli 1999	414	72.5	138/75	32%	no	DBP is associated with brain atrophy
Meyer 1999	224	59.5	NR	41.5%	yes	HTN is associated with brain atrophy & ventricular dilatation
Yamano 1999	78	63	Cases14 9/85 Controls 128/76	48%	yes	Better control of HTN is associated with less brain volume loss
Raz 2003	80	61	Cases 138/85 126/80 controls	50%	yes	Controlled HTN is associated with brain atrophy
Walters 2003	86 (60 TIA, 26 C)	71.7,	Cases 152,/86 controls 146/83	NR	yes	High DBP is associated with increased brain atrophy rate
Wiseman 2004	154	76.88	Cases 166/88 controls 132/74	66.8%	no	Moderate HTN is associated with smaller brain volume
Raz 2005	72	52.6	NR	14.9%	yes	HTN is associated with hippocampal atrophy
Gianaros 2006	134	60.7	131/78	31.3%	no	High SBP is associated with GMV loss in frontal & temporal lobe in men
Taki 2006	405	47	131/80	NR	no	SBP is associated with GM atrophy

Firbanks 2007	95	76.4	Cases 158/84 Controls 133/73	28.9%	no	HTN is associated with brain atrophy
Muller 2010	965	58	141/82	42%	no	Low SBP &PP is associated with cortical atrophy
Leritz 2011	115	68.3	133/74	49%	no	High BP is associated with cortical atrophy of frontal, temporal & occipital regions
Glodzik 2012	115	62.6	134/82	34%	no	HTN is associated with GMV loss
Studies that failed to find an association between hypertension or blood pressure and brain atrophy						
Ward 2005	114	54.2	132/79	NR	no	HTN is not associated with global brain atrophy
Du 2006	42	73.6	NR	16.6%	yes	HTN is not associated with brain atrophy
Nagai 2009	331	59	Cases 147/84, Controls 123/74	63.1%	yes	HTN is not associated with brain atrophy
Vlek 2009	331	58	138/80	63%	yes	HTN is not associated with brain atrophy
Burgmans 2010	93	62	126/76	38.7%	yes	HTN is not associated with brain atrophy
Chowdhury 2011	1108	61.8	123/71	0 %	no	HTN is not associated with brain atrophy
Davis 2012	79	69	139/NR	NR	no	SBP is not associated with TBM ,GMV loss
Gattringer 2012	287	66.6	142/85	52.9%	yes	HTN is not associated with brain atrophy

NR = not reported

There were 4 studies, including 386 participants, which reported relationships between ambulatory blood pressure and brain atrophy. All 4 studies reported a relationship between some aspect of ambulatory blood pressure and brain atrophy, details are given in Table 2-9.

Table 2.9 Results of studies of using 24-h ambulatory blood pressure monitoring.

Study author and year	N	Mean Age	Mean BP	BP parameters tested	Prevalence of hypertension	Longitudinal	Findings
Goldstein 2002	155	Older 71 Younger 61.1	Older 127/73 Younger 125/75	Casual, Wake Sleep, Wake and sleep variabilities	100%	no	Combination of high BP and greater variability is associated with brain atrophy
Goldstein 2005	121	66.2	Time1: 119/72 Time2: 126/74	Casual, Wake Sleep, Wake and sleep variabilities	8.2%	yes	Small increase in casual and 24 ambulatory BP associated with greater brain atrophy after 5 years.
Nagai 2008	55	72.7	148/82	Clinic SBP/ DBP Morning SBP /DBP 24-h SBP/DBP Awake SBP/ DBP Sleep SBP/ DBP Nocturnal SBP/DBP dipping (%)	100%	no	Sleep SBP is associated with TBM volume loss.
Nagai 2009	55	72.7	Atrophy 162/89, Non atrophy 143/80	Clinic SBP/ DBP Morning SBP /DBP 24-h SBP/DBP Awake SBP/ DBP Sleep SBP/ DBP Nocturnal SBP/DBP dipping (%)	100%	no	Clinic, 24 h, and sleep SBP, nocturnal SBP dipping, high nocturnal BP, and dipper pattern in SBP, are associated with intracranial volume loss.

Studies in special conditions – *i.e.*, depression, hemodialysis, obesity and high homocysteine level – generally did not find significant associations between blood pressure and reduced brain volume (Table 2-10). However, most of these studies were

small (<200 participants). The largest study showed a relationship between hypertension and atrophy in patients with diabetes.[71]

Table 2.10 Summary of studies in special patient populations.

Study author and year	N	Mean age	Mean BP	Prevalence of hypertension	Longitudinal	Findings
Studies in patients with diabetes						
Pirtilla 1992	416	NR	NR	26.2%	no	HTN is not associated with brain atrophy
Schmidt 2004	1252	68.9	NR	44.6%	no	Diabetics with HTN showed brain atrophy
Enzinger 2005	201	59.8	MR Follow-up139/86, No Follow-up 140/85	31.8%	yes	HTN is not associated with brain atrophy
Sakurai 2006	67	69.45	Cases135/74, Control110/65	NR	no	DBP is associated with frontal lobe atrophy
Brundel 2010	56	70	NR	75%	no	HTN is not associated with TBV, hippocampus and temporal lobe volume loss
Studies in patients with Alzheimer's disease						
Jack 1998	48	80.4	NR	47.9%	yes	HTN is not associated with brain atrophy
De Leeuw 2004	152	68.4	NoSVD 147/85 SVD 151/87	77.8%	no	HTN is not associated with hippocampal & subcortical atrophy
Seo 2012	385	72.1	NR	93.7%	no	HTN is associated with cortical thinning in frontal lobe
Studies in patients with Depression						
Colloby 2011	68	Cases 74.23	Cases137/80, Controls 131/75	6.1%, 6.4%	no	HTN is not associated with subcortical volumes loss
Studies in Hemodialysis patients						
Savazzi 1985	80	NR	NR	66.6% Predialysis	no	HTN is not associated with brain atrophy
Studies in patients with elevated Homocysteine levels						
Firbank 2010	97	79	144/77	71.5%	yes	DBP is associated with GMV loss, but not associated with atrophy rate
Studies in Obese patients						
Widya 2011	471	74.8	Cases158/87 Controls 155/84	NR	no	HTN is not associated with brain atrophy

Total 35 out of 55 studies looked for relationship between BP and regional brain atrophy, however different studies analyzed different regions. The most commonly measured regions were frontal, parietal, temporal lobe, gray matter, white matter, hippocampus and corpus callosum.

2.4 Discussion

A total of 55 original research articles were reviewed examining the relationship between hypertension and brain atrophy. No previous systematic reviews were identified. Most research studies found a relationship between higher BP and more brain atrophy. Most studies were conducted in older age groups. None of the studies suggested the atrophy of basal ganglia and cerebellum with hypertension, although these two are the common sites for presence of lacunar infarcts, microbleeds and intracerebral haemorrhages in hypertensive patients.

The studies reviewed did not establish the mechanism of the association between hypertension and brain atrophy. One potential mechanism by which hypertension could cause atrophy is by causing white matter hyperintensities (WMHs) of presumed vascular origin. Hypertension is a known risk factor for WMH,[84] and separate studies have also linked WMH with brain atrophy independent of age. Indeed, WMHs were measured and analyzed in some of the studies in this systematic review. However, in none of these studies was WMH analyzed as a potential mediator of the relationship between hypertension and brain atrophy. Future studies should seek to determine whether the occurrence of atrophy in hypertensive patients depends on the appearance

of WMH or silent brain infarcts. Some of these studies tried to establish an indirect relationship between increased WMH burden due to hypertension and brain atrophy.

A single study found that reduction in volume of the frontal and parietal cortices was correlated to lower, not higher, blood pressure in 85-year old non-demented men.[73] This study included the most elderly subjects of any of the studies. It is possible that, in the very elderly, low BP in patients with poor autoregulation could cause a fall in cerebral perfusion pressure, a reduction in cerebral blood volume and even ischemic injury, leading to atrophy. The relationship between blood pressure and atrophy could differ in the very elderly, but more studies are needed. Many studies involved hypertension assessments in the 1990's when the treatment of hypertension was not as effective compared to today. In no study was the relationship between use of antihypertensive medicines and brain atrophy established.

The timing of onset of atrophy after hypertension remains ill defined. Of 27 population-based or clinic/ad based studies with mean age >60, 21/27 (77.7%) reported a significant association while 6/27 (22.2%) failed to find an association. By comparison, of 11 population-based or clinic/ad based studies with mean age <60, 8/11 (72.7%) reported a significant association while 3/11 (27.2%) failed to find an association. Most of these studies linked mid-life hypertension with late life atrophy. One population based study that failed to establish a relationship did not report the mean age (age range 55-85years).[83] Two population-based studies [36, 41] found that BP measured in mid-life (mostly in the 50s) predicted the presence of brain atrophy on MRI done decades later, in the 70s; these studies demonstrate that hypertensive patients in mid-life are at

increased risk of developing brain atrophy in subsequent decades, but the timing of appearance of atrophy is unclear. Only one study [77] in middle aged subjects (<68) showed that mid-life SBP and DBP were related to mid-life atrophy determined by ventricular and sulcal widening. It is still unclear if midlife SBP, DBP and PP are related to regional brain atrophy.

Several studies 22/55 (40%) did not find a significant association between hypertension and brain volume. In many cases these were smaller studies investigating study hypotheses not related to hypertension, and therefore may not have had sufficient sample size to detected relationships between BP and brain atrophy.[79] [43] An alternate explanation is related to publication bias—that is, that studies investigating the hypothesis that hypertension leads to brain atrophy may be more likely to be published when the results are positive than when they are negative. The studies that analyzed BP and brain atrophy as covariates only, and not as the primary goal of the study, may better reflect the true relationship between BP and brain atrophy because their publication should not depend on the nature of that relationship. Unfortunately, we are not able to apply a formal statistical test for publication bias, such as Egger's test, because there was no consistent measure of effect size reported by all studies.

Of four studies that used ambulatory BP recordings, all found a relationship between some aspect of daily BP and brain atrophy. However, each study evaluated multiple BP measurements (e.g. casual BP, average sleep and wake BP, the presence or absence nocturnal 'dipping') without using any formal methods to control for multiple comparisons. For example, two studies found relationships between sleep BP and

atrophy while two others did not, and the reasons for the lack of reproducibility are not clear. One study was longitudinal; it found that longitudinal increase in sleep SBP variability was associated with more atrophy on follow-up MR after 5 years compared to baseline. The ambulatory BP studies were relatively small (sample sizes 55-155), probably reflecting the effort and expense of ambulatory BP recordings. To obtain consistent results regarding which aspects of ambulatory recordings predict brain atrophy, additional longitudinal studies will be needed.

Many studies investigated the relationship between BP and atrophy in specific brain regions. For the most part, these studies investigated specific hypotheses targeting single brain regions (*e.g.*, that higher BP would be associated with hippocampal atrophy) rather than comprehensively assessing multiple brain regions. In most cases, studies did not assess whether regional brain atrophy was out of proportion to global atrophy, as could occur if specific brain regions were more vulnerable to the effects of hypertension. The list of brain regions that exhibit BP-related atrophy is long: *e.g.,* prefrontal cortex, gray matter, white matter, corpus callosum, basal ganglia, hippocampus, ventricles, parietal, temporal and occipital lobes. Future studies should investigate the timing of atrophy, such that patterns of atrophy indicative of mild (early), moderate and severe target organ damage can be recognized, if they exist.

Many of the included studies were of only moderate quality according to Newcastle-Ottawa quality assessment scale. The most common limitation was that, except for the population-based studies, the study samples were not selected to be representative of the general population. Only 17/55 (30.9%) were longitudinal. However all studies

recorded BP according to acceptable standards. Reporting of the imaging methods was generally good, with the exceptions that there was variable reporting of the qualifications of study raters and the inter-rater reliability of the methods.

Strength of this systematic review is that a large number of studies have been published on the topic of hypertension and brain atrophy. However, some additional limitations also need to be acknowledged. There was no consistent measure of effect used across all studies; this is not surprising given the variety of ways in which hypertension or BP can be measured (*e.g.*, as SBP, DBP, mean BP, or based on ambulatory recordings) and in which metric was used to define atrophy (*e.g.*, as whole brain atrophy, ventricular volume or width, sulcal widening, *etc.*). We did not contact study authors to reanalyze their data to provide a consistent measure of effect because this would have been impractical for the study authors, given the time elapsed since their publication and the time that would have been needed to do new neuroimaging analysis. However, the absence of a consistent measure of effect meant that we could not do meta-analysis or formal statistical tests for publication bias. Articles published in languages other than English were not translated; however, there were few studies excluded for this reason. A small number of articles were excluded because they could not be retrieved. However, the abstracts of these articles described small non-population based studies published in minor journals; therefore, it is unlikely that the findings of these articles would have changed our overall results. While searching for literature, we also did not identify any other systematic review relevant to this topic. We believe that this systematic literature review will be a valuable addition to literature. We

searched two databases, Pubmed and Web of Science, to identify relevant studies. Due to time limitations other databases were be searched.

2.5 Conclusion

A systematic review of the literature supports an association between BP and brain atrophy. However, the mechanisms of this association—for example, whether it is driven by accumulation of WMH and silent brain infarction--has not been established definitively because of a dearth of longitudinal studies or mediation analyses. One study suggested that the relationship between BP and brain atrophy might inverse in the very elderly, such that higher BP might be associated with less, not more, atrophy in persons in their 80s. Future work is required to establish the longitudinal relationship between midlife hypertension, the timing of onset of atrophy and whether specific brain regions are more vulnerable than others. Most of the studies ignored the potential effect of antihypertensive treatments on brain volume. Future studies, including randomized controlled trials, are needed to determine the degree to which atrophy is preventable with adequate BP treatment.

Chapter Three: Brain Volume Analysis

3.1 Significance of Brain Volume Measurements

Cerebral atrophy is defined as a loss of neurons, glial cells and axons. Brain size can vary physiologically with changes in the external and internal environment of the human body. Dehydration, release of hormones, *e.g.*, corticosteroids, growth hormone and pregnancy [85] can affect the brain size temporarily but the number of brain cells remains constant under these conditions. Damage of brain cells due to ischemia or physical trauma causes permanent loss of brain tissue and cells, which causes permanent reduction in brain size and may cause changes in behavior, intellect, memory and attention. Brain volume can be reduced globally or in specific regions, *e.g.*, the hippocampus. Clinical symptoms of atrophy are usually related to the function of the affected region or regions. Global atrophy may be associated with symptoms of dementia such as impaired memory, attention, learning abilities and judgment. Regional atrophy gives specific symptoms, *e.g.*, hippocampal atrophy is associated with depression and cognitive decline,[37] involvement of temporal lobe may be associated with seizures [58] and cerebellar atrophy causes impaired balance and coordination. Various diseases, such as multiple sclerosis,[86] epilepsy, stroke and Alzheimer's disease,[87] cause brain atrophy although often through different mechanism. The brain

also shrinks with advancing age, however the rate of atrophy differs between healthy individuals and those with disease.[88]

Neurocognitive research studies and clinical trials often compare brain volumes between healthy and disease populations to establish the patterns of brain volume change between two groups.[89] Different diseases have different effects on structure and function of brain. Variable atrophy patterns are associated with different conditions. Normal aging, for example, usually causes generalized atrophy of the brain while hippocampal-specific atrophy is associated with Alzheimer's disease. Volume measurements can be used to measure the progression of disease and effect of treatment.[90] Therefore there has been considerable interest in the measurement of global and regional brain atrophy using neuroimaging. This chapter will briefly review principles of brain atrophy measurement using MR imaging, describe existing software tools for analysis of brain atrophy and provide an enhanced discussion of a specific software tool that was used to analyze brain atrophy in this study of the relationship between BP and brain atrophy.

3.2 Methods for Measuring Brain Volume

Measurement of brain volume can be categorized into global and regional volume methods that can be estimated with various quantitative or qualitative methods from computed tomography (CT) or MR images. MR provides better contrast between gray and white matter than CT, allowing easier separate estimation of gray and white matter volumes and more accurate estimation of brain volume than CT [87, 91, 92]. Magnetization-prepared rapid acquisition with gradient echo (MPRAGE) is a specific T1-

weighted MR imaging sequence that provides better contrast between gray and white matter than conventional spin-echo T1 sequence,[93] thus enabling automated tissue segmentation, surface reconstruction,[89] and tissue and structure specific volume measurements. Despite the clinical utility of MR in diagnosis of neuropathology, the clinical use of MR is less widespread compared to CT because MR is less available, perceived as being of higher in cost, has longer scan times and is limited in applicability because some patients cannot undergo MR because of contraindications (*e.g.*, claustrophobia or implanted metallic devices). Multi detector CT scanning is an alternative imaging modality in these circumstances.

3.2.1 Qualitative Assessment Methods

Qualitative visual assessments are based on evaluating sulcal widening, ventricular enlargement, and relative filling of the intracranial vault or a combination. Grading systems [27, 37, 55, 61, 77] have been developed for visual assessment of atrophy of specific structures such as the ventricles and medial temporal lobe. Visual rating can be done quickly but the inter- and intra-rater reliability can be poor without extensive experience and training, and small differences in volume cannot be distinguished.

3.2.2 Quantitative Assessment Methods

Quantitative methods deal with exact volume estimation, which may use manual, semi-automated or fully automated techniques. Manual tracing of brain regions is operator time-intensive and requires good neuroanatomical knowledge. The most basic manual method involves an individual manually tracing the entire brain or structures of interest through consecutive image slices on a computer and calculating volumes from the

number of voxels enclosed by the outline.[94] Another manual approach is to use stereology, which is a point counting method to calculate volume or surface area by systematically sampling the structure of interest to estimate volume or surface area. In this method, a grid of points is placed on area of interest and number of points within the structure is counted. Manual methods are time consuming, sometimes inaccurate and require comprehensive knowledge of neuroanatomy and practical experience.

Various automated and semi-automated techniques are freely available for research purposes; the most commonly used software packages include the Functional magnetic resonance brain imaging group Software Library (FSL),[95] FreeSurfer,[28] and Statistical Parametric Mapping (SPM).[96] These methods are fast, operator independent, reliable, repeatable and more precise in volume estimation than the manual methods.[94] FSL is automated software that is also used for brain volume calculation by segmenting the brain in to GM, WM and CSF after B1 field inhomogenities correction. The statistical parametric mapping (SPM) software, developed by members and collaborators of Welcome Trust Centre of Neuroimaging; University College London, analysis of functional and structural MR imaging. SPM is a voxel-based method of calculating brain volume. [97] Structural analysis can be done from segmentation of the brain however surfaces cannot be constructed using this software.[98]

FreeSurfer [28] is used for brain white matter and pial surface reconstruction and analysis, and for estimation of cortical thickness and sulcal depth.[99] FreeSurfer parcellates the brain into different regions, used in measurements of cortical thickness,

surface area, cerebral global and regional volumes.[13] Along with surface reconstruction, FreeSurfer can also segment the brain into cortical and non-cortical labels by using an automated algorithm that is based on variation in signal intensity, a probabilistic brain atlas and global position of the subcortical structures.[100] Brain volume can be calculated from these segmented structures. The brain is divided in to gray matter and white matter. The technique is fully automated, but manual input can be provided at multiple steps to correct problems with mislabelling.

There are differences between FS, FSL and SPM in the methods of segmentation and the outputs. FreeSurfer segments the brain by making binary mask, in which voxels are assigned a value of 1 if they belong to that structure and 0 if it does not belong to the structure. By contrast, both SPM and FSL use probabilistic segmentation, in which the voxel-wise output is a probability that the voxel belongs to a defined tissue class. All the three packages are used for automated volume measurements and all methods give different type of pathology assessment, FSL and SPM are mostly used in functional MR imaging (fMRI) assessment and FSL is also used in diffusion tensor imaging for evaluation of white matter tracts, while FreeSurfer has specific feature of cortical thickness measurement in addition to functional assessment [101] so the choice of method should be according to the requirement of the study.

Among these three software packages, FreeSurfer was selected for calculating brain volume in this study because it generates volumes in several brain regions of interest that may be affected by hypertension including the basal ganglia, has been validated in against manual measurement methods, [102] and has been shown to provide

consistent results across different MR vendors and magnetic strengths. [103] Additionally, in contrast to FSL and SPM FreeSurfer provides measurement of cortical thickness, which can be measured very precisely and therefore may be a sensitive indicator of volume differences.[104]

FreeSurfer uses spherical registration, explained in section 3.3.6, which gives a better estimate of brain cortex embedded in sulcal folds. This registration reduces the blurring of the cortex and metric distortion is also minimized.[99] Labelling of anatomical structures is not required for alignment instead whole brain curvature will be used for alignment of brain.

3.3 Analysis Pipeline for FreeSurfer Brain Atrophy Measurements

FreeSurfer processing involves eight key steps that are detailed below.

3.3.1 Talairach Registration

The high resolution 3-D T1 weighted MPRAGE sequence is registered to the Talairach brain atlas [105] using linear and affine registration, which corrects the difference in global brain sizes and minimizes internal anatomical variations in human brains. Talairach registration matches the subject brain to a reference brain by modifying its position, orientation and size. [106, 107] The Talairach atlas is labeled for relevant anatomical regions, *e.g.*, gray matter, white matter and Brodmann area. The subject brain is labeled according to Talairach atlas, allowing the algorithm to incorporate

probabilistic information about the likelihood of the specific tissue class according to voxel location. This baseline registration is used for placing seed points in the white matter in a later step, according to their location in Talairach space, which is needed to generate an accurate white matter surface.

3.3.2 Motion Correction

This is used to correct the motion artifacts of T1weighted images and to average multiple sequence acquisitions, if they were performed. Tissue segmentation accuracy may be improved by averaging multiple 3D T1 MPRAGE acquisitions, although in my study we only used a single acquisition to minimize the total scan time.

3.3.3 Signal Uniformity Correction

Variation in white matter signal intensity is present in all brain due to magnetic field inhomogeneity, variation in coils and physiological variation in human brain tissue; these variations in signal intensity are used for estimating bias in B1 field, which is a magnetic field generated by radiofrequency pulse. Intensity correction is done to remove the effect of the bias field by dividing voxel intensity by the bias field at that location.[105]

3.3.4 Intensity Normalization

This step normalizes the white matter signal intensity. Correction of intensity makes white matter mean intensity 110. Intensity normalization increases the contrast between white and gray matter, increasing the accuracy of the identification of the surface between the white matter and the gray matter.

3.3.5 Skull Stripping

After intensity normalization, signal from non-brain tissue, for example skull, fat, and skeletal muscle in the head, is removed. A preset template and threshold is used for this purpose, which categorizes the brain structures into CSF (*i.e.*, with low MR signal intensities), other extra cerebral tissue (fat, skull and spine) and brain parenchyma (*i.e.*, with higher MR signal intensities) and removes the skull around the brain. [105]

3.3.6 Surface Generation

The white matter signals are used as a baseline for creating the white matter surface on the basis of differences in the signal intensity of gray and white matter. Voxels are classified into white matter versus other cerebral structures on the basis of difference in signal intensity. Once white matter-gray matter surface is formed it is then computationally expanded outwards to define the pial surface on the basis of the difference in signal intensity between gray matter and cerebrospinal fluid (CSF).
The processing pipeline of FreeSurfer is given in Fig 3-1.

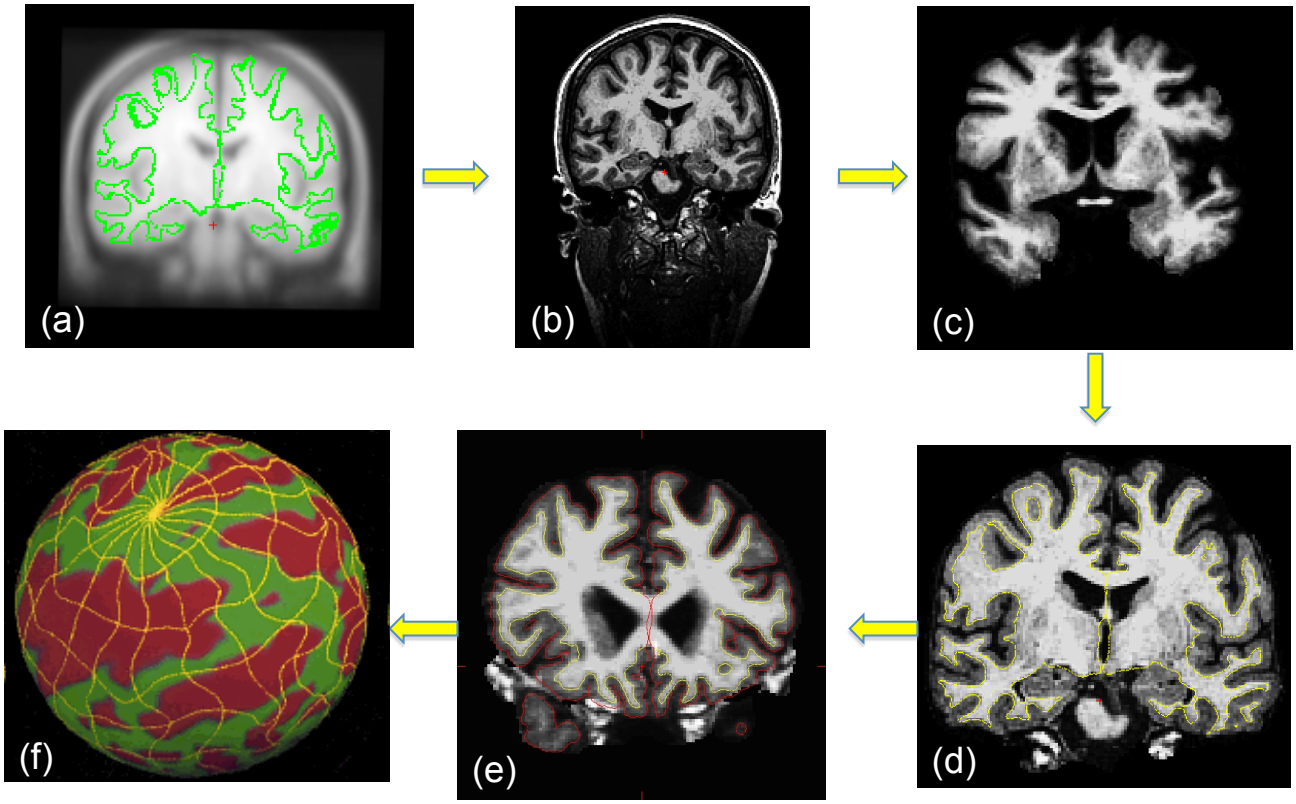


Figure 3.1 Analysis pipeline of FreeSurfer: (a) Talairach registration. (b) Image uploaded after signal uniform correction and intensity normalization (c) skull Stripping (d) white matter surface formation (e) pial surface formation (f) spherical registration [99].

3.3.7 Spherical Registration

For constructing precise surfaces spherical registration is used in FreeSurfer. The brain is registered to a spherical atlas that follows the sulcal and gyral pattern of the individual brain for generation of surfaces. Spherical registration is an important feature of

FreeSurfer which more accurately identified surfaces within the complex folds of brain.[108] (Fig 3-2)

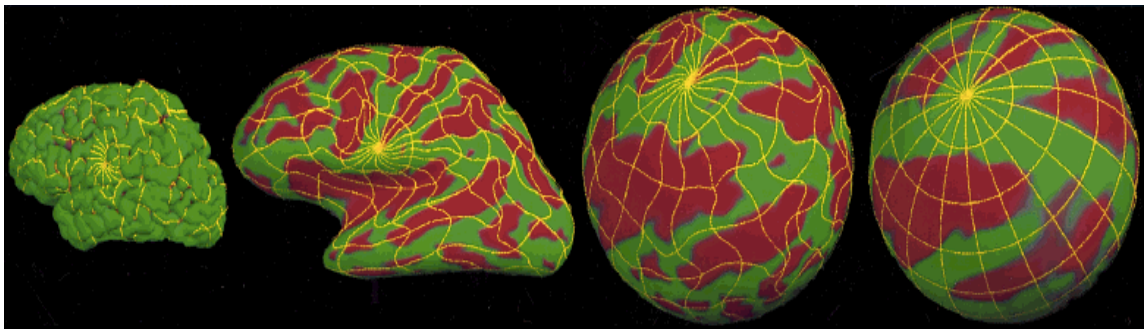


Figure 3.2 Example of spherical registration of cortical surfaces in FreeSurfer sulci and gyri are represented by red and green color. Reprinted with permission from Fischl et al. 1999 [99]

3.3.8 Segmentation and Parcellation

Segmented structures include the ventricles, thalami, and hippocampi.[100] Variation in signal intensity is not used alone for determination of segmentation because there is 12% overlap of signals between gray and white matter.[109] To overcome this problem labeling of brain is done on the basis of signal intensity, anatomical position of the structures and the neighbourhood.[109] Parcellation is a method of naming the each region of cortex that is also done with a probabilistic atlas. Parcellation helps in identification of cortical sub regions Additionally, FreeSurfer provides the volume occupied by parcellated regions such as the precunneus, prefrontal cortex, *etc*.[100]

3.3.9 Data Visualization

The final image can be visualized with overlaid white matter and pial surfaces and segmentations. The cerebral hemisphere can be inflated for further assessment such as white matter lesions.

3.4 Known Potential Errors in FreeSurfer Processing and Correction Methods

As with any automated method, some errors can occur during FreeSurfer processing. Visual inspection of critical steps is essential for quality control and accurate volume measurements. FreeSurfer allows manual intervention at several points to correct for processing errors.

3.4.1 Misregistration to the Talairach Atlas

Poor registration of the brain to the Talairach atlas can give a distorted image and can cause failure of further processing. The error in registration can be corrected by manually aligning the subject brain with the standard brain. Alignment should begin with the sagittal plane by aligning the corpus callosum, followed by aligning in coronal and axial planes.

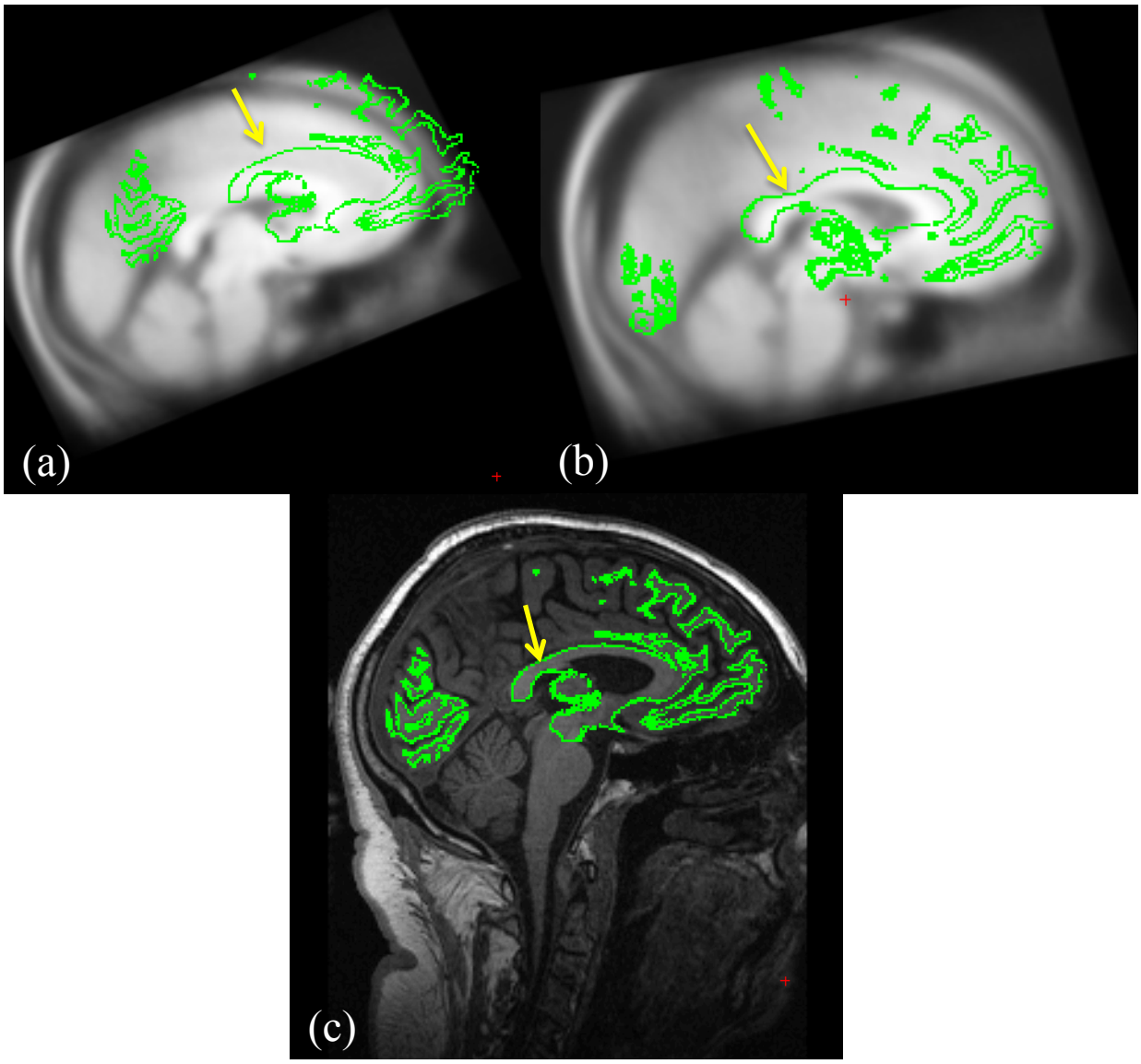


Figure 3.3 Example of the Talaraich misregistration correction: Sagittal images (a) misregistration of brain to Talairach atlas arrow is pointing corpus callosum (b) Manual correction of corpus callosum and (c) final corrected image after reconstruction (Images from PURE-MIND study)

3.4.2 Errors in Skull Stripping

Although most of the non-brain signal should be removed by FS, sometimes part of the dura, sagittal sinus, extra-cerebral fat, or cervical spine remains after skull stripping. Alternatively, over-aggressive skull stripping can result in erosion of part of brain parenchyma (*e.g.*, cerebellum or part of a cerebral hemisphere). Problems with the skull stripping are best appreciated in sagittal view. These problems can be overcome either by manually editing the brain mask slice by slice either erasing or copying from the original MR image as needed, or by adjusting the watershed threshold and running the reconstruction step again for skull strip adjustment until acceptable results are achieved.

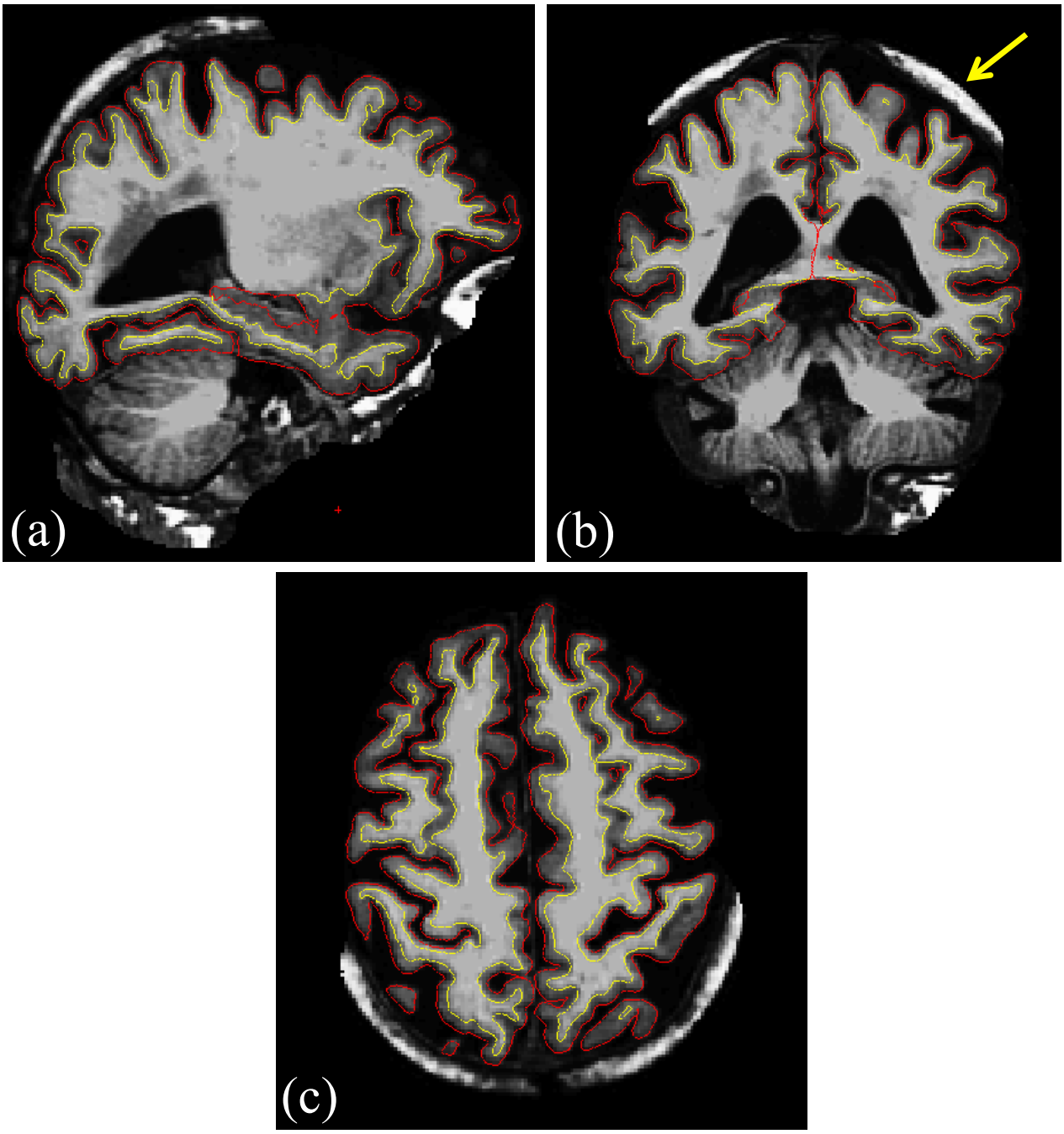


Figure 3.4 (a) Sagittal (b) coronal and (c) axial images showing residual areas of skull. Error in skull strip is marked with a yellow arrow. (Images from PURE-MIND study)

3.4.3 Errors in the White Matter Surface Identification

In the intensity normalization step, the average of all white matter voxels is assigned an intensity value of 110, which is the mean intensity of white matter. The image voxels then are rescaled using this intensity normalization. The white matter surface is generated by identifying the signal intensity of white matter, the signal intensity of neighbouring voxels, and its location in Talairach space. When true white matter regions are mislabelled with intensity less than 110, errors in boundary formation can result, where the white matter-gray matter surface extends inappropriately into the white matter or gray matter. To correct the boundaries one can manually place “control points” to manually label true white matter. Control points are added in to the locations that should be inside the white matter boundary but excluded during surface generation due to low signal intensity of white matter in that regions.

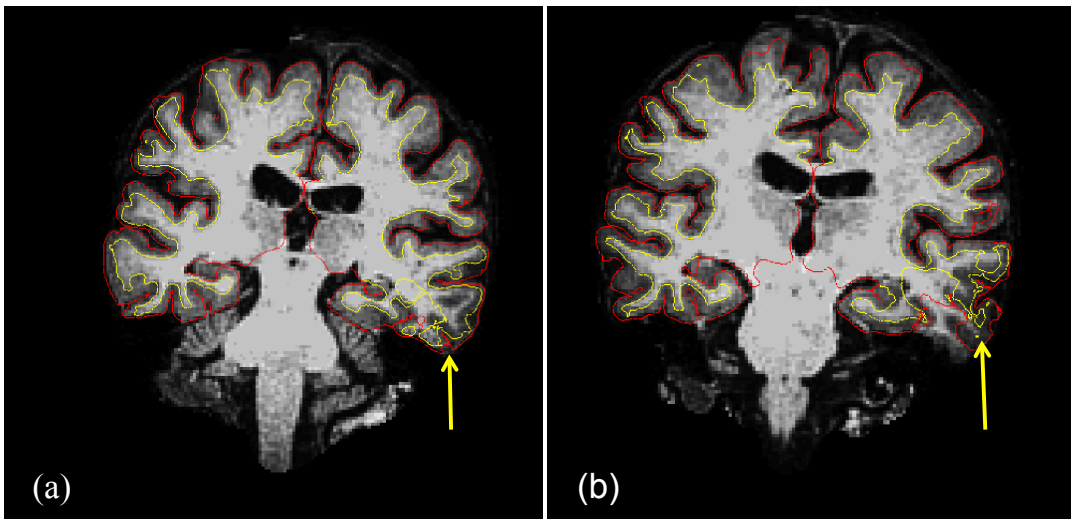


Figure 3.5 Errors in WM surfaces: Coronal images (a) arrow indicating incorrect white matter and (b) arrow indicating in accurate pial boundary in left temporal lobe. (Images from PURE-MIND study)

3.4.4 Errors in the Pial Surface Identification

Starting from the white matter surface, the variations in signal intensities of gray matter to cerebrospinal fluid (CSF) are used to create to pial surface. If the white matter boundary is not formed accurately, error occurs in pial surface formation because the white matter-gray matter surface once formed expand outside to follow intensity difference between gray matter and CSF to form pial surface. If higher intensity scalp fat is left over it may cause miscalculation of the intensities and result in stretching of pial and white matter boundaries outside the brain. The pial surface should neither overlap with the white matter surface nor extend outside the brain to include cerebellum, dura and superior sagittal sinus at any point.

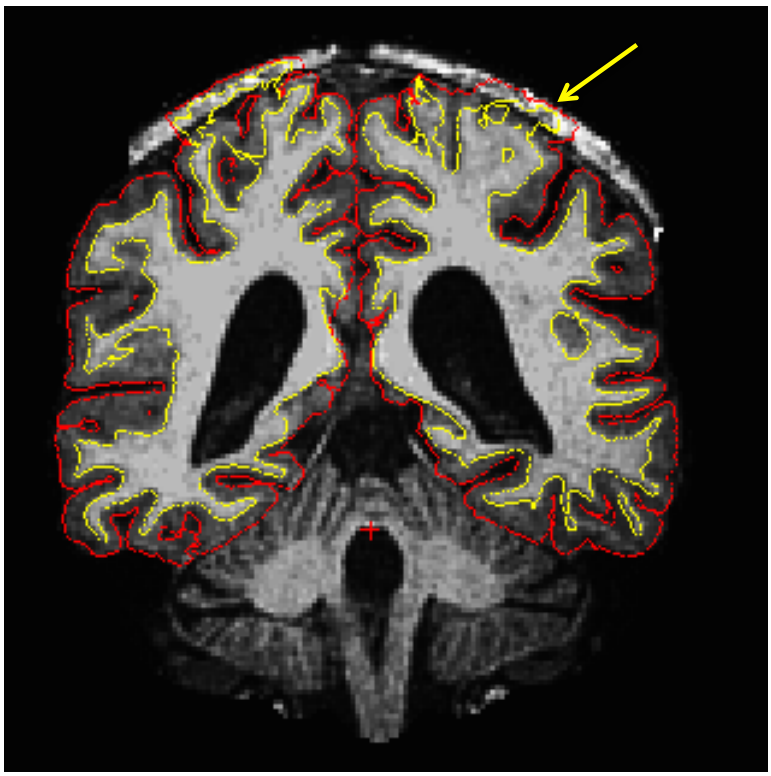


Figure 3.6 Errors in pial surfaces: Poor pial and white matter boundary in both frontal lobes extending to skull. (Images from PURE-MIND study)

3.4.5 White Matter Mis-segmentation

White matter lesions are hypointense on T1-weighted sequences and can be incorrectly identified as gray matter. When looking at an inflated surface view of the brain, these lesions appear as a hole or pit in the surface. If not corrected manually, these lesions can give inaccurate cortical thickness measurements.

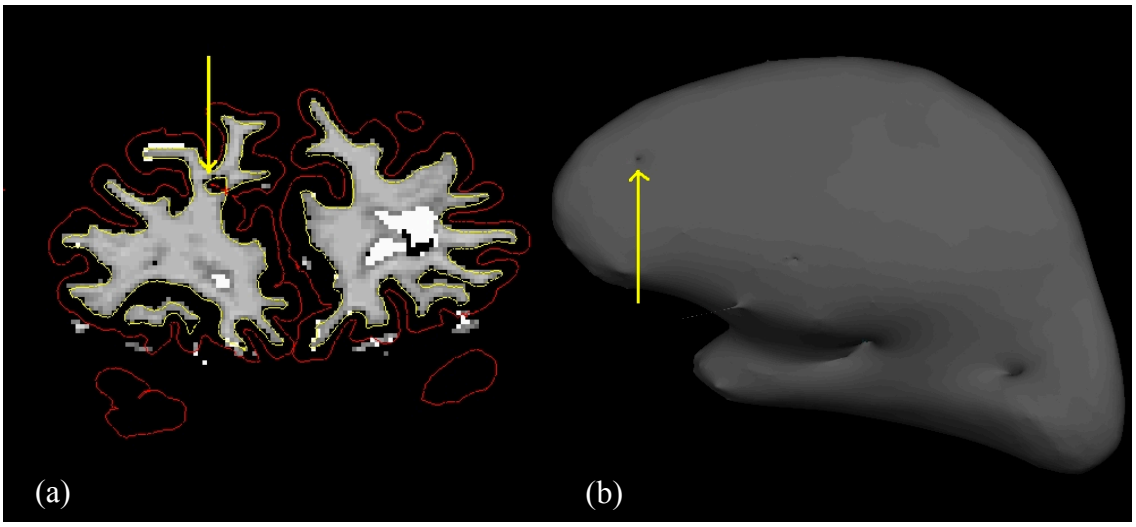


Figure 3.7 Hole due to WMH: (a) pial boundary is extended into white matter lesion indicating by arrow. (b) White matter lesion is visualized as hole on inflated brain—yellow arrow. (Images from PURE-MIND study)

3.4.6 Errors in Segmentation

A segmentation error is when one anatomical structure of brain is mislabelled as another structure. In particular, residual dura or superior sagittal sinus left over from incomplete skull stripping can be mislabelled as cerebral cortex.

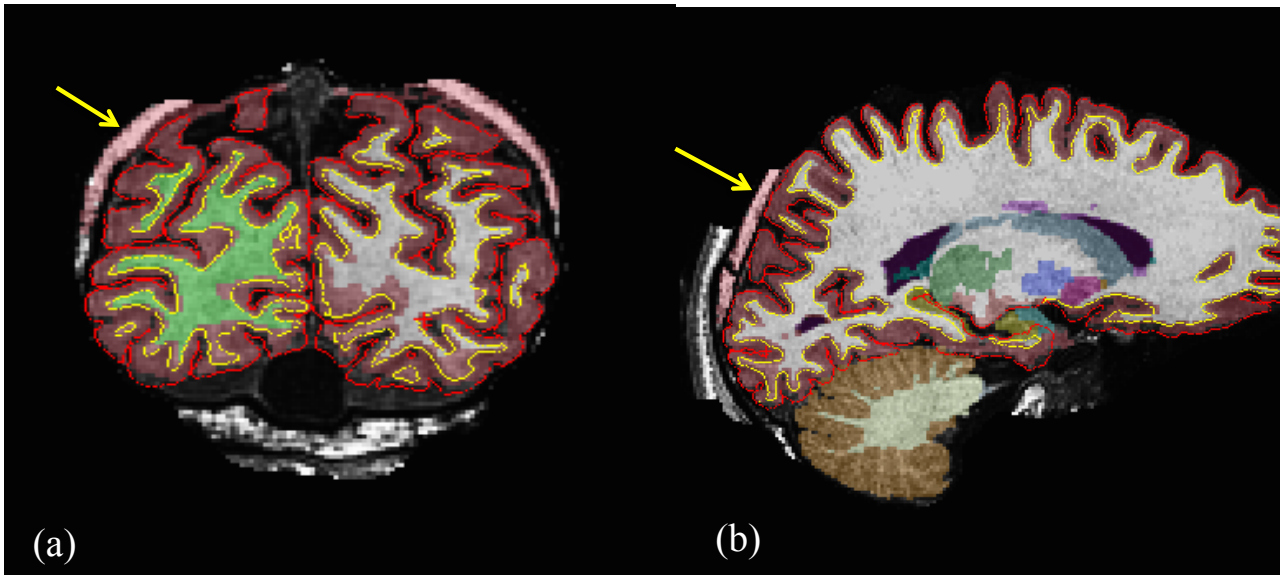


Figure 3.8 Errors in segmentation: Dura mater is mislabelled as cortex (arrow) in (a) coronal and (b) sagittal images. (Images from PURE-MIND study)

3.5 Feasibility and Accuracy of using FreeSurfer in the Multi-Centre PURE-MIND Study

3.5.1 Introduction

To address questions regarding BP and brain atrophy a subset of data from the larger Canadian population based study, PURE-MIND, were examined. This study is a longitudinal sub-study nested within international Prospective Urban Rural Epidemiologic (PURE) study that examines cerebrovascular diseases and the risk factors associated with them. To date, a total of 803 healthy individuals between 40 and 70 years of age, free from stroke and dementia have been recruited from four Canadian sites (Hamilton, Ottawa, Quebec and Vancouver). Participants were scanned with the same protocol on a 1.5 T GE MR machine, except in Hamilton where a 3 T GE machine

and Quebec where a 1.5 T Philips MR machine were employed. The full details about recruitment of study participants and study methodology are given in Chapter 4.

To determine the feasibility and accuracy of using FreeSurfer for determining global and regional brain volumes in a large, multi-centre study three factors were assessed: 1) the difference between automated FreeSurfer output and carefully manually edited volumes removing any remaining dura or skull from the skull stripped volumes (analyzing differences in resulting GM, WM, cortical, cerebral, cerebellar volume), 2) the amount of operator time required to make these manual edits and 3) the frequency and type of errors that led to discrepancies between automated output and careful visual inspection for mislabelling.

3.5.2 Methods

Twenty-eight participants (seven from each of the four study sites) were randomly selected for initial review and comprehensive manual editing, to assess differences between the automated output and manually edited output. High-resolution T1-weighted MR images were processed using FreeSurfer (version 5.1.0). Based on visual inspection of the 28 cases, the most common error was that some residual dura, venous sinus or skull remained visible in the skull-stripped volume. Therefore, an experiment was designed to remove all residual extra-cerebral tissue to test whether this had a significant impact on the final outputted volumes. First, all remaining extra-cranial tissues from the skull stripped images were manually removed. This step often required removal of some residual dura and superior sagittal sinus from the skull stripped images. After removal of all extra-cranial tissues, the manually corrected skull-

stripped MR image volume was reprocessed using FreeSurfer. Next, the 28 cases were reviewed for correct labelling of the white matter-gray matter surface. Control points were added for correcting in accurate white matter boundary in 9 cases. In each case the resultant image corrected the boundary where control points were added, but caused gross distortions in the boundary at other sites.

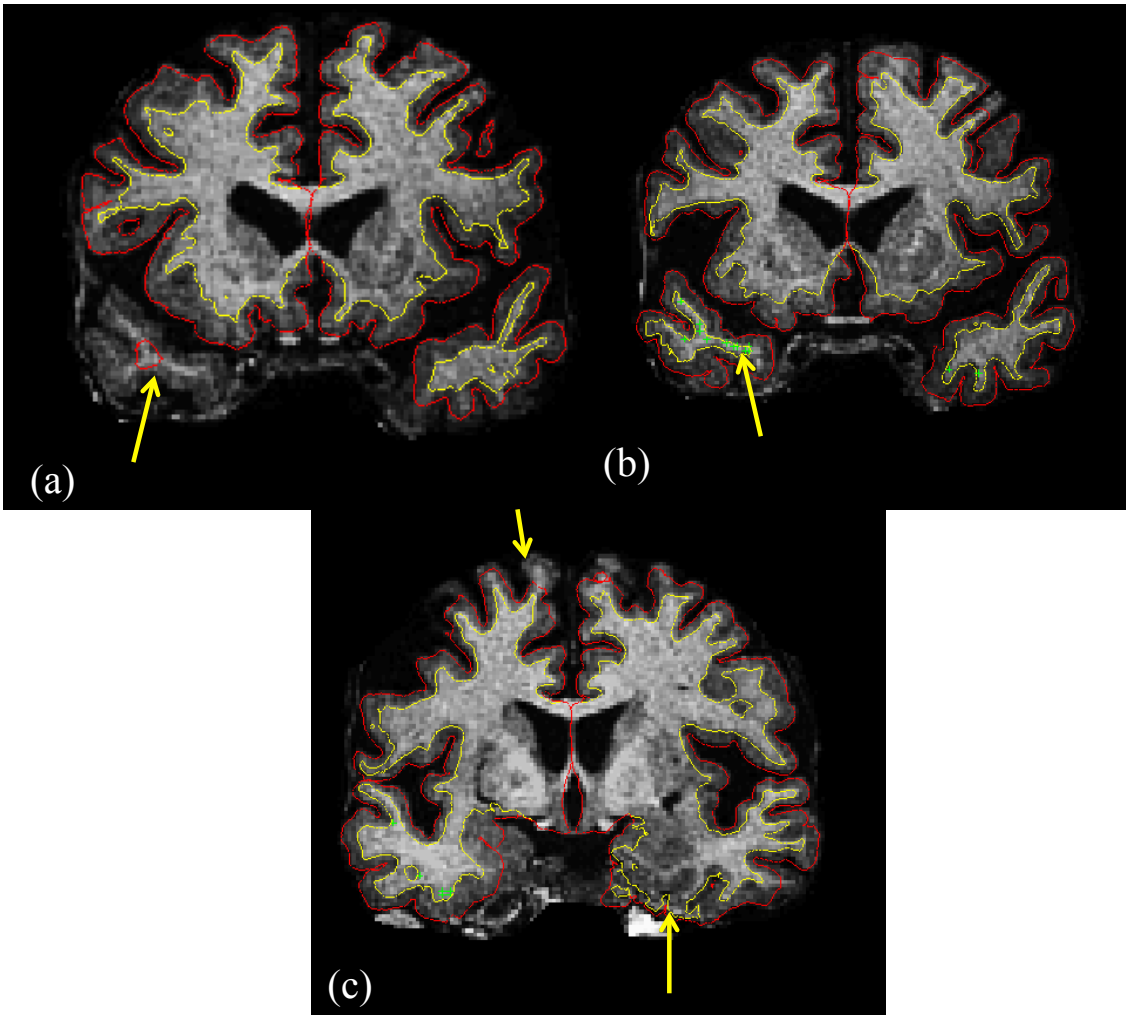


Figure 3.9 Example of failed control points for surface correction: Coronal images (a) White matter surface missing right temporal lobe (yellow arrow). (b) Control points were added shown by green + sign corrected the boundary (yellow arrow). (c) Reconstructed image showed distortion of boundary in left temporal and right frontal lobe (yellow arrows). (Images from PURE-MIND study).

3.5.3 Results

The time required for manual editing of each of the 28 selected cases varied from 30 minutes to 1.5 hours. The mean time required was 42.5 minutes and standard deviation was 7.65 minutes (Table 3-1).

Table 3.1 Average time required to manually edit seven cases from each site

Site	**N cases edited**	**Average Editing Time (±SD) (case/min)**	**Cases Reviewed**
Hamilton	7	40 ± 3.5	256
Vancouver	7	35 ±6.39	251
Quebec	7	45 ±9.5	251
Ottawa	7	50 ±11.21	46

The skull strip mask required manual edits to completely remove extra-cranial tissues in 24/28 cases. The mean of original volumes calculated from 28 cases are given in (Table 3-2) In these 24 cases, a mean 3.28 cm^3 (standard deviation 4.08 cm^3) of extra-cranial tissue were removed. By comparison, the difference in original and edited brain volumes is given in (Table 3-3). After reprocessing, all differences were non-significant except that cerebellum was significantly larger after manually removing all skull and reprocessing (mean difference 1.6 cm^3, standard deviation 1.6 cm^3, $p<0.001$). However, this difference was small (1.3% of original volume)

Table 3.2 Original volumes before manual complete skull strip and reprocessing of 28 cases

Variable	N	Mean	Std Dev	Minimum	Maximum
Cortical gray matter	28	439.3	36.3	349.6	495.8
White matter	28	471.0	51.1	382.8	559.8
Ventricles	28	26.5	15.9	8.3	71.7
Cerebellum	28	125.1	14.8	92.5	157.6
Basal ganglia	28	23.2	2.1	18.9	28.7

Table 3.3 Difference between edited and reprocessed volume minus the original volume

Variable	N	Mean	Std Dev	Minimum	Maximum
Cortical gray matter	28	0.6	3.4	-7.3	8.7
White matter	28	2.4	4.4	-2.4	13.7
Ventricles	28	-0.1	0.3	-0.9	0.6
Cerebellum	28	1.6	1.6	-1.3	5.4
Basal ganglia	28	-0.0	0.2	-0.5	0.3

All differences were non-significant except that cerebellum was significantly larger after manually removing all skull and reprocessing (mean 1.6 cm^3, standard deviation 1.6 cm^3, $p<0.001$). However, this difference was small (1.3% of original volume).

3.5.4 Discussion and Conclusions

Our results supported FreeSurfer as a feasible method for quantitative analysis of brain volume and cortical thickness for population-based studies. A validation study of 28

randomly selected cases showed only small differences between the automated volume calculations and the comprehensively manually edited volumes to produce the “perfect” labeled scan. The mean difference (0.94%) and standard deviation was less than the reported scan-rescan variance of FreeSurfer [110] and therefore unlikely to significantly affect the findings of our study. Based on the overall small differences between the automated output and the manually labeled output, we concluded that manual editing each FreeSurfer volume was not required, and instead adopted a quality assurance strategy of visually reviewing each case to identify and exclude cases with grossly visible errors explained in chapter 4. Our visual inspection of the final images suggested that the large majority were of moderate to good quality and useful for analytic purposes. Of the 793 cases that finished processing we excluded only 15/793 (1.9%) for errors.

FreeSurfer has advantages for large population study that volumes could be generated at once for whole brain, sub cortical regions and different cortical regions for all cases. FreeSurfer is a convenient method of calculating brain volumes for a large study dataset. Running FreeSurfer with specific Unix command require some learning time period however FreeSurfer wiki site is very helpful in that regard.

FreeSurfer has advantage of giving the cortical thickness measurement for brain volume calculation as compared to SPM and FSL. The rescan reliability of FreeSurfer for segmentation volume measurement is better than FSL.[110] Morey *et al*.[111] suggested FreeSurfer is more reliable than FSL for segmentation of the hippocampus,

while both techniques were equally reliable for the amygdala is equivalent in all methods.

Keller suggested FreeSurfer is an operator-independent and reliable method for calculation of global thalamic volume among healthy and diseased populations.[112] They suggested this fact by comparing volume calculated by FreeSurfer with manual stereological methods and both methods gave consistent results.

There are some limitations of FreeSurfer. FreeSurfer does not work well if extensive white matter pathology is present, which can result in defects in the white matter signal normalization and the white matter boundary. In the majority of cases, the surfaces along the medial temporal lobe, amygdala and hippocampus is not generated accurately; therefore alternative methods might be needed for studies focusing on these specific regions. The basis for this inaccuracy in the medial temporal lobe is because of the relatively thin amount of white matter in this region, which makes it difficult for the algorithm to determine the white matter-pial surface boundary, and magnetic field inhomogeneities related to distance from the MR head coil,.[113]

There are some limitations to our methods for quality assurance. We did not assess scan-rescan variability. Ideally, we would have scanned a subset of the same participants at each site to assess scanner differences. However, this was not feasible with such widely distributed study sites. Fortunately, the differences between automated output and manually edited output were similar across sites, and visually the data looked comparable. Our study of manual editing was limited to only 28 cases and therefore may not reflect the true range of variability in the overall sample of 803;

however, our comprehensive review of the 803 cases revealed major errors in only 1.9% of cases, consistent with the conclusions from our limited sample that FreeSurfer output is generally quite accurate.

3.6 Summary

FreeSurfer is a useful software tool for segmentation of brain MR images in population-based studies. Differences between automated FreeSurfer output and manually edited output is generally small. Nonetheless, all automated output should be visually inspected for gross errors which may occur in roughly 1.9% of cases.

Chapter Four: Relationships between MR-defined Brain Volumes and Blood Pressure in a Canadian Population-based Study

4.1 Introduction

Brain volume normally decreases with age.[62] Elderly people show smaller brain volumes and larger ventricular size as compared to younger people. After age, hypertension is considered to be an important potentially modifiable risk factor for brain atrophy.[27] As discussed in detail in Chapter 2, review of the literature on the effect of blood pressure on brain volume gave somewhat inconsistent results. While most studies suggested that hypertension was associated with brain atrophy, some studies failed to find an association between the two. Few studies looked comprehensively at the pattern of atrophy across different brain sub-regions. Few studies examined relationships with pulse pressure, which may be a significant factor reflecting arteriosclerosis, and few studies considered the potential modifying effect of blood pressure treatment with antihypertensives. Most studies focused on the elderly (defined as age >65 years old), or the relationship among blood pressure measured in mid-life blood (defined as age 40-65 years old) and brain atrophy in the elderly, without

assessing the relationship between blood pressure in mid-life and atrophy in mid-life. Therefore, the age at which blood pressure-related atrophy begins to appear is not well known. Finally, many studies involved patient assessments from 10-30 years ago, when population blood pressures were higher because targets for blood pressure control were higher than current practice.

Data was used from a Canadian population-based study, PURE-MIND, to determine whether hypertension or blood pressure was associated with MR-defined brain atrophy, controlling for age and sex, in 40-70 year old Canadians without history of stroke. The data from this study was used to address the following unresolved questions in the literature: a) whether midlife blood pressure or hypertension is associated with mid-life brain atrophy, b) whether pulse pressure is associated with brain atrophy, c) whether relationships between BP and atrophy are different according to whether antihypertensive medications are used, and d) whether there is a differential pattern of atrophy across brain sub-regions. Specifically, it was hypothesized that: a) SBP, DBP and pulse pressure would be associated with atrophy of the gray matter, white matter, prefrontal cortex, and basal ganglia, and enlargement of the ventricles; and b) that relationships between BP and atrophy would be present in persons taking vs. not taking BP medications, but that the most severe atrophy would be seen in patients with poorly controlled hypertension.

4.2 Methods

4.2.1 Population

The PURE-MIND study is a Canadian sub-study of the multinational Prospective Urban Rural Epidemiological study. PURE study methods and initial findings have been previously reported.[114] In brief, the PURE study was designed to investigate the social and environmental determinants of non-communicable diseases in 17, high-, middle- and low-income countries including Canada, with a focus on identifying risk factors for diseases in urban and rural populations. In Canada, participants in the PURE study were recruited based on residence within pre-specified communities, defined as geographical areas whose inhabitants are generally expected to have shared cultural and social characteristics and usage of goods and services.[115] Communities were defined using Canada Post forward sortation areas (three-digit postal code areas). To be broadly representative of the Canadian population, fifty-five communities were selected in three Canadian provinces (British Columbia, Ontario and Quebec) to include a variety of age groups, ethnicities and socioeconomic status from different regions. Participants were recruited from urban and rural locations centered around 4 Canadian cities: Vancouver (British Columbia), Hamilton (Ontario), Ottawa (Ontario) and Quebec City (Quebec). For adequate representativeness of each community, all community residents were contacted through a participation invitation letter. Participants were contacted by telephone within two weeks of receiving the letters and screened if willing to participate to include in the study. PURE inclusion criteria were: 35-70 years old at the beginning of the study in 2007, fluent in either English or French, were ambulatory,

were not pregnant, had no plans to migrate for next 4 years and provided informed consent by themselves. All eligible participants were invited. A total 10, 455 Canadian resident were recruited between 2006-2009.

Canadian PURE participants returning for 3-year follow-up, during 2010 and 2012, were invited to participate in the PURE-MIND sub-study. The PURE-MIND study aimed to recruit 800 men and women. PURE-MIND study inclusion criteria were: age ≥40, no history of stroke or dementia, no history of other neurological diseases of the brain, and no contraindications to MR imaging such as pacemaker or other metallic implants.

4.2.2 Assessments

In PURE, a detailed history was collected about cardiovascular and other risk factors at baseline and three-year follow-up. Details were obtained about medical history, lifestyle moods and medication use including antihypertensives. Sitting BP was recorded from the right arm two times after an interval of five minutes. Height, weight and waist circumference was taken for each participant.

Brain MR imaging was performed at each of the 4 recruitment sites on their own scanner (3 at 1.5 T, 1 at 3 T). Details of the site MR and acquisition protocols are in the Table 4-1 and 4-2. Following MR sequences were obtained for each participants axial T2-weighted (T2w), proton-density weighted (PD), fluid attenuated inversion recovery (FLAIR), and T2*-weighted gradient-recalled echo (GRE), diffusion-weighted as well as a high-resolution T1-weighted (T1w) for volumetric analysis. The Alzheimer's Disease Neuroimaging Initiative (ADNI) phantom was scanned in all sites at the start and end of

study to compare the MR data quality, obtained within same and across different sites.[116, 117]

Table 4.1 Details of MR scanners used in data collection of PURE-MIND Study

Site	Tesla	Manufacturer	Model
A. Hamilton	3.0 T	GE Medical Systems	Signa Excite
B. Ottawa	1.5 T	GE Medical Systems	Signa HDxt
C. Quebec	1.5 T	Philips Medical Systems	Acheiva
D. Vancouver	1.5 T	GE Medical Systems	Signa HDxt

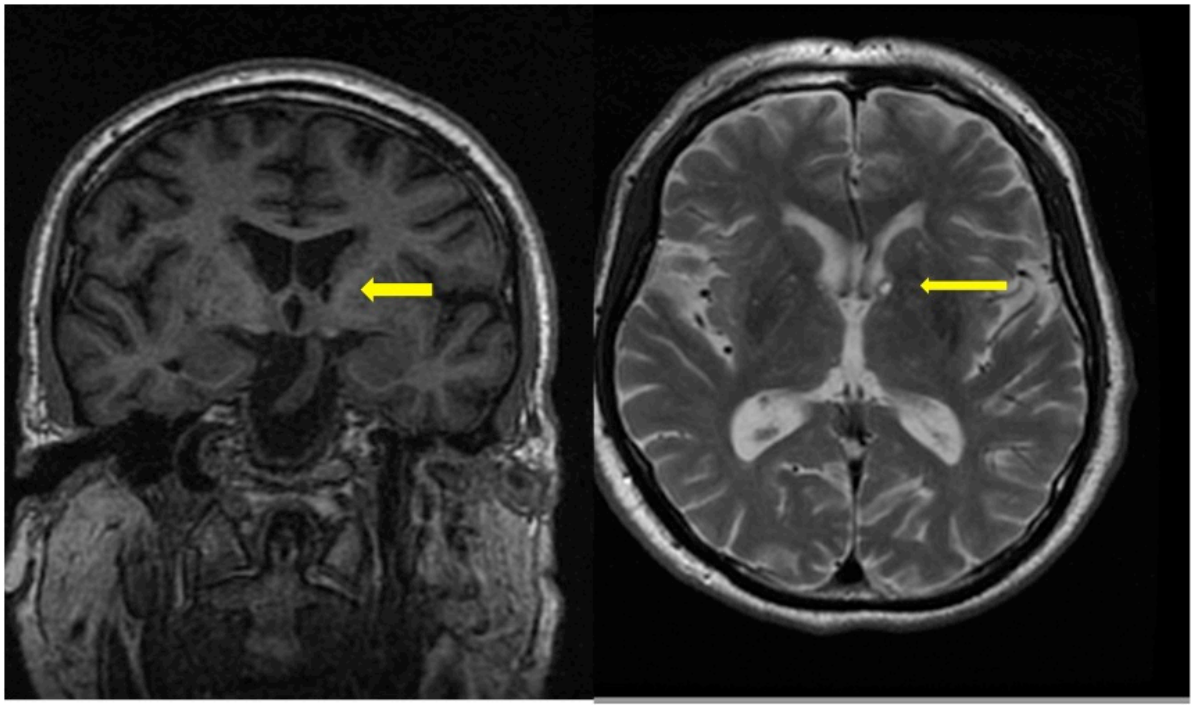
Table 4.2 MR protocol used in data collection of PURE-MIND Study

Sequence	**Site**	**TR (ms)**	**TE (ms)**	**Reconstructed Voxel size (mm^3)**	**FOV**	**Acquisition Matrix**	**Additional details**
3D T1w	A	5	2	1.0 x 1.0 x 1.0	256	256 x 256	α= 8 °, TI=650 ms
	B	8	3	0.94 x 0.94 x 1.0	240	256 x 256	α = 8 °, TI=650 ms
	C	9.5	4	0.87 x 0.87 x 1.0	195	196 x 187	α = 8 °
	D	8	3	0.94 x 0.94 x 0.8	240	256 x 256	α = 12 °, TI= 400 ms
2D FLAIR	A	9002	149	0.94 x 0.94 x 3.5	240	256 x 256	TI = 2250 ms
	B	9002	155	0.47 x 0.47 x 3.5	240	256 x 160	TI = 2250 ms
	C	9000	140	0.55 x 0.55 x 3.5	240	256 x 286	TI = 2250 ms
	D	9002	142	0.94 x 0.94 x 3.5	240	256 x 256	TI = 2250

							ms
2D PD/T2	A	3300	8/82	0.94 x 0.94 x 3.5	240	256 x 256	ETL = 12
	B	3300	13/92	0.94 x 0.94 x 3.5	240	256 x 256	ETL = 12
	C	3300	10/95	0.45 x 0.45 x 3.5	230	288 x 326	ETL = 12
	D	3317	10/78	0.94 x 0.94 x 3.5	240	256 x 256	ETL = 12
2D T2*w (GRE)	A	1000	20	0.94 x 0.94 x 5.0	240	256 x 256	α = 18 °
	B	700	20	0.94 x 0.94 x 1.0	240	256 x 160	α = 18 °
	C	1282	23	0.90 x 0.90 x 3.5	230	256 x 205	α = 18 °
	D	1170	19	0.94 x 0.94 x 3.5	240	256 x 256	α = 18 °
2D DWI	A	14000	94	1.25 x 1.25 x 3.5	240	192 x 192	*b*=1000, 11 directions
	B	12000	82	0.94 x 0.94 x 3.5	240	128 x 128	*b* = 1000
	C	14000	72	0.93 x 0.93 x 3.5	207	96 x 81	*b* = 1000
	D	14000	89	1.46 x 1.46 x 3.5	280	192 x 192	*b* = 1000

All MR scans were analyzed at the Seaman Family MR Research Centre at Foothills Medical Centre in Calgary, Alberta. The presence of lacunar infarcts, microbleeds and white matter hyperintensities (WMH) were reported by a neurologist (EES) or a radiologist (JM) according to consensus guidelines.[30] Brain infarcts were defined on the basis of size (lesions ≥3 mm diameter), shape (wedge) and signal intensity, which appear as with CSF-like hypointensity on T1-weighted sequence and hyperintensity on the T2-weighted sequences (Fig 4-1), consistent with previously published characteristics. [118] Infarcts may have central hypointensity on fluid

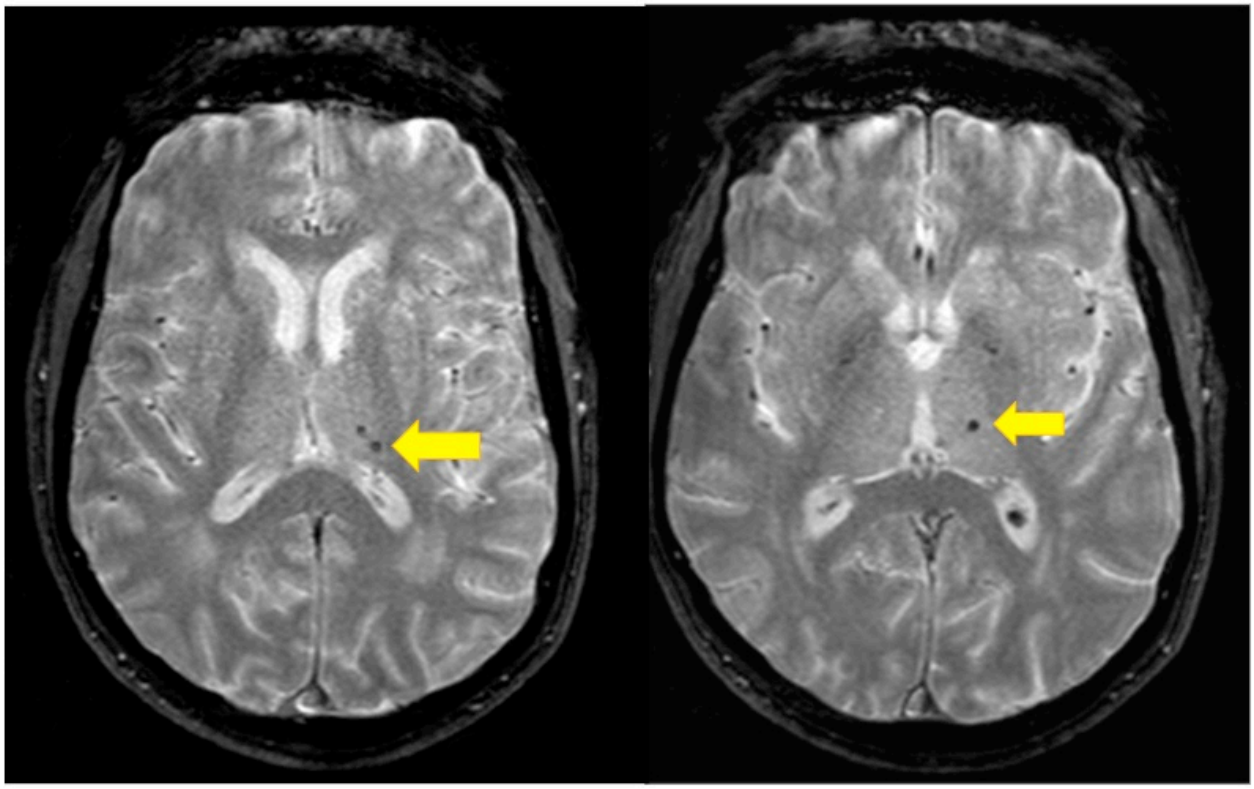
attenuation inversion recovery (FLAIR) sequence with a T2 hyperintense rim but not in all cases.



(a) (b)

Figure 4.1 Example of lacunar infarct a) coronal T1W and (b) Axial T2W MR scans showing infarct in right caudate nucleus (arrows). (Images from PURE-MIND study)

Brain microbleeds are defined as small round foci of hypointensity on the T2*-weighted GRE sequence (Fig 4-2), consistent with published reporting standards. [119] The location and size of each lesion type was be recorded on standardized forms.



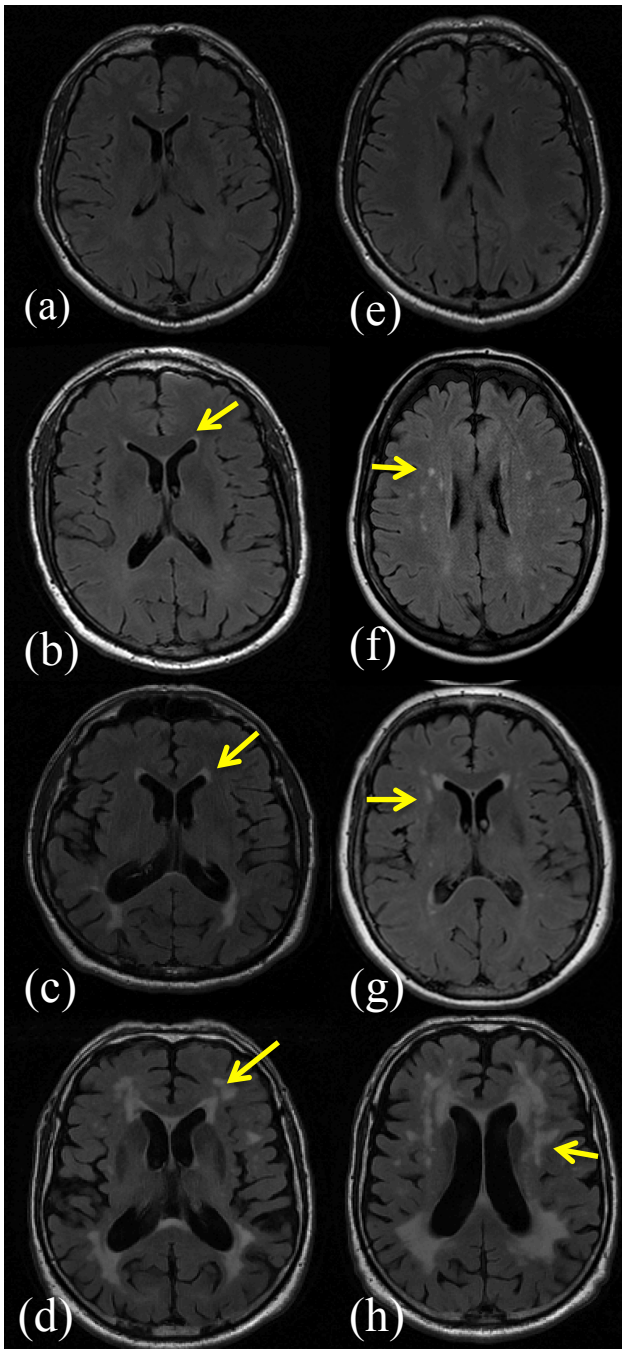
(a) (b)

Figure 4.2 Example of cerebral microbleeds. (a, b) axial T2 GRE sequence showing microbleeds in left thalamus (arrows) (Images from PURE-MIND study)*

The inter-rater reliability of the two reviewers (EES, JM), determined by comparison of independent review of 30 cases was kappa of 0.79.

Quantomo software (Cybertrials, Calgary, Canada) was used for quantitative volume estimates of WMHs. It is a semi-automated seed-growing method, in which seeds are placed in each WMH and a computer algorithm determines the boundary between WMH and normal tissue in 3D. Volumes of WMH generated by Quantomo were validated on 30 stroke patients in separate work, which were comparable to manual tracings with high inter-rater reliability (intra-class correlation coefficient 0.98). WMH

were also rated qualitatively using the Fazekas scale, in which periventricular and subcortical WMH are separately graded based on extent of WMH from 0-3 (Figure 4-3).



a-d) Example of Periventricular Hyperintensity grade 0-3. (a) Grade 0=No lesion (b) Grade1= pencil thin hyperintensity (c) Grade 2=Halo around ventricles (d) WML extending in to deep WM

e-h) Example of Sub-cortical Hyperintensity grade 0-3.
(e) Grade 0=No lesion (f) Grade1= punctate foci (g) Grade 2= beginning confluence of foci (h) large confluent lesions

Figure 4.3 Example of White matter Lesions grades Axial FLAIR sequence showing severity of WMH according to Fazekas Score.

Regional brain volume measurements, cortical thicknesses and intracranial volumes were calculated using FreeSurfer software (see Chapter 3).[109, 120] Dr. McCreary processed the data using FreeSurfer. I reviewed the accuracy of output for all 803 cases to decide whether FreeSurfer output was acceptable for further analysis. The surfaces and segmentations were judged separately and assigned a four-point qualitative grade according to the visual accuracy of the labeling: grade 1—not acceptable for further use due to obvious mislabelling of a significant proportion of the brain surfaces or segmentations, grade 2—acceptable despite minor errors in the surfaces or segmentations, usually related to mislabelling of the pial surface to include minor amounts of dura along the anterior frontal lobe or temporal lobe, grade 3—optimal FreeSurfer output, where visual interpretation matches FreeSurfer output with the exception of the minor segmentation or surface errors in the anterior temporal lobes that are an acknowledged limitation of the FreeSurfer method and grade 4—No error in the image (Figure 4-4 and Figure 4-5)

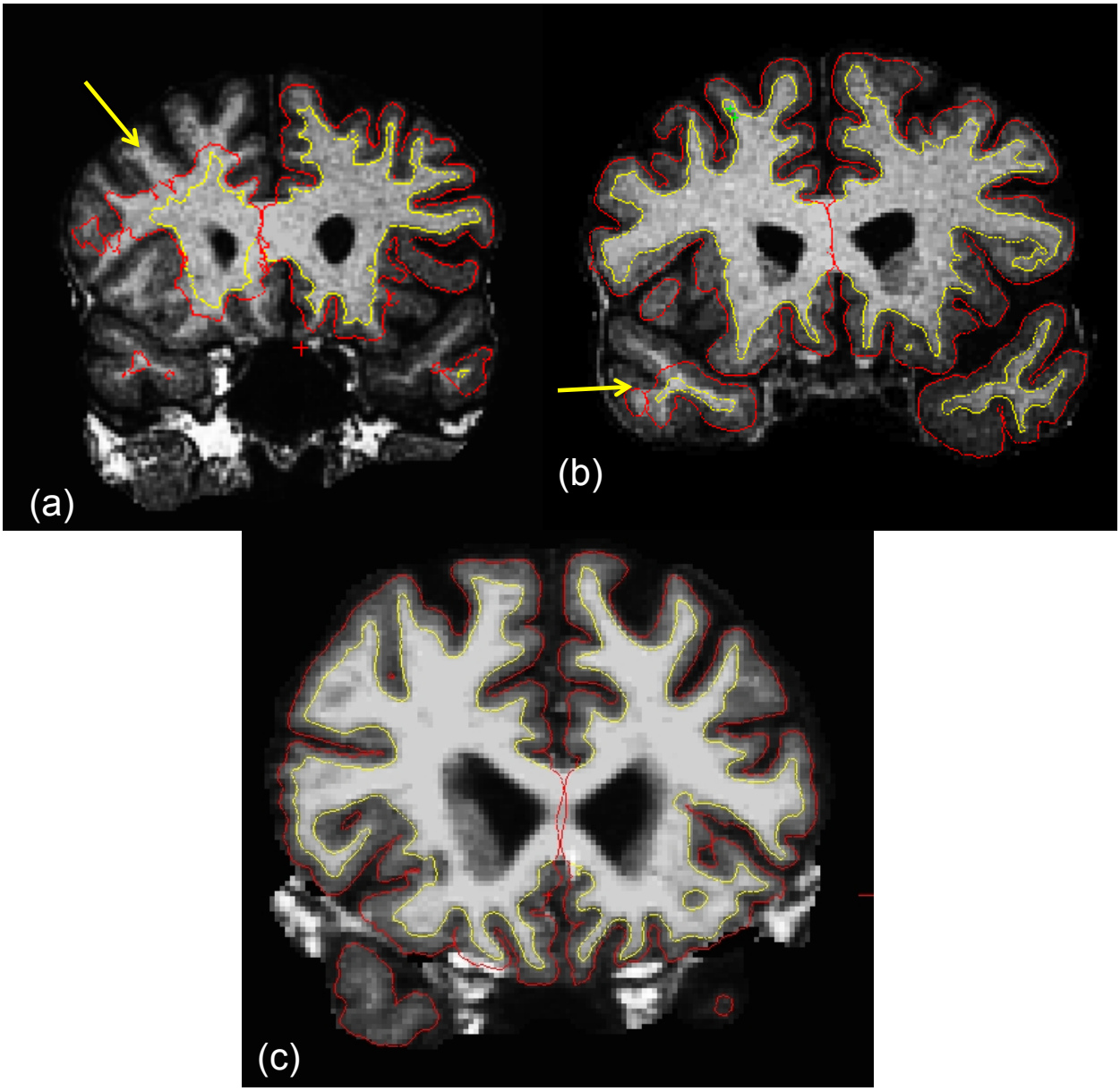


Figure 4.4 Surface quality check. (a) Example of poor quality surface grade 1, in which surfaces are missing lot of brain parenchyma (arrow) (b) Intermediate quality grade 2, surface is missing part of temporal lobe (arrow) (c) Good quality surface grade.(Images from PURE-MIND study)

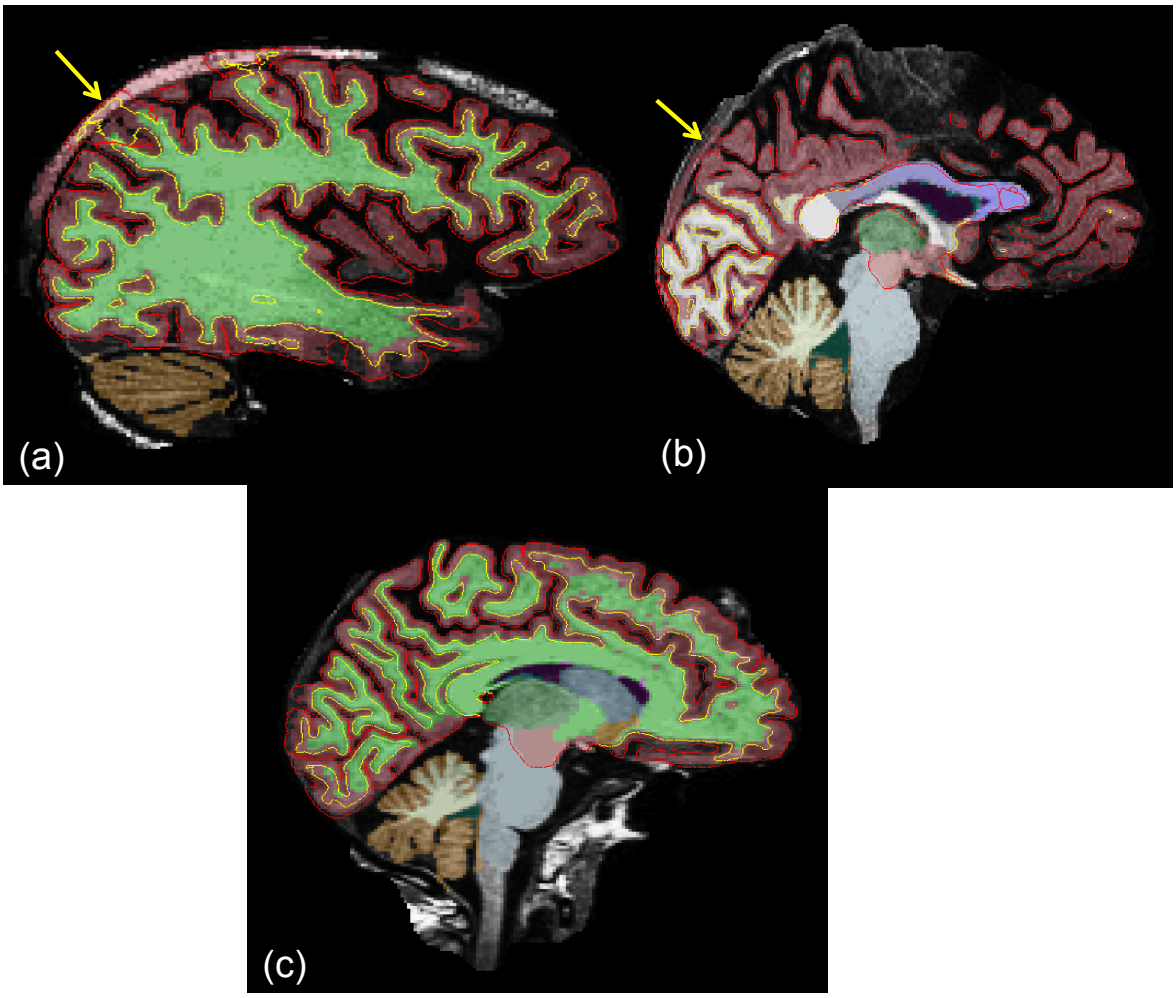


Figure 4.5 Segmentation Quality. (a) Poor quality Segmentation grade 1, skull strip is labelled as cortex (arrow) (b) Intermediate quality grade 2, part of superior sagittal sinus is labelled as cortex (arrow) (c) Good quality grade 3 (Images from PURE-MIND study)

In our study the failure rate of the software—i.e. cases where the software failed to complete processing steps was only 1.2%. A small number of scans, 25/803 (3.1%), were excluded for inaccurate surface labels and failure of processing in FreeSurfer. To eliminate normal variation in brain size due to differences in head size related to height and sex among different participants, we normalized all volumes to the subject average

intracranial volume (1,448.9 cm^3); and brain volumes are presented in cm^3 relative to this average total intracranial volume.

Based on clinical knowledge and our systematic review of the literature (Chapter 2), we hypothesized *a priori* that selected brain regions would be atrophied in the setting of hypertension and higher blood pressures. These regions were prefrontal cortex, basal ganglia, cerebellum, and total gray and white matter. In each case, right and left hemispheres were combined for analysis.

The prefrontal cortex volume was chosen as a region of interest based on findings of prior studies.[48, 73] The frontal lobe controls executive functions and memory, and frontal lobe damage may mediate many of the symptoms of vascular dementia [67]. Seo suggested that hypertensive subjects showed cortical thinning in supplementary motor area, superior, inferior, medial frontal gyrus, anterior cingulate cortex, superior, middle temporal gyrus and lingual gyrus.[72] The prefrontal cortex was defined based on 11 regions parcellated by FreeSurfer (Table 4-3).

Table 4.3 Regions of Prefrontal cortex parcellated in FreeSurfer

Regions
Caudal anterior cingulate
Rostral anterior cingulate
Caudal middle frontal
Rostral middle frontal
Frontal pole
Superior frontal
Lateral orbitofrontal
Medial orbitofrontal
Pars opercularis
Pars orbitalis
Pars triangularis

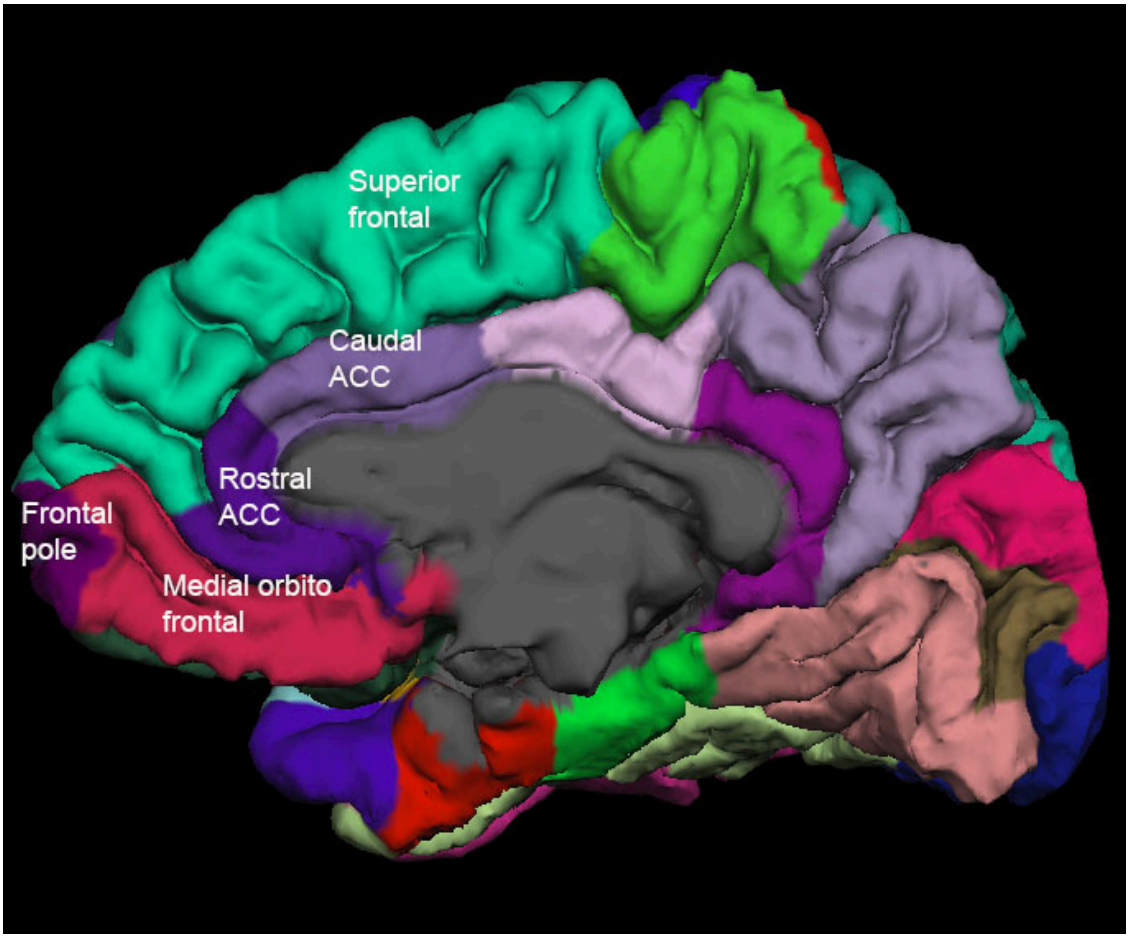


Figure 4.6 Frontal lobe regions parcellated in FreeSurfer medial part of cerebral Hemisphere. (Images from PURE-MIND study)

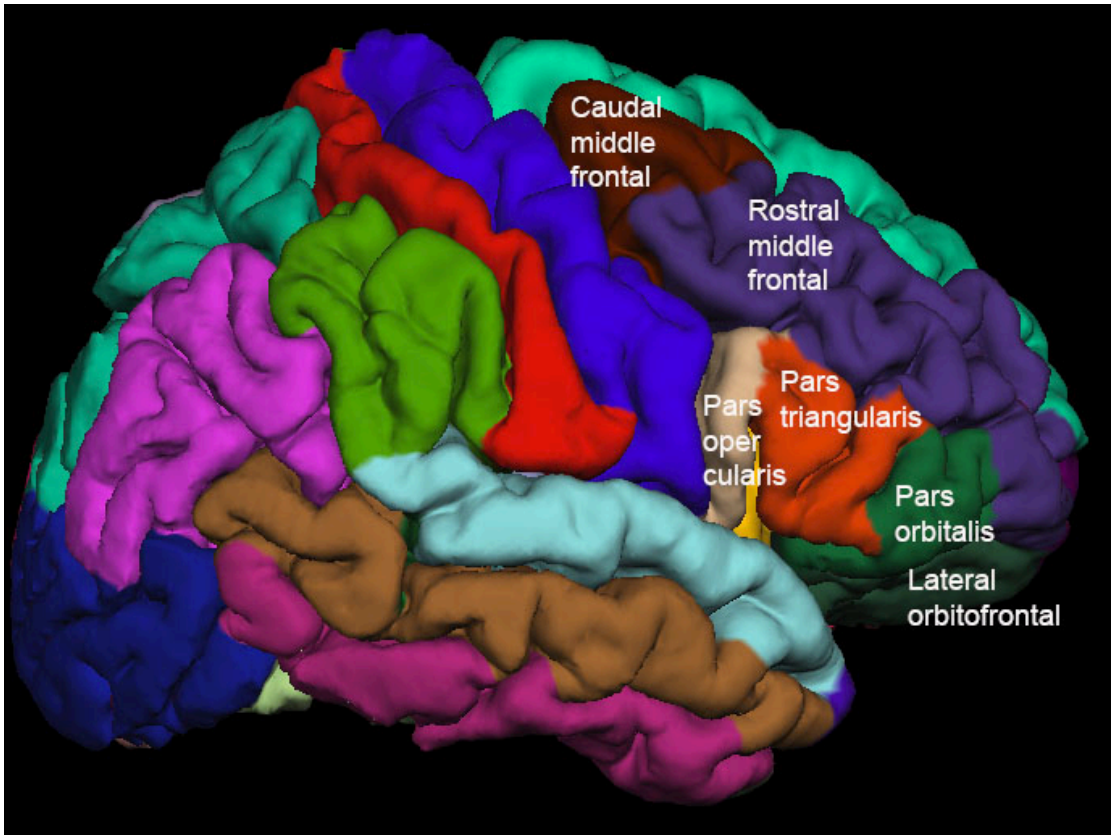


Figure 4.7 Frontal lobe regions parcellated in FreeSurfer lateral part of cerebral Hemisphere. (Images from PURE-MIND study)

The *basal* ganglia region of interest was chosen because hypertension is a known risk factor for microbleeds and small subcortical infarcts in the basal ganglia and thalami.[121] Basal ganglia volumes were calculated from these FreeSurfer segmented regions: pallidum, caudate and thalamus

The *ventricular volume* was chosen as a region of interest because shrinkage of brain structures causes dilatation and widening of ventricles, which is a indirect evidence of atrophy. Many researchers studied relationships between hypertension and

ventricular dilatation.[55, 61, 66, 71, 77, 122] The ventricular volume was calculated as sum of the segmented volumes of the lateral ventricles, third and fourth ventricle.

The *cerebellum* was analyzed based on the sum of cerebellar gray matter and white matter volumes calculated by FreeSurfer. We chose the cerebellum as a region of interest because hypertension has been associated with cerebellar microbleeds [123] and a previous study suggested that cerebellar gray matter volume is reduced in elderly hypertensive subjects.[49]

FreeSurfer also calculated total supratentorial cortical gray matter and white matter volume. Volumes of cerebral and cerebellar cortex with deep nuclei are added to give total gray matter volume. The total brain parenchyma volume is sum of total gray matter volume, cortical and cerebellar white matter volume and brain stem volume.

Average cortical thickness was calculated as the average of all 68 regions of interest from both hemispheres generated by FreeSurfer (Table 4-4), weighted by the surface area occupied by each region.

Table 4.4 Regions included in measurement of average cortical thickness from FreeSurfer

Banks of superior temporal sulcus
Caudal anterior cingulate
Caudal middle frontal
Cuneus
Entorhinal
Fusiform
Inferior parietal
Inferior temporal
Isthmuscingulate
Lateral occipital
Lateral orbitofrontal
Lingual
Medial orbitofrontal
Middle temporal

Parahippocampal
Paracentral
Parsopercularis
Parsorbitalis
Parstriangularis
Pericalcarine
postcentral
Posteriorcingulate
Precentral
Precuneus
Rostral anterior cingulate
Rostral middle frontal
Superiorfrontal
Superiorparietal
Superiortemporal

Supramarginal
Frontal pole
Temporal pole
Transverse temporal
Insula

4.2.3 Statistical Analysis

Continuous variables were described as means and standard deviations, or medians and interquartile ranges, as appropriate with significance testing by analysis of variance (ANOVA) or Kruskal-Wallis test. The following brain regions were examined based on *a priori* hypotheses that they would be related to blood pressure: cortical gray matter volume, surface area and thickness, prefrontal cortical gray matter, surface area and thickness, basal ganglia volume, cerebellar volume and ventricular volume. Of 803 participants, 25 had brain volumes with poor segmentation (3.1%) and were excluded, leaving 778 participants for analysis. For other variables, data were missing in <1%.

Blood pressure was categorized as: a) normotensive (SBP <120 mmHg and DBP <80 mmHg), b) pre-hypertensive (SBP 120-139 mmHg or DBP 80-89 mmHg without hypertension), or c) hypertensive (SBP >140 mmHg or DBP >90mmHg or history of hypertension including antihypertensive medication use.[2] The purpose of this

classification was to compare the results of normotensive group with the pre-hypertensive and hypertensive groups to identify the association of hypertension with brain atrophy. Poorly controlled hypertension was defined as either SBP >140 mmHg or DBP >90 mmHg despite the use of antihypertensive medicines. The relationships between SBP, DBP and pulse pressure with brain volumes were examined.

Linear mixed models were used to determine the relationships between hypertension categories and brain volumes. To account for the study-sampling scheme, study site was included as a random variable in the models. Multivariable-adjusted linear mixed models were used to determine whether blood pressure category or blood pressures were associated with brain volumes, controlling for age and sex. Age was entered as a linear continuous covariate in the models, except for supratentorial hemispheric white matter volume in which there was a mild acceleration of volume loss with increasing age. Therefore, models of white matter volume included a quadratic as well as linear term for age. Blood pressures were entered as linear continuous covariates, after first viewing scatterplots of blood pressures vs. brain volumes to exclude the possibility of non-linear relationships. To determine whether relationships between blood pressures and brain atrophy differed in those taking antihypertensives vs. those not taking antihypertensives, we tested interaction terms in the fully adjusted models. Statistical analyses were performed (by EES) using SAS version 9.3 (Cary, USA). A *p*-value of <0.05 was considered significant.

4.3 Results

Among 803, 10 cases failed to complete initial processing steps of FreeSurfer either because of motion artifacts in the high-resolution T1-weighted images or the reason could not be determined. In these cases no further analysis could be done.

Surface based scoring method of brain images were used for inclusion of participants in the final volume calculation. Among 793 cases that completed FreeSurfer processing, 81 cases were found to have errors during surface reconstruction. The rest of cases were of good quality and labelled as grade-4.

Twelve cases were assigned quality grade 1, meaning that they were judged to be inadequate for further analysis without correction. In 2/12 (16.6%) cases were corrected by adjustment of the watershed threshold, which enabled more accurate removal of skull and dura, producing acceptable results. However, in the other 10/12 (83.3%) cases no corrections were possible and therefore they were removed from analysis: in one case a brainstem tumour caused artifact that preventing processing.

A total of 25 cases were scored grade 2, indicating mild errors there were judged visually to be acceptable for study statistical analyses. In 20/25 (80%) cases, FreeSurfer output was improved by reprocessing after placing control points 2/25 (8%) or adjusting the watershed zone to improve the skull stripping step 18/25 (72%), while in 5/25 (20%) cases no improvements could be made.

Another 44 cases had minor problems with either the pial surface including some part of dura and/or sagittal sinus or pial surface miss small part of frontal and temporal lobe

and scored grade 3. After all review and corrections, a total of 778 of 803 (96.8%) scans were judged to be usable for data analysis.

Demographic characteristics of study participants in relation to blood pressure category are given in Table 4-5. The overall mean SBP and DBP in the study population was 126±16 mmHg and 79±10 mmHg. The mean age of hypertensive participants was 60.8 years (SD ±7.6 years), of pre-hypertensives was 58.2 years (SD ± 8.1 years) and of normotensives was 55.2 years (SD ±7.2 years). The hypertensive group consisted of 269 persons, of whom 141 (52%) had a self-reported history of hypertension and 128 (48%) had measured blood pressure >140 mmHg/90 mmHg despite no self-reported history of hypertension. The mean SBP and DBP were 135±17 mmHg and 82±10 mmHg among the 141 self-reported hypertensives. Of these 141 hypertensive subjects, 117 (83%) were taking antihypertensive medications. There were 47 subjects with poorly controlled hypertension, defined as measured BP >140/90 despite known history of hypertension, and mean SBP and DBP in this group was 151±12 mmHg and 91±8 mmHg. Among the 128 subjects that had unrecognized high blood pressure despite no known history of hypertension, the mean SBP and DBP were 145±10 mmHg and 89±8 mmHg, indicating that the blood pressure elevation was relatively mild for most subjects. A total of 11.5% and 7.1% of hypertensive subjects showed infarcts and microbleeds respectively. The median volume of white matter lesions was 2.0 mL in hypertensive subjects, 1.6 mL in pre-hypertensive subjects and 1.4 mL in normotensive subjects ($p < 0.001$).

Mean volumes of hemispheric gray and white matter, prefrontal cortex, basal ganglia, cerebellum and ventricles are presented in Table 4-6, according to blood pressure category. In univariate analysis, there was a relationship between brain atrophy and the presence of pre-hypertension or hypertension ($p < 0.001$).

Multivariable-adjusted relationships between blood pressure category (normotensive, pre-hypertensive and hypertensive) and brain volumes are shown in Table 4-7. After adjusting for age and sex, there were no differences in brain volumes according to blood pressure category. Patients with poorly controlled blood pressure did not have more atrophy than other subjects.

Table 4-8 shows the difference in volume, surface area or thickness for each 10 mm Hg increase in either SBP, DBP or pulse pressure, adjusted for age and sex. Interactions with antihypertensive medication use were not significant ($p > 0.05$; data not shown), indicating that the relationships between blood pressures and brain volumes were not different in participants taking antihypertensive medications versus participants not taking antihypertensive medications.

Table 4.5 Characteristics of study participants according to blood pressure category

Characteristic		**Normotensives <120/90 n=221**	**Prehypertension 120-139/80-89 n=288**	**≥140/90 or history of hypertension n=269**	**P value**
Age		55.3±7.3	58.2±8.0	60.8±7.6	<0.001
Female sex		81%	48%	53%	<0.001
SBP		110±7	127±7	140±15	<0.001
DBP		69±6	79±6	86±10	<0.001
PP		40±7	48	54±12	<0.001
History of hypertension		0	0	53%	<0.001
Taking antihypertensive medications		1.4%	2.4%	44.2%	<0.001
Diabetes		3.2%	6.3%	8.2%	0.07
Coronary artery disease		1.4%	2.8%	5.2%	0.05
Smoking	Never	54.3%	53.3%	53.6%	0.75
	Former	36.7%	40.1%	40.1%	
	Current	9.1%	6.6%	6.4%	
BMI		24.2 [21.8, 27.1]	26.0 [23.4, 29.7]	27.9 [25.3, 31.4]	<0.001
Waist:hip ratio		0.80 [0.76, 0.87]	0.86 [0.80, 0.94]	0.88 [0.82, 0.94]	<0.001
SBI		2.7%	9.0%	11.5%	0.001
CMBs		3.6%	4.9%	7.1%	0.22
WMH (mL)		1.4 [0.8, 2.5]	1.5 [0.9, 3.0]	2.0 [1.2, 4.1]	<0.001
MoCA <26		21.4%	29.1%	30.2%	0.05

SBP = systolic blood pressure; DBP =diastolic blood pressure; PP= pulse pressure; BMI = body mass index; WMH = white matter hyperintensity; SBI= silent brain infarct ; CMB= cerebral microbleeds; MoCA = Montreal Cognitive Assessment tool.

Among the 141 with a history of hypertension, 117 (83%) were taking antihypertensive medications.
Values are percentages, mean±standard deviation, or median [25th percentile, 75th percentile]. Statistical testing by analysis of chi-square test, analysis of variance or Kruskal-Wallis test as appropriate.
Forty-nine participants had poorly controlled hypertension, defined as a measured SBP >140 or DBP >90 mmHg despite being on antihypertensive medications. A small number of normotensive or prehypertensive participants were taking antihypertensive class medications for other reasons—e.g. beta blockers for treatment of palpitations.

Table 4.6 Measured brain volumes by hypertensive group.

Structure	<120/90 n=221	Prehypertension 120-139/80-89 n=288	≥140/90 or history of hypertension n=269	P value
Cortical gray matter	450.3±34.3	440.5±36.3	438.0±36.5	<0.001
Prefrontal cortex	122.6±8.9	120.0±10.3	118.7±9.8	<0.001
Hemispheric white matter	476.2±32.9	470.3±38.1	464.8±40.3	0.004
Basal ganglia	23.5±1.8	23.1±2.0	23.0±1.9	0.008
Cerebellum	130.4±13.7	126.2±13.1	125.5±13.6	<0.001
Ventricles	17.7±8.3	21.2±11.1	22.0±11.3	<0.001

Values are mean± standard deviation. Brain volumes were normalized to the average intracranial volume (1,448.9 cm^3). 778 participants had usable MR data for volume calculations.
Mean cortical thickness was 2.48±0.3mm and mean cortical surface area was 1,629 cm^2.
Mean prefrontal cortical thickness was 2.53±0.3mm and mean prefrontal surface area was 481 cm^2.

Table 4.7 Relationships between blood pressure categories and brain volumes

	Total cortical gray matter			Prefrontal cortex gray matter			Hemispheric white matter (mL)	Basal Ganglia (mL)	Cerebellum (mL)	Ventricles (mL)
BP category	Volume (mL)	Surface Area (cm^2)	Thickness (mm)	Volume (mL)	Surface Area (cm^2)	Thickness (mm)				
<120/90	Ref	Ref	Ref	Ref	Ref	Ref	Ref	Ref	Ref	Ref
120-139/<90	-1.0 (-5.8, 3.8)	-5.3 (-21.4, 10.9)	-0.01 (-0.04,0.05)	-0.5 (-2.0, 1.0)	-2.2 (-8.0, 3.5)	-0.01 (-0.04, 0.06)	-2.1 (-8.2, 4.1)	-0.02 (-0.30, 0.34)	-1.1 (-3.4, 1.2)	0.9 (-0.9, 2.6)
≥140/90 or history of hypertension	-2.5 (-8.4, 1.2)	-5.2 (-22.9 11.5)	-0.009 (-0.05,0.04)	-1.2 (-2.7,0.4)	-3.2 (-9.1,2.7)	-0.003 (-0.05, 0.04)	-1.9 (-8.2, 4.5)	0.11 (-0.22, 0.44)	-1.0 (-3.4, 1.4)	0.5 (-1.3, 2.3)
Poorly controlled hypertension vs. the rest	-2.9 (-10.6,4.7)	-1.5 (-2.7, 2.4)	0.02 (-0.06,0.09)	0.3 (-2.1, 2.6)	1.7 (-7.5,10.9)	0.03 (-0.05, 0.11)	-9.1 (-19.0,0.7)	-0.14 (-0.7, 0.4)	-2.4 (-6.1, 1.2)	1.4 (-1.4, 4.2)

Ref =reference category. Table cells show the difference in volume, surface area or thickness compared to the reference category (<120/90), adjusted for age and sex

Table 4.8 Relationships between blood pressures and brain volumes

	Total cortical gray matter			Prefrontal cortex gray matter			Hemi-spheric White matter (mL)	Basal Ganglia (mL)	Cereb-ellum (mL)	Ventricles (mL)
BP measurement	Volume (mL)	Surface Area (cm^2)	Thickness (mm)	Volume (mL)	Surface Area (cm^2)	Thickness (mm)				
SBP	-0.2 (-1.5, 1.0)	-2.5 (-6.8, 1.7)	0 (-0.01,0.01)	-0.2 (-0.5,0.2)	-0.9 (-2.4, 0.6)	0 (-0.01,0.02)	-1.4 (-3.0, 0.2)	0.03 (-0.05,0.12)	-0.4 (-1.0, 0.2)	0.2 (-0.2, 0.7)
DBP	-0.7 (-2.6, 1.2)	-1.3 (-7.7, 5.0)	0.01 (-0.01,0.03)	-0.4 (-1.0,0.2)	-0.9 (-3.2,1.3)	0.01 (-0.01,0.03)	-1.5 (-3.9, 0.9)	0.1 (-0.02,0.23)	-0.2 (-1.1, 0.7)	0.3 (-0.3, 1.0)
PP	0.2 (-1.7, 2.1)	-4.6 (-11.0,1.9)	0 (-0.02,0.02)	0 (-0.6,0.6)	-1.1 (-3.4, 1.1)	0 (-0.02,0.02)	-1.7 (-4.2, 0.7)	-0.03 (-0.16, 0.10)	-0.7 (-1.6, 0.2)	0.2 (-0.5, 0.9)

Table cells show the difference in volume, surface area or thickness for each 10 mm Hg increase in pressure, adjusted for age and sex. SBP, systolic blood pressure; DBP, diastolic blood pressure. Interactions between blood pressure and use of antihypertensive medications were tested and were negative (p>0.05), indicating that the relationships between pressures and brain parameters were not

different in participants taking antihypertensive medications vs. participants not taking antihypertensive medications. Values in brackets are showing upper and lower limit of 95% confidence interval

4.4 Discussion

Quantitative automated methods were used to determine the relationship between brain volume and hypertension in healthy community dwelling participants free of stroke and dementia, most of whom were middle-aged (*i.e.*, <65 years old). Brain volumes were smaller in pre-hypertensives and hypertensives in univariate analysis but these relationships disappeared after controlling for age and sex, likely because of confounding by age, which was related to both higher prevalence of hypertension (Table 4-5) and lower brain volumes (data not shown). After controlling for age and sex, no significant relationship between SBP, DBP, or pulse pressure and atrophy of cortical gray matter, white matter, ventricles, basal ganglia or cerebellum was noted. These associations were absent even though we reproduced known associations between hypertension and MR imaging manifestations of cerebrovascular diseases—infarcts, microbleeds and white matter lesions (Table 4-5). The data suggest that BP is not associated with brain atrophy in a predominantly middle-aged Canadian population. No significant difference in brain volumes were found in poorly controlled hypertensives those who are taking antihypertensive medication or not taking medications. It may be that the mean duration of hypertension was not long enough to result in brain atrophy in this population, that our imaging methods were not sensitive enough to brain volume differences (although we used state of the art image processing methods that should be more sensitive than prior studies), or that improved population control of hypertension

compared to prior decades has attenuated the relationship between blood pressure and brain atrophy.

My results differ from many previous studies, which suggested that high BP levels are associated with brain atrophy.[51] In the Framingham Heart Study, midlife blood pressure was associated with atrophy of hippocampus and global brain in later life.[41] Similarly, in the Rotterdam Scan Study, higher and lower DBP readings in mid-life was associated with greater global brain atrophy in old age.[27] However, in the 3C study no significant association was seen between hypertension and brain atrophy, similar to the PURE-MIND study. The systematic literature review on BP and brain atrophy was tabulated in Chapter 2 of this thesis. Most of the population-based studies found association between blood pressure and brain atrophy unlike my results. Only one population based study [60] found positive association between BP and brain atrophy in the midlife, high SBP was associated with a significant reduction in gray matter volumes in the temporal lobe in hypertensive than normotensives.Three studies showed that DBP is associated with cortical atrophy: Heijer [27] found 0.52-units /year (CI 0.23-0.82) greater cortical atrophy with increase in DBP after controlling for age, Hoogendam found a borderline positive association between higher DBP and greater brain atrophy [56] and a third study also showed an association between higher DBP and greater brain atrophy in an older population.[57] Higher SBP was also related to ventricular atrophy [55] and to posterior part of corpus callosum.[53] Chen established a borderline association between frontal lobe gray matter atrophy and BP.[36]

Explanations for the differences between my study results and prior studies must be considered. One possible explanation is that average BP in PURE-MIND population was lower than the previous studies. Mean blood pressure in our sample population and in the hypertensive subjects was in the low range compared to prior studies, and the prevalence of hypertension was low (see Chapter 2, Table 2-7), indicating very good BP control or very mildly elevated pressures in my study population. A second potential explanation is that it may take a long period of time for atrophy to develop in hypertensive patients. There is a strong possibility that long standing hypertension causes change in the brain in the late life as a majority of previous studies found a positive relationship between hypertension and brain atrophy in older age groups [72, 81-83, 124]. In our cross-sectional study, most participants were <65 years old. Unfortunately, the PURE study does not contain information on the duration of hypertension. The least likely potential explanation is that FreeSurfer was not sensitive enough to detect brain volume differences. This seems unlikely given that FreeSurfer has been validated as a reliable and accurate method for brain volume calculation (see Chapter 3 for details). However, there may be other more subtle effects of hypertension on brain microarchitecture and function that could not be detected in this study. For example, a population-based study recently identified changes in white matter structure using diffusion tensor imaging in young subjects (mean age 39.2 years) with hypertension.[60]

The strength of our study is that we have a large number of people in our sample who are representative of the Canadian population. Qualified reviewers reviewed all the

MR images and brain volumes were included in the study after passing them through the quality control check and the quantitative analysis of brain volume. However, there are also limitations to consider. BP and brain volume were measured in a cross-sectional manner. Longitudinal studies are needed to see if subjects with elevated blood pressure develop atrophy over time. We did not have ambulatory BP recordings, which may be more closely linked with end-organ damage than averaged daytime sitting measurements. Another limitation is that we had few older participants our study group; therefore, the study results are applicable to a middle-aged population and may not reflect relationships in the elderly. Additional studies are needed to identify the relationship between BP and brain atrophy in elderly Canadians. We did not have information on hypertension duration and did not have information on antihypertensive medication class; therefore we could not analyze those variables.

4.5 Clinical Implications

The data failed to establish a significant association between the midlife hypertension and brain atrophy. However it is still a very important clinical observation. This study suggests that brain atrophy does not begin for quite some time after the diagnosis of hypertension. However other structural changes in the brain like WMH, microbleeds and infarct may appear in the early stages. There may be a considerable window for initiation of hypertension treatment to prevent atrophy in later life.

Chapter Five: Conclusions and Future Directions

5.1 Conclusions

This final chapter summarizes and provides concluding remarks regarding the relationship between hypertension, blood pressure and brain atrophy, based on systematic review of the previous literature (Chapter 2) and original research on the relationship between blood pressure and regional brain atrophy in 778 community-dwelling Canadians in the community-representative PURE-MIND study (Chapter 4).

Systematic review provided a strong motivation for this thesis project hypotheses that higher blood pressure would be associated with greater atrophy of the brain cortical gray matter, supratentorial white matter, basal ganglia and cerebellum. Most of the studies that met pre-specified inclusion criteria for review reported positive associations between higher blood pressure and presence of hypertension, and the presence of global or regional brain atrophy. However, we did note limitations of previous studies, including that they were mostly cross-sectional, that many were not population-based, that methods and metrics for reporting and analyzing brain atrophy varied greatly, and that neuroimaging methods were often incompletely specified. I did not find any randomized controlled trials that reported on longitudinal changes in brain atrophy according to different blood pressure treatment regimens; therefore, it remains unknown

whether atrophy can be prevented with treatment of hypertension. Most previous studies did not consider the possibility that hypertension treatment could modify the relationships between blood pressure and brain atrophy. Finally, most studies did not attempt to determine whether hypertension-related brain atrophy was associated with cognitive consequences.

Despite the prior evidence of relationships between hypertension, evidence of associations between blood pressure and brain atrophy was not found in PURE-MIND community-representative study despite using sensitive, modern neuroimaging methods. Interactions between blood pressure and anti-hypertensive treatment were tested and failed to find that atrophy was only present in poorly controlled hypertension. In sum, contemporary data from a relatively large cohort suggest that the relationship between blood pressure and brain atrophy in middle-aged Canadians is either weak or absent.

In light of original research findings which failed to confirm the thesis hypotheses, and are in conflict with much of the previous literature, some additional discussion is warranted. The potential explanations for the negative findings of the PURE-MIND study were discussed previously (Chapter 4, pages 105 to 106). The most likely explanation for findings is that we studied a younger population with relatively low blood pressures; most of the hypertensive participants had controlled blood pressure on medication or only very mildly elevated blood pressure. Alternative explanations are also possible advancement in treatment of hypertension with newer pharmaceutical agents from previous decades could be more beneficial effects in control of the disease and reduce

the incidence of associated complications. Follow up imaging of same participants in 5 years may establish the association between hypertension and blood pressure.

Limitations of original research study include the cross sectional design of the study, and the lack of older participants. These limitations should be considered when designing additional studies to investigate the relationship between blood pressure and brain atrophy.

5.2 Future Research Directions

The current study has established with high confidence that hypertension is not associated with brain atrophy in middle-aged Canadians sampled from the community. The prevalence of hypertension and poorly controlled hypertension were low in this study. However, we cannot exclude the possibility that mid-life hypertension, even if mild, could contribute to later life complications such as stroke and dementia in the 70s and 80s, as has been observed in other population-based studies. Elderly subjects were not included in this study. Therefore, it will be important to perform follow-up assessments in PURE-MIND to analyze whether baseline blood pressure, and changes in blood pressure over time, predict later-life brain atrophy and cognitive decline.

Larger studies would be needed to determine the relationships between brain atrophy and specific antihypertensive medication use. Ideally, these studies would be done as sub-studies of randomized controlled trials, testing whether specified antihypertensive protocols reduce the rate of brain atrophy in comparison to control groups.

Despite the large sample size of this study, there were relatively few participants that had poorly controlled hypertension. A case control design, comparing poorly controlled

hypertension to controlled hypertension and normotensives, could be considered to obtain more information in this subgroup that may be at higher risk for brain atrophy. Ideally, such a study would be nested within a large population-based study, so that the poorly controlled antihypertensive would be representative of poorly controlled hypertension in the overall population.

In this thesis the relationship between hypertension, blood pressure and brain-related clinical impairments such as cognitive impairment was not analyzed. Although the PURE-MIND study contains assessments of cognition, gait, mood and activities of living, analyses of these outcomes were outside the scope of my thesis. Additional work is ongoing by other team members to analyze the relationships between brain atrophy and these functional assessments. Future work should determine whether hypertension or blood pressure is associated with these clinical impairments. Even though blood pressure was not related to brain atrophy, it is possible that hypertension causes clinical impairments independent of the amount of atrophy. As explained in chapter 1 page 2, blood pressure causes structural changes in brain by initiating arteriosclerosis and micro hemorrhages; these changes damages the white matter tracts and brain cells and could lead to cognitive impairment and dementia.

5.3 Implications for Clinical Care

My study suggested that good control of BP might prevent complications of brain atrophy in midlife. However, it is not yet known whether control of BP will result in a reduced incidence of brain atrophy in later life. Improved awareness of hypertension, healthier lifestyles and more effective drugs may have all contributed to better

population control of blood pressure. However hypertension related complications such as stroke and dementia are increasing because of our aging population. Therefore, renewed attention to blood pressure control is still warranted.

Appendix I

Pre-specified Form for Data Extraction in Meta-analysis

Study lead author
Year of publication
Journal
Title
Overall N
Study type: RCT, cohort study, case control study
Prospective vs retrospective data collection
Setting: population, clinic
N with hypertension
Overall mean systolic blood pressure
Overall SBP SD
Overall mean diastolic blood pressure
Overall DBP SD
Cross-sectional vs longitudinal measurement of BP
Antihypertensive use reported (yes or no)
Mean age
Age range
Age standard deviation
Imaging Modality (CT or MR Imaging)
Atrophy measurement method Qualitative vs. Quantitative
Global atrophy reported (yes vs. no)
Regional atrophy reported and regions measured (list)

Which region were measured
Cross-sectional vs longitudinal comparison of atrophy
Univariate association between BP and brain atrophy
Explanation of Relationship
Multivariable-adjusted
Model covariates (list all)
BP meds considered as confounder (yes vs no)
BP meds considered as effect modifier (yes vs no)
Comments
exclude/include

Appendix II

Copyright release form for Fig 3.2

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