

HEART-BRAIN AXIS AND CARDIOVASCULAR RISK FACTORS IN YOUNG ADULTS

by

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Dedications

For Aoife, Hamish Wolf and my Family

Abstract

Lower indices of ideal cardiovascular health, such as hypertension, obesity, smoking, alcohol consumption and physical inactivity, are known risk factors for midlife cardiovascular and cerebrovascular disease. Whether modifiable cardiovascular risk factors, and novel early life exposures such as preterm birth, influence the structure and integrity of the heart-brain axis in young adults is less well studied.

This thesis investigates the heart-brain axis using detailed brain and cardiac imaging, exercise testing and ambulatory monitoring. Purposive recruitment established a cohort of young adults with a range of gestational ages and blood pressures. The detailed phenotypic measures were used to investigate associations between cardiac measures, brain volumes, brain cortical thickness, white matter hyperintensity lesions and brain vascular measures to understand factors influencing the heart-brain axis in early life.

It was demonstrated that cardiovascular risk profiles are associated with structural differences across the heart-brain axis. Healthier profiles of ideal cardiovascular health indices are associated with lower left ventricular mass to left ventricular end diastolic volume ratios, higher left ventricular stroke volumes and higher brain vessel density and vessel calibre. In addition, healthier cardiovascular profiles were associated with lower number of white matter hyperintensity lesions and higher brain blood flow. In addition, there was evidence of cortical-thalamic associations with cardiac function at rest. These associations were then tested under exercise stress and found evidence that cortical-thalamic structures are associated with indicators of peak exercise performance. There was also evidence of cortico-thalamic associations with submaximal cardiac output and myocardial strain.

The thesis then adopts best practice guidelines to design intervention strategies to promote ideal cardiovascular health in young adults. A systemic review of randomised control trials demonstrated an evidence gap, making it difficult to recommend specific lifestyle interventions for sustained cardiovascular risk reduction in young adults. However, beneficial short-term reduction in clinic blood pressures was shown to be associated with exercise intervention. Meta-analysis demonstrated supervised, moderate-to-high intensity exercise and frequent contact between participants and intervention teams as the most effective intervention components.

The preliminary data on the heart-brain axis and evidence assimilated in the systematic review have informed development of a randomized control trial to test the effectiveness of exercise intervention on ambulatory blood pressure and remodelling across the heart-brain axis. The combined work will inform future primary prevention strategies and identify biomarkers of heart-brain health to track in young adults.

List of Publications

Original Manuscripts Contributing to D.Phil. Thesis

1.Trial of Exercise to Prevent HypeRtension in young Adults (TEPHRA) a randomized controlled trial: study protocol. **Williamson W,** Huckstep O.J, Frangou E, Mohamed A, Tan C, Alsharqi M, Bertagnolli M, Lapidaire W, Newton J, Hanssen H, McManus R, Dawes H, Foster C, Lewandowski A.J, Leeson P. *BMC Cardiovascular Disorders* 2018;**18**:208. 0 citations accessed - 07/02/2019

2. Association of Cardiovascular Risk Factors with MRI Indices of Cerebrovascular Structure and Function and White Matter Hyperintensities in Young Adults. **Williamson W,** Lewandowski AJ, Forkert ND, et al. JAMA. 2018;320(7):665–673. doi:10.1001/jama.2018.11498. 2 citations accessed - 07/02/2019

3. Will Exercise Advice Be Sufficient for Treatment of Young Adults With Prehypertension and Hypertension? A Systematic Review and Meta-Analysis. **Williamson W,** Foster C, Reid H, Kelly P, Lewandowski AJ, Boardman H, Roberts N, McCartney D, Huckstep O, Newton J, Dawes H, Gerry S, Leeson P. Hypertension. 2016 Jul;68(1):78-87. doi:10.1161/HYPERTENSIONAHA.116.07431. Epub 2016 May 23. 26 citations accessed - 07/02/2019

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Clinical cardiovascular risk during young adulthood in offspring of hypertensive pregnancies: insights from a 20-year prospective follow-up birth cohort. Davis EF, Lewandowski AJ, Aye C, **Williamson W,** Boardman H, Huang RC, Mori TA, Newnham J, Beilin LJ, Leeson P. BMJ Open. 2015 Jun 23;5(6):e008136. doi: 10.1136/bmjopen-2015-008136. 50 citations accessed - 07/02/2019

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1 Introduction

1.1 Heart-Brain Health

A working definition of optimal heart-brain health could be considered the functional capacity to complete daily tasks across domains of thinking, moving and feeling, with the ability to perceive, interpret, and react to external environments[1]. Underpinning functional capacity is the physiological capacity to meet the associated energy demands, and therein, heart and brain health become intrinsically related[2]. Brain function is dependent on aerobic metabolism and requires adequate delivery of oxygen and glucose. Therefore, a regular blood supply, dependent on an adequate cardiac output and a healthy, functional, vascular tree, is required to meet the metabolic demands of the brain[3]. Many brain activities are energy intensive, ranging from resting state metabolism with basal synaptic activity and maintenance of ionic gradients across cell membranes to gene expression, protein synthesis and associated neuronal cellular regulation and axonal transportation[4]. Having minimal to no energy stores, neurons and glia cells are vulnerable to disruption to the regular energy supply. Therefore, brain blood flow and vascular function are tightly regulated to protect the energy demands of the brain[5]. Physiological capacity to rise above resting state function and meet flux in regional brain metabolic demand is regulated by neurovascular coupling, which ensures that focal increase in brain activity is matched by an increase in brain blood flow[6]. Therefore, the task of maintaining optimal heart-brain health can be simplified to that of protecting the capacity to meet the energy requirements of desired activities of daily living.

1.1.1 Heart-Brain Vascular Health in Young Adults and Future Risk of Disease

Functional capacity across the heart-brain axis begins to peak in late adolescence and then starts to decline from as early as the mid-twenties. Peak oxygen uptake, a measure of cardiovascular performance, declines at a rate of 5% to 10% per decade[7,8]. Cerebral metabolism and cerebral blood flow also decline at similar rates of 5% per decade[9,10]. The paralleled decline in heart-brain indices may suggests a similar pattern of biological aging associated with reduced efficiency in energy utilisation, oxygen uptake and vascular dysfunction (**Figure 1-1**).

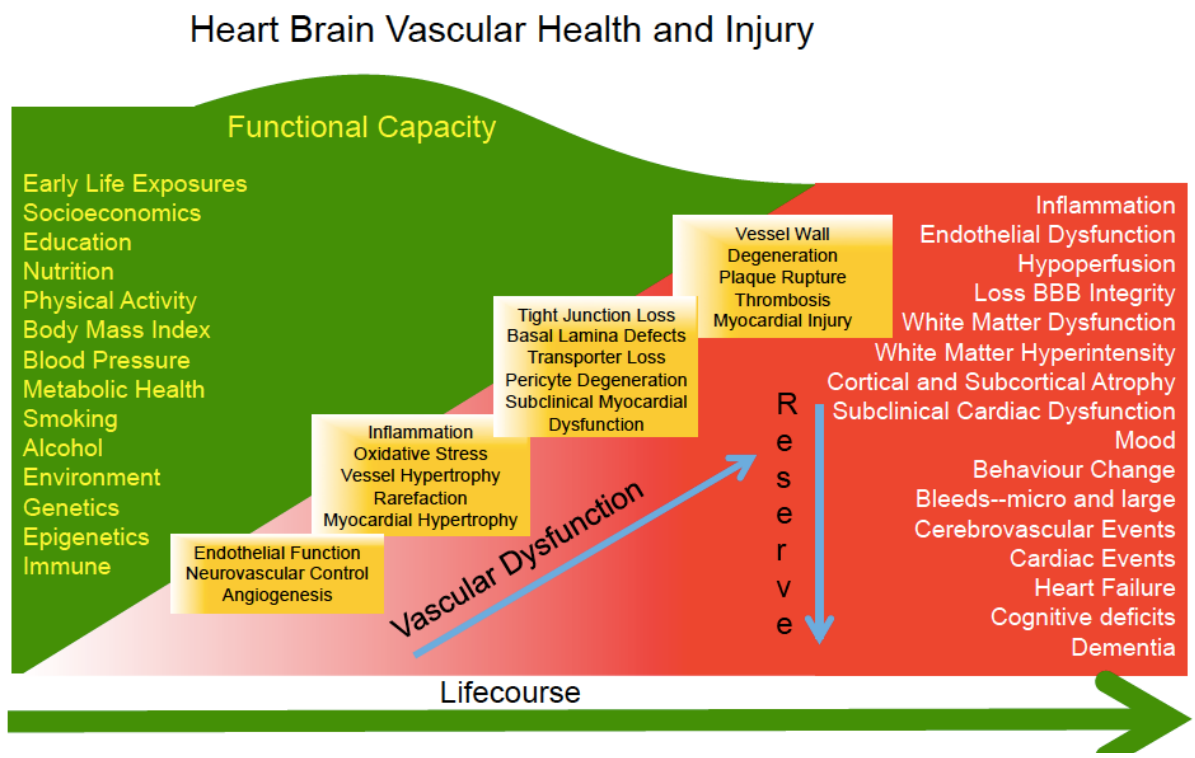

Figure 1-1 Heart-Brain Vascular Dysfunction Across the Lifecourse

Major predictors of resilience to illness and injury across the heart-brain axis are the developmental and physiological peaks reached in early adult life. Low cardiovascular

fitness in early adult life is an independent predictor of midlife risk of stroke and heart failure[11,12]. Furthermore, lower cognitive scores in adolescence are an independent predictor of midlife risk of stroke, heart failure and cardiovascular events[13]. In addition, higher educational attainment is a major protector against cardiovascular disease and dementia[14,15].

Indices of ideal cardiovascular health are consistently reported as strong determinants of future heart-brain health, but they may also impact achieved physiological and developmental peak and accelerate the rate of decline with age[16-18]. Longitudinal population studies have demonstrated that modifiable risk factors in childhood and adolescence can trigger pathological processes, with evidence of vascular dysfunction and early atherosclerotic change observed in major vessels of young adults[19,20]. Modifiable risk factors such as; hypertension, obesity, metabolic dysfunction, physical inactivity, smoking and alcohol consumption are attributable for over 40% of cardiac and cerebrovascular disease[1,15,21].

1.1.2 Heart-Brain Disease and Ideal Cardiovascular Health

Overall, the United Kingdom scores poorly on modifiable indices of ideal cardiovascular health with some of the worst figures in Europe: high rates of obesity (38% of men BMI > 35 kg/m^2)[22,23]; high rates of uncontrolled hypertension (1 in 3 adults reaching treatment threshold for hypertension, only 27% adequately treated)[24]; 1 in 6 smoking[25]; high rates of physical inactivity (39% of adults physically inactive)[26]; and some of the highest rates of alcohol consumption in the world (daily average 3-4 units)[27].

The current patterns of ideal cardiovascular health in UK place the population at risk of a renewed surge in cardiovascular disease. A similar trajectory has already been observed in the USA, which has witnessed 3-fold increase in cerebrovascular events in the under 55 year old population in the last 20 years[28]. There has also been a significant increase in the prevalence of heart failure in young adult populations[29]. There is growing concern that the burden of modifiable risk factors may start to contribute to a decline in life expectancy, and indeed, there is recent evidence of a reduction in life expectancy in the United Kingdom[30].

1.1.2.1 Cardiac Dysfunction and Disease

There is an established evidence base that demonstrates that early life exposures contribute to mid-life cardiac dysfunction and disease. In the Cardiovascular Risk in the Young Finns study, higher scores for ideal cardiovascular health during adolescence was associated with up to 30%-40% lower risk of coronary artery calcification and carotid intima media thickness in middle age[17,19]. The Coronary Artery Risk Development in Young Adults Study (CARDIA) also looked at coronary calcium scores in middle age adults who were recruited as young adults and identified similar risk trajectories[31]. Then, in a slightly older cohort, the Framingham Heart study demonstrated that being in the lowest tertile of ideal cardiovascular health at age 40 years was associated with an 11% risk of an ischaemic cardiovascular event in the following 10 years[32-34]. Incidence of acute cardiac events remains common in the UK population with 200,000 cases per year and up to 2.3 million people living with coronary heart disease[35].

An unexpected shift in population cardiovascular health has been the increased incidence of heart failure in young adults and there is uncertainty of the aetiology[29,36]. Swedish[37] and Danish[38] longitudinal studies report a 50% increased incidence of heart

failure in young adults and a similar trend is observed in the United Kingdom. Diagnosis of heart failure from 2002 to 2014 recorded on the Clinical Practice Research Datalink (CPRD), has increase nearly 2-fold in the under 55 year old population; the risks being highest in males from lower socio-economic groups[29]. In the Swedish cohorts low cardiovascular fitness, low musculoskeletal strength and increased body mass index were major risk factors for heart failure in young adults[11,39]. Low fitness was associated with near 2-fold increased risk of heart failure in young adults, while obesity was associated with a 6-to-10 fold increased risk.

There is some evidence from the Cardiovascular Risk in Young Finns Study and CARDIA studies that suggest left ventricular mass and left ventricular end diastolic volumes decrease with increase in ideal cardiovascular health[40]. However, remodelling of the left ventricle needs to be interpreted with reference to the stimulus and mechanism of remodelling. Increased left ventricular mass can be related to hypertension, obesity and cardiovascular fitness. To date the literature exploring cardiac modelling in association with cardiovascular fitness in young adults is dominated by studies reporting training response in military recruits and athletes[41,42]. These studies have observed an increase in left ventricular mass and end diastolic volume. However, none of the discussed studies have indexed cardiac mass with reference to volume. Left ventricular remodelling in response to cardiovascular risk exposures is multi-dimensional, with effects on mass, geometry and function. Further research is required to identify and understand cardiac phenotypes that may be contributing to the increase in heart failure incidence in younger adults and cardiac remodelling associated with modifiable exposures.

1.1.2.2 Stroke

In older adults, lower indices of ideal cardiovascular health are associated with up to 3-fold increase risk of stroke[43]. Similar patterns are observed in young adult populations. In a large case control study completed in Germany with 2125 stroke cases and 8500 controls, population-attributable risk of physical inactivity and hypertension accounted for 60% of the total number of strokes[44]. Globally, the estimate is that physical inactivity and hypertension accounts for 27.1% of the 2 million strokes that occur annually in young adults aged 18 to 55 years old[45,46]. Similar to the observations relating to heart failure, Swedish longitudinal cohort studies report increased risk of stroke associated with low cardiovascular fitness and low musculoskeletal strength measured in young adult life[12]. Population level surveillance provides an indication of the risk factors contributing to stroke incidence. However, there is major concern that current stroke prevention strategies are not reaching individuals at risk in early and midlife. Since 2007, the average age for first stroke in the United Kingdom has fallen by 3 years for men and women, with over a third of all strokes now occurring under 65 years old[47,48]. This is causing a major burden on society, as young people of working age are more likely to be unemployed after a stroke and the estimated annual hospital costs for stroke care alone is £26 million per year[47].

1.1.2.3 Cognitive Decline and Dementia

At a population level, evidence suggests that environmental and behavioural risk exposures in early to mid-life are stronger predictors of later life risk of dementia[1,15,49,50]. Longitudinal imaging studies suggest that over 58% of dementia cases may be triggered by mid-life vascular dysfunction[51,52]. Supporting these findings is the evidence that indices of ideal cardiovascular health measured in young adult life are associated with cognitive decline and markers of cerebral atrophy in mid-life[16,21,53]. However, there is limited understanding of cerebrovascular changes occurring in young adult life in association with ideal cardiovascular health. Furthermore, there is no understanding of the relationship with markers of cardiac structure and function. Therefore, there is currently a deficit in reliable tools to provide early and midlife risk assessment for future brain health. Providing evidence based approaches to support targeted primary prevention is a research priority[54]. In the UK, 300,000 new cases of dementia are diagnosed annually, and it is estimated that 2 million people will be living with dementia by 2040[55]. The current cost of dementia care in UK approaches £26 billion, which is higher than the combined cost for treatment of cancer and chronic heart disease[15,56].

1.2 Heart-Brain Axis – Homeostasis and Maintenance of Functional Capacity

1.2.1 Brain Neurovascular Unit

The functional integrity of neural connectivity and cerebrovascular reactivity is dependent on cellular and molecular co-ordination across the neurovascular unit[6]. The neurovascular unit is a complex anatomical and function system which refers to the interaction and interface between blood vessels, vascular endothelial cells, smooth muscle cells, neurons, astrocytes, pericytes and perivascular cells within the brain[2,4,5]. A primary function of the neurovascular unit is neurovascular coupling, which is the interaction between hemodynamic factors, neuronal activity, and cell-to-cell signalling. Neurovascular coupling maintains basal metabolic function and up-regulates vascular and metabolic response to match regional brain activity[2,4-6].

Haemodynamic responses associated with neurovascular coupling are mediated by vasodilation and vasoconstriction of the brain microvascular network. These responses are co-ordinated by complex interactions involving synaptic activation, neurotransmitter release from astrocytes and interneurons, and feed-forward and feedback regulation along neural paths between basal forebrain and brainstem[2,4-6]. A variety of molecular signals carry the stimulus for brain vascular vasoconstriction and vasodilation. These include partial pressure of carbon dioxide; neurotransmitters such as noradrenaline, glutamate and GABA; also nitric oxide and by-products of metabolism such as NADH, lactate and prostaglandins[2,4-6]. Regional endothelial response also regulates vascular tone in response to mechanical force, sheer stress and neurotransmitters within the brain microvascular network[57,58]. The endothelial cellar layer, and associated endothelial functions, may be the physiological system most vulnerable to modifiable risk factors and explain the interdependent relationships across the heart-brain axis.

1.2.2 Blood-Brain-Barrier

The endothelial cell layer within the neurovascular unit is the main interface of the blood-brain-barrier and the regulation of molecular exchange between the circulating blood and brain. In the brain, tight junction complexes adjoin endothelial cells to prevent the bidirectional exchange of hydrophilic substances across the blood brain barrier[59]. Transport channels located in the endothelial cell membrane regulate movement of metabolites across the blood-brain-barrier. Transport channels such as the GLUT transporters and aquaporins; regulate glucose uptake and facilitate removal of CO_2 and NH_3 to maintain metabolic haemostasis[59,60]. Neurogenesis and repair also depends on endothelial transporters, such as, LRP-1 and ABCB1 regulated channels are involved in the transport of lipids and lipoproteins and removal of waste products such tau and β-amyloid[57]. Integrity of the endothelial cell layer and efficient up-regulation and down-regulation of transport channels across the blood-brain-barrier function are essential for neuronal growth, repair and protection against toxic insult and hypoxic injury[2]. It maybe that cardiovascular risk exposures impair normal endothelial function within the neurovascular unit[4,61-63]. The accumulated burden of lower indices of ideal cardiovascular health may lead to multiple hits along different cell receptors and transport channels that lead to neurovascular endothelial dysfunction.

Endothelial cells and astrocytes produce a number of growth factors such as brain-derived neurotrophic factor (BDNF), insulin-like growth factor, and vascular endothelial growth factor (VEGF), which support the sprouting of new vessels, the migration of neuronal progenitor cells and oligodendrocytes and differentiation of neuronal cell lines[64,65]. The endothelial cells also provide the required energy and regulation of metabolism for growth and repair. Therefore, the neurovascular unit and the

microvascular network provide the scaffolding and essential growth factors for neurogenesis and thus determine the potential for neuroplasticity and the ability to recover following acute brain injury. Furthermore, neuroplasticity and neuronal repair are also dependent on functional immune signalling across the blood-brain-barrier and appropriate systematic vascular responses to support cell migration and regulation of inflammatory responses[2,6,61]. This includes the up-regulation of circulating endothelial progenitor cells (EPC) and release of pro-angiogenic factors to stimulate vascular homeostasis and maintain the integrity of the endothelial cells within the neurovascular unit[66,67]. Therefore, the functional integrity of the neurovascular unit to respond appropriately to both toxic stimuli or injury, and positive extrinsic stimuli with potential for neuroplasticity is dependent on systemic vascular and endothelial function[2,6]. Accumulated exposure to modifiable risk factors may impair the regulatory pathways co-ordinated by the neurovascular unit and endothelial function, and with time lead to impaired cerebral blood flow and white matter injury detectable on magnetic resonance imaging.

1.2.3 Brain Vascular Network

The internal carotid artery and vertebral arteries merge in the base of the skull to form the circle of Willis. Then the anterior, middle and posterior cerebral arteries branch off the circle of Willis to give rise to the pial arteries and arterioles, which provide the majority of the blood supply to the brain (**Figure 1-2**)[65]. The periventricular white matter, basal ganglia and the brainstem are supplied by penetrating arterioles arising directly from the circle of Willis and the proximal branches. This may make these areas more vulnerable to cardiac and vascular dysfunction. On the brain cortical surface, the pial arteries and arterioles form a rich anastomotic network of vessels. From the

cortical surface, the pial arteries and arterioles then penetrate down into the brain parenchyma to provide blood supply to the deep brain tissues[6].

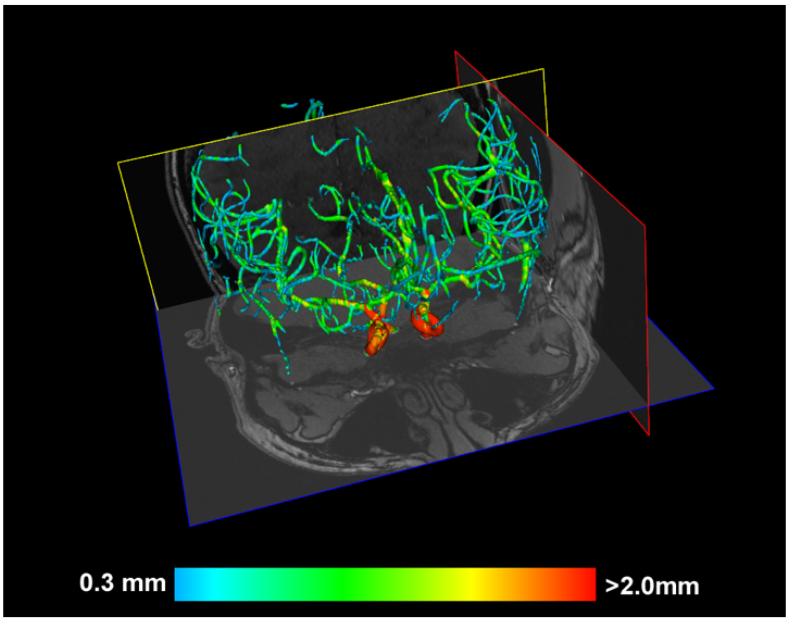

Figure 1-2 Time of Flight Magnetic Resonance Angiography of Cerebral Vessels

Time of Flight Magnetic Resonance Angiography (MRA) displaying the arterial anatomy of the cerebrovasculature. The colours represent the calibre of the vessel between 0.3 mm and 2 mm.

Pial arterioles are the only vessels to be able to penetrate the cortex, and in doing so they become interrelated with the neuron basal lamina, leading to the formation of the blood brain barrier. Pial vessels have a smooth muscle cell layer, which surrounds a layer of endothelial cells. As the pial cells branch into the brain substance, the pial arterioles are surrounded by an extension of subarachnoid space filled with cerebrospinal fluid, which forms the perivascular space[2,6]. As the pial capillaries perforate further into the brain, the perivascular space disappears, the vessel basement membrane and the glial basement membrane fuse and envelop around the astrocytes. Then, the smooth muscle cells of the capillaries are replaced by pericytes,

exposing more of the endothelial layer, and this becomes the blood-brain-barrier interface of the neurovascular unit[59,65,68].

The density of the brain capillary network means that the average inter-capillary distance is less than 40μm and approximates to each neuron having a discrete blood supply[69]. Therefore, the integrity and homeostasis of the neurovascular unit is intricately coupled to endothelial function and vascular health. It is estimated that penetrating arterioles and microcirculation contribute approximately 40% of brain vascular resistance while the pial surface vessels contribute 21% and the large cerebral vessel arteries contribute 39% of segmental vascular resistance[6]. Therefore, 60% of the variation in cerebral blood flow may be determined by the larger cerebral blood vessels outside of the brain parenchyma[2,6]. The regional contributions to cerebral blood flow highlight the potential risk to brain health from decline in the integrity and function of the major vessels. The morphology of brain vasculature, including vessel density, vessel calibre and vessel tortuosity can be assessed using time of flight magnetic resonance arteriography. It is uncertain whether variance in morphological characteristics in young adult relates to modifiable risk factors.

1.2.4 Cardiovascular contribution to Brain Blood Supply

In the resting state, regional cerebral blood flow closely approximates to oscillations in the blood flow measured in the middle cerebral artery suggesting a major cardiac impulse during functional magnetic resonance imaging and contribution to resting cerebrovascular haemodynamic function[70]. Relative distribution of cardiac output to the brain decreases with age and there is increasing evidence that decline in brain health in older adults is associated with lower left ventricular function[9,10,71-74]. However, there is less understanding of the cardiac contribution to brain vascular health and

cerebral blood flow in young adults. There are a number of pathological examples where dysfunctional cerebral blood flow directly relates to cardiac function. This is most apparent in cases of acute heart failure that resolve with treatment and this is most dramatic in the example of cardiac transplantation in paediatric and young adult populations where cerebral blood flow returns to normal[75].

Experimental models have been able to explore the continuum between variance in cardiovascular capacity and cerebral blood supply. Higher cardiovascular function in exercise trained older adults has been associated with higher cerebral blood flow, with cortical areas of the precuneus and posterior cingulate most affected[76-78]. Exercise detraining has also been associated with reduction in cerebral blood flow in older athletes[79]. The observed associations between cardiovascular performance and cerebral blood flow may identify a co-dependence across the heart-brain axis in these areas or highlight common vulnerability to vascular and metabolic dysfunction. The cortical associations with cardiac function in younger adults have not been described. Furthermore, there is limited understanding of the associations between cardiac function and indices of vascular brain health in younger adults.

1.2.5 Neuro-cardiac control

Cardiovascular response across the heart-brain axis is regulated by the autonomic nervous system[80,81]. Sympathetic and parasympathetic activity facilitates rapid response to emotional, environmental, physical and psychological stimuli. There is a growing body of multidisciplinary research that describe a central autonomic network[81-90]. Cortical areas commonly identified during neuroimaging associated with cardiovascular response include the prefrontal cortex with projections to the anterior and posterior cingulate cortex, the precuneus, and the insular cortex[80,81,84,91-93].

Subcortical nuclei, including the caudate and thalamus, have established projections to these cortical areas and have also been found to be active in the central autonomic network[92-102]. Thalamic nuclei have also been demonstrated to have synchronous activity with the cardiac cycle at rest and increased functional activity during cardiovascular stressor models[103]. Furthermore, direct stimulation of basal ganglia nuclei and thalamic nuclei during neurosurgical procedures for movement disorders have been associated with increased heart rate and blood pressure responses[85,104]. Primate and rodent studies have tracked vagal projections to the thalamus and basal ganglia nuclei, and these help to explain how direct stimulation of these areas elicits cardiovascular response[103,105,106].

Brainstem nuclei have traditionally been thought of as primary regulators of cardiac and vascular function. Animal studies have facilitated increased understanding of the projections of brain stem nuclei to higher subcortical and cortical areas that may form circuits involved in the regulation of cardiovascular activity[106-109]. There is an evolving matrix of different receptor and neurotransmitter driven anticipatory, inhibitory and stimulatory functions that may contribute to central autonomic control and cardiovascular response to exercise. Experimental models exploring projections and function of the locus coeruleus, a brain stem nucleus and neural hub for central autonomic control demonstrate the complexity of the regulatory pathways[101,110,111]. Direct stimulus of the locus coeruleus is demonstrated to regulate vagal function and heart rate[112,113]. Furthermore, other independent studies demonstrate that locus coeruleus activation also produces improved cortical information transmission and sensory processing through enhanced thalamic selectivity to the cortex and regulation of neurovascular coupling and cerebral blood flow[110,111].

Functional imaging studies are helping to assimilate the understanding from across the research field by demonstrating the neural networks involved in cardiorespiratory function[78,98,101,114-118]. This work includes identification of cortical hubs within the somatosensory and interoceptive networks active in regulation of cardiorespiratory responses to exercise[98]. These studies support animal models that demonstrate selective persistence of activity between cortical and subcortical nuclei that may be a trained response and facilitate anticipatory and pre-excitatory cardiorespiratory response[108]. Structural and functional imaging studies demonstrate differences between trained and sedentary individuals in brain areas, including the thalamus, insula and the posterior cingulate and precuneus cortical areas, which are related to heart rate, blood pressure and exercise response[78,98,101,114-118]. Differences in cortical thickness in the posterior cingulate and precuneus areas may even predict training response, and influence perceptions of exertion during exercise[83,116,119,120].

A number of studies are now attempting to manipulate higher cortical and subcortical regulation of central autonomic function to explore determinants of exercise capacity and cardiovascular function[121]. Transcranial stimulation has been used in trained athletes to explore effects on resting indices of heart rate variability an indicator of vagal tone and exercise response. At rest, transcranial stimulation over the temporal and insular cortex increased parasympathetic activity measured with heart rate variability in trained athletes[122]. The same stimulus was then demonstrated to reduce heart rate and perceptions of exertion in trained athletes during submaximal exercise, which was associated with an overall improvement in peak performance[123]. However, the same results could not be reproduced in sedentary individuals[124]. Meta-analysis of transcranial stimulation studies identifies a small, positive effect on exercise performance but also rates the quality of the studies as poor and there was significant

heterogeneity[125]. In clinical settings, studies investigating parasympathetic activity in relation to mood disorders report improved heart rate variability at rest following transcranial stimulation over the dorsolateral prefrontal cortex[126,127]. There appears to be considerable heterogeneity in autonomic response patterns across populations and better understanding of characteristics of heart-brain phenotypes could improve intervention and help predict response.

In animal models, modulation of the activation of dorsal vagal motor nuclei (DVMN) demonstrates the potential importance of central autonomic control on cardiovascular function. Genetic silencing of brainstem vagal preganglionic nuclei in the dorsal vagal motor nucleus resulted in impaired peak exercise capacity[128]. When vagal tone was increased, via optogenetic recruitment of dorsal vagal motor nuclei, cardiac contractility and exercise performance improved[128]. These animal models point towards a strong vagal regulatory component in the modulation of cardiac reactivity and exercise capacity. The same research group, then investigated the associations of heart rate recovery, an indicator of vagal recruitment, with measures of cardiovascular capacity in clinical population. In a sample of 1200 older adults undergoing pre-operative cardiopulmonary exercise testing, there was a graded response between heart rate recovery and independent measures of aerobic performance including peak oxygen uptake, ventilatory anaerobic threshold and oxygen pulse[128]. It seems that cardiac vagal preganglionic neurons innervate the myocardium, and dorsal vagal motor nuclei neurons directly contribute to the autonomic control of electrical and contractile properties of the left ventricle. Therefore, the experimental data from human and animal models demonstrate vagal tone to be a primary determinant of cardiovascular performance. This expanding literature is helping to provide a theoretical framework to

investigate the functional and anatomical networks that compose the central autonomic network and higher regulatory control of cardiovascular function.

1.3 Burden of Young Adult Cardiovascular Risk Exposures and Interactions Across Heart-Brain Axis

1.3.1 Preterm Birth

Young adults with a history of preterm birth have increased risk of hypertension, stroke, heart failure and cardiovascular mortality[129-132]. In the United Kingdom, 5% to 13% of all live births are born preterm, before 37 weeks gestation[133]. Advances in neonatal care have resulted in a dramatic increase in survival from preterm birth. In the United Kingdom an estimated 2 million young adults below the age of 40 years were born preterm[134].

Preterm birth exposes the immature heart-brain axis to extraurterine conditions. This has a significant multisystem effect, with a shift from placental to left-sided cardiac circulation, pulmonary ventilation, increased oxidative stress, introduction of oral nutrition and extrinsic stressors associated with neonatal intensive care such as medical intervention and pharmacological treatment[135]. Evidence from animal models and studies in preterm infants suggests that the combined exposures lead to altered cardiac and cerebrovascular development[135-139].

The cerebrovascular system and white matter may be particularly vulnerable to preterm delivery. Neonatal imaging studies suggest reduced tortuosity across the circle of Willis and risk of periventricular white matter injury[139]. The third trimester is an important period of cerebrovascular remodelling[64,65,140]. Vessel density in the gray matter

increases incrementally in the third trimester in parallel with cortical network development to approach adult levels of brain vessel density[64,65,140]. There is also a degree of vessel remodelling; with increased brain vessel tortuosity and decline in white matter vessel density during the third trimester[64,65,140]. The remodelling of the vascular tree is stimulated by angiogenic stimuli and hypoxic induced activation of HIF 1α, and these stimuli may be inhibited by neonatal healthcare practice and preterm birth[65]. The extrinsic physiological stressors, increased cardiac workload, and exposure to pain in the neonatal environment may also influence subcortical development and functional connectivity development, and this in turn may influence cardiovascular response in adult life[136,141,142].

Cardiovascular phenotyping studies have observed that young adults born preterm may have evidence of endothelial dysfunction and altered cardiac structure and function[143-145]. MRI imaging suggest altered geometric cardiac shape, increased left ventricular mass, reduced myocardial relaxation and reduced left ventricular stroke volumes with normal ejection fraction in association with preterm birth[144,145]. The available literature suggests that preterm born adults may be a risk group for reduced functional capacity across the heart-brain axis, with potential vulnerability to the effects of modifiable risk factors[146]. To date no studies have explored structure and function across the heart-brain axis in young adults adjusted for gestational age and modifiable risk factors. Such studies, aimed at understanding the interactions across the heart-brain axis and association with modifiable risk factors in relation to gestation age, may help to segment risk populations and target interventions.

1.3.2 Young Adult Hypertension and Blood Pressure

Lifetime risk of cardiovascular and cerebrovascular events is associated with modest increase in blood pressure above 115 mmHg[147]. Acknowledgement of the wider spectrum of risk associated with blood pressure previously considered to be normotensive has lead the American College of Cardiology (ACC) and American Heart Association (AHA) to revise blood pressure (BP) guidelines. The revised guidelines introduce a category of elevated blood pressure, defined as clinic measured systolic blood pressure of 120 mmHg to 129 mmHg and diastolic blood pressure less than 80 mmHg. They also reclassify stage 1 hypertension as systolic blood pressure of 130 mmHg to 139 mmHg or diastolic blood pressure of 80 mmHg to 89 mmHg. The previous threshold for stage 1 hypertension of blood pressure above 140/90 mmHg is now classified by the ACC/AHA as stage 2 hypertension[148]. However, at the individual level and especially in younger adults there is debate regards risk associated with isolated systolic blood pressure. In part, the debate has been driven by observation of pulse amplification of brachial blood pressures and when central blood pressures are measured in younger adults the central blood pressure may be up to 10 mmHg or more lower than the brachial measure[149]. In the limited studies with available central blood pressures the evidence supports central blood pressure as a risk indicator[150]. However, previous observation in younger adults has been limited by sample size of studies and duration of longitudinal follow-up to hard endpoint. Recent evidence from the HELIUS study does identify that isolated systolic hypertension is associated with pulse amplification but that the majority of participants with isolated systolic hypertension still had elevated blood pressure on central measure[151]. Therefore, there may be less need

for concern about over estimating a young adult's cardiovascular risk by relying on brachial blood pressure.

Since the publication of the new AHA hypertension guidelines there has been renewed interesting in studying risk of young adult hypertension. The reclassification has dramatically increased the number of young adults classed as hypertensive. Up to 40% of young adults could now be classified as hypertensive. The revised guidelines have led to controversy with regards appropriate screening and treatment approaches for young adults[152,153]. A major barrier to evidence-based practice for young adults has been the bias in pharmacological and non-pharmacological cardiovascular prevention trials skewed towards recruitment of older adults and higher risk cardiovascular populations. Currently, the available data suggests no additional benefit or potential harm associated with pharmacological antihypertensive interventions in younger adults with mild hypertension associated with lower 10-year cardiovascular risk[154]. However, cardiovascular risk calculators have only limited validity in young adult populations, and have predominantly been developed using data from populations aged 40 to 79 years old[155]. Furthermore, lifetime risk for young adults with hypertension may still be high[156,157]. Therefore, applying current treatment algorithms to young adults may be inappropriate; there is both risk of under estimation of risk and adverse events secondary to limited understanding of the associated heart-brain phenotypes. However, this does not diminish the potential risk the individual and highlights the need for development of sustainable lifestyle interventions and alternative treatment options.

The available prospective evidence from young adult populations demonstrate future risk of cardiovascular events in association with elevated blood pressure. Cardiovascular event rates in 148 204 men aged 35 to 44 years screened for the Multiple Risk Factor Intervention Trial (MRFIT) was over 2-fold higher for men with

blood pressure between 130-139/85-89 mmHg (18.4 events per 10 000 person years) compared with blood pressure less than 120/80 mmHg (8.8 events per 10 000 years)[158]. In the CARDIA study, elevated blood pressure, stage 1 and stage 2 hypertension, defined as per the 2017 ACC/AHA guidelines, and recorded during at least 2 study visits, before age 40 years, were associated with increased risk of a cardiovascular events including coronary heart disease, transient ischaemic attack, stroke and heart failure[152]. Risk associated with stage 2 hypertension was 3-fold higher in comparison with the normotensive group (hazard ratio 3.49, 95%CI 2.42 to 5.05). There was also evidence, from the CARDIA study of cardiac events occurring within 10 years of confirmed hypertension, therefore still in a young age group[152]. Increased risk of all-cause mortality was also associated with stage 2 hypertension. However, the absolute event rate was still comparatively low in the CARDIA study and the future risk beyond midlife follow-up remains uncertain[152].

Given the risk of cardiovascular events and mortality but also the risk of complications associated with current treatment options there are calls for new research in young adult populations with hypertension and early cardiovascular risk[154]. This includes recommendations to develop new risk stratification tools and targeted intervention trials to guide management strategies[152]. Medical imaging may offer new directions for risk assessment and outcome measures to use in management of young adult cardiovascular risk. Markers of cardiac remodelling such as left ventricular hypertrophy and subclinical indicators of atherosclerotic disease such as IMT and coronary artery calcification could be used as potential screening tools and outcome measures to target in primary prevention trials[19,20,31,159,160]. However, the evidence-base to support the use of coronary calcification scores, as screening tools in young adults, is limited, less than 10% of individuals under 40 years will have any calcification[156,157]. In addition, the

presence of coronary calcium in isolation may not be a sensitive screening tool[161]. A combination of using ideal cardiovascular risk scores and coronary artery calcification may improve feasibility as a screening tool, but this may still be limited to use in midlife[32].

Imaging-based screening for subclinical atherosclerotic markers may not be sensitive enough to use in younger adult populations. There may still be a significant lag phase between risk exposures and atherosclerotic change, and the specificity of measuring coronary calcification may not be comprehensive enough as an imaging strategy to improve cardiovascular risk phenotyping. Magnetic resonance imaging across the heart-brain axis may provide more sensitive tools to use in young adult populations to improve cardiovascular risk phenotyping. Evidence from the Framingham and CARDIA studies demonstrates decline in indices of brain white matter microstructure and cerebral volume in association with elevated blood pressure in middle age adults[36,162-164]. However, there is uncertainty of the duration between exposure and remodelling and limitations with regards normal reference ranges for cerebral volumes to facilitate clinical application. It is also uncertain whether blood pressure and cardiovascular risk indices in younger adults under the age of 40 years is associated with MRI indices of brain health. There is also evidence to suggest that more comprehensive blood pressure assessment including exercise blood pressure response and ambulatory blood pressure may be needed to improve risk stratification in young adults. Exercise blood pressure response at age 25 in the CARDIA study is associated with midlife cerebral atrophy[53].

The vasculature across the heart-brain axis is most vulnerable to exposure to hypertension. Vessel walls and the myocardium experience significant mechanical stress stimulating hypertrophic remodelling of smooth muscle cells and

cardiomyocytes. There is up-regulation of extracellular metalloproteases, increased deposition of collagen and fibronectin, and extravasation of plasma proteins and macrophages around capillaries and arterioles that leads to inflammatory responses. This is associated with loss of vascular and myocardial compliance and increase in stiffness, which may reduce shear stress response. These changes are common across the vascular tree but lead to different effects, ranging from luminal narrowing and atherosclerotic change in vessels, fibrotic changes in the myocardium, disruption of the blood brain barrier, and white matter damage including loss of microstructural integrity and white matter hyperintensity lesions, which are most marked around the periventricular areas[4,165,166]. There is also significant microvascular rarefaction across the vascular tree, which is exacerbated by age[4,165,166]. Chronic hypertension exposure is also associated with endothelial dysfunction across the vascular network[4,165,166]. The combined structural and functional changes lead to disruption of vascular haemodynamics and associated risk of tissue hypoperfusion as well as increased risk of prothrombotic and atherosclerotic environments across the heart-brain axis[1,166]. The cerebrovasculature is particularly vulnerable, with microvascular dysfunction leading to potential impairment in neurovascular coupling, cerebral autoregulation and disruption of the blood brain barrier[4,165].

Given the recent data from the CARDIA study there is now stronger evidence to support intervention to manage young adult blood pressure above 130/80mmHg. This thesis adopts blood pressure classification and terms developed in the recent AHA guidelines, with blood pressure below 130/80 mmHg being classified as ideal blood pressure.

1.3.3 Obesity and Lipid Dysfunction

Obesity and dyslipidaemia during adolescence and young adult life are predictive of cognitive decline with lower scores in executive function when measured in midlife; currently in the UK 1 in 3 of the population between 16 and 24 years old are overweight or obese[22,28,39]. The prevalence of obesity increases with age with 60% overweight or obese at age 44 years, with body mass index at age 50 a major predictor of future brain health[167]. Elevated BMI in adolescence and young adult life is associated with up to a 5-fold increased risk of midlife hypertension and diabetes, 2-fold increased risk of carotid artery atherosclerosis, and up to 5-fold increased risk of angiography-proven coronary heart disease in midlife[17,19,40,168].

Animal models describe the potential underlying mechanisms for neurovascular decline associated with lipid dysfunction and BMI[4,6]. Early exposure to behavioural lifestyle risk factors leads to dysfunction in cerebral energy metabolism, which is associated with reduced integrity of the blood brain barrier driven by reduced expression of lipid protein receptor (LRP1) and lipoprotein lipase (LPL)[169-171]. The impaired lipid channel transport compromises growth and repair with increased risk of neuronal injury. The impaired neurogenesis during young adult exposure may lead to reduced peak functional capacity and risk of midlife decline in function integrity[16]. By midlife accumulated neuronal injury leads to disruptive plaque formation and global disruption in functional networks and subsequent decline in cognitive function and behaviours[4]. As the pathological processes accumulate, these may continue to exacerbate behavioural neglect, with an on-going decline in physical activity, maintenance of adequate nutrition and progressive decline in activities of daily living and then, later in life, dementia[172,173].

1.3.4 Physical activity and cardiovascular fitness

Currently, 39% of adults in the United Kingdom are physically inactive, with an estimated economic cost of £7.4 billion and accountable for 1 in 6 deaths[35,167]. In younger adults, there are significant gender differences in participation, 70-75% of males age between 16 and 44 years self-report attainment of aerobic guidelines while only 65% of women report recommended levels. Less than 50% of men and less than 35% of women under 44 years old achieve both aerobic and strength training guidelines[35,167].

Physical activity and exercise participation have multisystem effects that combine to influence the efficiency of energy and substrate delivery across the heart-brain axis[174]. The metabolic benefits are driven by improved insulin sensitivity, increased expression of glucose transporters, mitochondrial growth, improved mitochondrial efficiency, and more efficient efflux of metabolic by-products[174]. There are also complex metabolic bone, hepatic and renal adaptations; which may benefit the heart-brain axis including: acid-base regulation, up-regulation of thermoregulation capacity, increased efficiency in sodium and potassium transport, regulation of adiposity, and improved lipid metabolism. The cardiopulmonary adaptations associated with exercise lead to improved oxygen uptake, which is associated with adaptations across numerous vascular beds with common features of increased vessel density, increased vessel compliance and up-regulation of endothelial function[175-177]. Regular exercise also leads to functional network development associated with learning and motor skill development[94,96,117,178-181]. The combined effects are believed to provide cognitive and vascular resilience and optimise neurogenesis and recovery from neuronal injury[182,183].

Major mechanisms of exercise adaptation across the heart-brain axis include enhanced eNOS driven vascular endothelial function, improved immune function and enhanced inflammatory regulation[174]. A common regulator of exercise adaptations across the heart-brain axis is the myoendocrine function of musculoskeletal system[184]. Examples of immune and inflammatory regulation include IL-6 and BNDF effects on fat oxidation and regulation of adipokine function to reduce TNF-alpha and pro-inflammatory activity[174,184]. Other examples include the role of the myokine irisin, which suppresses vascular inflammation, enhances endothelial function and also improves insulin sensitivity and glucose tolerance[174,184]. Circulating irisin may also drive increased expression of neuronal BDNF and the irisin/BDNF pathway is suggested as the mechanistic link between exercise participation and strengthened learning and cognitive function[185,186].

Seminal exercise trials demonstrated the cardiac benefits of exercise via endothelium-dependent vasodilatation in patients with stable coronary artery artherosclerosis. Exercise intervention proved to significantly reduce rates of coronary stenosis and improved myocardial perfusion[187,188]. Atherosclerotic plaque rupture is a major pathological process for both ischaemic myocardial events and cerebrovascular stroke. However, individuals with higher cardiovascular fitness and concomitant coronary atherosclerotic plaque are observed to have lower lipid inclusion and more fibrotic plaques, which suggests higher plaque stability[161,189,190].

Musculoskeletal expression of irisin may also be contributing to the increased arterial plaque stability. Experimental models demonstrate that plaque stability associated with exercise is not an eNOS driven pathway, but rather, appears to be regulated by multifactorial anti-inflammatory pathways which include reduced macrophage involvement and reduced TNF alpha in both cardiac and cerebral vascular beds[184-186].

There is also reduced matrix metalloproteinase activity and increased smooth muscle cell content of coronary plaques in association with exercise training in animal models. The coronary action of irisin also appears to reduce vascular inflammation and endothelial dysfunction via suppression of oxidized low-density lipoprotein and activation of reactive oxygen species pathways and inflammatory gene expression[184-186,191].

In the brain there are observations of increased vessel density, maintained cerebral vascular perfusion and increased neurogenesis in association with higher cardiovascular fitness[78]. In addition to the eNOS pathways that regulate cerebral vascular adaptation, recent experimental studies have identified a number of other potential regulatory pathways that may be stimulated by regular exercise[192]. These include a lactate sensitive receptor HCAR1 that mediates release of brain endothelial VEGFA and increases vessel density, down regulation of matrix metalloproteinase and improved basal luminal integrity that is protective against ischaemic injury, and increased activation of the angiotensin-converting enzyme 2, which enhances local cerebral perfusion and vessel density[192,193]. In addition to established cardiorespiratory adaptations to exercise, there is increased appreciation from human and animal models of central regulation of autonomic function and functional connectivity adaptations to exercise that may mediate health benefits across the heart-brain axis.

1.3.5 Smoking

Approximately 20% of the young adult population continue to smoke tobacco and 4% of hospital admissions and 16% of all deaths are attributable to smoking[167]. Oxidized free radicals in tobacco smoke are believed to be the main drivers of vascular pathology, which follows a pattern of endothelial dysfunction, accelerated

atherosclerotic change and prothrombotic environment. Young adults are not protected against the negative effects of smoking. The Cardiovascular Risk in Young Finns study has reported increased carotid IMT in association with smoking in young adults and studies in adolescent populations demonstrate early endothelial dysfunction[16,17,20]. The CARDIA study also demonstrated that smoking in young adults was associated with a trend towards increase in left ventricular mass and increased ventricular wall stress consistent with myocardial stiffness[31,36,194]. The LARGE study also reported increased left ventricular mass in young adults associated with smoking exposure, however, this was a military cohort undergoing training and the effects of baseline cardiovascular fitness were not adjusted for in analysis[195]. The cerebrovascular impacts of smoking include disruption of the blood brain barrier, disruption to auto-regulation, which in older adults has been associated with white matter injury, and reduced cerebral perfusion[162,196-198]. It is uncertain what effects smoking has on cerebrovascular indices in younger adults.

1.3.6 Alcohol consumption

Recent studies demonstrate graded increased risk of all-cause mortality, stroke, fatal hypertensive disease, heart failure and fatal aortic aneurysm per additional 100 g/week higher alcohol consumption (approx.14 units). On average, a 40 years old male drinking 200g per week (approx. 28 units) might have a 2-3 years shorter life-expectancy compared to drinking within the guidelines (14 units or 112 g/week)[199]. In the Whitehall study, there is also a relatively linear increased risk of dementia with consumption above 14 units[200]. Alcohol consumption in the British Isle's continues to occur at dangerous levels, and the controversy around the role of alcohol in society continues. In total 58% of adults regularly drink alcohol and this increases to 79% of

those earning more that £49,000[167]. The most dangerous drinking is occurring in late middle age, but it is unlikely that public policy to target this age group will change in the near future[167].

It may be more effective to focus alcohol policy on alcohol behaviours in adolescents and younger adults and protect against the potential negative effects on the developing brain. Over 70% of 15 year olds have had alcohol in the last 12 months and 23% report being drunk in the last 4 weeks[167]. There is longitudinal evidence of risk of alcohol consumption in young adults. The Cardiovascular Risk in the Young Finns study reports increased carotid IMT with all alcohol levels and the CARDIA study reports increased coronary calcium scores at midlife follow-up[201,202]. However, it is uncertain what effects alcohol consumption may be having on the cerebrovascular and cardiac function on younger adults. MRI imaging in older adults reported reduced subcortical volumes and loss of white matter microstructural integrity in association with alcohol consumption[203]. Regards cardiac structure, the CARDIA study reports an association between alcohol consumption and mid-life left ventricular mass and function but no young adult associations[204]. The LARGE study reported simple bivariable association between left ventricular mass and alcohol consumption; however, this association did not remain significant in multivariable analysis[195].

1.4 Primary Prevention for Heart-Brain Health

1.4.1 The argument for early intervention

Lower indices of ideal cardiovascular health across the life-course are consistently reported as strong determinants of future heart-brain health[162]. However, in older adults the strength of association declines within the 10 years of clinical diagnosis of

cerebrovascular event or cognitive decline[43,162]. This is consistent with the recent trajectory analyses reported from the Whitehall study, where behavioural related indices of physical activity and body mass index decline 8 to 14 years prior to diagnosis of cognitive decline and dementia[172,173,200,205]. It is suggested that these observations describe a prodromal period of subclinical disease. Recent work published from the Alzheimer's Disease Neuroimaging Initiative (ADNI) dataset would support this conclusion, demonstrating that the aetiology of dementia started in midlife, 3 to 4 decades before onset of symptoms[51,52,206,207]. The primary feature common to future diagnosis was brain vascular disorganisation, including dysfunction in indices of perfusion and resting state metabolism.

The work on the ADNI study also suggested that there were differences in individual potential for neuroplasticity that could be equated as the quotient of energy required to achieve favourable remodelling. Therefore, at the individual level, there may be significant heterogeneity in the type of intervention that would be most effective to deliver the required quotient of energy for positive heart-brain remodelling. The baseline functional capacity across the heart-brain axis may determine the type and intensity of intervention, the appropriate outcome measure and likelihood for success. This may explain why the majority of lifestyle interventions in older adults have performed relatively poorly as regards improvement in the primary physiological or brain health related outcomes. The growing evidence of early and midlife determinants of future heart-brain health and interaction with modifiable risk factors suggests that early and midlife interventions are need to achieve primary prevention and protect future heart-brain health.

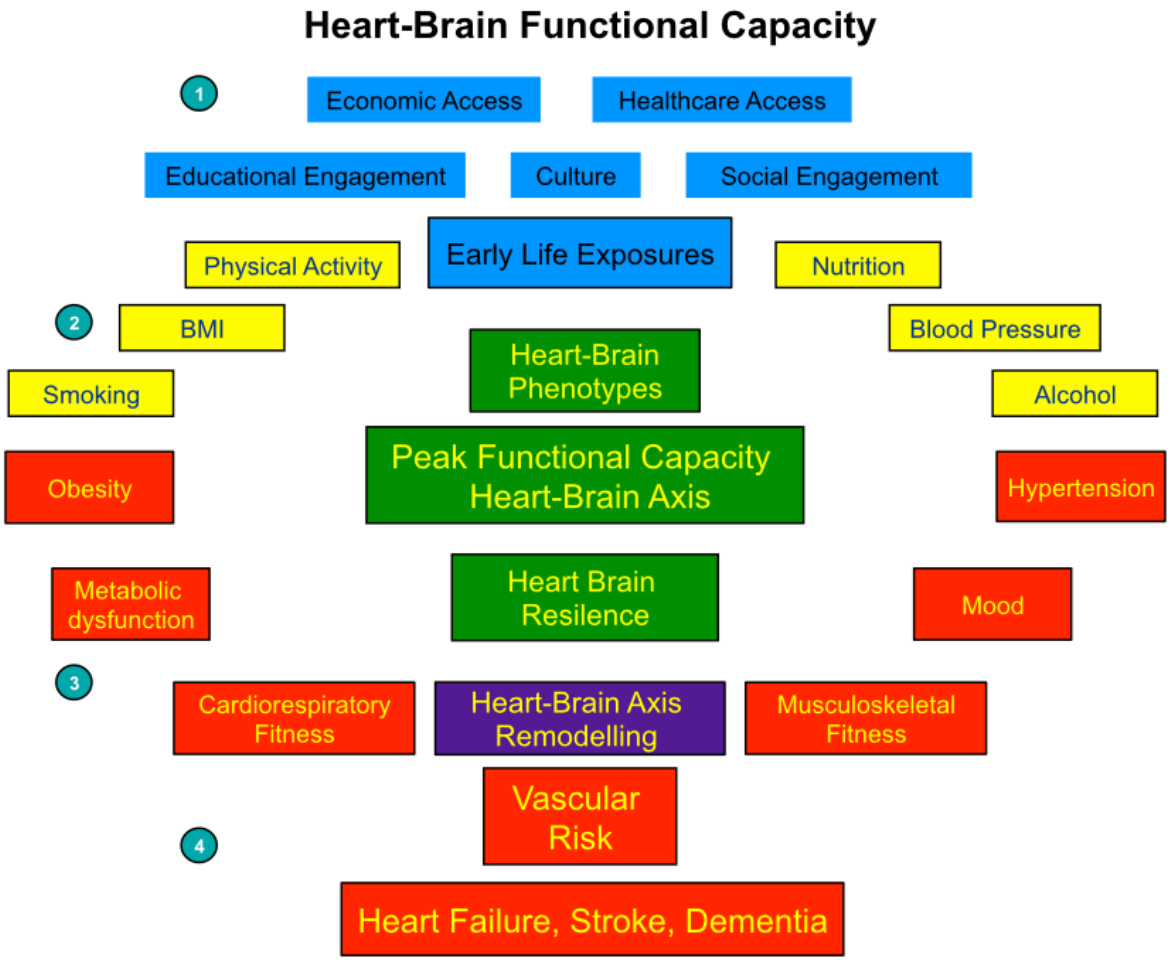

Figure 1-3 Targets and Stages of Prevention to Promote Heart-Brain Health
The figure outlines stages of prevention to promote and protect heart-brain health. Stage 1 is primordial prevention, Stage 2 describes primary prevention, Stage 3 is secondary prevention and Stage 4 is tertiary prevention to combat accelerated decline of heart-brain function in vulnerable groups.

Figure 1-3 outlines stages of prevention to promote and protect heart brain health. Phase 1 would focus on primordial prevention (Stage1), targeting societal and macro-level problems that may shape the development of the heart-brain axis. This could include strategies to reduce preterm birth to reduce risk of developing features of preterm cardiovascular phenotypes. Stage 2 describes primary prevention strategies to promote and maintain peak functional capacity across the heart-brain axis. This

could then enhance cognitive and vascular reserve and resilience to aging or injury. Primary prevention strategies could be improved by understanding the distinctions in heart-brain phenotypes between high risk and flow risk populations and the interactions with modifiable risk factors. Stage 3 is secondary prevention to reduce decline in heart-brain capacity and prevent pathological remodelling in the presence of known modifiable risk factors such as obesity, hypertension and metabolic dysfunction. Secondary prevention in chronic disease groups could benefit from personalised tailored intervention to address the complexity of risk factors and barriers to promoting ideal health behaviours. Barriers to intervention and risk reduction may include pathological remodelling across the heart-brain axis. Understanding heart-brain driven barriers could facilitate strategies to improve compliance and efficacy of interventions. Stage 4 is tertiary prevention to combat accelerated decline in populations with potentially the greatest needs and precarious independence and function. Understanding the heart-brain axis could help target feasible and pragmatic interventions that accommodate existing loss of function. Better understanding of the interactions across the heart-brain axis may also identify specific intervention strategies and biological targets for intervention.

1.4.2 Segmented Audiences and Early Outcome Markers

Major barriers to early intervention are the remoteness between risk exposures and hard endpoints and the scale of population burden of modifiable risk factors. A population approach for targeting primary prevention in young adults lacks individual specificity and has a long lead-time before observed benefits in reduced disease incidence. This means that it requires long-term vision and planning. An alternative

approach is to segment and target at-risk audiences and use interim measures of heart-brain health to target as intervention outcomes. Understanding the heart-brain axis in young adult populations and interactions with modifiable risk factors has the potential to identify young adults with unique risk phenotypes that would benefit from targeted intervention to protect future heart-brain health. Medical imaging and advances in magnetic resonance image acquisition and processing offer the potential for highly sensitive, quantitative measures of heart-brain health and vascular integrity decades in advance of clinical symptoms. The objective of this study is therefore to use multimodal imaging in young adults with a range of cardiovascular risk exposures to investigate heart-brain associations and interactions with modifiable risk factors. The aim is to strive towards individualised assessment of risk and personalized prevention plans and to identify potential measures to use as interim outcome markers of heart-brain health.

1.4.3 Intervention Planning

The Medical Research Council (MRC) has developed guidance on the design and implementation of complex interventions[208]. There are also established recommendations and strategies for application of social marketing techniques in healthcare to optimise recruitment, segmentation of target audiences and delivery of interventions. The design and delivery of a primary prevention intervention to promote heart-brain health in young adult populations will benefit from following established guidance for complex intervention implementation. The MRC guidance follows phases of 1., defining components of the intervention, 2., defining trial and intervention design, 3., addressing methodological issues of the trial to guide trial design, 4, promoting effective implementation[208].

Practical steps described to align with the phases of trial development include identifying existing evidence. The available epidemiological evidence, as explained above, highlights the fact that targeting young adults with elevated blood pressure may be a feasible step towards reducing early risk of cardiovascular and cerebrovascular disease. In addition, young adults with preterm birth may be more likely to be hypertensive and have lower resilience to low indices of ideal cardiovascular health, which may exacerbate cardiovascular risk. Current National Institute of Clinical Evidence (NICE) guidance recommends lifestyle intervention as the primary intervention in young adults with hypertension[209]. This includes increase in cardiovascular fitness and weight loss. Structured exercise interventions offer the potential for benefit in cardiovascular fitness and weight loss and are recommended in older adults. However, there is limited trial evidence on cardiovascular prevention interventions in young adults with hypertension[165,166]. Therefore, additional work is required to complete a systematic review of the available evidence and identify key intervention components for cardiovascular risk reduction in young adults with hypertension and elevated cardiovascular risk. The systematic review will provide the theoretical understanding needed of how previous interventions cause change, so that potentially weak components of the intervention can be strengthened.

MRC guidance also recommends modelling and piloting the processes involved in the study, which includes review of outcome measures[208]. As mentioned previously, there is uncertainty about the interim outcome markers to use in young adult populations as indicators of heart-brain health. In addition, a single outcome measure may not capture the heterogeneity in baseline functional capacity across the heart-brain axis or the potential unintended consequences of intervention, which might explain some of the underlying determinants of risk and health outcome. This includes understanding the

physiological mechanisms up-regulated or down-regulated by intervention. Completing a detailed cross-sectional study investigating the heart-brain axis in young adults with a range of cardiovascular risk factors provides the platform to pilot study processes involved in a trial and refine appropriate outcome measures. A cross sectional study combined with a systematic review of available trial evidence will also inform the appropriate sample size to target.

The systematic review and completion of the cross-sectional study will help to reduce implementation failure and assess feasibility of the trial. Young adults with elevated blood pressure and preterm birth may be hard to identify, and the feasibility of intervention is dependent on timely intervention. Therefore effective advertising and screening strategies are required to recruit the target audience, which can be explored in the setting of the cross-sectional study. The cross-sectional study also offers the opportunity to gain invaluable public feedback to inform the design and components of an intervention.

1.5 Thesis Objectives and Hypotheses

Objective 1: To examine the relationships between modifiable cardiovascular risk factors and cerebrovascular structure, function and white matter integrity in young adults. Then to test the hypothesis that indices of ideal cardiovascular health will be associated with brain vessel morphology, white matter hyperintensity lesion counts and cerebral blood flow in young adults.

Objective 2: To explore the extent to which gestational age and modifiable risk factors relate to cardiac structure and function in young adults. Then to test the hypothesis that indices of cardiac structure and function are independently associated with white matter hyperintensity lesion counts and perfusion.

Objective 3: To examine relationships between thalamic and basal ganglia volumes, cortical thickness, and cardiac function at rest and during exercise in young adults. The hypothesis is that in young adults, thalamic volume is associated with MRI indices of cardiac function at rest and markers of cardiovascular performance during exercise, independent of cardiovascular fitness and early life exposures such as preterm birth.

Objective 4. To perform a systematic review and meta-analysis to evaluate the quality of the evidence base and effectiveness of exercise intervention to reduce blood pressure in young adult populations. This will test the feasibility and potential efficacy of exercise intervention to improve modifiable cardiovascular risk indices in young adults with elevated cardiovascular risk to protect future heart-brain health.

Objective 5. To complete the MRC phases of complex intervention development and design a study protocol to test the effectiveness of exercise intervention for primary prevention of cardiovascular risk in young adults with elevated blood pressure.

2 YACHT Study Design

2.1 Summary Overview

A cross-sectional observational study was completed between August 2014 and May 2016. Participants between 18 and 40 years were recruited through purposive active and passive recruitment including local advertising, invitation from local birth cohort studies and invitation from the Oxford University Hospital Hypertension Service. Strategies were designed to recruit young adults with heterogeneity in risk factors known to be present in young adult populations including traditional risk factors such as hypertension and more novel factors such as gestational age. Participants were excluded if they had history of a previous cardiovascular or cerebrovascular event, renal dysfunction or metabolic disease including diagnosis of familial hyperlipidaemia. Participants with secondary causes of hypertension such as renal vascular disease, vascular anomalies or adrenal dysfunction were excluded following assessment in the Oxford University Hospital Hypertension Service. Measurements were completed at the Oxford Cardiovascular Clinical Research Facility and Oxford Centre of Clinical Magnetic Resonance Research, John Radcliffe Hospital, University of Oxford, United Kingdom. Final data collection was completed on the 31st of May 2016.

2.2 Sponsor and Ethical Approval

The South Central Research Ethics Committee for the National Health Service Health Research Authority (NHS HRA) approved the study (14/SC/0275). All participants gave written informed consent.

2.3 Eligibility

2.3.1 Inclusion Criteria

Eligible participants were defined by the following criteria:

- Be willing and able to give informed consent for participation in the study
- Be willing and able to complete study visits at the John Radcliffe Hospital, Oxford, United Kingdom
- Be able (in the investigator's opinion) and willing to comply with all study measures and requirements
- Be male or female, from 18 to 40 years old
- Have verified birth history: preterm birth (<37 weeks) or full-term birth (>37 weeks)

2.3.2 Exclusion Criteria

The participant was not eligible to enter the study if any of the following applied:

- Unwilling or unable to give informed consent for participation in the study.
- Pregnant or lactating during the course of the study
- Simultaneous participation in another human or clinical randomized trial (if there is any possibility of compromising health, safety, or well-being, or any possible compromise of study data)
- Planning to donate blood during the study duration

- BMI >40 kg/m^2
- Major contra-indications to exercise
- Evidence of cardiomyopathy
- Evidence of inherited cardiac conduction abnormalities
- Evidence of congenital heart disease or significant chronic disease relevant to cardiovascular status.
- A permanent pacemaker
- History of Family Hyperlipidaemia/Hypercholesterolemia
- Any other significant disease or disorder which, in the opinion of the investigator, might influence the participant's ability to participate in the study.
- Contraindications to Magnetic Resonance Imaging:
  - shrapnel injuries
  - metal clips in blood vessels of the brain
  - other metal or electronic implants adversely affected by the magnetic field
  - an injury to the eye involving fragments of metal
  - unsuitable for MRI based on responses on the MRI safety screening form.

2.3.3 Eligibility Assessment

As a clinically trained study investigator, who held an honorary clinical contract with the Oxford University Hospitals Trust I discussed eligibility for study participation with individuals who had been provided with the participant information leaflets, and expressed an interest in the study. I provided them with an opportunity to ask any

questions relating to study participation. I assessed study eligibility face-to-face when possible and if required during telephone calls to potential participants. During any conversation I asked for verbal consent to ask potential participants about their medical history and birth history. This was to clarify if there were significant contraindications, diseases or disorders which, might have influenced the individual's ability to participate in the study.

2.4 Recruitment

Participants between 18 and 40 years old were recruited through purposive active and passive recruitment including local advertising, invitation from local birth cohort studies and invitation from the Oxford University Hospital Hypertension Service.

Three main recruitment approaches were implemented and described below:

1. Recruitment from the Oxford University Hospital Hypertension Service.
2. Contacting those who gave consent to be contacted for research from previous ethically approved studies including invitation following participation in a local birth cohort study, the Exercise and Blood Pressure Study Oxfordshire (EXPRESSO).
3. Open advertising targeting Oxford residents

1. Oxford University Hospital Hypertension Service.

NICE guidance recommends young people under the age of 40 with hypertension have specialist review to guide management[209]. Young people referred to the Oxford University Hospital Trust Hypertension service were invited to participate in the study.

As a member of the clinical team working in the adult hypertension service I identified potential participants from recent referral lists maintained by the clinical administration

team. I reviewed referrals made to the service made between August 2013 and August 2015 to identify potential eligible participants. Potential participants were then sent the study information sheets along with an invitation letter that I had signed along with the lead physician for the service Professor Paul Leeson. After a minimum of two weeks, I then followed up the letters with a telephone call to discuss an individual's potential involvement. If required I then gained the individual's verbal consent to correspond via email and to send them more information and answer any additional questions.

Eligible participants, willing to participate in the study were then invited to attend the Oxford Cardiovascular Clinical Research Facility to complete the informed consent process. Participants were asked for their consent to access their medical records including birth and neonatal records. Birth and neonatal records were only reviewed if the individual was born and cared for in Oxfordshire. In absence of birth records self-reported birth history was used to verify birth history. If possible participants were asked to confirm their birth and pregnancy details, such as birth weight and gestational age with their parents.

2. Recruitment from EXPRESSO Study and other ethically approved studies.

Participants from previous ethically approved studies, that expressed interest to participate in future studies and consented to being contacted were made aware of the study and invited to participate. Individual's taking part in the Exercise and Blood Pressure Study Oxfordshire (EXPRESSO) were invited to take part in the study. I designed and established the EXPRESSO study with the support of Professor Paul Leeson as the principal investigator. Our patient and participant involvement (PPI) group informed the study design. I led a workshop with the PPI group discussing our interest in understanding the associations between birth history and future

cardiovascular risk. I asked the PPI group how best to recruit young adults with known histories of preterm birth to explore relationships between lifestyle exposures and birth history. The PPI group's recommendation was to establish a local cohort of young adults via contact tracing and invitation from the local birth records. I subsequently wrote the design for the study, secured ethical approval and initiated recruitment for the study; the study is currently on going. The South Central Research Ethics Committee for the National Health Service Health Research Authority (NHS HRA) approved the study (14/SC/0281). Individuals taking part in the EXPRESSO study were informed about the YACHT2 study at the time of their study visit and if interested were provided with the study information. Following provision of study information and after a minimum of 24 hours the research team contacted interested individuals. Individuals were contacted via email or telephone call to explore if they were interested to also participate in the study. I answered any questions individuals asked and if willing I invited the participants to schedule a study visit and to complete the informed consent process.

3. Open advertising targeting Oxford residents

Open advertising in the local community was used to invited participants to participate in the study. Posters were placed in University of Oxford Colleges and on notice boards at the Oxford Brookes University. This included research department notice boards at the John Radcliffe Hospital, Oxford. University email bulletins were approached to share an invitation email with their readership.

Where persons express an interest in the study the initial contact was followed up (according to the person's wishes) by a telephone call, letter, e-mail or if necessary by meeting the person. Interested individuals were provided with the full study information

sheets, which were sent with an accompanying cover letter. Following provision of the study information sheets individuals were contacted after a minimum of 24 hours and given an opportunity to talk through any questions. I answered any questions the potential participant may have had and confirmed that the potential participant was suitable for recruitment. Eligible participants were then invited to schedule a visit to complete the study consent process at the Oxford Cardiovascular Clinical Research Facility.

2.5 Informed Consent

Individuals willing to participate in the study were provided with the full participant information leaflets detailing the study information in advance of the informed consent process. Only after participants had a minimum of 24 hours to review the study information were they invited to provide consent to participate. The participant personally signed and dated the approved version of the informed consent form before any study specific procedures were performed. Written and verbal versions of the participant information leaflet and informed consent form were presented to the participants detailing the nature of the study; what it involved for the participant; and any side effects or any risks involved in taking part. It was clearly stated that the participant was free to withdraw from the study at any time for any reason without prejudice to future care, and with no obligation to give the reason for withdrawal.

Candidates were asked for their consent to access their medical records including birth and neonatal records. In absence of obstetric records, self-reported birth history was used to verify birth history. As appropriate, participants were asked to confirm their birth and pregnancy details with appropriate input from family or guardian(s).

2.6 Participant Confidentiality

The study staff ensured that the participants' anonymity was maintained at all times. Only the assigned study ID number on the case report form and in the electronic database identified participants. All study documents were stored securely and only accessible by study staff and authorised personnel. The study complied with the Data Protection Act.

2.7 Sample Size

Purposive recruitment was used to recruit a sample of young adults for the YACHT study with a range of cardiovascular fitness, gestational ages at birth and blood pressures. This included recruitment of preterm born participants from local birth registers and invitation from local attendance at the local hypertension clinic.

To guide recruitment a series of power calculations were completed. Differences in measures between lowest and highest cardiovascular risk tertile groups were used as the comparison to provide an estimate for the sample size to target during recruitment. Peak oxygen uptake was used as the primary cardiovascular outcome measure for the sample calculations. Previous studies have demonstrated greater that 10 ml/kg/min difference in peak vo2 between highest and lowest tertiles of populations. This would require a sample size of 30 participants per group. To be more conservative a difference between 3 and 6 ml/kg/min between tertiles was used. This equated to a target sample of 40 participants per group. This is based on a simple between group comparison using a two-sided significance level of $p=0.05$ and power of 80%.

As variability of cerebrovascular measures in young adults was unknown prior to the study this calculation was powered to identify a 0.70-SD difference in vessel density,

vessel calibre, and white matter lesion count between lowest and highest cardiovascular risk tertile groups. Arterial spin labeling (ASL) MRI imaging was added during the course of the study to provide a subgroup of 52 participants, recruited sequentially for an estimated 80% power to detect a 10% difference in cerebral blood flow. These calculations recommended 40 participants per group powered to 80% at a significance level of p=0.05. Recruitment was continued to over 120 participants in the MRI group to accommodate for drop out and missing data secondary to image quality.

3 YACHT Study Measures and Population Description

3.1 Overview

The core study measures were completed during a 4-5 hour visit, participants had the option to complete measures on a single day or split over two days, at the participants' convenience. Participants were invited to complete a study questionnaire during the visit or alternatively to take the questionnaire home to return by pre-paid envelope. The questionnaire provided self-report measures of lifestyle behaviours such as physical activity, smoking and alcohol consumption.

To measure structure and function across the heart-brain axis participants were invited to have non-invasive brain and cardiac imaging. Cardiac imaging included echocardiogram and magnetic resonance imaging (MRI). To measure metabolic status participants were invited to provide blood samples to measure inflammatory, angiogenic and metabolic profiles. To provide a measure of cardiovascular fitness and peak exercise capacity participants were asked to complete cardiopulmonary exercise testing and stress echocardiography (optional subgroup). The study visit concluded with participants being provided with blood pressure and physical activity monitors to wear. These monitors provided objective measures of ambulatory blood pressure and physical activity while participants went about their normal activities.

3.2 Participant Preparation

Participants were provided with a participant information leaflet prior to the study to explain the study visit and measures. Participants were asked to attend the Oxford Cardiovascular Clinical Research facility at the John Radcliffe Hospital, having fasted for a minimum of 4-6 hours prior to the study visit. Participants were instructed to wear comfortable clothing and bring clothes to exercise in. Participants were also instructed that facilities were available to shower after completion of the exercise testing.

3.3 Study Measures

3.3.1 Lifestyle Questionnaire

The questionnaire consisted of validated questions and self-report tools developed in independent studies. The questionnaire was piloted prior to use to confirm comprehension and ease of completion. The questionnaire included self-reported smoking history and current frequency, alcohol consumption, brief questionnaire of nutritional choices. Questions also included, self-reported medical history, birth history, exercise levels, and indicators of social-economic status including years in education, educational level achieved, occupation and indication of financial stability. Physical activity questions included the Recreational Physical Activity Questionnaire (RPAQ)) and determinants of activity including environmental perceptions. Questionnaire items also provided a measure of self-efficacy, enjoyment and motivation to engage with physical activity. The questionnaire includes a measure of health related quality of life captured through the SF-8 Health Survey question elements, a validated questionnaire tool. These questionnaire elements were combined into one questionnaire document and took less than 40 minutes to complete.

Participants were provided with the questionnaire at the start of their visit and encouraged to complete the questionnaire during the visit. If required questionnaires were taken home to completed and returned in a prepaid envelope.

3.3.2 Anthropometric Measures

Using an integrated height and weight measurement station (Seca, Birmingham, United Kingdom), height was measured to the nearest centimetre and weight measured to the nearest 0.1 kg with footwear removed and participants wearing light clothing. Manual waist circumference was measured 2 cm above the iliac crest, and manual hip measurements were taken at the point of widest overall girth near the level of the greater trochanter of the femur or mid-buttock.

Participants had their blood pressure checked after 5 minutes rest using the automated mode of a validated sphygmomanometer (Dinamap V100, GE Healthcare, Chalfont St. Giles, United Kingdom). Three blood pressure readings were taken at intervals of 1 minute. The mean of the second and third readings was used as the clinic blood pressure reading.

3.3.3 Brain Magnetic Resonance Imaging

Individuals underwent a multimodality MRI scan (3.0T Trio Tim, Siemens, Munich, Germany). The Brain MRI protocol included: T1-weighted structural imaging (TR/TE = 2040/4.7 ms, flip angle 8°, FOV 200 mm, voxel size 1.0 mm isotropic); T2-weighted FLAIR (TR/TE = 9000/90 ms, flip angle 150°, FOV 220 mm voxel size 1.1 x 0.9 x 3.0 mm); Diffusion Weighted Imaging (DWI) (TR/TE = 8900/95 ms, 2.0 mm isotropic resolution, multiband echo-planar imaging (EPI), 64 slices, 64 diffusion weighted

directions, FOV 192 mm, b-value 1500s/mm^2, five non-diffusion weighted images, b-value 0s/mm^{2,} with one b0 volume acquired in the reverse phase encoded direction); Time-of-Flight (TOF) MRA (TR/TE = 23/8 ms, flip angle 10°, FOV 300 mm voxel size 1.6 x 1.2 x 5.0 mm); and in a subgroup of participants, multi-delay vessel-encoded pseudocontinuous Arterial Spin Labelling (ASL), identical to a previously published protocol[210] (subgroup n=52).

Brain vessel segmentation was completed on TOF MRA using previously described automated segmentation tools[211,212]. Binary segmentations were used to determine vessel density, calibre and tortuosity. Processing of the TOF MRA images was completed by study collaborator Dr Nils Forkert, University of Calgary.

I use the Brain Intensity AbNormality Classification Algorithm (BIANCA) from the FMRIB Software Library (FSL) to segment white matter hyperintensity (WMH) lesions and associated volumes. BIANCA is a fully-automated, supervised method for WMH detection[213,214]. BIANCA classifies image voxels based on their intensity and spatial features, where the intensity features were extracted from T2-weighted FLAIR, T1-weighted and DTI fractional anisotropy (FA) images, FA images were generated using DTI tools, FSL topup, FSL eddy and DTIFit[215,216]. I manually segmented WMH lesions on 10 participant image datasets to use as the training set for BIANCA, these were independently verified by Dr David McClean, Consultant Radiologist and Dr Timo Siepmann a Consultant Neurologist (TS) who were blinded to the participant risk profile. Lesion count was selected as the most sensitive outcome of white matter change in young adults in whom a single lesion, independent of volume, could be considered abnormal[217]. I used a minimum lesion size of 1 mm^3 to extract lesion count and volume from the BIANCA outputs.

I processed T1-weighted images using FMRIB Software Library (FSL) tools[215] to generate gray and white matter segmentations. I processed the diffusion weighted imagines using established DTI tools, FSL topup, FSL eddy and DTIFit[215,216,218,219] to generate fractional anisotropy (FA) images and mean diffusivity (MD) images. I completed subcortical segmentation using FSL MIST (Multimodal Image Segmentation Tool (MIST)) a fully automated segmentation tool. MIST takes advantage of unique MRI contrast properties of the subcortical nuclei to optimise shape and boundary recognition across T1, T2 FLAIR and FA images.

To estimate cortical thickness I also processed the T1-weighted images using the CIVET 2.1.0 pipeline developed at the Montreal Neurological Institute (MNI). Images were aligned to the Montreal Neurological Institute (MNI) template using a multi-scale, 9 parameter transformation. The images were corrected for non-uniformities using the nonparametric nonuniformity intensity normalization (N3) method. The N3 spline distance used was 125 mm. The partial volume tissue classification is completed with iterative correction and a subcortical gray tissue class to distinguish deep gray from cortical gray matter. Cortical thickness was calculated at 81,924 vertices (the perpendicular distance between gray and white matter surfaces) across the cortex via the t-laplace metric.

A subgroup of 52 participants also had multi-delay vessel-encoded pseudocontinuous Arterial Spin Labelling (ASL), identical to a previously published protocol[210]. I processed the ASL images using a previously described analysis pipelines to estimate cerebral blood flow and blood arrival time[210,220]. Gray matter masks were used to calculate the average cerebral blood flow after linear registration of the ASL MRI to the T1-weighted MRI dataset.

3.3.4 Cardiac Magnetic Resonance Imaging

Cardiac steady-state free-precession cine sequences was used to acquire localization images, followed by optimized left ventricular horizontal and vertical long-axis cines. Left ventricular short-axis cine stack was obtained with standardized basal slice alignment, 7-mm slice thickness, 3-mm inter-slice gap. Image acquisition parameters for the steady-state free-precession images were as follows: echo time 1.5 ms; repetition time 3.0 ms; flip angle 60°.

All cardiovascular magnetic resonance imaging was prospectively ECG gated with a precordial 3-lead ECG and acquired during end-expiration breath holding. The short- and long-axis steady-state free-precession images were stored on a digital archive for post processing, which was undertaken as detailed below.

Image analysis for left ventricular volumes and mass was performed offline on the short-axis cine stack with Argus (Siemens Healthcare, Erlangen, Germany) as previously described[144,145]. Left ventricular short-axis epicardial and endocardial borders were manually contoured at end diastole and end systole to allow automated calculation of left ventricular mass and volumes. Mass represents the following: (end-diastolic epicardial−endocardial volume)×1.05. Stroke volume is end-diastolic volume minus end-systolic volume, and ejection fraction is given by the following: (stroke volume/end-diastolic volume)×100%. Wall thickness was measured on the midventricular short-axis slice at end diastole, and internal and external cavity diameters were measured on the midventricular short-axis slice at end diastole between the septum and inferolateral wall.

In addition to gross volumetric measures of systolic left ventricular function (ejection fraction and stroke volume), systolic function and cardiac rotational movement, was measured on the basis of myocardial deformation parameters assessed with TomTec 2D Cardiac Performance Analysis MR (TomTec Diogenes, Germany). The endocardial borders of the left ventricular steady-state free-precession horizontal long-axis cine and basal, mid, and apical left ventricular short-axis cines were manually contoured on the end-diastolic frame. The software then tracked the motion of related features adjacent to this endocardial line such as the cavity-tissue boundary or individual tissue patterns over the cardiac cycle to produce endocardial strain parameters[221]. The averages of segmental strains from the short-axis planes were used for endocardial circumferential strain. To maintain standardization of Cardiac MRI image analysis Dr Adam Lewandowski led the image analysis. I completed analysis on a test sample of 30 participant datasets to familiarize with analysis methods and outputs.

3.3.5 Blood Samples

All blood samples were collected from the antecubital fossa by venipuncture or indwelling venous catheter. After collecting fasting, at-rest blood samples (approximately 50 ml), participants were provided a standardized snack of 2 cereal bars. A further venous blood sample (approximately 100 ml total) was taken after exercise testing. Blood samples were centrifuged, separated within 30 minutes, and stored at -80°C. Fasting blood biochemistry was measured at the Oxford John Radcliffe Hospital Biochemistry Laboratory using routine validated assays with clinical level quality controls. Insulin resistance was calculated using the homeostasis model

assessment calculator. Additional circulating biomarkers were quantified using standard commercially available assays[143].

3.3.6 Resting Echocardiography

Resting transthoracic echocardiography was performed in the left lateral decubitus position using a commercially available Philips iE33 or Philips EPIQ 7C cardiology ultrasound machine. British Society of Echocardiography guidelines were followed for collection of a standard clinical imaging dataset to quantify left and right ventricular dimensions and systolic and diastolic function as well as review valvular function[222]. Echocardiography analysis was completed off-line in accordance with standard guidelines from the European Association of Echocardiography and the American society of Echocardiography[223-225] by a single blinded investigator to minimise bias and intra-observer variation. The Philips Xcelera 3.3 and TomTec Image Arena 4.6 software suites were utilised for image analysis. End-diastole was set at the point of mitral valve closure and end systole was set at the point of minimum LV cavity size, LV endocardial borders were manually contoured at end-diastole and end-systole to allow calculation of end diastolic volume (EDV) and end systolic volume (ESV). Resting EFs were then calculated via the bi-plane Modified Simpson's Method using apical 4-chamber and apical 2-chamber views while sub-maximal exercise EFs were calculated by the single-plane Modified Simpson's Method using apical 4-chamber views. I completed training in echocardiography image acquisition with the Oxford Cardiovascular Clinical Research Facilities (CCRF) Core Echocardiography Lab to ensure I could lead image acquisition on participant study visits. To maintain standardisation image analysis was completed by the CCRF echocardiography core lab.

3.3.7 Cardiopulmonary Exercise Testing

Participants completed resting spirometry testing prior to performing a peak cardiopulmonary exercise test on a seated stationary cycle ergometer (Ergoline GmbH, Germany) using a validated incremental protocol with respiratory gases collected and measured (Metalyzer 3B, Cortex Biophysik, Germany). Heart rate was recorded using continuous electrocardiogram monitoring, rate of perceived exertion was recorded every two minutes and blood pressure was recorded every four minutes using a manual mercury sphygmomanometer, with dynamic echocardiography imaging collected in a subgroup. After one quiescent minute of resting measurements, participants were instructed to maintain a rate of 60 revolutions per minute during the active portion of test, which begins with a two-minute warm-up with a 20-watt workload. After the warm-up period, workload was increased to 35 watts. To normalise test duration to approximately 8-12 minutes, participants who reported higher activity or fitness levels had their workload increased to 75 watts after the warm-up period. Workload automatically increment by 15 watts each minute and participants cycled continuously until safety termination criteria were met or exhaustion prevented them from maintaining at least 50 revolutions per minute. Participants then completed a two-minute cool down period at 35 watts maintain revolutions per minute of their preference. Following the cool down period, participants were transferred to a reclined treatment bed for post-exercise venous blood sampling. I completed analysis of the cardiopulmonary exercise data offline and blinded to the participant details. Breath by breath data was averaged over 15 seconds and peak exercise parameters reported as the highest averaged values over sequential 30-second periods and V-slope method applied to calculate submaximal ventilatory anaerobic threshold[226].



Figure 3-1 Demonstration of Cardiopulmonary Exercise Testing

3.3.8 Stress Echocardiography

After completing CPET testing, participants had a 30-min recovery period during which they were offered water and an additional cereal bar. Submaximal exercise stress echocardiography was then performed on study subjects for 3 min at 40%, 60%, and 80% of their peak exercise wattage achieved during CPET testing. After participants exercised at each prescribed workload for 2 min, and steady-state heart rate was verified, apical 4-chamber views oriented for optimal imaging of the LV and pulsed wave Doppler imaging of mitral inflow were collected at each stage using either a Philips iE33 or Philips EPIQ 7C cardiology ultrasound machine (Philips Healthcare,

Surrey, United Kingdom). Participants were given a 3-min rest period between each stage.

Off-line image analysis was only performed on images where heart rate was within 5% of maximum achieved steady-state heart rate for a given stage. Cardiac output indexed to body surface area (BSA) (ml/min/m^2) was calculated by multiplying heart rate in beats per minute (BPM) by stroke volume indexed to BSA, measured using echocardiography.

3.3.9 Objective Measure of Physical Activity (7 days)

A wrist worn accelerometer (Axivity AX3) similar in design to a wrist worn watch was provided for patients to wear from the end of their study visit. Wrist worn accelerometers have high compliance and reliability and are validated measures of physical activity. Participants were asked to wear the accelerometer for 7 days. Stamped addressed envelopes were provided for the return of monitoring devices. The raw accelerometer data was processed by Dr Aiden Doherty using established methods[227].

3.3.10 Ambulatory Blood Pressure (24 Hours)

24-hour ambulatory blood pressure monitoring was initiated at the end of the study visit using validated, automated oscillometric, ambulatory devices (TM-2430, A&D Instruments or equivalent). Correct cuff size was chosen based on arm circumference. Subjects were instructed to remain still during measurements. Measurements were automatically taken every 30 minutes during daytime and then hourly from 11:00PM to 7:00AM. Subjects were asked to complete a diary documenting hours asleep and

awake. Participants returned monitoring devices using prepaid envelopes, if monitors were not returned after 2 weeks participants were provided with a telephone call to ask for the devices to be returned. Compliance with returning self-monitoring devices remained high throughout the study. I validated and quality checked the ambulatory recordings and extracted data for analysis.

3.4 General Statistical Methods

Statistical analysis was undertaken using Statistical Product and Service Solutions (SPSS) Version 22 (Armonk, New York, U.S). If normally distributed, results are presented as mean ± standard deviation for continuous variables, otherwise median and interquartile range. For categorical variables, number and percentage are presented. All tests were 2-sided, P-values<0.05 were considered statistically significant with no adjustment for multiple comparison. Comparison between groups for continuous variables was performed with a 2-sided, independent-sample Student's *t* test. All multivariable analysis included residual analysis to assess model assumptions. Due to multiplicity of testing all results were considered exploratory. Results are presented as point estimates and 95% confidence intervals stated in units appropriate for the cardiac and brain measures, r^2 values are presented when indicated. Graphpad Prism 7 software was used for statistical figures and mean with 95% confidence intervals presented. The point estimate refers to the magnitude of change in the dependent variable per unit change in the independent variables. The strength of association and clinical interpretation of the point estimate will be discussed in the text. Where possible, the point estimate will be scaled in the text to a meaningful clinical unit. For example the unit change in vessel morphology per 10 mmHg of blood pressure. Given the exploratory nature of the analysis r^2 values are presented unadjusted and as an indicator of variance in the dependent variable explained by the

independent variable. The r^2 values are not include as a measure of effect size. Discussion on effect size of the observed associations will be commented on with reference to point estimates and potential clinical relevance with reference to the published literature.

3.5 Study Population Demographic

In total 129 participants underwent cardiac MRI imaging with brain MRI indices available on a total of 125 participants. The mean age of participants was 24.5±4.6 years, 64 participants were female (50%), the mean gestational age was 36.6±4.2 weeks, educational attainment was high with 88 completing University level education (68.2%), 31 participants had prior history of hypertension of which 21 were on anti-hypertension medications (16.2%) (Table 3.1).

The 52 participants with available ASL cerebral blood flow data shared a comparable demographic profile as the overall study group mean age 24.6±5.0 years, 22 females (42%), mean gestational age 37.2±3.6 weeks and 10 participants were taking anti-hypertension medications.

The 53 participants with available exercise stress echocardiography data also shared a comparable demographic profile as the overall study group mean age 24.2±5.0 years, 24 females (45%), mean gestational age 37.1±4.2 weeks, mean peak oxygen uptake 38.5±9.3 ml/min/kg and 8 participants were taking anti-hypertension medications.

Table 3.1 Study Population Demographics and Cardiovascular Risk Profile

Demographics	
Age, mean (SD), years	24.5 (4.6)
Female, n, (%)	64 (50%)
Gestational Age, mean (SD), weeks	36.6 (4.2)
Smoking, n, (%)	22 (17.0)
Smokers' median pack years (Q1-Q3)	2.7 (6.7)
Alcohol, n, (%)	97 (75.0)
Alcohol consumers' median drinks per week (Q1-Q3)	4.0 (4.0)
Hypertension Diagnosis, n, (%)	31 (24.0)
Taking Hypertension Medication, n (%)	21 (16.2)
FHx Stroke or CHD, n, (%)	10 (7.7)
Education Level	
Completed University, n, (%)	88 (68.2)
Anthropometrics	
Height, mean (SD), m	1.73 (0.1)
Weight, mean (SD), kg	70.7 (13.1)
BMI, mean (SD), kg/m^2	23.6 (3.7)
Blood pressure, mean (SD), mmHg	
Resting Systolic	121.0 (12.3)
Resting Diastolic	71.0 (9.3)
Ambulatory Awake Systolic	129.5 (12.3)
Ambulatory Awake Diastolic	76.6 (8.3)
Ambulatory Sleep Systolic	110.1 (14.1)
Ambulatory Sleep Diastolic	62.0 (8.5)
Peak Exercise Systolic	174.3 (24.6)
Peak Exercise Diastolic	87.0 (12.6)
Cardiovascular Fitness	
Peak VO_2, mean (SD), ml/kg/min	37.0 (9.4)
Peak Respiratory Exchange Ratio, mean (SD)	1.2 (0.06)
VPA, median (Q1-Q3), hours per week	0.69 (1.27)
MVPA, median (Q1-Q3), hours per week	14.9 (8.5)
Biochemistry	
Total Cholesterol, mean (SD), mmol/L	4.4 (0.75)
LDL, mean (SD), mmol/L	2.52 (0.67)
HDL, mean (SD), mmol/L	1.44 (0.29)
TChol:HDL ratio, mean (SD)	3.18 (0.86)
Triglyceride, median (Q1-Q3), mmol/L	0.87 (0.61)
Blood Glucose, mean (SD), mmol/L	4.9 (0.4)
Insulin, mean (SD), pmol/L	43.1 (30.0)
HOMA-IR, mean (SD)	0.82 (0.57)
HsCRP, median (Q1-Q3), mg/L	0.55 (1.1)

Abbreviations: FHx, Family History, BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; Peak VO_2, Peak Oxygen Uptake; VPA, Vigorous Physical Activity; MVPA, Moderate to Vigorous Physical Activity; LDL, low density lipoprotein; HDL, high density lipoprotein; T Chol: total cholesterol; HsCRP, highly sensitive C reactive protein; HOMA-IR, homeostatic model assessment of insulin resistance; WMH, white matter hyperintensity. Brain blood arrival time and cerebral blood flow data was available in 52 participants.

4 Young Adult Cerebrovascular Risk – Cardiovascular risk factors and associated impact on MRI indices of cerebrovascular health

4.1 Abstract

Risk of stroke and brain atrophy in later life relate to levels of cardiovascular risk in early adulthood. However, it is unknown whether indices of ideal cardiovascular health such as blood pressure, smoking, alcohol consumption and cardiovascular fitness relate to brain MRI indices of cerebrovascular health in young adults.

The objective of this chapter is to examine relationships between modifiable cardiovascular risk factors and cerebrovascular structure, function and white matter integrity in young adults. It will then test the hypothesis that indices of ideal cardiovascular health will be associated with brain vessel morphology, white matter hyperintensity lesion counts and cerebral blood flow in young adults.

Bivariable and multivariable associations between cardiovascular risk factors and brain MRI indices were studied. In addition, participants were categorized from 0 to 8, for the number of modifiable cardiovascular risk factors at recommended levels, based on the following criteria: BMI <25 kg/m^2; highest tertile cardiovascular fitness and/or physical activity; alcohol <8 drinks/week; non-smoker for >6 months; blood pressure on awake ambulatory monitoring <130/80 mmHg; a non-hypertensive diastolic response to exercise (peak diastolic blood pressure <90 mmHg), total cholesterol <200mg/dL; and fasting glucose <100mg/dL.

Cardiovascular risk factors were correlated with cerebral vessel density (vessels/cm^3), calibre (μm) and tortuosity, brain white matter hyperintensity lesion count (number) and volume (mm^3), and in a subgroup (n=52) brain blood arrival time (seconds) and cerebral blood flow (ml/100g/min) assessed by brain magnetic resonance imaging.

Low combined scores of modifiable cardiovascular risk factor categorized as ideal was associated with lower vessel density and vessel calibre and higher white matter hyperintensity lesions. In addition, amongst the 52 participants with available data, cerebral blood flow was lower in association with lower ideal cardiovascular health scores.

These preliminary findings provide evidence of subclinical changes in brain MRI indices of vascular health in association with modifiable cardiovascular risk factors. Further research is needed to verify these findings and determine clinical importance.

4.2 Introduction

A life-course approach to understand risk of cardiovascular disease is well established[20] and it is accepted that changes in cardiac and vascular structure that underlie this risk emerge very early in life[145,228]. Whether modifiable cardiovascular risk factors and novel early life exposures such as preterm birth, influence the early cerebrovasculature is less well studied.

Cardiovascular risk closely relates to cerebrovascular injury and cognitive decline in older adults[162]. MRI markers of cerebral injury in mid-life, including white matter hyperintensity lesions, are associated with future stroke, dementia and all-cause mortality[229]. Progression of white matter hyperintensity lesions is faster in association with metabolic dysfunction and hypertension[196]. Experimental studies have demonstrated that cardiovascular risk factors result in remodelling of the brain vasculature, including vessel rarefaction, lower vessel calibre and cerebral blood flow[230]. Elevated blood pressure, dyslipidemia and low fitness in early adulthood are known to predict brain health in older adult life[12,19,231]. Whether cerebrovascular morphological changes are already evident in young adults and correlate with white matter hyperintensity lesions and risk factors at this age, is unclear.

Advances in brain MRI allow automated segmentation and analysis of vessel morphology, white matter hyperintensity lesions[211,213] and blood flow[210]; thus making it possible to estimate brain vascular and structural status for an individual[211,213]. Therefore, the objective of the current chapter was to use multi-modality brain imaging to test the hypothesis that indices of ideal cardiovascular health will be associated with brain vessel morphology, cerebral blood flow and white matter hyperintensity lesion counts in young adults.

4.3 Statistical Analysis

Existing literature on risk predictors of brain health was used to define an a priori set of potential correlates of MRI brain health in young adults[12,44,53,162,164]. These were grouped as: 1) non-modifiable, including age, sex, gestational age, and 2) modifiable, including systolic blood pressure, body mass index (BMI), peak exercise capacity (oxygen uptake ml/min/kg), peak exercise diastolic blood pressure, weekly vigorous activity level, alcohol consumption, smoking history, lipid profile, glucose and insulin resistance, and current hypertension medication.

In a priori analysis, bivariable and multivariable analysis was completed to investigate correlation between the defined cardiovascular risk markers, brain vessel morphology and indices of vascular health including; white matter hyperintensity lesions and cerebral perfusion. In multivariable models, to reduce multiple testing and potential interactions between the variables, the model was restricted to an a priori subset of variables: resting systolic blood pressure, BMI, peak oxygen uptake as a measure of cardiovascular fitness, alcohol consumption and smoking. These variables were selected as modifiable risk factors that could be readily assessed within a clinical environment and potentially in a primary care setting. This model was adjusted for non-modifiable factors including age, sex and gestational age. In addition to the a priori model, an explorative stepwise regression was completed as a comparative analysis to identify independent modifiable risk factors related to brain MRI indices. Cardiovascular risk variables were included in the stepwise regression analysis if they were identified in bivariable analysis to have an association with brain MRI indices with p value of 0.1 or less. These models were completed as a forced entry regression analysis adjusted for age, sex and gestational age at birth.

In post hoc analysis, the individuals' combined cardiovascular score from across 8 risk factors, was used as a metric of overall modifiable cardiovascular health. The relationships between the individuals' modifiable cardiovascular score and brain imaging findings were studied using linear regression adjusted for age and sex. Comparison between brain imaging findings was made between groups of participants in the lowest and highest tertiles for the cardiovascular score.

4.4 Demographic profile and distribution of Brain MRI vascular indices: Vessel Morphology, White Matter Hyperintensity Lesion Burden, and Cerebral Perfusion

Table 4.1 Brain MRI indices of vessel morphology, white matter hyperintensity lesion burden and cerebral perfusion

Brain MRI Vessel, Perfusion and White Matter Parameters	
Vessel density, mean (SD), vessels/cm^3	8.3 (1.41)
Vessel calibre, mean (SD), μm	531 (36)
Vessel tortuosity, mean (SD)	1.49 (0.088)
WMH total lesion count, mean (SD)	20.9 (7.9)
WMH total lesion volume, mm^3 mean (SD)	622.1 (272.8)
WMH periventricular lesion count, mean (SD)	13.3 (6.9)
WMH periventricular lesion volume, mm^3 mean (SD)	446.6 (228.7)
WMH deep lesion count, mean (SD)	9.1 (5.8)
WMH deep lesion volume, mm^3 mean (SD)	173.4 (166.6)
Brain Blood Arrival Time (SD), seconds	1.01 (0.08)
Cerebral Blood Flow (SD), ml/100g/min	60 (11.5)

Table presents mean values and standard deviation. Abbreviations: WMH, white matter hyperintensity. Brain blood arrival time and cerebral blood flow data was available in 52 participants

A total of 125 participants completed the brain MRI protocol and cardiovascular risk assessment measures. The demographic and cardiovascular risk profiles of the study population are presented (Table 3.1).The mean values and standard deviation for MRI indices of vessel morphology, white matter hyperintensities and cerebral perfusion are presented in Table 4.1.

The distributions of MRI brain outcomes between lowest and highest quintile of the respective measures are presented in Table 4.2.

Table 4.2 Quintile distribution of brain vascular indices and mean difference between top and bottom quintile

Brain Measure	Quintile 1 Mean (SD)	Quintile 5 Mean (SD)	Mean Difference between 1st and 5th quintile of respective measure Mean ((95%CI)	Relative percentage difference between 1st and 5th quintile of respective measure
Brain Vessel Density, vessels/cm^3	6.6 (0.62)	10.5 (0.95)	3.8 (3.3 to 4.4)	37% lower vessel density
Brain Vessel Calibre, µm	504 (56)	587 (47)	82 (61 to 103)	14% lower vessel calibre
Brain Vessel Tortuosity	1.40 (0.02)	1.64 (0.05)	0.241 (0.22 to 0.27)	15% lower tortuosity
Brain white matter hyperintensity lesion count, number	34.5 (4.5)	11 (2)	23 (21 to 25)	3 fold higher in first quintile
Brain Blood Arrival Time, seconds	1.31 (0.03)	1.08 (0.02)	0.22 (0.2 to 0.25)	20% slower arrival time in the first quintile
Cerebral Blood Flow, ml/min/100g	46 (4.7)	78 (8)	32 (25 to 38)	41% lower cerebral blood flow in the first quintile

The 52 participants with available cerebral blow flow data shared a comparable demographic profile as the overall study group (mean age 24.6 ±5.0 years, 42% female (n=22), gestational age 37.2±3.6 weeks, and 10 participants were on anti-hypertension medications (19.2%).

4.5 Results

4.5.1 Association between non-modifiable and modifiable risk factors and Brain MRI Indices of Vessel Morphology

Table 4.3 Associations between cardiovascular risk factors and MRI indices of brain vessel morphology

	Vessel Density (vessels/cm^3)		**Vessel Calibre (µm)**		**Vessel Tortuosity**	
	Point Estimate (95 %CI)	P value	Point Estimate (95 %CI)	P value	Point Estimate (95 %CI)	P value
Gestational Age, weeks	-0.001 (-0.06 to 0.06)	.98	-0.1 (-2.0 to 1.0)	.88	***0.005 (0.001 to 0.009)***	***.007***
SBP, mmHg	***-0.03 (-0.05 to -0.004)***	***.02***	-0.4 (-1.0 to 2.0)	.15	-0.00003 (-0.001 to 0.001)	.97
DBP, mmHg	-0.015 (-0.04 to 0.013)	.31	-0.4 (-1.0 to 0.3)	.26	-0.000025 (-0.002 to 0.002)	.98
BMI, kg/m^2	**-0.10 (-0.16 to -0.02)**	**.01**	-1.0 (-3.0 to 1.0)	.33	-0.001 (-0.004 to 0.005)	.75
VPA, hours per week	0.10 (-0.17 to 0.39)	.42	1.0 (-6.0 to 8.0)	.73	-0.002 (-0.015 to 0.02)	.81
Alcohol, drinks per week	-0.10 (-0.025 to -0.008)	.31	-0.1 (-1.0 to 1.0)	.70	-0.001 (-0.002 to 0.003)	.61
Smoking, pack year	***0.20 (0.06 to 0.30)***	***.004***	3.0 (-0.2 to 6.0)	.06	-0.005 (-0.13 to 0.002)	.15
Peak Vo2, ml/kg/min	0.01 (-0.02 to 0.04)	.5	0.4 (-0.2 to 1.0)	.19	0.001 (-0.001 to 0.003)	.16
Peak Ex DBP, mmHg	***-0.02 0.04 to -0.003)***	***(-.047***	***-1.0 (-1.4 to -0.4)***	***<.001***	-0.001 (-0.001 to 0.002)	.36
Cholesterol, mmol/L	-0.3 (-1.0 to 0.05)	.09	-6.0 (-16 to 3)	.18	0.01 (-0.013 to 0.033)	.39
Cholesterol/HDL,	***-0.40 (-0.69 to -0.06)***	***.02***	-3.0 (-10.0 to 5.0)	.52	0.009 (-0.01 to 0.03)	.37
Glucose, mmol	-0.2 (-0.8 to 0.4)	.55	-8.0 (-25.0 to 8.0)	.31	0.003 (-0.037 to 0.044)	.87
Insulin, pmol/L	-0.01 (-0.02 to 0.0004)	.06	-0.2 (-0.4 to 0.02)	.07	0.0002 (-0.0004 to 0.001)	.53
Hypertension treatment	0.75 (-0.01 to 1.5)	.05	10 (-9.0 to 31.0)	.27	-0.004 (-0.05 to 0.04)	.87
Nocturnal SBP, mmHg	***-0.03 (-0.05 to -0.013)***	***.001***	-0.4 (-1.0 to 0.1)	.14	0.00035 (-0.001 to 0.002)	.58
Nocturnal DBP, mmHg	***-0.04 (-0.07 to -0.007)***	***.016***	-0.5 (-1.3 to 0.3)	.22	0.001 (-0.001 to 0.003)	.27

Abbreviations and units: SBP, systolic blood pressure (mmHg); BMI, body mass index (kg/m^2); VPA, Vigorous Physical Activity (hours per week); Alcohol (units); Smoking (pack years); Peak VO_2, Peak Oxygen Uptake (ml/kg/min); Ex DBP, Peak exercise diastolic blood pressure (mmHg), Cholesterol/HDL ratio, ratio total cholesterol/high density lipoprotein; HOMA-IR, homeostatic model assessment of insulin resistance, Hypertension treatment, prescription medications for hypertension (yes/no)**.** The point estimate refers to the magnitude of change in the vessel morphology variable per unit change in the non-modifiable and modifiable variables.

Table 4.4 A priori multivariable model of associations between cardiovascular risk factors and MRI indices of vessel morphology

	Brain Vessel Density (vessels/cm^3)		**Brain Vessel Calibre (µm)**		**Brain Vessel Tortuosity**	
	Point Estimate (95 %CI)	**P value**	**Point Estimate (95 %CI)**	**P value**	**Point Estimate (95 %CI)**	**P value**
Gestational Age, weeks	-0.02 (-0.09 to 0.05)	.51	-1.0 (-3.0 to 1.0)	.25	***0.006 (0.001 to 0.01)***	***.016***
SBP, mmHg	***-0.026 (-0.05 to -0.002)***	***.035***	***-0.67 (-1.0 to -0.05)***	***.03***	0.0003 (-0.001 to 0.002)	.66
BMI, kg/m^2	***-0.11 (-0.19 to -0.03)***	***.01***	-1.0 (-3.0 to 1.0)	.20	-0.0006 (-0.005 to 0.006)	.81
Alcohol, drinks per week	-0.03 (-0.9 to 0.04)	.40	-1.0 (-2.0 to 0.1)	.50	-0.001 (-0.006 to 0.003)	.53
Smoking, pack years	***0.22 (0.09 to 0.36)***	***.002***	3.0 (-1 to 6.0)	.12	-0.008 (-0.017 to 0.001)	.09
Peak Vo2, ml/kg/min	-0.012 (-0.05 to 0.02)	.51	-0.26 (-1.0 to 1.0)	.58	0.001 (-0.002 to 0.003)	.61
Model Statistics	**R^2=0.20 p =.003**		**R^2=0.17 p=.015**		R^2=0.10 p=.22	

Abbreviations and units: SBP, systolic blood pressure (mmHg); BMI, body mass index (kg/m^2); Alcohol (drinks per week); Smoking (pack years); Peak VO_2, Peak Oxygen Uptake (ml/kg/min); Model adjusted for age, sex and gestational age at birth. The point estimate refers to the magnitude of change in the vessel morphology variable per unit change in the non-modifiable and modifiable variables. The point estimate refers to the magnitude of change in the vessel morphology variable per unit change in the non-modifiable and modifiable variables.

Association between risk factors (clinic and ambulatory blood pressure, BMI, smoking pack years, exercise diastolic blood pressure, lipid profile, hypertension treatment) and brain vessel density and vessel calibre are presented in Table 4.3. In the a priori multivariable cardiovascular risk models; systolic blood pressure (-0.26 vessels/cm^3 per 10mmHg, 95%CI -0.5 to -0.02, p=0.04), smoking (2.2 vessels/cm^3 per 10 pack years, 95%CI 0.9 to 3.6, p=0.002) and body mass index (-0.11 vessels/cm^3 per 1kg/m^2, 95%CI -0.19 to -0.03, p=0.01) were independently associated with vessel

density, while vessel calibre only correlated with systolic blood pressure (-6.7μm per 10mmHg, 95%CI -10.0 to -0.5, p=0.03) (Table 4.4). Vessel tortuosity only varied with gestational age in both bivariable and adjusted models (0.005 unit tortuosity change/gestational week, 95%CI 0.001 to 0.009, p=0.007) (Table 4.3, Table 4.4).

Table 4.5 Stepwise multivariable model of associations between cardiovascular risk factors and brain vessel density

	Brain Vessel Density (vessels/cm^3)	
	Point Estimate (95 %CI)	**P value**
Nocturnal MAP, mmHg	-0.06 (-0.08 to -0.03), r^2=0.15	**.00003**
Smoking, pack years	0.22 (0.09 to 0.34), r^2=0.09	**.0007**
Cholesterol HDL Ratio	-0.44 (-0.7 to -0.1), r^2=0.06	**.0047**
Hypertension Treatment	1.0 (0.2 to 2.0), r^2=0.05	**.014**
Model Statistics	**R^2=0.33 p =.000006**	

Abbreviations and units: MAP, mean arterial blood pressure (mmHg); Smoking (pack years); Cholesterol/HDL ratio, total ratio cholesterol/high density lipoprotein; Hypertension treatment, participant taking prescription medications for hypertension (yes/no). Adjusted for age, sex and gestational age. The point estimate refers to the magnitude of change in the vessel morphology variable per unit change in the non-modifiable and modifiable variables.

In stepwise multivariable regression built from the observed bivariable associations (Table 4.3) and adjusted for age, sex and gestational age; nocturnal mean arterial blood pressure (-0.6 vessels/cm^3 per 10mmHg, 95%CI -0.8 to -0.3, r^2=0.15, p=0.00003), smoking (2.2 vessels/cm^3 per 10 pack years, 95%CI 0.9 to 3.4, p=0.0007) and cholesterol HDL ratio (-0.44 vessels/cm^3 per unit, 95%CI -0.7 to -0.1, p=0.0047) and use of hypertension treatment (1.0 vessels/cm^3 95%CI 0.2 to 2.0, p=0.014) were independently associated with vessel density (Table 4.5).

In stepwise multivariable analysis vessel calibre was only correlated with exercise diastolic blood pressure (-9.7μm per 10mmHg, 95%CI -14.0 to -4.0, p=0.0002), adjusted for age, sex and gestational age.

4.5.2 Association between non-modifiable and modifiable risk factors and Brain MRI indices of white matter hyperintensity lesion count and volume

The total number and volume of white matter hyperintensity lesions in young adults are associated with measures of blood pressure and smoking and alcohol behaviours. Bivariable analysis identified smoking to be associated with 8 lesions per 10 pack years (95%CI 0.15 to 14.4, p=0.02) and lesion volumes of 32 mm^3 per pack year (95%CI 10.2 to 52.7, p=0.004). Alcohol consumption was associated with 4 lesions per 10 weekly alcoholic drinks (95%CI 0.3 to 8.0, p=0.03) and lesion volumes of 127.0 mm^3 per 10 weekly alcoholic drinks (95%CI 8.2 to 247.0, p=0.04). Nocturnal systolic blood pressure was associated with 1.3 lesions per 10 mmHg (95%CI 0.2 to 2.4, p=0.02) and lesion volumes of 46 mm^3 per 10 mmHg (95%CI 10.2 to 81.1, p=0.01). Nocturnal diastolic blood pressure was associated with 2.6 lesions per 10mmHg (95%CI 1.0 to 4.3, p=0.002), and lesion volumes of 74 mm^3 per 10 mmHg (95%CI 18.1 to 130, p=0.01). Exercise diastolic blood pressure was also associated with lesion number in bivariable analysis 1.3 lesions per 10mmHg, (95%CI 0.07 to 2.4, p=0.04) (Table 4.6). In regional bivariable analysis, the associations between periventricular lesion count and volume drives the global white matter hyperintensity associations with cardiovascular risk factors. There were no significant associations between deep white matter lesion count and volume and cardiovascular risk factors.

Table 4.6 Associations between cardiovascular risk factors and MRI indices of total white matter hyperintensity lesion count and volume

Total brain white matter hyperintensity lesion count	Point Estimate (95 %CI)	P value
Gestational Age, per week	-0.22 (-0.56 to 0.12)	.21
SBP, per mmHg	0.07 (-0.05 to 0.20)	.26
DBP, per mmHg	0.1 (-0.05 to 0.26)	.18
BMI, per unit kg/m^2	-0.04 (-0.44 to 0.36)	.84
VPA, per hours per week	-0.07 (-1.60 to 1.50)	.93
Alcohol, per drink per week	***0.42 (0.03 to 0.80)***	***.034***
Smoking, per pack year	***0.79 (.15 to 1.44)***	***.017***
Peak Vo2, per ml/kg/min	-0.05 (-0.20 to 0.11)	.54
Peak Ex DBP, per mmHg	***0.125 (.007 to 0.244)***	***.038***
Cholesterol, mmol/L	-0.05 (-2.1 to 2.0)	.96
Cholesterol/HDL Ratio, per unit	-0.21 (-1.98 to 1.56)	.82
Glucose, per mmol	-2.8 (-6.27 to 0.73)	.12
Hypertension treatment	2.26 (-1.6 to 6.2)	.25
Nocturnal SBP, per mmHg	***0.13 (0.02 to 0.24)***	***.018***
Nocturnal DBP, per mmHg	***0.26 (0.095 to 0.43)***	***.002***
Total brain white matter hyperintensity lesion volume	**Point Estimate (95 %CI)**	**P value**
Gestational Age, per week	6.26 (-5.1 to 17.6)	.28
SBP, per mmHg	2.25 (-1.9 to 6.4)	.29
DBP, per mmHg	3.76 (-1.3 to 8.8)	.14
BMI, per unit kg/m^2	2.0 (-11.2 to 15.3)	.76
VPA, per hours per week	-19.8 (-70.5 to 31.0)	.44
Alcohol, per drink per week	***12.7 (0.82 to 24.7)***	***.036***
Smoking, per pack year	***31.5 (10.2 to 52.7)***	***.004***
Peak Vo2, per ml/kg/min	0.51 (-4.6 to 5.6)	.84
Peak Ex DBP, per mmHg	-0.06 (-4.1 to 3.9)	.98
Cholesterol, mmol/L	8.2 (-59.3 to 75.7)	.81
Cholesterol/HDL Ratio, per unit	9.5 (-49.3 to 68.3)	.75
Glucose, per mmol	-59.6 (-188.1 to 68.8)	.36
Hypertension treatment	69.9 (-59.8 to 199.5)	.29
Nocturnal SBP, per mmHg	***4.6 (1.02 to 8.11)***	***.012***
Nocturnal DBP, per mmHg	***7.4 (1.81 to 13.1)***	***.01***

Abbreviations and units: SBP, systolic blood pressure (mmHg); BMI, body mass index (kg/m^2); VPA, Vigorous Physical Activity (hours per week); Alcohol; Smoking (pack years); Peak VO_2, Peak Oxygen Uptake (ml/kg/min); Ex DBP, Peak exercise diastolic blood pressure (mmHg), Cholesterol/HDL ratio, ratio total cholesterol/high density lipoprotein; HOMA-IR, homeostatic model assessment of insulin resistance, Hypertension treatment, taking prescription medications for hypertension (yes/no)**.** The point estimate refers to the magnitude of change in the vessel morphology variable per unit change in the non-modifiable and modifiable variables.

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Table 4.7 Associations between cardiovascular risk factors and MRI indices of total white matter hyperintensity lesion count and volume in a priori multivariable models

	Total WMHL Count		**Total WMHL volume (ml)**	
	Point Estimate (95 %CI)	P value	Point Estimate (95 %CI)	P value
Gestational Age, weeks	-0.25 (-0.61 to 0.11) r^2=0.014	.17	3.8 (-8.2 to 15.8) r^2=0.003	.53
SBP, mmHg	0.051 (-0.09 to 0.2) r^2=0.004	.46	0.65 (-3.9 to 5.2) r^2=0.001	.78
BMI, kg/m^2	-0.24 (-0.68 to 0.21) r^2=0.008	.29	-5.9 (-20.8 to 9.1) r^2=0.005	.44
Alcohol, drinks per week	***0.39 (0.01 to 0.77) r^2=0.032***	***.04***	10.7 (-2.1 to 23.5) r^2=0.02	.10
Smoking, pack years	***0.96 (0.25 to 1.66) r^2=0.06***	***.009***	***27.6 (3.9 to 51.3) r^2=0.04***	***.023***
Peak Vo2, ml/kg/min	-0.008 (-0.21 to 0.19) r^2=0.00005	.94	-0.79 (-7.5 to 5.9) r^2=0.0004	.81
Model Statistics	Model Statistics r^2=0.12 p=.06		Model Statistics r^2=0.10 p=.12	

A priori multivariable models of associations between modifiable cardiovascular risk factors and MRI indices of white matter hyperintensity, adjusted for age, sex, and gestational age at birth. The point estimate refers to the magnitude of change in the vessel morphology variable per unit change in the non-modifiable and modifiable variables.

In multivariable analysis, smoking and alcohol consumption remained independently associated with total white matter hyperintensity lesion count explaining between 3% and 6% of the lesion burden (Table 4.7, Table 4.8). Smoking history was also independently associated with lesion volume in multivariable analysis explaining 4% of the lesion volume. Nocturnal blood pressure, calculated as the mean arterial pressure, was independently associated with both total white matter hyperintensity lesion count and lesion volume (2.0 lesions per 10 mmHg 95%CI 0.5 to 3.5, p=0.01, and 56.0 mm^3 lesion volume per 10 mmHg 95%CI 41.0 to 108.0, p=0.03) (Table 4.8). Overall, the a priori cardiovascular risk models provided weak predictive models for white matter hyperintensity characteristics (Table 4.7). The stepwise cardiovascular risk models for white matter hyperintensity lesion count and volume maintained

stronger multivariable model statistics than the a priori model and explained 16% of variance in lesion count (r^2=0.16, p=0.005) and 13% of the variance in lesion volume (r^2=0.13, p=0.014) (Table 4.8) adjusted for age, sex and gestational age at birth.

Table 4.8 Independent cardiovascular risk factors associated with MRI indices of total white matter hyperintensity lesion count and volume following multivariable stepwise regression

	Total WMHL count		**Total WMHL volume (ml)**	
	Point Estimate (95 %CI)	**P value**	**Point Estimate (95 %CI)**	**P value**
Nocturnal MAP, mmHg	***0.20 (0.05 to 0.35), r^2=0.06***	***.01***	***5.6 (0.41 to 10.8), r^2=0.04***	***.033***
Smoking, pack years	***0.78 (0.07 to 1.5), r^2=0.04***	***.016***	***25.3 (0.60 to 50.0), r^2=0.04***	***.038***
Alcohol, drinks per week	***0.39 (0.03 to 0.78), r^2=0.04***	***.048***	.	**.**
Model Statistics	**Model Statistics r^2=0.16 p =.005**		**Model Statistics r^2=0.13 p=.014**	

Multivariable model built with stepwise regression, included variables identified in bivariable analysis with p value < 0.1 in Table 4.6, and retained in model if p value < 0.1. Model adjusted for age, sex, and gestational age at birth. MAP; mean arterial blood pressure. The point estimate refers to the magnitude of change in the vessel morphology variable per unit change in the non-modifiable and modifiable variables.

4.5.3 Association between non-modifiable and modifiable risk factors and brain MRI indices of cerebral perfusion

To explore whether cerebral blood flow varied with cardiovascular risk factors, a subgroup (n=52) analysis was performed in those with cerebral blood flow measures The mean cerebral blood flow was 60 ml/100g/min (SD 11.5) and the mean blood arrival time 1.01 seconds (SD 0.08). Higher body mass index correlated with slower blood arrival time 0.01 seconds per 1kg/m^2 (95%CI 0.001 to 0.05, p=0.001) and lower cerebral blood flow -1.1 ml/100g/min per 1kg/m^2 (95%CI -2.0 to -0.1, p=0.03). Cerebral blood flow was also lower in correlation with anti-hypertensive medication 11

ml/100g/min (95%CI -18.4 to -3.2, p=0.007) (Table 4.9). Cerebral blood flow was also lower in association with elevated nocturnal diastolic blood pressure 6ml/100g/min per 10 mmHg (95%CI 3 to 10, p=0.001) (Table 4.9). Body mass index remained independently associated with blood arrival time in multivariable analysis. In the a priori cardiovascular risk model BMI was associated with 0.013 seconds slower arrival time per unit increase in body mass index (95% CI 0.006 to 0.02, r^2=0.17, p<0.001). The relative variance in blood arrival time explained by body mass index remained similar in stepwise multivariable analysis (0.008 seconds per unit increase body mass index 95% CI 0.003 to 0.012, r^2=0.14, p=0.002). BMI was the only risk factor independently associated with blood arrival time. Overall, the a priori cardiovascular risk model explained up to 48% of the variance in brain blood arrival time (r^2=0.48, p=<.001).

Table 4.9 Associations between cardiovascular risk factors and MRI indices of cerebral perfusion

Blood Arrival Time (n=52)	Point Estimate (95 %CI)	P value
Gestational Age, per week	0.003 (-0.003 to 0.01)	.27
SBP, per mmHg	0.001(-0.01 to 0.03)	.14
DBP, per mmHg	-0.001 (-0.003 to 0.002)	.54
BMI, per unit kg/m^2	***0.011 (0.001 to 0.05)***	***.001***
VPA, per hours per week	0.001(-0.023 to 0.026)	.91
Alcohol, per drink per week	-0.00015 (-0.0015 to 0.0015)	.88
Smoking, per pack year	-0.001 (-0.011 to 0.009)	.84
Peak Vo2, per ml/kg/min	0.002 (-0.001 to 0.004)	.17
Peak Ex DBP, per mmHg	-0.001 (-0.003 to 0.001)	.48
Cholesterol, mmol/L	0.002 (-0.03 to 0.03)	.89
Cholesterol/HDL Ratio, per unit	0.02 (-0.005 to 0.045)	.11
Glucose, per mmol	0.01 (-0.04 to 0.06)	.70
Hypertension treatment	-0.02 (-0.08 to 0.04)	.46
Nocturnal SBP, per mmHg	0.002 (-0.0002 to 0.003)	.08
Nocturnal DBP, per mmHg	0.001 (-0.002 to 0.004)	.46
Cerebral Blood Flow (n=52)	**Point Estimate (95 %CI)**	**P value**
Gestational Age, per week	-0.441 (-1.3 to 0.45)	.32
SBP, per mmHg	-0.213 (-0.484 to 0.082)	.16
DBP, per mmHg	-0.26 (-0.60 to 0.07)	.12
BMI, per unit kg/m^2	***-1.06 (-0.1 to -2.01)***	***.03***
VPA, per hours per week	1.52(-2.02 to 5.06)	.86
Alcohol, per drink per week	0.01 (-0.23 to 0.25)	.93
Smoking, per pack year	-0.395 (-1.8 to 1.01)	.58
Peak Vo2, per ml/kg/min	0.065 (.27 to 0.4)	.70
Peak Ex DBP, per mmHg	-0.178 (-0.48 to 0.13)	.24
Cholesterol, mmol/L	-2.87 (-7.2 to 1.5)	.19
Cholesterol/HDL Ratio, per unit	-2.15 (-5.8 to 1.5)	.24
Glucose, per mmol	0.04 (-7.7 to 7.8)	.99
Hypertension treatment	***-10.8 (-3.2 to 18.4)***	***.007***
Nocturnal SBP, per mmHg	-0.23 (-0.47 to 0.01)	.06
Nocturnal DBP, per mmHg	***-0.62 (-0.97 to -0.27)***	***.001***

Abbreviations and units: SBP, systolic blood pressure (mmHg); BMI, body mass index (kg/m^2); VPA, Vigorous Physical Activity (hours per week); Alcohol (drinks per week); Smoking (pack years); Peak VO_2, Peak Oxygen Uptake (ml/kg/min); Ex DBP, Peak exercise diastolic blood pressure (mmHg), Cholesterol/HDL ratio, ratio total cholesterol/high density lipoprotein; HOMA-IR, homeostatic model assessment of insulin resistance, Hypertension treatment, taking prescription medications for hypertension (yes/no). The point estimate refers to the magnitude of change in the perfusion variable per unit change in the non-modifiable and modifiable variables.

In multivariable analysis, only nocturnal mean arterial blood pressure, remained independently associated with cerebral blood flow 5.3ml/100g/min lower per 10 mmHg (95%CI 2.0 to 8.7, r^2=0.20, p=0.002) adjusted for age, sex and gestational age.

Overall, the stepwise model explained 24% of the variance in cerebral blood flow (r^2=0.24 p=0.026) with the model including the variables age, sex and gestational age at birth in addition to nocturnal blood pressure. The a priori cardiovascular risk model did not explain variance in cerebral blood flow (r^2=0.15 p=0.51).

4.5.4 Association between optimal cardiovascular health scores and MRI indices of cardiovascular risk

The cardiovascular score provided a cumulative score for each of the following factors: high cardiovascular fitness scored as top tertile of peak oxygen uptake (>=110% predicted peak oxygen uptake) or participating in >=75 minutes vigorous physical activity per week; not smoking in last 6 months; alcohol <8 drinks/week; ambulatory awake blood pressure <130/80mmHg; body mass index <25kg/m^2; fasting total cholesterol <200 mg/d; fasting blood glucose <100 mg/dL; and diastolic blood pressure at peak exercise <= 90mmHg.

Healthier categories on the modifiable cardiovascular score correlated with vessel morphology. Each additional healthier category of risk factor was associated with a 0.3 vessels/cm^3 higher vessel density (95%CI 0.1 to 0.5, p=0.003) and 8μm greater vessel calibre (95%CI 3.0 to 13.0, p=0.01). Similarly, white matter hyperintensity lesion count correlated with the cardiovascular score with 1.6 fewer white matter hyperintensity lesions per additional healthier category of risk factor (95%CI to -3.0 to -0.5, p=0.006). In addition, the cardiovascular score correlated with total volume of white matter hyperintensity adjusted for brain size, with 51 mm^3 lower white matter hyperintensity lesion volume per additional healthier category of risk factor (95%CI to -87 to -15 mm^3 p=0.006). Differences in vessel morphology and white matter

hyperintensity lesions between tertiles of the study group, divided based on the cardiovascular score, are presented in Figure 1. Cerebral blood flow was 2.5ml/100g/min higher for each additional healthier category of the cardiovascular score (95%CI 0.16 to 4.89, p=0.03). There was no significant correlation between blood arrival time and the cardiovascular scores.

Healthier categories on the modifiable cardiovascular score correlated with vessel morphology. Each additional healthier category of risk factor was associated with a 0.3 vessels/cm^3 higher vessel density (95%CI 0.1 to 0.5, p=0.003) and 8μm greater vessel calibre (95%CI 3.0 to 13.0, p=0.01). Similarly, white matter hyperintensity lesion count correlated with the cardiovascular score with 1.6 fewer white matter hyperintensity lesions per additional healthier category of risk factor (95%CI to -3.0 to -0.5, p=0.006). In addition, the cardiovascular score correlated with total volume of white matter hyperintensity adjusted for brain size with 51 mm^3 lower white matter hyperintensity lesion volume per additional healthier category of risk factor (95%CI to -87 to -15 mm^3 p=0.006). Differences in vessel morphology and white matter hyperintensity lesions between tertiles of the study group, divided based on the cardiovascular score, are presented in Figure 1. Cerebral blood flow was 2.5ml/100g/min higher for each additional healthier category of the cardiovascular score (95%CI 0.16 to 4.89, p=0.03). There was no significant correlation between blood arrival time and the cardiovascular scores.

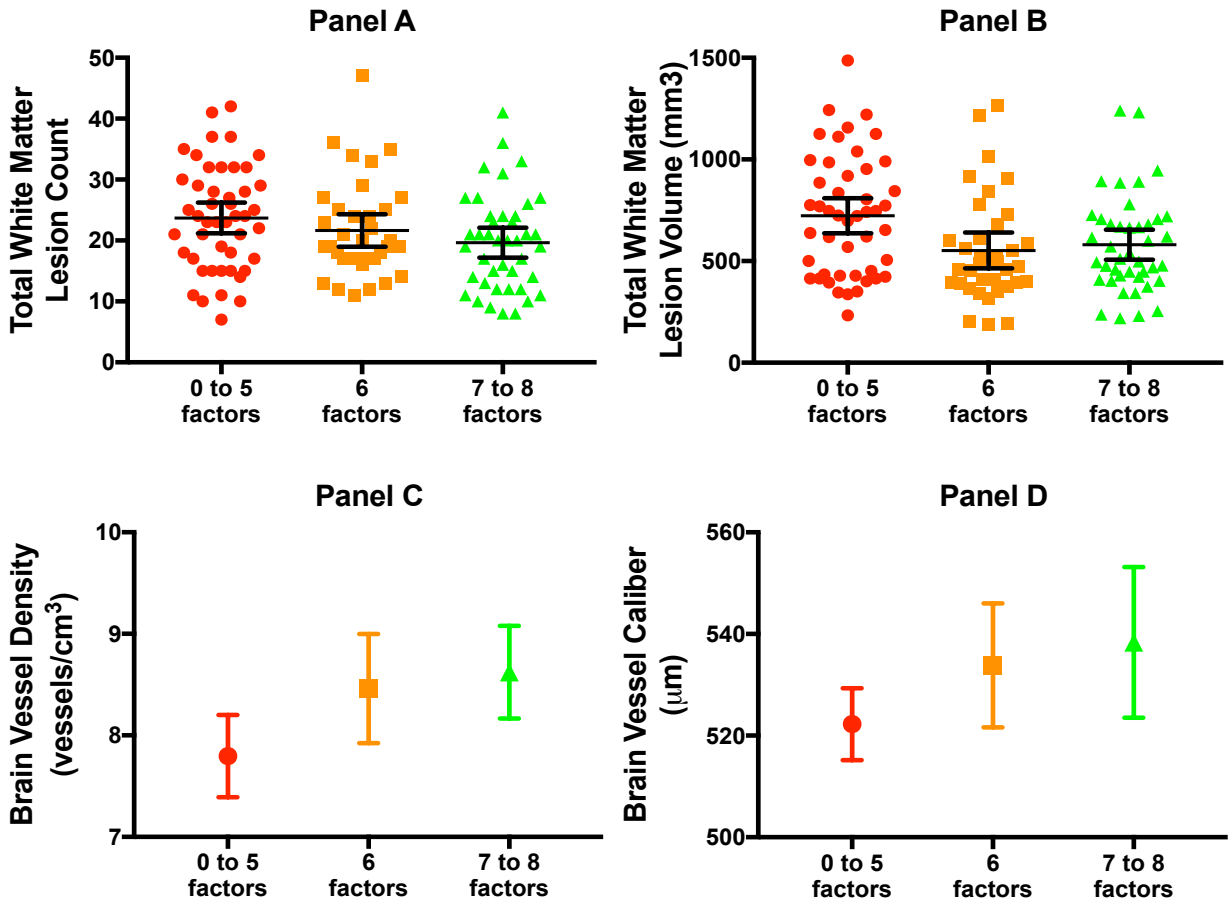


Figure 4-1 Comparison of white matter lesion burden and vessel morphology between groups of participants based on their modifiable cardiovascular score.

Panel A presents the white matter lesions counts and panel B presents the white matter hyperintensity lesion volumes for individual participants and associated group mean and 95% CI, Panels B and C present the mean group values and 95% CI for brain vessel density and brain vessel calibre.

Figure 4-1 presents a post hoc comparison between groups of participants who score 0 to 5 positive factors (n=47), 6 factors (n=36) and 7 to 8 positive factors (n=42). The groupings were defined to approximate tertiles of the combined cardiovascular score. Adjusting for age and sex, participants with 7 to 8 healthier categories of risk factor have a mean vessel density 11% higher than participants with 0 to 5 healthier categories of risk factor (Panel C, 8.6 vessels/cm^3 (SD 1.39) vs 7.8 vessels/cm^3 (SD

1.21) p=0.007), a mean vessel calibre 3% higher (Panel D, 538μm (SD 21) vs 522μm (SD 45) p=0.02) and on average 20% lower white matter hyperintensity lesion counts (Panel A, 19.6 lesions (SD 7.8) vs 23.5 lesions (SD 8.6) p=0.03) and 23% lower volume of white matter hyperintensity lesion volumes (Panel B, 581.0 mm^3 SD (238.0) vs 715.0 mm^3 (SD 291.0) p=0.04)

4.6 Discussion

4.6.1 MRI indicators of cerebrovascular risk in young adults

Modifiable risk exposures during early to mid-life are major drivers for cerebrovascular disease and dementia[21,205]. This has been demonstrated in the Framingham cohorts where mid-life ideal cardiovascular health was the strongest predictor of future stroke and dementia[162]. Similar associations are observed between modifiable risk factors and MRI indices of brain vascular health such as cerebral perfusion and white matter hyperintensity lesions in older adults[163,232]. This is the first study to report on evidence of brain vascular changes in young adults in relation to modifiable risk factors. The study supports the argument for early to mid-life promotion of vascular risk reduction to protect future brain health and prevent future dementia[1,50]. Barriers to tailored primary prevention in young adults include the remoteness of the hard endpoints and lack of sensitive prediction tools for brain health. There is interest to identify early biomarkers that are on pathological pathways impacting future brain health. A primary focus of this activity is to drive drug discovery and provide early outcome markers for pharmacological intervention. However, if the same approaches were used to frame the interactions of modifiable risk factors directly on structural and function indices of brain health then this offers the potential to provide sensitive biomarkers for prevention. It would then be possible to drive improvement in prevention intervention

and directly quantify physical effects decades in advance of clinical symptoms. In this cross-sectional analysis, the observed associations with measures of vessel morphology, white matter hyperintensity lesion and cerebral blood flow confirms that MRI may offer sensitive tools and new biomarkers to evaluate brain vascular health in young adults. Therefore, these preliminary findings may provide a platform to drive innovation in cardiovascular prevention research to promote and protect brain health in young adults.

4.6.2 Independent associations between modifiable risk factors and indices of cerebrovascular risk

Modifiable risk factors such as blood pressure, BMI, smoking, alcohol consumption and lipid profile are known to drive systemic vascular disease in young adults in part through biological vascular disorders including endothelial dysfunction and oxidative stress[233-235]. The current study suggests the cerebrovasculature may be similarly affected. In this young adult study, the observed patterns of potentially modifiable associations group across areas of cardiovascular and metabolic health including: blood pressure, BMI, lipid profile and behavioural risk factors of smoking exposure and alcohol consumption. This is the first study to provide evidence that smoking and alcohol consumption are associated with white matter hyperintensity lesions in young adults. It also suggests potential of vascular remodelling in response to environmental stressors, with higher vessel density associated with smoking. The higher vessel density may be an adaptive response to tissue hypoxia, which may also be a contributor to the observed association between smoking and white matter hyperintensities lesion characteristics. Smoking and alcohol are established targets

for primary prevention efforts in adolescent and young adult populations. This preliminary evidence reinforces the importance of continuing those strategies.

Overall, the United Kingdom and Ireland score poorly on modifiable indices of ideal cardiovascular health, with some of the worst figures in Europe. The findings from the current study supports the prediction that we may be on the same trajectory as the USA which has witnessed 3-fold increase in cerebrovascular events in the under 55 population in the last 20 years[28].

4.6.3 Potential cerebrovascular disease burden from these findings in young adults

To date, studies tracking changes in brain vascular measures have largely focused on the transition from middle age to older adulthood. Cerebral blood flow is estimated to decline over the life course[10] with risk of dementia in older adults being 2 to 3 fold higher in those whose cerebral blood flow is below 55 ml/100g/min[232]. Vascular dementia has also been associated with lower vascular density in brains of adults who have an early diagnosis of disease[236]. In the current study, young adults in the lowest tertile for the modifiable cardiovascular score had approximately 1 vessel/cm^3 lower vessel density and a mean value for cerebral blood flow of 55 ml/100g/min, which is in the bottom 40% of the current study population. Therefore, the distribution of MRI findings observed in the current study raises the potential that some individuals may be starting to diverge onto different risk trajectories for brain vascular health in early adulthood. Furthermore, levels of cerebral blood flow associated with an increased risk of dementia are evident in some young adults. No participants had clinically significant white matter hyperintensity lesion volumes but lesion count was up to 4 lesions lower in the highest tertile of optimal status of modifiable risk factors.

The difference in white matter lesion burden between lowest and highest tertiles of modifiable risk scores was around 20%. No longitudinal outcome studies have tracked white matter hyperintensities from similar age groups, but the typical rate of progression of white matter hyperintensity lesions per year in older populations is 10% to 20%[197,237] Adverse modifiable cardiovascular risk factors are major determinants of white matter hyperintensity progression[238], with small lesions increasing in size or clustering into confluent lesions[239]. Accumulation of lesions from an early age might explain why, by mid-life, white matter hyperintensity lesion volume is an established predictor of future stroke and dementia risk[229]. The longitudinal relationships between vessel morphology, cerebral perfusion and white matter lesion burden are uncertain. However, the patterns observed in the current study may suggest that resilience of the white matter, and potential to withstand risk exposures, may be influenced by the vascular morphology of an individual. The vascular haemodynamic interaction with structural integrity may be major determinants of future risk for brain health. In longitudinal studies the risk of dementia associated with cerebral hypoperfusion was strongest in association with white matter hyperintensity lesion burden[232].

4.6.4 Implications of suboptimal cardiovascular risk profiles in at risk young adults

4.6.4.1 Preterm Birth

In this study, gestational age was associated with vessel tortuosity, consistent with previous reports in infants[240], but not other cerebrovascular measures. Further work is needed to understand whether this was because participants were largely born late preterm or because cardiovascular risk profile overwhelms this early exposure.[236] The regional analysis of white matter hyperintensity lesions count demonstrates that there

may be increased periventricular lesion counts in association with earlier gestational age. It may be that preterm birth may change the microstructural integrity of the white matter and lower the resilience to adverse exposures. This is the first study to explore the association between modifiable risk factors and white matter hyperintensities in relation to gestational birth in young adults. Longitudinal research is required to explore white matter development and remodelling across the life-course in relation to environmental exposures. Longitudinal population studies have identified increased risk of early and midlife adult stroke in association with preterm birth. Further work is required to understand the interaction of modifiable risk factors with these observed risk patterns and how to implement sustainable intervention in preterm born adults.

4.6.4.2 Young Adult Hypertension

This is the first study to report changes in brain vessel morphology in association with elevated blood pressure in young adults. Previous studies have demonstrated change in white matter integrity with higher blood pressure with predicted decline in trajectory from early adulthood[163]. In the current study it was nocturnal blood pressure that maintained the strongest association with vessel density and white matter changes in multivariable models. In addition, peak exercise diastolic blood pressure, a known predictor for future cerebral atrophy, was associated with lower brain vessel density in bivariable analysis and was the only risk factor associated with vessel calibre in multivariable analysis. The observed associations with different measures of blood pressure highlight some of the potential challenges with blood pressure screening in young adults. Simple clinic measures of blood pressure may provide false reassurance. Furthermore, the observation that hypertension treatment in young adults is associated with patterns of not only increased vessel density but also cerebral

hypoperfusion raises concern about potential harms of pharmacological management of hypertension in young adults. Recent longitudinal observations and trial evidence highlights that there may be limited benefit and increased risk of adverse events associated with pharmacological management of blood pressure in young adults[154,165]. The current study confirms that elevated blood pressure in young adulthood is associated with decline in indices of brain vascular health. However, more research is needed to develop diagnostic protocols in young adults and caution is required with regard to appropriate intervention.

4.7 Conclusions

This is a preliminary, exploratory study involving young adults without clinical evidence or symptoms of cerebrovascular disease. However, modifiable cardiovascular risk factors were associated with MRI indices of cerebral vessel structure and function and white matter hyperintensities. Further research is needed to verify these findings and determine future clinical importance.

5 Young adult cardiac risk across the heart-brain axis: cardiac structure and function and association with white matter hyperintensities and cerebral blood flow

5.1 Abstract

Subclinical cardiac dysfunction is more readily identified in early and mid-life with cardiac MRI (CMR). It is uncertain whether indices of cardiovascular structure and function may be associated with subclinical brain vascular dysfunction in young adults. Modifiable risk factors and early life-exposures such as preterm birth may contribute to vascular dysfunction across the heart-brain axis.

The objective of this chapter is to explore the extent to which gestational age and modifiable risk factors relate to cardiac structure and function in young adults. It will then test the hypothesis that indices of cardiac structure and function are independently associated with white matter hyperintensity lesion counts and perfusion.

The chapter includes analysis of cardiac MRI indices and vascular brain MRI indices. Cardiovascular phenotyping included cardiopulmonary exercise testing and detailed assessment of modifiable cardiovascular risk factors. Cardiac output recorded using stress echocardiography with apical 4 chamber views was available in subgroup of participants (n=53). Cardiovascular risk factors were correlated with MRI indices of cardiac structure and function. In addition, participants' ideal cardiovascular health scores, capturing modifiable risk factors at recommended levels, were correlated with

cardiac MRI indices. Multivariable analysis was completed to explore associations between cardiac indices and brain MRI indices of white matter hyperintensity lesions count and cerebral blood flow (subgroup n=52).

The findings identify that modifiable cardiovascular risk factors are associated with cardiac MRI indices of structure and function in young adults. Left ventricular end diastolic volume and stroke volume are independently associated with white matter hyperintensity lesion counts and cerebral blood flow. Cardiovascular capacity to increase cardiac output during exercise is associated with lower burden of white matter hyperintensity lesions. Further research is needed to verify these findings and determine clinical importance.

5.2 Introduction

Cardiac structure and function are sensitive indicators of brain health in older adults. Across the life-course, disruption to the heart-brain axis, as result of myocardial events, can lead to acute decline in cognitive function and cerebrovascular injury[3,72,74,241-243]. In longitudinal population studies, increased ventricular mass and lower cardiac output in populations who were free of cerebrovascular disease at baseline, have been demonstrated as risk factors for future stroke, dementia and all cause mortality[49,74]. Furthermore, clinical heart failure is an established risk factor for dementia, but earlier in this continuum are observations that subclinical left ventricular dysfunction is associated with changes in cerebral volume and cognitive decline[3,72,74,241-243].

Subclinical cardiac dysfunction in early adult life may be precursors for brain vascular dysfunction and reduced cerebral blood flow[72,242,244]. Observational studies in older adults that demonstrated that lower resting measures of cardiac function are related

to lower cerebral blood flow and higher white matter hyperintensity lesions, point towards the potential for common vascular aetiological pathways for stroke and dementia[74,197,232,242,245,246]. The vascular origins for Alzheimer's and dementia has been supported by recent work completed using data from the longitudinal Alzheimer's Disease Network Initiative (ADNI) in USA demonstrated brain vascular disorganisation up to 30 years before the onset of symptoms was common to 60% of Alzheimer's cases[51,52,206,207]. Remodelling of cardiac phenotypes may also be contributing to observed changes in brain vascular haemodynamics in mid-life.

Modifiable risk factors have been identified as major determinants of variance in cardiovascular structure and function across the life-course[32,40,43,162,247]. However, no studies have investigated how modifiable risk factors may be influencing the heart-brain axis in younger adults. Therefore, the objectives of the current chapter were to use multimodal imaging across the heart-brain axis to explore the extent to which gestational age and modifiable risk factors relate to cardiac structure and function. Then to test the hypothesis that indices of cardiac structure and function were independently associated with white matter hyperintensity lesion counts and perfusion.

5.3 Statistical Analysis

The analysis template developed in chapter 4 to explore risk predictor association for brain vascular health was applied to cardiac MRI indices [12,44,53,162,164]. The variables were grouped as: 1) non-modifiable, including age, sex, gestational age, and 2) modifiable, including systolic blood pressure, body mass index (BMI), peak exercise capacity (oxygen uptake ml/min/kg), peak exercise diastolic blood pressure, weekly

vigorous activity level, alcohol consumption, smoking history, lipid profile, glucose, insulin and insulin resistance, and current hypertension medication.

Bivariable and multivariable analysis was completed to investigate correlation between the defined cardiovascular risk markers and cardiac MRI indices of left ventricular end diastolic volume (LVEDV), mass and geometry (Left Ventricular mass to End Diastolic Volume (LVEDV) ratio) and function (Left Ventricular Stroke Volume (LVSV) and Left Ventricular Cardiac Output (LVCO). To remain consistent with analysis presented in chapter 4 the multivariable models comparing association between cardiovascular risk exposures and cardiac MRI indices were restricted to the a priori set of cardiovascular risk factors; gestational age, clinic SBP, BMI, alcohol consumption, smoking and cardiovascular fitness measured as peak oxygen uptake. All multivariable models were adjusted for age and sex. The individuals' combined cardiovascular score presented in chapter 4 was then used as a metric of overall modifiable cardiovascular health. The relationships between the individuals' modifiable cardiovascular score and cardiac MRI indices of structure and function were studied using linear regression adjusted for age and sex.

Bivariable analysis was then completed to examine the associations between cardiac MRI indices and brain MRI indices of total and regional white matter hyperintensity lesion count. Multivariable analysis using forced entry regression models were used to examine the associations between cardiac MRI indices and white matter hyperintensity lesions count, adjusted for the potential confounding effects of risk exposures identified as factors influencing ideal cardiovascular health (gestational age, blood pressure, BMI, alcohol consumption, smoking, cardiovascular fitness, fasting glucose and cholesterol).

To examine the relationship between brain vascular structure and cardiac function on indices of brain vascular health, multivariable analysis was then expanded to examine the multivariable associations between features of brain vessel morphology (brain vessel density and tortuosity) and cardiac left ventricular volumes with measures of white matter hyperintensity lesion count and brain cerebral flow. Models were adjusted for age, sex and cardiovascular risk exposures, identified as factors influencing ideal cardiovascular health, listed above. Lesion count was selected as the most sensitive outcome of white matter change in young adults, in whom a single lesion, independent of volume, could be considered abnormal[217]. Minimum lesion size used in analysis was 1 mm^3.

In a post hoc analysis, the associations between cardiac output during submaximal exercise and white matter hyperintensity lesion count were explored in the subgroup (n=53) of participants with available stress echocardiography during exercise. The associations were adjusted for multivariable cardiovascular risk exposures.

5.4 Demographic profile and distribution of Cardiac MRI indices

Table 5.1 Study group characteristics for Cardiac MRI indices of structure, volume and function

Cardiac MRI Measures	
Left Ventricular Mass, mean (SD), g	107.4 (23.9)
Left Ventricular Length, mean (SD) mm	95.9 (10.6)
Left Ventricular End Diastolic Volume, mean (SD), ml	145.1 (34.5)
Left Ventricular Stroke Volume, mean (SD), ml	91.9 (23.5)
Left Ventricular Ejection Fraction, mean (SD), %	63.6 (4.7)
Left Ventricular Cardiac Output, mean (SD), ml	6083 (1593)
Left Ventricular Mass/End Diastolic Volume, mean (SD)	0.77 (0.20)

Table presents mean values and standard deviation. Abbreviations: WMH, white matter hyperintensity, SD, Standard Deviation

129 participants underwent cardiac MRI imaging with vascular brain MRI indices available on 125 participants. The demographics and cardiovascular risk profile of the sample are described in chapter 3.The mean values and standard deviation for cardiac MRI indices are presented in Table 5.1.

Table 5.2 Association between cardiovascular risk factors and cardiac MRI indices

	LV Stroke Volume indexed BSA		**LV End Diastolic Volume index BSA**	
	Point Estimate (95 %CI)	**P value**	**Point Estimate (95 %CI)**	**P value**
Gestational Age, weeks	***1.2 (0.89 to 1.58)***	***<.001***	***1.7 (1.2 to 2.2)***	***<.001***
SBP, mmHg	0.01 (-0.14 to 0.16)	.89	-0.07 (-0.27 to 0.13)	.48
DBP, mmHg	-0.13 (-0.31 to 0.04)	.14	***-0.36 (-0.61 to -0.11)***	***.005***
BMI, kg/m^2	-0.30 (-0.77 to 0.18)	.22	-0.60 (-1.2 to 0.1)	.05
VPA, hours per week	***2.60 (0.91 to 4.3)***	***.003***	***3.5 (0.93 to 6.1)***	***.008***
Alcohol, drinks per week	*0.18 (-0.03 to 0.40)*	*.09*	0.27 (-0.36 to 0.91)	.40
Smoking, pack years	-0.32 (-1.2 to 0.54)	.46	-0.59 (-1.64 to 0.46)	.27
Peak Vo2, ml/kg/min	***0.62 (0.47 to 0.76)***	***<.001***	***0.95 (0.74 to 1.1)***	***<.001***
Peak Ex DBP, mmHg	***-0.13 (-0.26 to -0.003)***	***.049***	***-0.23 (-0.42 to -0.04)***	***.019***
Cholesterol, mmol/L	***-3.3 (-5.6 to -1.1)***	***.004***	***-4.6 (-7.9 to -1.3)***	***.006***
Cholesterol/HDL Ratio	***-2.3 (-4.2 to -0.40)***	***.02***	***-3.18 (-6.0 to -0.34)***	***.028***
Glucose, mmol	-2.8 (-6.8 to 1.1)	.15	***-6.3 (-12.1 to -0.47)***	***.034***
Hypertension treatment	-3.2 (-7.9 to 1.5)	.18	***-8.7 (-15.5 to 1.9)***	***.012***
Nocturnal SBP, mmHg	0.07 (-0.06 to 0.20)	.31	0.06 (-0.13 to 0.24)	.54
Nocturnal DBP, mmHg	-0.11 (-0.32 to 0.11)	.33	-0.30 (-0.6 to 0.01)	.058
	LV Mass to EDV		**LV Cardiac Output**	
	Point Estimate (95 %CI)	**P value**	**Point Estimate (95 %CI)**	**P value**
Gestational Age, weeks	***-0.03 (-0.038 to -0.025)***	***<.001***	***159.0 (97.0 to 220.0)***	***<.001***
SBP, mmHg	***0.003 (0.001 to 0.006)***	***.02***	33.0 (10.0 to 55.0)	.12
DBP, mmHg	***0.006 (0.002 to 0.009)***	***0.002***	11.5 (-18.0 to 41.0)	.44
BMI, kg/m^2	0.002 (-0.007 to 0.01)	.62	64.0 (-6.0 to 134.0)	.12
VPA, hours per week	-0.03 (-0.07 to 0.009)	.14	30.5 (-278.0 to 338.0)	.85
Alcohol, drinks per week	-0. 0004 (-0.009 to 0.009)	.93	***37.0 (1.0 to 73.0)***	***.04***
Smoking, pack years	0.001 (-0.14 to 0.06)	.86	-14.9 (-136.0 to 106.0)	.81
Peak Vo2, ml/kg/min	***-0.007 (-0.01 to -0.003)***	***<.001***	***45.0 (16.0 to 74.1)***	***.003***
Peak Ex DBP, mmHg	0.002 (-.0003 to 0.005)	.08	-3.04 (-25.2 to 19.1)	.79
Cholesterol, mmol/L	***0.07 (0.03 to 0.12)***	***.003***	***-429.5 (-812.2 to -46.8)***	***.028***
Cholesterol/HDL Ratio	***0.04 (0.001 to 0.08)***	***.04***	50.3 (-282.0 to 383.0)	.77
Glucose, mmol	***0.09 (0.004 to 0.17)***	***.04***	-222.5 (-902.0 to 456.8)	.52
Hypertension treatment	0.07 (-0.03 to 0.2)	.15	96.5 (-688.0 to 881.0)	.81
Nocturnal SBP, mmHg	0.001 (-0.002 to 0.004)	.48	***25.0 (4.3 to 45.7)***	***.02***
Nocturnal DBP, mmHg	15.3 (-18.3 to 48.9)	.37	15.3 (-18.3 to 48.9)	.37

Abbreviations and units: SBP, systolic blood pressure (mmHg); BMI, body mass index (kg/m^2); VPA, Vigorous Physical Activity (hours per week); Alcohol (drinks per week); Smoking (pack years); Peak VO_2, Peak Oxygen Uptake (ml/kg/min); Ex DBP, Peak exercise diastolic blood pressure (mmHg), Cholesterol/HDL ratio, ratio total cholesterol/high density lipoprotein; HOMA-IR, homeostatic model assessment of insulin resistance, Hypertension Rx participant taking prescription medications for hypertension (yes/no)**.** LVEDV indexed for body surface area. LVSV indexed for body surface area. The point estimate refers to the magnitude of change in the left ventricular variables per unit change in the non-modifiable and modifiable variables.

5.5 Results

5.5.1 Association between non-modifiable and modifiable risk factors and Cardiac MRI indices of cardiovascular risk

Bivariable associations between risk factors and cardiac MRI indices of structure and function are presented in Table 5.2. Early gestational age at birth is associated with smaller left ventricular volumes, smaller more globular hearts, with increased left ventricular mass to end-diastolic volume and lower cardiac output. These patterns remain consistent in multivariable analysis; highlighting gestational age as an independent predictor of cardiac structure and function in young adults.

Left ventricular mass to end diastolic volume was elevated in association with higher blood pressure, lower cardiovascular fitness, deranged lipid profile and higher fasting glucose. In multivariable analysis systolic blood pressure (0.03 units higher per 10 mmHg increase in SBP, 95%CI 0.01 to 0.05, r^2=0.03, p=0.01), cardiovascular fitness (0.004 units lower per ml/kg/min increase in peak VO_2, 95%CI -0.008 to -0.001, r^2=0.02, p=0.02) and gestational age (0.03 units lower per additional week of gestation, 95%CI -0.037 to -0.024, r^2=0.37, p<0.0001) remained independently associated with left ventricular mass to end diastolic volume ratio. Overall, the a priori cardiovascular risk model explains up to 53% of the variance in left ventricular mass to end diastolic volume (model statistic r^2=0.53, p<0.0001). The a priori cardiovascular risk model is also a strong predictor of left ventricular stroke volume (model statistic r^2=0.56, p<0.0001) and left ventricular end diastolic volume (model statistic r^2=0.57, p<0.0001). In the multivariable analysis peak oxygen uptake and gestational age remained independently associated with left ventricular volumes. Left ventricular end diastolic volume was higher in association with peak oxygen uptake (0.8 ml/m^2 per ml/kg/min, 95%CI 0.56 to 1.03, r^2=0.17, p<0.0001) as was left ventricular stroke volume 0.5 ml/m^2

per ml/kg/min (95%CI 0.37 to 0.70, r^2=0.15, p<0.0001). Full term birth was associated with higher left ventricular end diastolic volume 1.3 ml/m^2 per additional week of gestation (95% CI 0.9 to 1.75, r^2=0.14, p<0.0001) and higher left ventricular stroke volume 1.0 ml/m^2 per additional week of gestation (95%CI 0.71 to 1.3, r^2=0.17, p<0.0001).

5.5.2 Association between optimal cardiovascular health scores and MRI indices of cardiovascular risk

Healthier categories on the modifiable cardiovascular score developed in chapter 4 correlated with left ventricular MRI indices of volume and mass, adjusted for age, sex and gestational age. Each additional healthier category of risk factor was associated with 2.8 ml/m^2 higher left ventricular end diastolic volume (95%CI 1.37 to 4.2, p=0.0001), 1.5 ml/m^2 higher left ventricular stroke volume (95%CI 0.47 to 2.48, p=0.004), and 0.02 lower mass to end diastolic volume ratio (95%CI -0.04 to -0.004, p=0.02). There were no significant associations between number of healthier categories of risk factor and left ventricular cardiac output (-36.0 ml 95% CI -226.0 to 154.0), p=0.71).

Adjusting for age, sex and gestational age, participants with 7 to 8 healthier categories of risk factor have a mean left ventricular stroke volume 2.9% higher, (mean difference 1.83 ml/m^2 95%CI 0.18 to 3.5, p=0.03), mean left ventricular end diastolic volume 3.5% higher, (mean difference 3.5 ml/m^2 95%CI 1.2 to 5.7, p=0.003), and mean left ventricular mass to end diastolic volume 2.8% lower (mean difference 0.024 units 95%CI -0.05 to -0.002, p=0.004) than participants with 0 to 5 healthier categories of risk factor.

5.5.3 Association between cardiac MRI indices and brain MRI indices of white matter hyperintensity lesion burden

Table 5.3 Bivariable association between cardiac MRI indices of structure and function and brain MRI indices of white matter hyperintensity lesion count

	Total WMHL count		**Periventricular WMHL count**		**Deep WMHL count**	
	Point Estimate (95 %CI)	P value	Point Estimate (95 %CI)	P value	Point Estimate (95 %CI)	P value
LV EDV	-0.04 (-0.09 to -0.001)	**.046**	-0.04 (-0.07 to -0.001)	**.04**	-0.009 (-0.4 to 0.02)	.58
LV SV	-0.06 (-0.12 to 0.003)	**.06**	-0.05 (-0.10 to 0.006)	**.08**	-0.014 (-0.06 to 0.032)	.55
LVM/EDV	7.1 (-0.24 to 14.5)	**.058**	8.3 (2.2 to 14.4)	**.008**	-0.55 (-5.87 to 4.78)	.84
LV CO	-0.001 (-0.002 to 0.0004)	.25	-0.001 (-0.002 to 0.000001	.05	0.0002 (-0.004 to 0.001)	.50

LV, left ventricle; LVM, left ventricular mass; CO, Cardiac Output; EDV, end diastolic volume; SV, stroke volume. WMHL, white matter hyperintensity lesion count. The point estimate refers to the magnitude of change in the WMHL count per unit change in the left ventricular variable.

In simple bivariable analysis higher resting left ventricular end diastolic volume was associated with lower total (-0.4 lesions per 10 ml EDV 95%CI -0.09 to -0.001, p=0.046) and periventricular white matter hyperintensity lesion count (-0.4 lesions per 10 mls EDV 95%CI -0.07 to -0.001, p=0.04). Higher left ventricular mass to end diastolic volume ratio was associated with higher periventricular white matter hyperintensity lesion count (8.3 lesions per unit increase 95%CI 2.2 to 14.4, p=0.008) with trend towards significant association with total white matter hyperintensity lesion count (7.1 lesions per unit increase 95%CI -0.24 to 14.5, p=0.058). Left ventricular stroke volume at rest also trended towards significance with total (-0.6 lesions per 10 mls SV 95%CI -1.2 to 0.03, p=0.06) and periventricular (-0.5 lesions per 10 mls SV 95%CI -1.0 to 0.06, p=0.08) white matter hyperintensity lesion count. There were no significant associations between cardiac MRI indices and deep white matter hyperintensity lesion count.

Measures of left ventricular volume remained independently associated with white matter lesion count after being indexed for body surface area and controlled for cardiovascular risk factors. In multivariable analysis indexed left ventricular stroke volume (-2.4 lesions per 10 ml/m^2, 95% CI -4.6 to -0.2, r^2=0.035, p=0.03) and indexed left ventricular end diastolic volume (-1.7 lesion per 10 ml/m^2, 95% CI -3.2 to -0.1, r2=0.033, p=0.037), were independently associated with total white matter hyperintensity lesion count and trended towards significant association with periventricular lesion count. In multivariable analysis, left ventricular cardiac output at rest and left ventricular mass to end diastolic volume were not independently associated with measures of white matter hyperintensity lesion count. Multivariable analysis confirmed that there was no independent associations between MRI indices of cardiac structure and function with deep white matter hyperintensity lesion count in this study population.

5.5.4 Association between brain vascular structure, left ventricular volumes and brain MRI indices of white matter hyperintensity lesion count

The associations between indices of brain vessel morphology, resting left ventricular volumes with indices of white matter hyperintensity lesion count and cerebral perfusion were compared in multivariable analysis. Vessel density and tortuosity were included as independent measures of vessel morphology. Vessel calibre was excluded from analysis as it was co-linear with vessel density and may vary acutely. Indexed left ventricular stroke volume and indexed left ventricular end diastolic volume maintained similar properties in the multivariable analyses. Given the cardiac MRI data is collected supine and at rest, the results for left ventricular end diastolic volume are presented as a measure of potential functional capacity.

Table 5.4 Multivariable model demonstrating associations between vessel morphology, LV end diastolic volume and MRI indices of WMH lesion counts and cerebral blood flow

	Total WMH lesion count		**Cerebral Blood Flow**	
	Point Estimate	P value	Point Estimate	P value
Vessel Tortuosity	-13.4 (-31.8 to 5.0), R^2=0.02	.15	23.2 (-10.8 to 57.3), R^2=0.03	.17
Vessel Density	***-1.5 (-2.8 to -0.3), R^2=0.05***	***.018***	***3.1 (0.7 to 5.5), R^2=0.09***	***.013***
LV EDV indexed BSA	***-0.20 (-0.35 to -0.02), R^2=0.044***	***.03***	***0.46 (0.15 to 0.76), R^2=0.13***	***.005***
Model Statistics	**R^2=0.34, P=.001**		**R^2=0.60, P=.003**	

Adjusted for age, sex, gestational age and ideal cardiovascular health indices. The point estimate refers to the magnitude of change in WMHL and CBF per unit change in vessel morphology or LVEDV. LVEDV, left ventricular end diastolic volume; WMHL, white matter hyperintensity lesion count; CBF, cerebral blood flow.

Higher left ventricular end diastolic volume and brain vessel density are independently associated with lower total white matter hyperintensity lesion counts and increased cerebral blood flow (Table 5.4). Higher indexed left ventricular end diastolic volume was independently associated with lower total white matter hyperintensity lesion count (-2.0 lesion per 10 ml/m^2, 95%CI -3.5 to -0.2, r^2=0.044, p=0.03) and higher cerebral blood flow (4.6 ml/100g/min, 95%CI 1.6 to 7.5, r^2=0.13, p=0.005). Left ventricular end diastolic volume and brain vessel density explain similar relative variance in total white matter hyperintensity lesion burden (4% to 5%), while left ventricular end diastolic volume explained more of the variance in cerebral blood flow compared with vessel density (13% vs 9%). The associations observed suggests that white matter may be sensitive to physiological plasticity and cardiovascular capacity to respond to environmental stressors. To explore this hypothesis a post hoc analysis was completed to study the association between cardiac output during submaximal exercise and number of white matter hyperintensity lesion counts in the subgroup with available stress echocardiography data.

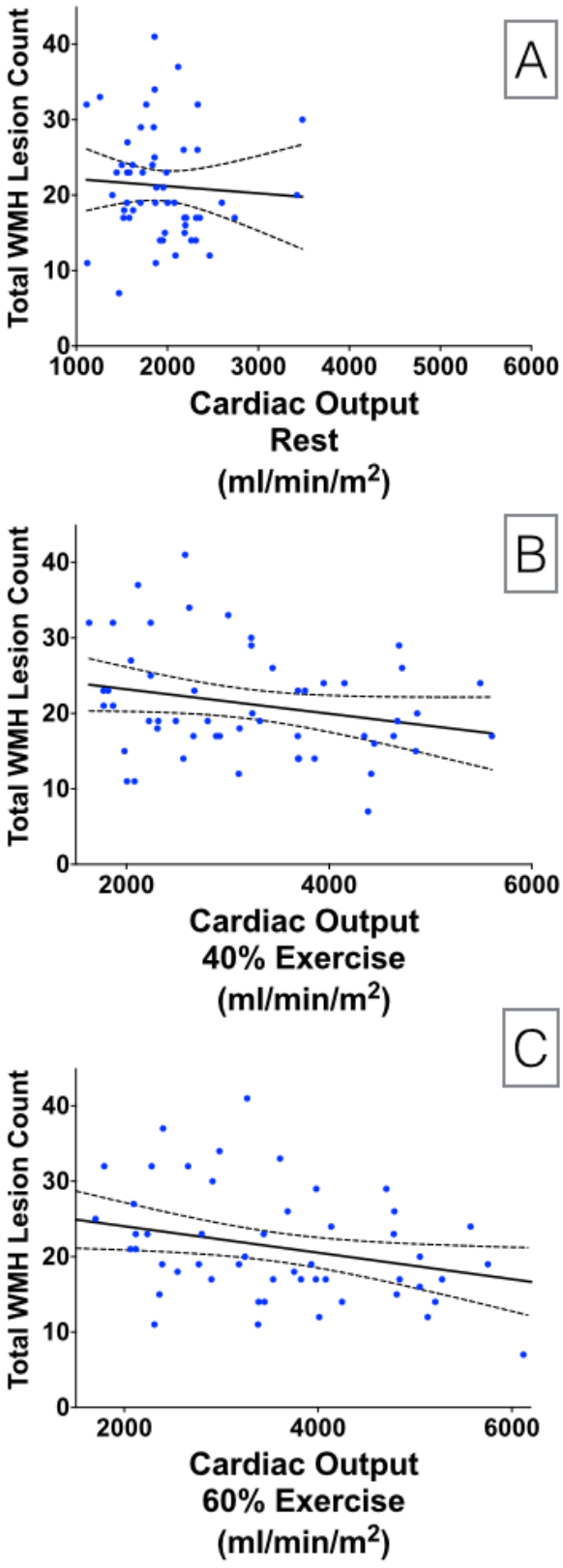


Figure 5-1 Association between total white matter lesion count and cardiac output during exercise

Figure 5-1 plots the associations of submaximal cardiac output from rest to 60% intensity with white matter lesion count. In multivariable models adjusted for cardiovascular risk factors, age, sex and gestational age, cardiac output is not associated with lesion count at rest, but the strength of association increases with exercise. Cardiac output at 60% exercise appears to be a sensitive indicator of white matter hyperintensity lesion count, and explains 15% of the variance in WMHL Table 5.5.

Table 5.5 Cardiovascular stress model for risk of white matter hyperintensity lesions: associations of white matter lesion count with change in indexed cardiac output from rest to moderate intense exercise in subgroup (n=53) with exercise stress echocardiography

	Total WMH lesion count		**Periventricular WMH lesion Count**	
A- Rest				
	Point Estimate	P value	Point Estimate	P value
Cardiac Output	-0.001 (-0.005 to 0.003) R^2=0.003	.68	-0.0002 (-0.004 to 0.004) R^2=0.0001	.92
Model Statistics	R^2=0.44 P=.01		R^2=0.39 P=.03	
B - 40% exercise workload				
	Point Estimate	P value	Point Estimate	P value
Cardiac Output	***-0.004 (-0.008 to -0.0001) R^2=0.08***	***.045***	***-0.004 (-0.007 to -0.001) R^2=0.10***	***.02***
Model Statistics	R^2=0.32 P=.17		R^2=0.39 P=.06	
C- 60% Exercise Workload				
	Point Estimate	P value	Point Estimate	P value
Cardiac Output	***-0.004 (-0.006 to -0.001) R^2=0.15***	***.008***	***-0.003 (-0.005 to -0.001) R^2=0.13***	***.01***
Model Statistics	R^2=0.38 P=.08		R^2=0.42 P=.04	

Models adjusted for age, sex, gestational age, indices of ideal cardiovascular health. The point estimate refers to the magnitude of change in WMHL per unit change in cardiac output. WMHL, white matter hyperintensity lesion count.

5.6 Discussion

5.6.1 Cardiovascular risk factors and indices of cardiac structure and function in young adult

This chapter confirms previously reported associations between history of preterm birth and cardiac structure and function with evidence of suboptimal cardiac MRI indices in association with earlier gestational age[144,145]. However, this study goes further than previous analysis, being able to control for the influence of modifiable cardiovascular risk factors, to demonstrate the independent association between cardiac structure and function and preterm birth. The inclusion of a broad set of cardiovascular risk factors in multifactorial analysis also goes further than previous observations in young adults to report on the potential modifiable determinants of cardiac indices of health, which are independent of preterm birth and potential targets of intervention in younger adults regardless of early life exposures. Specifically, maintaining higher cardiovascular fitness and a healthier overall profile on the ideal cardiovascular health score is associated with more favourable cardiac MRI indices. In younger adult populations, with normal cardiac ejection fraction and resting cardiac output, indices of left ventricular end diastolic volume and the ratio of left ventricular mass to end diastolic volume may be suitable markers to track cardiovascular risk burden and effectiveness of intervention. In previous young adult studies left ventricular end diastolic volume has been demonstrated to be modifiable with intervention and a determinant of peak exercise and cardiovascular capacity[42,248,249]. Left ventricular mass to end diastolic volume provides a marker of left ventricular mass that accommodates for left ventricular dimension. The potential importance of this indicator in the context of young adults is that it may help to distinguish pathological

versus physiological cardiac remodelling. Left ventricular mass often increases in presence of hypertension and obesity with evidence of concentric left ventricular hypertrophy[49,159]. However, exercise training may also increase cardiac mass in young adults[248-250]. In the current study left ventricular mass to end diastolic volume was higher in association with higher blood pressure, higher cholesterol, and higher fasting glucose and lower in association with cardiovascular fitness.

5.6.2 Cardiac Function, Brain Vessels and White Matter Hyperintensities

In this study, resting measures of left ventricular volumes were independently associated with total white matter hyperintensity lesion count. Resting left ventricular end diastolic volume is a primary determinant of cardiac output and could be considered a proxy measure of peak functional cardiac capacity. The observed association at rest suggested white matter hyperintensity lesion counts might relate to cardiac capacity under stress. This was then confirmed in the subgroup with stress exercise cardiac echocardiography. Cardiac output at 60% exercise workload explained up to 15% of the variance in white matter hyperintensity lesion count.

This is the first study to demonstrate that resting and exercise measures of cardiac function relate to white matter hyperintensity lesion counts in young adult populations. Lower cardiac index in older adult populations, who were free of cerebrovascular disease at baseline has been demonstrated as a risk factor for future dementia[49,72,74,250]. Furthermore, studies in older adults have demonstrated that lower resting measures of cardiac function are related to lower cerebral blood flow and white matter hyperintensity lesions[71,72,75,251]. In the current study, indexed left ventricular end diastolic volume explained up to 13% of the variance is cerebral blood flow in the subgroup with available data. This current study demonstrates that the associations

between cardiac functional measures, white matter hyperintensity lesion counts and cerebral blood flow are independent of brain vessel morphology. However, brain vessel density is also an independent predictor of total white matter hyperintensity lesion count and cerebral blood flow, with higher vessel density associated with higher cerebral blood flow and lower lesion count.

The observed associations in this cross-sectional study suggest equal importance of the integrity of the brain vascular tree and cardiac function to cerebral blood flow and presence of white matter hyperintensity lesions. The results provide evidence of a complex structural and functional heart-brain axis in young adults. Maintaining higher cardiac fitness and vascular health may be major determinants of brain vascular resilience and capacity for neurovascular auto-regulation under stress conditions.

The observation that capacity to achieve higher cardiac output across increased exercise intensity is associated with lower white matter hyperintensity lesion counts suggests that cardiovascular function may be inherently related to brain vascular health and dynamic regulation of brain vascular function. Furthermore, the observed interactions of modifiable risk factors across the heart-brain axis suggest a need for combined focus on heart-brain health in cardiovascular disease management. Protection of future brain health, in older adult life, may benefit from reduction in sub-clinical cardiac dysfunction in early and mid-life. Impaired neurovascular coupling and reduced vascular reserve in early to mid-life driven by changes in cardiac function and brain vascular structure may be early stages of the pathophysiological pathways that lead to mid-life stroke and late life cognitive decline and dementia.

5.6.3 Potential cardiovascular burden in young adults

In the current study, the differences in cardiac MRI indices between the lowest and highest tertile for the modifiable cardiovascular scores are equivalent to up 10 years accelerated aging[159,247,252]. The expected 10 year, age associated, decline in indexed left ventricular diastolic volumes and indexed stroke volume would be estimated as 3 ml/m^2 and 1.7 ml/m^2. The observed differences between tertiles in the current study were 2.9 ml/m^2 for left ventricular end diastolic volume and 1.8 ml/m^2 left ventricular stroke volume. There is also a comparable difference in left ventricular mass indexed for end diastolic volume, with an observed 3.0% difference between the lowest and highest tertiles for the modifiable cardiovascular scores. In longitudinal studies tracking cardiovascular remodelling, lower ideal cardiovascular health scores in young adults associated with 10% to 16% difference in left ventricular mass 20 years later and increased risk of left ventricular hypertrophy[33,40,247]. In adults over 60 years old 10% difference in left ventricular mass is associated with up to 20% increased risk of stroke and 40% increased risk for heart failure in subsequent 5 years follow-up[33,242,252]. The observations in the current study clearly identify that younger age is not protective against the adverse effects of modifiable risk factors with earlier than anticipated separation in risk trajectories that track to future risk of heart failure and dementia.

Longitudinal epidemiological studies have identified that lower cardiovascular fitness in young adults is association with increased 10 year risk of stroke and risk of heart failure in mid-life[11,12]. The deep cardiovascular phenotyping completed in the current study offers new insight into the potential aetiology of this mid-life cardiac dysfunction and stroke risk. The sensitivity of cardiac and brain MRI indices has made it feasible to identify the structural and functional interactions across the heart-brain axis and the

influence of modifiable risk factors in young adults. Furthermore, the observations identify potential modifiable targets to promote future heart and brain health. In early adulthood, cardiac structure and function has potential to remodel to intervention and this may be lost with age[248-250,253,254]. In young adult training studies, intensive exercise training is associated with 2% to 3% favourable change in cardiac MRI indices, capable of reversing the differences between tertiles observed in the current study[42,248-250,253,254]. Longitudinal studies have demonstrated that in early and middle age it remains possible to transition from low to high ideal cardiovascular health and gain the associated cardiac structural and functional characteristics and reduce the associated cardiovascular risks[19,255]. In trials delivering intensive exercise training sedentary middle age adults achieved up to 17% in improvement in indexed left ventricular volume[248-250,253,254]. Benefits of improved ideal cardiovascular health can be attained at any age, with evidence of cardiac remodelling in older adults. However, relative benefits are more modest than maintaining life-long exercise and there is impaired ability to improve left ventricular compliance in sedentary participants over 65 years old[1,40,247-250,253,254].

5.6.4 Implications of suboptimal cardiac function in at risk young adults

5.6.4.1 Preterm Birth

Preterm birth and the associated preterm cardiovascular phenotype may be contributing to the increased incidence of heart failure observed in younger adult populations[29]. In high-income countries there has been a dramatic increase in survival from early preterm birth, with over 70% surviving from extreme preterm birth below 28 weeks[133,134]. However, the majority of preterm deliveries are between 32 and 36

weeks, accounting for up to 80% of preterm births. Risk of heart failure is up to 36% higher in association with moderate preterm birth between 32 and 36 weeks, 3 fold higher between 28 to 31 weeks and up to 17 fold higher with extreme preterm birth below 28 weeks delivery[146,256]. In the current study gestational age explained between 14% and 17% of variance in left ventricular volume and over a third of the variance in left ventricular mass to end diastolic volume. These features of smaller, heavier ventricles with reducing gestational age have previously been reported[144,145]. However, this is the first study to report the independent association with indices of ideal cardiovascular health, which highlights the importance of potential modifiable targets in young adult populations born preterm. Of note, maintaining higher cardiovascular fitness was independently associated with increased left ventricular volumes and reduced left ventricular mass to end diastolic volumes. Further research is required to understand the remodelling potential and training adaptation of the preterm cardiovascular phenotype. The observed associations with ideal cardiovascular health indices highlight the importance of prevention of accumulated burden of modifiable risk factors in preterm born young adults. The observation in the current study of association between lower left ventricular capacity and higher burden of white matter hyperintensity lesion count may have greater significance for preterm born young adults with evidence of reduced cardiovascular capacity[137,138,257,258]. However, in the current study, global white matter hyperintensity lesion count was not related to preterm birth. Further longitudinal research and explorative work is required to explore the interactions of the heart-brain axis and the longitudinal association with cerebrovascular disease and cognitive function in relation to preterm birth.

5.6.4.2 Young Adult Hypertension

In the last 20 years, incidence of heart failure and stroke has increased in young adult populations[28,29]. Increased prevalence of hypertension and accumulated burden of multiple cardiovascular risk factors has occurred in parallel with the increase in cardiovascular disease. In the USA, from 2003 to 2012 the prevalence of having 3 to 5 cardiovascular risk factors (listed as hypertension, lipid disorders, diabetes, tobacco use or obesity), increased from 1 in 10 to 1 in 6 for young adults hospitalised with acute stroke aged 18 to 34 years old[28,29,153]. In the context of primary prevention, young adults with hypertension identify a population with increased risk of other modifiable risk factors. Therefore, an important message for health care professionals when identifying high blood pressure in young adults would be to consider other modifiable risk factors, such as lipid dysfunction and fasting metabolic state. Higher blood pressure, fasting glucose and cholesterol were negatively associated with left ventricular end diastolic volume and positively associated with left ventricular mass to end diastolic volume. This is then reflected in the tertile analysis of ideal cardiovascular health scores. The analysis in chapter 4 also demonstrated the associations between indices of ideal cardiovascular health and brain vessel density. Furthermore, the independent associations between lower vessel density and lower left ventricular volumes with higher number of white matter hyperintensity lesion count suggests that young adults with hypertension are target populations for primary prevention to protect future brain health. However, current screening and treatment guidelines fail to adequately discuss cardiovascular risk assessment and management in young adult populations[148,154,209]. The presented analysis highlights that risk assessment in young adult populations may require more detailed measures such as exercise testing, cardiac echocardiography and ambulatory monitoring[152,155]. Further research is

required to translate the current findings into primary prevention screening and treatment plans in young adult populations. This includes understanding more about the added benefit of more detailed phenotyping using magnetic resonance imaging. In the context of community and primary care settings the important message to health professionals is please do not ignore young adult hypertension, assess other modifiable risk factors and offer smoking and alcohol session advice when appropriate, encourage lifestyle modification for blood pressure and consider medical management for lipid dysfunction if lifestyle intervention is unsuccessful or not sustained.

5.7 Conclusions

In this analysis, modifiable cardiovascular risk factors are associated with imaging indices of cardiac structure and function. Furthermore, cardiac indices of structure and function are independently associated with white matter hyperintensity lesion counts and cerebral blood flow. The observed associations suggest that cardiovascular capacity to respond to an external stressor is associated with vascular indices of brain health. Further research is needed to verify these findings and determine clinical importance.

6 Heart-Brain Axis: Cardiovascular associations with brain cortical thickness and subcortical volumes

6.1 Abstract

Subcortical volumes and cortical thickness decline with age and in later life are associated with reduced cardiac function. There is also experimental evidence to support hypotheses for central control of cardiovascular capacity. These associations have not been studied in younger adults.

The objective of this chapter was to examine relationships between thalamic and basal ganglia volumes, cortical thickness, and cardiac function at rest and during exercise in young adults. The hypotheses are that in young adults, thalamic volume will be associated with MRI indices of cardiac function at rest and markers of cardiovascular performance during exercise, independent of cardiovascular fitness and early life exposures such as preterm birth. To support this hypothesis the chapter explores for evidence of cortical-thalamic pathways associated with cardiac function.

Multimodality automated-segmentation of subcortical basal ganglia and thalamic volumes, in addition to values for white matter microstructure were extracted using FSL pipelines. The CIVET MNI-pipeline was used to generate cortical thickness data. Statistical analysis was completed to study associations between cardiovascular measures with subcortical volumes and cortical thickness. Data from exercise stress echocardiograms with apical 4 chamber views was available on a subgroup (n=52) thus allowing comparison of associations with dynamic measures of cardiac output and myocardial strain.

Thalamic volume was associated with resting cardiac MRI indices of left ventricular stroke volume and left ventricular end diastolic volume. Indexed cardiac output at rest and across submaximal exercise increased with thalamic volume. Cortical thickness over the precuneus and posterior cingulate positively associated with thalamic volume and cardiac function. In multivariable analysis, thalamic volume and cortical thickness over the left temporal and right frontal area were positively associated with peak exercise capacity. To support these findings there was evidence of cortico-thalamic associations with submaximal cardiac output and myocardial strain on stress echocardiography.

The analysis provides preliminary evidence of cortical-thalamic associations with measures of cardiac function at rest and during exercise. The patterns of cortical thickness associated with thalamic volume and cardiac function suggest common functional pathways, which are unexplored in the causal pathways of cardiac dysfunction. The findings also provide support for central regulation of performance at peak exercise.

6.2 Introduction

There is limited understanding of the structural and functional associations across the heart-brain axis in early adult life. The combined peak structural and functional integrity across the heart brain axis is believed to be reached in early adult life. Peak cardiovascular function and cerebral vascular function then decline at rates of 5% to 10% per decade[7,8]. Cortical thickness, basal ganglia volumes and thalamic volume decline in parallel with cardiovascular function. Thalamic volume declines at an estimated rate of 0.69% per year and maintains a strong linear trend with advancing age (r=-0.65)[259-261]. Cortical volume loss which is driven by decline in cortical thickness

is greatest over precuneus (-0.64%) and cingulate cortex (-0.71%) in younger adults[259-261]. However, decline in white matter and hippocampal volumes start later around the age of 50 years old and follows an accelerated trajectory over the age of 60 and the older life-course[259-261]. Age associated decline in cerebral volumes of older adults has been related to decline in cardiovascular function[49,72]. A dominant disease model in older adults is that cardiac dysfunction is causal of decline in brain health.

However, neural-cardiac development reached in young adulthood and the associated cortical-thalamic function may be determinants of peak exercise capacity, cardiac function and future trajectories for age associated decline[11-15]. Furthermore, cardiac and vascular response across the heart-brain axis is regulated by the central autonomic nervous system[80,81]. There is a growing body of multidisciplinary research that describe the central autonomic network[81-90]. Cortical areas commonly identified during neuroimaging associated with cardiovascular response include the prefrontal cortex with projections to the anterior and posterior cingulate cortex, the precuneus, and the insular cortex[80,81,84,91-93]. Subcortical thalamic nuclei are a central hub of the autonomic network with established projections to the same cortical areas [92-102]. Thalamic nuclei have also been demonstrated to have synchronous activity with the cardiac cycle at rest and increased functional activity during cardiovascular stressor models[103]. Furthermore, direct stimulation of thalamic nuclei during neurosurgical procedures for movement disorders have been associated with increased heart rate and blood pressure responses[85,104].

Myocardial dysfunction and ventricular remodelling are risk factors for cerebrovascular decline and associated with neurodegenerative disease[49,72,74,259-262]. However, there is increased appreciation of potential common pathological mechanisms for cardiac and cerebrovascular disease. It is highly plausible for bi-directional cause and effect

of pathological processes across the heart-brain axis[173,263-265]. In young adults, acute vascular injury and neurodegenerative[266] changes in the thalamus have resulted in cardiac dysfunction[94,267,268]. Furthermore, loss of thalamic volume and thalamic functional connectivity are associated with high-risk phenotypes of sudden cardiac death in association with epilepsy[269-271]. In experimental studies, thalamic nuclei have been shown to be regulators of central cardiac function and haemodynamic response[84,86,88,93,94,104,272]. Therefore, basal ganglia and thalamic nuclei may be regulators of central cardiac function[31-40,82,84-90,92-95,100,104,115,117,119,178,272-274] and potential mediators of remodelling across the heart-brain axis[259-262,275]. Furthermore, functional imaging studies suggest that cortical areas across the somatosensory and interoceptive networks may regulate peak exercise capacity[98].

The objective for the current chapter is to complete an explorative analysis of the associations between basal ganglia volume, thalamic volume, cortical thickness and indices of cardiovascular function and performance. The hypotheses are that in young adults, thalamic volume will be associated with MRI indices of cardiac function, independent of cardiovascular fitness and early life exposures such as preterm birth. Then, to support a hypothesis for central regulation of cardiac performance in young adults there will be evidence of higher cortical-thalamic pathways associated with peak exercise performance and cardiac function on stress echocardiography.

6.3 Rationale for studying subcortical volume and cortical thickness as brain health measures

Trajectories of age associated declines in total cerebral volumes, subcortical volumes and cortical volume are well established[259-261]. Measures of cerebral volume and accelerated biological decline are also being explored as potential risk predictors for all-cause mortality and cognitive decline[276]. Understanding early and midlife trajectories of subcortical volumes and association with cardiovascular indices offers potential to explore new metrics of heart-brain health in young adult populations[277]. Accelerated midlife decline in cerebral volumes has been associated with early life cardiovascular risk exposures[53]. Furthermore, cardiac dysfunction in midlife and older adults is associated with accelerated cerebral atrophy[49,74]. However, there is limited understanding of structural association across the heart-brain axis in younger adults.

The human brain is a complex network of interlinked regions. Brain networks are comprised of central neural hubs that are rich in connection between neural areas including over physically long distances[278]. There are a number of highly connected central neural hubs that have been described as the brain rich club[279]. This includes hubs within the precuneus, superior frontal, superior parietal cortex and the subcortical hippocampus, putamen and thalamus[278,280]. The rich club phenomenon is present when the hubs of a network are more densely connected among themselves than nodes of lower degrees and peripheral to the central connections[278,280]. Structural hubs with rich connections may provide the anatomical framework to facilitate the integration of task and time dependent functional networks[281].

An emerging view is that cortical thickness represents functional network organization[282]. With increase in cortical thickness and volumes association with increased connectivity within defined neural hubs. Supporting this concept, developmental studies suggest that tissue volume in the thalamus and cortex reflect thalamocortical connectivity and is dependent on the growth and integrity of white matter tracts[283,284]. Furthermore, the number of neurons in topographically connected thalamic and cortical regions is closely related[285]. There is also good correlation between functional and structural connectivity within resting states[281]. In addition, cortical thickness across functional networks predicts cognitive function[282,286]. Cortical thickness over the default mode, salience and somatomotor network appear to be particularly sensitive to age associated thinning which is a mediator of cognitive decline[282]. Decline in thalamocortical volume from early adulthood may therefore reflect loss of structural complexity.

Cortical thickness has been identified as the dominant cortical morphological feature influenced by age and associated with cognition[259-261,287]. Cortical thinning is also the main determinant of age associated cortical volume loss[259,287]. Previous studies support cortical thickness as an index of age related brain structural integrity[259-261,287]. It is believed that subcortical volumes and cortical thickness may represents a marker of neural structural complexity[282]. There is potential that modifiable risk factor and adverse exposures in early life may influence the peak complexity of neural integration and density of the connections within functional networks[288,289]. Then with age and continued exposure to vascular risk factors that there may be accelerated decline in neuronal complexity[290]. Age associated decline in cortical thickness and subcortical volume is believed to because of a reduction in the number of dendritic spines,

synapses and reduced intracortical myelination rather than a reduction in neuronal number[282].

Advances in neuroimaging allow automated segmentation and extraction of thalamic volume[291]. Therefore, the thalamus may be a midbrain regulator of cardiac function that may be more readily measured in clinical settings in comparison to deep brainstem nuclei and functional connectivity[269,270]. If thalamic volume is a marker of heart-brain health then, strategies to reduce later life burden of heart failure and dementia may be targeted decades earlier[259,260,262,275]. However, no previous studies have investigated the association between basal ganglia and thalamic volumes and cardiovascular indices. Furthermore no studies have investigated associations between cardiovascular function and cortical thickness.

6.4 Statistical Analysis

The first analysis completed was to investigate the bivariable associations between modifiable and non-modifiable risk factors and thalamic and basal ganglia volumes. Then multivariable analysis was completed to identify independent associations between basal ganglia and thalamic volumes with cardiac MRI indices of structure and function. Multivariable analysis was completed using forced entry linear regression and general linear modelling with residual analysis completed to assess model assumptions. In multivariable analysis thalamic volumes were the only subcortical nuclei to be independently associated with cardiac MRI indices. To explore the full extent of cardiac and thalamic association, analysis was expanded to include

association with cardiac MRI indices ventricular length and strain patterns. Multivariable analyses were adjusted for age and sex and because of their known associations with cardiac function gestational age and peak oxygen uptake. When indicated, analysis was also indexed for brain size or body surface area.

The SurfStat script for MATLAB was used to complete analysis of the associations between thalamic volume and measures of cardiac function with brain cortical thickness. Multivariable models were constructed to identify the areas of cortical surface independently related to thalamic volumes adjusted for age, peak oxygen uptake and gestational age. The associations between cardiac indices and cortical thickness were also modelled adjusting for thalamic volumes to identify cortical areas independently associated with resting and peak cardiovascular measures. Cortical thickness values from individual vertices, with significant associations in multivariable models, were extracted for additional analysis.

To investigate the hypothesis of higher cortical-thalamic pathways associated with regulation of cardiac function, cortical thickness values and thalamic volumes were combined in multivariable analysis in prediction models for indices of peak exercise performance. Cortical thickness values associated with peak oxygen uptake, independent from thalamic volume, were extracted from vertices identified in multivariable cortical surface analysis. Multivariable models were constricted to examine cortical-thalamic associations with peak exercise performance markers including: peak oxygen uptake as a global cardiovascular measure reproducible as an indicator of heart and brain health; peak wattage as an indicator of power output and muscular function; peak heart rate, as an indicator of peak sympathetic drive; peak oxygen uptake as an indirect measure of cardiac output; oxygen uptake efficiency slope, as a composite measure of cardiopulmonary and metabolic function; and heart

rate recovery an indicator of central autonomic control. Models included variables of age, sex, gestational age, vigorous physical activity, left ventricular diastolic volume and baseline respiratory function (FEV1/FVC forced expiratory volume in one second (FEV1) over forced vital capacity (FVC)) as independent predictors of peak exercise performance and adjusted for confounders of brain size and modifiable risk factors (smoking, alcohol consumption, blood pressure and body mass index). To further explore the hypothesis of cortical-thalamic association with exercise and cardiac performance multivariable analysis was completed using sub-maximal exercise data reported from the ventilatory anaerobic threshold during the cardiopulmonary exercise test. This analysis included investigation of associations with peak oxygen uptake, wattage, heart rate and oxygen pulse recorded at the time of transition across the ventilatory anaerobic threshold during peak cardiopulmonary exercise testing. Also modelled were the cortical-thalamic associations with cardiac measures recorded during stress echocardiography in a subgroup of participants with available data.

6.5 Study Population

129 participants underwent cardiac MRI imaging and 125 participants completed multimodal brain MRI imaging. After quality checks, 124 image sets were available for cortical surface analysis and 121 image sets were available for subcortical segmentation applying MIST which used four images (T1, T2 FLAIR, DTI FA and DTI MD). The sample description is comparable to that included in other analysis. The mean age of participants was 24.4±4.6 years, 58 participants were female (48%), the mean gestational age was 36.6±4.3 weeks, educational attainment was high with 84 completing University level education (69.4%), 29 participants had prior history of hypertension of which 21 were on anti-hypertension medications (17.3%).

Table 6.1 Study group characteristics for brain and cardiac MRI measures and cardiopulmonary exercise measures

Cardiac MRI Measures	
Left Ventricular Mass, mean (SD), g	108 (24.5)
Left Ventricular Length, mean (SD) mm	95.9 (10.7)
Left Ventricular End Diastolic Volume, mean (SD), ml	145 (34.5)
Left Ventricular Stroke Volume, mean (SD), ml	92.2 (23.6)
Left Ventricular Ejection Fraction, mean (SD), %	63.7 (4.6)
Left Ventricular Cardiac Output, mean (SD), ml	6102 (1590)
Global Peak Systolic Circumferential Strain, mean (SD)	-18.1(2.72)
Basal Peak Systolic Circumferential Strain, mean (SD)	-17.37(3.38)
Mid Peak Systolic Circumferential Strain, mean (SD)	-18.2 (2.58)
Apical Peak Systolic Circumferential Strain, mean (SD)	-21.24 (4.17)
Brain MRI Measures	
Thalamic Volume, mean (SD), cm^3	12.44 (1.26)
Caudate and Accumbens Volume (SD), cm^3	7.10 (0.77)
Putamen Volume (SD), cm^3	8.41 (0.98)
Globius Pallidum Volume (SD), cm^3	2.66 (0.27)
White Matter Fractional Anisotropy	0.341 (0.012)
White Matter Mean Diffusivity	0.0007 (0.000014)
Cardiopulmonary Exercise Measures	
Peak VO_2, mean (SD), ml/kg/min	37.3 (9.6)
Peak Respiratory Exchange Ratio, mean (SD)	1.18 (0.06)
Peak Wattage, mean (SD), watts	226 (68)
Peak Ventilatory Equivalent, mean (SD), L/min	102 (37.0)
Oxygen Uptake Efficiency Slope, mean (SD)	2.8 (0.85)
Peak Oxygen Pulse, mean (SD), ml/beat	14.1 (4.3)
Peak Cardiovascular Product, mean (SD), mmHg.bpm	32420 (5234)
Ventilatory Anaerobic Threshold, mean (SD), ml/min/kg	20.1 (6.0)
Heart Rate Recovery, mean (SD), bpm	20.4 (6.1)

Abbreviations: BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; Alcohol (drinks per week), Peak VO_2, Peak Oxygen Uptake.

6.6 Results

6.6.1 Associations between cardiovascular risk factors and subcortical volumes

Table 6.2 Associations between cardiovascular risk factors and basal ganglia and thalamic volumes

	Thalamic Volume (mm^3)		Caudate Volume (mm^3)	
	Point Estimate (95 %CI)	**P value**	**Point Estimate (95 %CI)**	**P value**
Gestational Age, weeks	***95.1 (43.2 to 147)***	***.0004***	2.2 (-30.9 to 35.4)	.89
SBP, mmHg	10.7 (-7.7 to 29.2)	.25	-4.2 (-15.4 to 7.0)	.46
DBP, mmHg	-1.85 (-25.6 to 21.9)	.88	**-22.0 (-35.9 to -8.2)**	**.002**
BMI, kg/m^2	39.8 (-16.7 to -96.3)	.17	13.9 (-20.5 to 48.4)	.43
VPA, hours per week	108.1 (-137.4 to 353.5)	.39	74.7 (-74.0 to 223.5)	.32
Alcohol, drinks per week	18.5 (-40.3 to -77.4)	.53	-5.3 (-41.1 to 30.5)	.77
Smoking, pack years	-2.7 (-100.2 to 94.7)	.96	-2.9 (-62.0 to 56.2)	.92
Peak Vo2, ml/kg/min	***51.4 (29.3 to 73.6)***	***.00001***	***31.5 (18.1 to 44.9)***	***.00001***
Peak Ex DBP, mmHg	-15.8 (-33.5 to 1.80)	.08	-6.8 (-17.6 to 3.9)	.21
Cholesterol, mmol/L	-172.5 (-487.3 to 142.3)	.28	-81.2 (-272.6 to 110.2)	.40
Cholesterol/HDL Ratio	64.7 (-204.4 to 333.9)	.64	54.4 (-108.7 to 217.5)	.51
Glucose, mmol	-91.4 (-640.2 to 457.4)	.74	24.6 (-308.5 to 357.7)	.88
Hypertension treatment	-89.0 (-721.0 to 542.6)	.78	-332.5 (-711.0 to 46.1)	.08
Nocturnal SBP, mmHg	4.0 (-13.1 to 21.1)	.65	6.8 (-3.5 to 17.2)	.19
Nocturnal DBP, mmHg	-24.0 (-50.7 to 3.0)	.08	-7.9 (-24.3 to 8.6)	.34
	Putamen Volume (mm^3)		**Pallidum Volume (mm^3)**	
	Point Estimate (95 %CI)	**P value**	**Point Estimate (95 %CI)**	**P value**
Gestational Age, weeks	34.3 (-7.7 to 76.2)	.11	***16.6 (5.2 to 27.9)***	***.005***
SBP, mmHg	12.8 (-1.4 to 26.9)	.08	1.1 (-2.9 to 5.1)	.58
DBP, mmHg	-4.1 (-22.5 to 14.3)	.66	-3.84 (-8.9 to 1.2)	.14
BMI, kg/m^2	33.2 (-10.5 to 76.9)	.14	10.2 (-1.9 to 22.2)	.10
VPA, hours per week	65.1 (-125.3 to 255.5)	.50	36.5 (-15.9 to 88.9)	.17
Alcohol, drinks per week	8.78 (-36.9 to 54.4)	.71	-1.7 (-14.4 to 10.9)	.79
Smoking, pack years	-0.24 (-75.7 to 75.3)	.99	3.38 (-17.5 to 24.3)	.75
Peak Vo2, ml/kg/min	***23.0 (4.9 to 41.2)***	***.01***	***9.05 (4.16 to 13.9)***	***.0004***
Peak Ex DBP, mmHg	-7.3 (-21.1 to 6.4)	.29	-2.7 (-6.5 to 1.2)	.17
Cholesterol, mmol/L	-55.9 (-300.8 to 189.1)	.65	-34.8 (-102.5 to 32.8)	.31
Cholesterol/HDL Ratio	178.2 (-27.8 to 384.3)	.09	30.5 (-27.0 to 88.1)	.29
Glucose, mmol	0.24 (-425.1 to 425.6)	.99	-36.7 (-154.4 to 80.9)	.54
Hypertension treatment	-141.2 (-630.1 to 347.6)	.57	-10.2 (-145.8 to 125.5)	.88
Nocturnal SBP, mmHg	***13.1 (0.08 to 26.1)***	***.05***	***4.1 (0.46 to 7.7)***	***.03***
Nocturnal DBP, mmHg	-4.4 (-25.5 to 16.7)	.68	-0.82 (-6.7 to 5.0)	.78

Abbreviations and units: SBP, systolic blood pressure (mmHg); BMI, body mass index (kg/m^2); VPA, Vigorous Physical Activity (hours per week); Alcohol (drinks per week); Smoking (pack years); Peak VO_2, Peak Oxygen Uptake (ml/kg/min); Ex DBP, Peak exercise diastolic blood pressure (mmHg), Cholesterol/HDL ratio, ratio total cholesterol/high density lipoprotein; HOMA-IR, homeostatic model assessment of insulin resistance, Hypertension treatment, prescription medications for hypertension (yes/no). The point estimate refers to the magnitude of change in the subcortical volume per unit change in the non-modifiable and modifiable variables.

Bivariable associations between risk factors and basal ganglia and thalamic volumes are presented in (Table 6.2). Early gestational age at birth is associated with smaller thalamic volumes, and smaller pallidum volumes. Higher cardiovascular fitness, measured by higher peak oxygen uptake was associated with higher volumes in all reported basal ganglia and thalamic volumes.

6.6.2 Association between Subcortical Volumes and Cardiac Structure and Function

Table 6.3 Associations between subcortical volumes and cardiac structure and function

	LVM		**LV EDV**		**LV CO**		**LV EF**	
	Point Estimate (95 %CI)	**P value**	**Point Estimate (95 %CI)**	**P value**	**Point Estimate (95 %CI)**	**P value**	**Point Estimate (95 %CI)**	**P value**
Thalamus Volume cm^3	0.85 (-3.5 to 5.2)	.70	***5.0 (2.5 to10.0)***	***.039***	***300 (20.5 to 580)***	***.036***	***1.47 (0.52 to 2.4)***	***.003***
Caudate Volume cm^3	-1 (-8.0 to 6)	.77	3.5 (-4.0 to 11)	.36	332 (-100 to 764)	.131	1.3 (0.52 to 2.4)	.08
Putamen Volume cm^3	0.5 (-5 to 6)	.83	0.9 (-5.0 to 7.0)	.76	176 (-161 to 514)	.30	0.6 (-0.6 to 2.0)	.317
Pallidum Volume cm^3	-3.0 (-23.0 to 17)	.70	6.2 (-16.0 to 28)	.58	290 (-970 to 1551)	.65	3.0 (-1.3 to 7.0)	.18

Associations between volume of subcortical nuclei and cardiac MRI indices adjusted for age, sex, brain size, peak oxygen uptake and gestational age. Abbreviations LV=Left Ventricular LVM=Left Ventricular Mass, EDV=End Diastolic Volume, CO=Cardiac Output, EF=Ejection Fractions. The point estimate refers to the magnitude of change in the left ventricular variables per unit change in subcortical volume.

In multivariable analysis there are no observed associations between volumes of the caudate nuclei, putamen and globus pallidum with cardiac MRI indices of structure or function (Table 6.3).

Table 6.4 Associations between MRI indices of cardiac structure and function and thalamic volume

	Point Estimate per cm^3 increase in Thalamic Volume (95 %CI)	P value	R^2
Left ventricular mass (g)	0.85 (-3.5 to 5.2)	0.70	0.001
Left ventricular length (cm)	1.05 (-0.47 to 2.5)	0.17	0.007
Left ventricular stroke volume (ml)	5.8 (2.4 to 9.1)	**<0.001**	**0.04**
Ejection fraction (%)	1.47 (0.52 to 2.4)	**0.003**	**0.08**
Cardiac output (ml)	300 (20.5 to 579.7)	**0.036**	**0.03**
Global Circumferential Strain Short Axis	-0.65 (-0.08 to -1.2)	**0.026**	**0.04**
Basal Circumferential Strain Short Axis	-0.36 (-1.1 to 0.4)	0.34	0.008
Mid Circumferential Strain Short Axis	-0.57 (-0.01 to -1.13)	**0.04**	**0.03**
Apical Circumferential Strain Short Axis	-1.2 (-0.38 to -2.1)	**0.005**	**0.06**

Associations between volume of subcortical nuclei and cardiac MRI indices adjusted for age, sex, brain size, peak oxygen uptake and gestational age. Abbreviations LV=Left Ventricular LVM=Left Ventricular Mass, LVL=Left Ventricular Length, EDV=End Diastolic Volume, SV=Stroke Volume, CO=Cardiac Output, EF=Ejection Fractions. Table presents point estimates from multivariable models adjusted for age, sex, peak oxygen uptake, gestational age and brain size. The point estimate refers to the magnitude of change in the left ventricular variables per unit change in thalamic volume.

Thalamic volume was not associated with cardiac mass or length but was associated with left ventricular stroke volume (5.8 ml per 1 cm^3 thalamic volume, 95%CI 2.4 to 9.1, p<0.001), ejection fraction (1.47% per 1 cm^3 thalamic volume, 95%CI 0.52 to 2.4, p=0.003), cardiac output (300 ml per 1 cm^3 thalamic volume, 95%CI 20.5 to 579.7, p=0.036) and global circumferential strain (-0.65 per 1 cm^3 thalamic volume, 95%CI -0.08 to -1.2, p=0.026) (Table 6.4). On regional assessment, thalamic volume associations with myocardial strain were present in the mid ventricle (-0.57 per 1 cm^3 thalamic volume, 95%CI -0.01 to -1.13, p=0.04) and apex (-1.2 per 1 cm^3 thalamic volume, 95%CI-0.38 to -2.1, p=0.005) but not in the basal segments.

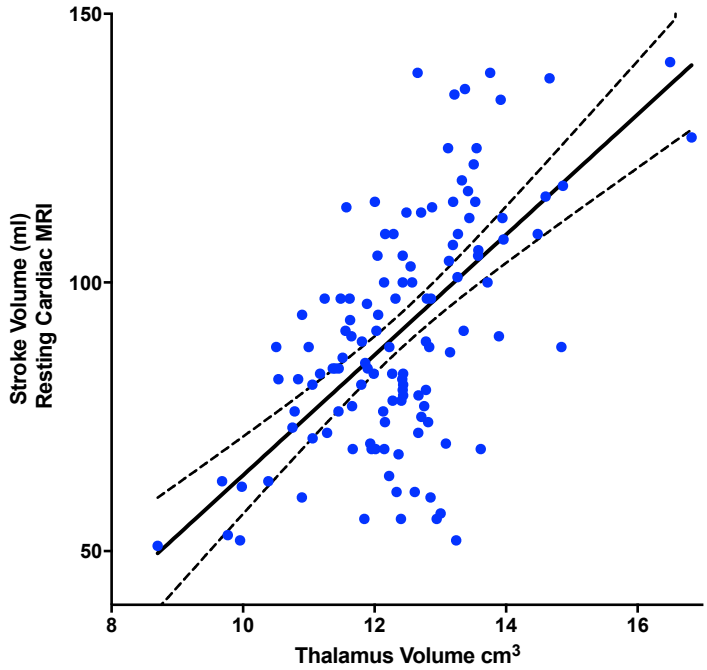


Figure 6-1 Association between total thalamic volume and left ventricular stroke volume

Figure presents association between thalamic volume and left ventricular stroke volume measured by cardiac MRI, stroke volume increases 5.8 ml per 1cm^3 increase in thalamic volume (95% CI 2.4 to 9.1, p<0.001) adjusted for age, sex, gestational age, peak VO_2 and brain size.

6.6.3 Cardiovascular and Thalamic Associations with Cortical Thickness

There were positive associations between thalamic volume and areas of cortical thickness (Figure 6.2) that co-located with areas associated with resting measures of left ventricular end diastolic volume and left ventricular stroke volume including; precentral gyrus, precuneus, mid cingulate, posterior cingulate, lingual, calcarine, parahippocampal and temporal areas (Figure 6.2).

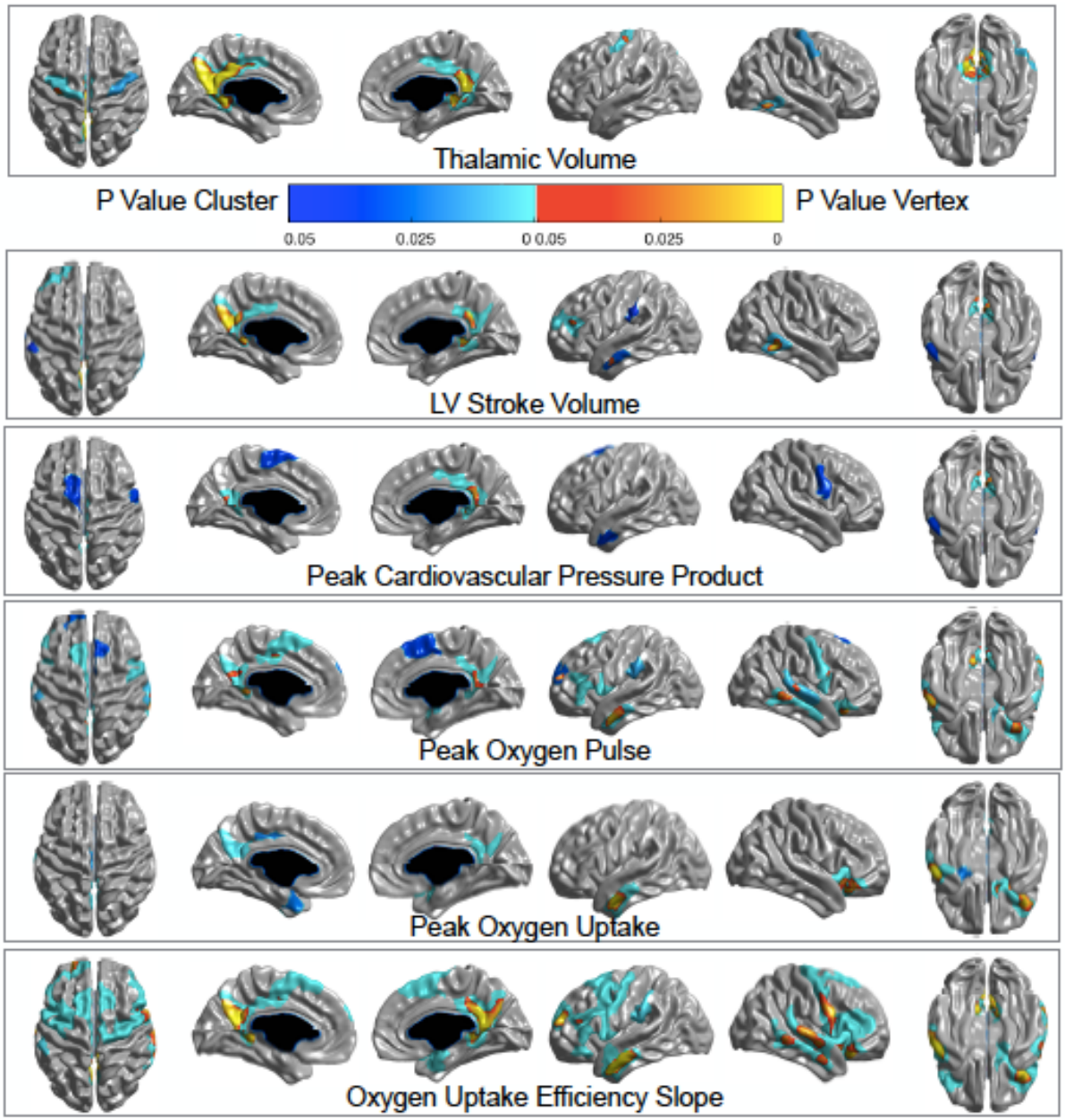


Figure 6-2 Thalamic and cardiovascular associations with cortical thickness.

In multivariable models, thalamic volume remained positively associated with cortical thickness over areas of the mid and posterior cingulate, precuneus, lingual and calcarine cortex adjusting for peak oxygen uptake, age and gestational age (Table 6.5). Thalamic associations were strongest over the left precuneus, left posterior cingulate and left calcarine areas. The table also presents cortical vertices associated with left ventricular end diastolic volume and left ventricular stroke volume.

Table 6.5 Areas of brain cortical thickness and related cortical vertices with strongest association with thalamic volume and cardiac left ventricular stroke volume and end diastolic volume

	Cortical Area		T Statistics	MNI Co-ordinates			P Value
Thalamus*				x	y	z	
	Precuneus	Left	6.12	-4.0	-57.0	21.0	**0.00002**
		Right	4.97	3.0	-49.0	20.0	**0.002**
	Posterior Cingulate	Left	5.69	-3.0	-46.0	31.0	**0.0001**
	Mid Cingulate	Left	4.80	-4.0	-36.0	35.0	**0.005**
	Lingual	Left	4.81	-12.0	-38.0	1.0	**0.005**
		Right	4.30	12.0	-53.0	8.0	**0.03**
	Calcarine	Left	5.67	-3.0	-59.0	14.0	**0.0001**
Left Ventricular End Diastolic Volume+							
	Precuneus	Left	6.17	-5	-58	24	**0.0001**
		Right	4.78	2	-50	27	**0.005**
	Posterior Cingulate	Left	5.59	-3	-48	26	**0.0002**
	Lingual	Left	5.38	-9	-44	3	**0.0005**
		Right	5.31	13	-54	8	**0.0006**
	Calcarine	Left	4.66	-13	-51	5	**0.008**
		Right	4.67	11	-55	12	**0.008**
	Temporal	Left Mid	4.86	-62	-2	-24	**0.004**
		Right Inferior	5.21	59	-49	-9	**0.001**
	Frontal	Left Superior	4.44	-24	58	19	**0.02**
		Left Mid	4.23	-44	40	19	**0.04**
Left Ventricular Stroke Volume+							
	Precuneus	Left	5.13	-5	-58	25	**0.001**
		Right	4.46	3	-49	21	**0.02**
	Posterior Cingulate	Left	4.69	-3	-48	26	**0.007**
		Right	4.70	3	-50	30	**0.007**
	Lingual	Left	4.69	-10	-41	2	**0.007**
		Right	4.94	11	-42	0	**0.003**
	Calcarine	Left	4.92	-9	-43	4	**0.003**
	Parahippocampal	Right	4.28	14	-38	-5	**0.03**
	Temporal	Left Mid	4.33	-62	-2	-24	**0.03**
		Right Inferior	4.84	59	-49	-9	**0.004**
	Frontal	Left Mid	4.30	-44	41	18	**0.03**

*Association adjusted for age, gestational age and peak oxygen uptake; +Unadjusted association.

Association between cortical thickness and left ventricular end diastolic volume and left ventricular stroke volume were strongest over the left precuneus. The peak association between cortical thickness over the left precuneus and left ventricular end diastolic volume was located at the MNI co-ordinate x = -5, y= -58, z = 24. Cortical thickness at this location increases 0.0013 mm per 1ml increase in left ventricular end

diastolic volume 95% (0.0006 to 0.002), p<0.0008, adjusted for age, gender, gestational age and peak oxygen uptake (**Figure 6-3**). This association maintained significance when further adjusted for total brain volume (p=0.02). However, when the model is adjusted for thalamic volume, left ventricular end diastolic volume only trends towards significant association with precuneus cortical thickness (0.0007 mm per ml LVEDV, 95% CI -0.00009 to 0.002, p=0.08). In the full multivariable model, thalamic volume is independently associated with cortical thickness: 0.00003 mm per mm^3 thalamic volume, 95% CI 0.000006 to 0.00005, p = 0.013 at MNI co-ordinate x=-5, y=-58, z=24

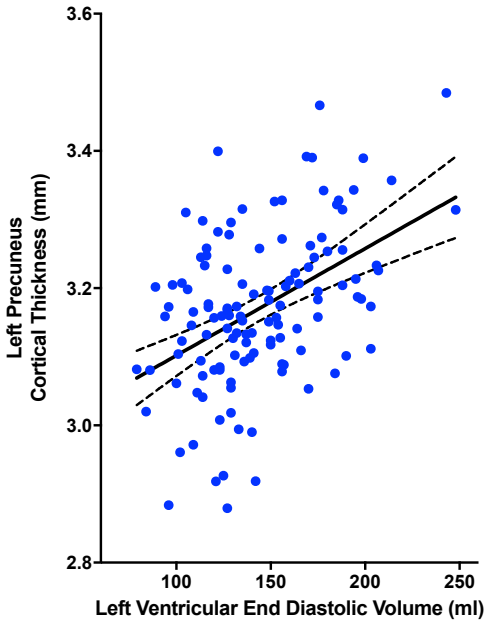
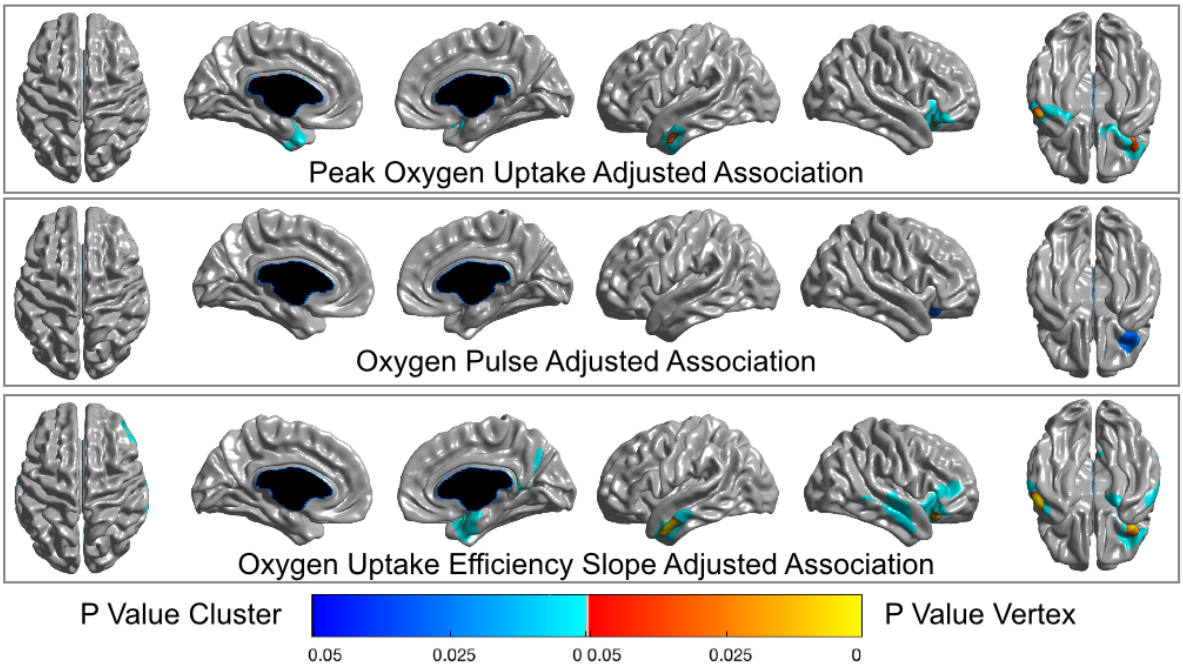


Figure 6-3 Association between cortical thickness over the left precuneus and left ventricular end diastolic volume

In multivariable analysis, adjusting for age, gender, gestational age, peak oxygen uptake and brain volume, left ventricular stroke volume was positively associated with cortical thickness over the left precuneus 0.0012 mm per 1ml increase in left ventricular stroke volume (95% 0.00008 to 0.002, p=0.036). Independent association between left precuneus cortical thickness and left ventricular stroke volume was lost

when thalamic volume was included in multivariable analysis (0.0008 mm per 1ml increase in left ventricular stroke volume 95% CI -0.0004 to 0.002, p=0.20).

Areas of cortical thickness associated with measures of cardiovascular performance during peak exercise are presented in rows 3 to 6 of **Figure 6.2**. Peak cardiovascular pressure product (a measure of cardiac workload), peak oxygen pulse (a measure of cardiac output) and oxygen uptake efficiency during exercise share positive associations with cortical thickness over motor areas including the primary motor cortex and precentral gyrus. There were no observed associations between primary cortical motor areas and left ventricular stroke volume and left ventricular end diastolic volume. In addition, there are no associations between cortical thickness and left ventricular measures of mass and length. The associations between motor areas and measures of cardiovascular performance during exercise are no longer observed when adjusted for thalamic volume. The observed associations between left ventricular stroke volume and left ventricular end diastolic volume and cortical thickness are no longer observed when adjusted for thalamic volume. The observations suggest the associations between cortical thickness and left ventricular volumes may be mediated through thalamic volume. However, the cortical associations with left ventricular volumes do not appear to be related to cardiac size or motor function.



Cortical Area		T Statistics	MNI Co-ordinates			P Value
Peak Oxygen Uptake			x	y	z	
Temporal	Left Inferior	4.62	-50	0	-33	0.00003
Frontal	Right Inferior Orbital	3.96	37	22	-16	0.001
Peak Oxygen Pulse						
Frontal	Right	4.24	37	28	-22	0.037
Oxygen Uptake Efficiency						
Temporal	Left Mid	5.19	-60	-1	-27	0.001
Frontal	Right Inferior Orbital	4.77	40	-24	-18	0.005

Figure 6-4 Areas of cortical thickness associated with cardiovascular performance measures and MNI co-ordinates of cortical vertices with strongest association after adjusting for thalamic volume.

Cortical associations with peak oxygen uptake, oxygen uptake efficiency slope and peak oxygen pulse, when adjusted for thalamic volume, are clustered in areas of the left mid temporal and left inferior temporal area, the right frontal cortex over the inferior orbital area and the right insular area (**Figure 6-4**). In additional multivariable models, further adjusting for modifiable risk factors, age, gender, gestational age, thalamic

volumes and brain size, the observed clusters of cortical thickness associated with peak oxygen uptake remained consistent with the image presented in row one of **Figure 6-4**. However, the individual vertices that held significant association were reduced to peaks within the areas of the left inferior temporal area and right frontal cortex (Figure 6-5). The associations with oxygen uptake efficiency were reduced to the area of the right insular and inferior orbital area of the right frontal cortex with significance maintained at the cluster level as opposed to individual vertices. In the multivariable model there were no observed cortical associations with oxygen pulse.

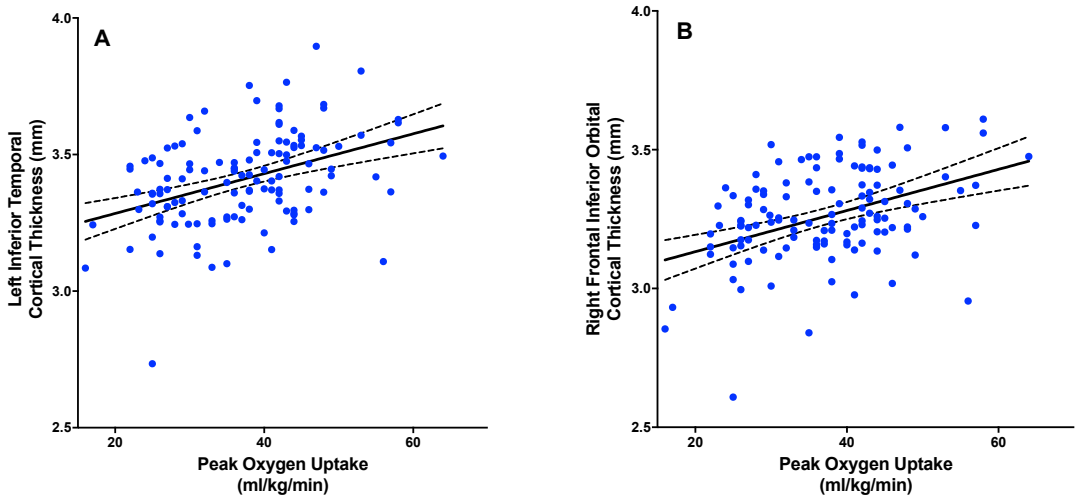


Figure 6-5 Associations from multivariable analysis of peak oxygen uptake and cortical thickness at vertices over the left inferior temporal area and right frontal inferior orbital area.
Peak A, MNI co-ordinates, x=-50, y=0, z=-33: cortical thickness increases 0.009 mm per 1ml/kg/min increase in peak oxygen uptake, 95% CI 0.005 to 0.013, p=0.00003. Peak B, MNI co-ordinates x=37, y=22, z=-16; cortical thickness increases 0.008 mm per 1ml/kg/min increase in peak oxygen uptake, 95% CI 0.004 to 0.012, p=0.001. Associations adjusted for age, sex, gestational age, indices of ideal cardiovascular health (smoking, alcohol, blood pressure (nocturnal map), body mass index, glucose, cholesterol) thalamus volume and total brain size.

6.6.4 Cortical-thalamic associations with Peak Exercise Measures

Table 6.6 Cortical and thalamic associations in multivariable analysis with peak cardiovascular performance during cardiopulmonary exercise testing

Peak Oxygen Uptake (ml/kg/min)	Point Estimate, (95% CI)	P value	R^2
Vigorous Physical Activity	1.84 (0.58 to 3.1)	**0.005**	**0.03**
Left End Diastolic Volume	0.26 (0.15 to 0.37)	**<0.0001**	**0.07**
Thalamic Volume	2.0 (0.17 to 3.0)	**0.027**	**0.02**
Left Inferior Temporal Cortical Thickness	14.3 (6.9 to 21.7)	**0.0002**	**0.05**
Peak Wattage (watts)			
Vigorous Physical Activity	11.4 (2.8 to 20.0)	**0.01**	**0.022**
Left End Diastolic Volume	2.0 (1.23 to 2.72)	**<0.0001**	**0.087**
Thalamic Volume	13 (4.0 to 22.0)	**0.006**	**0.026**
Left Inferior Temporal Cortical Thickness	58.7 (8.6 to 109.0)	**0.022**	**0.017**
Peak Heart Rate (bpm)			
Vigorous Physical Activity	1.42 (-1.0 to 3.85)	0.25	0.009
Left End Diastolic Volume	-0.15 (-0.37 to 0.06)	0.16	0.014
Thalamic Volume	0.4 (-2.0 to 3.0)	0.76	0.001
Left Inferior Temporal Cortical Thickness	18.8 (4.6 to 33.0)	**0.01**	**0.050**
Oxygen Pulse (ml/bpm)			
Vigorous Physical Activity	0.44 (-0.03 to 0.92)	0.066	0.008
Left End Diastolic Volume	0.13 (0.09 to 0.17)	**<0.0001**	**0.090**
Thalamic Volume	0.81 (0.3 to 1.3)	**0.002**	**0.022**
Left Inferior Temporal Cortical Thickness	3.75 (0.98 to 6.53)	**0.0085**	**0.016**
Oxygen Uptake Efficiency Slope			
Vigorous Physical Activity	0.09 (-0.03 to 0.20)	0.145	0.008
Left End Diastolic Volume	0.02 (0.01 to 0.03)	**<0.0001**	**0.059**
Thalamic Volume	0.15 (0.02 to 0.27)	**0.025**	**0.019**
Left Inferior Temporal Cortical Thickness	1.1 (0.41 to 1.80)	**0.002**	**0.036**
Heart Rate Recovery (bpm)			
Vigorous Physical Activity	1.4 (0.14 to 2.65)	**0.03**	**0.016**
Left End Diastolic Volume	0.06 (-0.5 to 0.17)	0.298	0.075
Thalamic Volume	1.4 (0.07 to 3.0)	**0.039**	**0.011**
Left Inferior Temporal Cortical Thickness	4.4 (-2.9 to 11.8)	0.24	0.021

Models adjusted for age, sex, gestational age, FEV1/FVC, brain size, and modifiable risk factors (smoking, alcohol, blood pressure, body mass index). The point estimate refers to the magnitude of change in the exercise variables per unit change in thalamic volume, vigorous physical activity, left end diastolic volume and left inferior temporal cortical thickness.

Thalamic volumes and cortical thickness over the left inferior temporal area are independently associated with measures of cardiovascular performance at peak exercise in multivariable analysis (Table 6.6). Cortical thickness extracted from the vertices over the right inferior orbital area of the frontal cortex were removed from the model as they are not independent from thickness over the inferior temporal cortical area. Thalamic volume and cortical thickness over the left inferior temporal area are

positively, and independently, associated with peak oxygen uptake, peak wattage, peak oxygen pulse and oxygen uptake efficiency slope achieved during cardiopulmonary exercise testing (Table 6.6).

Figure 6.6 presents the association between thalamic volume and peak oxygen pulse (0.8 ml/beat per 1 cm^3 increase in thalamic volume (95%CI 0.3 to 1.3, p=0.039, r^2=0.02). Cortical thickness over the left inferior temporal area was the only variable independently associated with peak heart rate, 2 bpm per 0.1 mm increase in cortical thickness (95% CI 0.5 to 3.3, p=0.01, r^2 =0.05). Thalamic volume was the only independent variable associated with heart rate recovery (1.4 bpm per 1 cm^3, (95% CI 0.5 to 3.3, p=0.01, r^2 =0.05). Associations adjusted for age, sex, gestational age, FEV1/FVC, brain size, and modifiable risk factors (smoking, alcohol, blood pressure, body mass index).

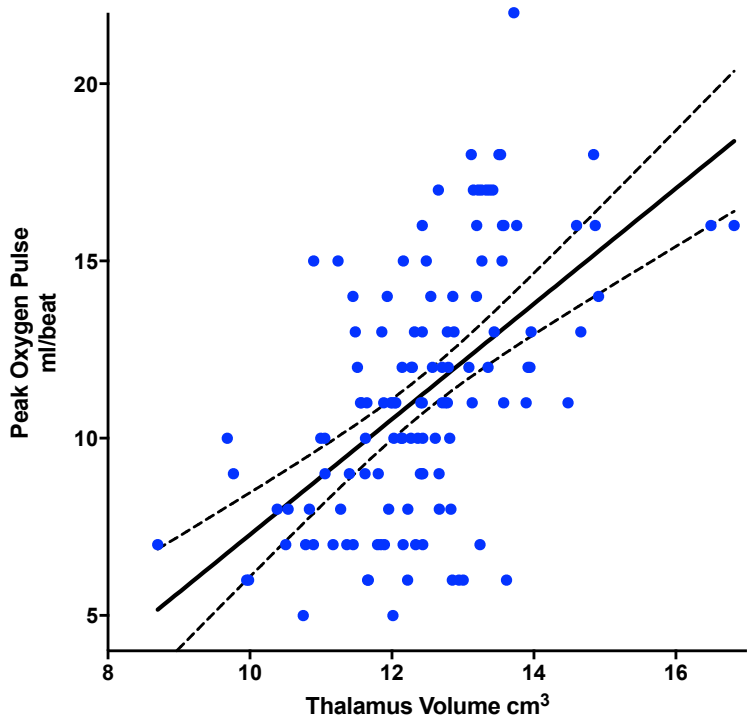


Figure 6-6 Association between oxygen pulse measured at peak exercise and total thalamic volume

6.6.5 Cortical and Thalamic Associations with Submaximal Cardiac Response during Cardiopulmonary Exercise Testing and Stress Exercise Echocardiography

Table 6.7 Thalamic volume and inferior temporal cortical thickness associations with submaximal cardiovascular measures reported at the ventilatory anaerobic threshold

Oxygen Uptake (ml/kg/min)	Point Estimates, (95% CI)	P value	R^2
Vigorous Physical Activity	1.5 (0.53 to 2.4)	**0.003**	**0.05**
Left End Diastolic Volume	0.18 (0.09 to 0.26)	**<0.0001**	**0.082**
Thalamic Volume	0.3 (-0.7 to 1.3)	0.610	0.001
Left Inferior Temporal Cortical Thickness	6.95 (1.2 to 12.6)	**0.018**	**0.03**
Wattage (Watts)			
Vigorous Physical Activity	6.86 (0.48 to 13.2)	**0.04**	**0.023**
Left End Diastolic Volume	1.1 (0.53 to 1.65)	**<0.0001**	**0.075**
Thalamic Volume	4.3 (-3.0 to 11.0)	0.213	0.008
Left Inferior Temporal Cortical Thickness	44.2 (6.75 to 81.6)	**0.021**	**0.03**
Heart Rate (bpm)			
Vigorous Physical Activity	-0.35 (-4.24 to 3.54)	0.86	0.0003
Left End Diastolic Volume	0.043 (-0.30 to 0.38)	0.80	0.001
Thalamic Volume	-2.0 (-6.0 to 2.0)	0.34	0.008
Left Inferior Temporal Cortical Thickness	15.4 (-7.4 to 38.2)	0.18	0.015
Oxygen Pulse (ml/bpm)			
Vigorous Physical Activity	0.58 (0.19 to 0.98)	**0.004**	**0.02**
Left End Diastolic Volume	0.12 (0.09 to 0.16)	**<0.0001**	**0.11**
Thalamic Volume	0.52 (0.09 to 9.4)	**0.017**	**0.013**
Left Inferior Temporal Cortical Thickness	2.29 (0.98 to 6.53)	0.053	0.008

Models adjusted for age, sex, gestational age, FEV1/FVC, brain size, and modifiable risk factors (smoking, alcohol, blood pressure, body mass index). Units: vigorous physical activity, hours per week, left ventricular end diastolic volume, ml/m^2, thalamic volume ($1cm^3$), inferior temporal cortical thickness mm^2. The point estimate refers to the magnitude of change in the exercise variables per unit change in thalamic volume, vigorous physical activity, left end diastolic volume and left inferior temporal cortical thickness.

In multivariable analysis of submaximal cardiopulmonary exercise measures recorded at the ventilatory anaerobic threshold (Table 6.7), thalamic volume was only independently associated with oxygen pulse with 0.52 ml/beat increase per 1 cm^3 increase in thalamic volume (95%CI 0.09 to 9.4, p=0.0017, r^2=0.013). Unlike the peak exercise associations, left inferior temporal cortical thickness was not associated with submaximal oxygen pulse or heart rate at the ventilatory anaerobic threshold. Left inferior temporal cortical thickness was independently associated with oxygen uptake (0.7 ml/kg/min per 0.1 mm of cortical thickness) and wattage (4.4 watts per 0.1 mm of cortical thickness) recorded at the ventilatory anaerobic threshold (Table 6.7).

Multivariable analysis of sub-maximal exercise stress echocardiography was completed to explore cardiac associations with thalamic volume and cortical thickness over the left inferior temporal area. Indexed cardiac output during 3-minute exercise intervals at 40% (mean 3204 ml/m^2 (SD 1074), 60% (mean 3589 ml/m^2 (SD 1230), and 80% (mean 4014 ml/m^2 (SD 1241), of peak exercise intensity was positively associated with thalamic volume (Table 6.8). The relative association increased with exercise intensity with thalamic volume explaining 9% of the variance in cardiac output at 40% exercise (0.27 ml/m^2 per 1 mm^3 thalamic volume, 95%CI 0.08 to 0.47, p=0.008, r^2=0.09) and 14% of the variance at 80% exercise intensity (0.36 ml/m^2 per 1 mm^3 thalamic volume, 95%CI 0.06 to 0.66, p=0.006, r^2=0.14). The thalamic associations were independent of cortical thickness over the left inferior temporal area, which was positively associated with cardiac output at 40% (1.7 ml/m^2 per 1µm cortical thickness, 95%CI 0.42 to 3.0, p=0.01, r^2=0.09) and 60% exercise intensity (1.7 ml/m^2 per 1µm cortical thickness, 95%CI 0.1 to 3.4, p=0.04, r^2=0.07).

Table 6.8 Associations of thalamic volumes and inferior frontal cortical thickness with indexed cardiac output measured during stress echocardiography at intervals of 40%, 60% and 80% exercise intensity

	Indexed Cardiac Output (ml/m^2) Point Estimate (95% CI)	P value	R^2
40% Exercise Intensity			
Thalamic Volume (mm^3)	0.27 (0.08 to 0.47	**0.008**	**0.09**
Inferior Frontal Cortical Thickness (µm)	1.7 (0.42 to 3.0)	**0.01**	**0.09**
60% Exercise Intensity			
Thalamic Volume (mm^3)	0.35 (0.11 to 0.61)	**0.006**	**0.12**
Inferior Frontal Cortical Thickness (µm)	1.7 (0.1 to 3.4)	**0.038**	**0.07**
80% Exercise Intensity			
Thalamic Volume (mm^3)	0.36 (0.06 to 0.66)	**0.008**	**14.0**
Inferior Frontal Cortical Thickness (µm)	1.2 (-0.71 to 3.3)	0.199	0.04

Cardiac output indexed for body surface area. Associations presented from multivariable analysis adjusted for age and sex. Associations remain consistent with additional adjustment for gestational age and ideal cardiovascular health score. The point estimate refers to the magnitude of change in the cardiac output per unit change in thalamic volume and left inferior temporal cortical thickness.

Peak systolic longitudinal strain, at rest and prior to exercise testing had a mean value of -19.7% (SD 2.9). During the first exercise stress interval at 40% exercise intensity, which was approximately 30 minute after the peak exercise cardiopulmonary exercise test, the mean strain was higher than baseline with a value of -21.5% (SD 3.1), longitudinal strain then declined with subsequent stress exercise bouts (60% intensity -20.7% (SD 3.2), 80% intensity -19.8% (SD 2.8)). Thalamic volume was associated with the peak strain achieved at 40% exercise and the relative increase in peak longitudinal strain between baseline and 40% exercise intensity (-0.001% of mean difference in strain per mm^3, 95% CI -0.002 to -0.0002, p=0.014, adjusted for age and sex). The difference in strain was equivalent to 5% mean difference in strain per 100 mm^3 of thalamic volume. However, as exercise testing progressed thalamic volume was associated with lower strain patterns. Strain at 60% exercise intensity compared with 40%, was 0.001% lower per mm^3, 95% CI 0.00009 to 0.002, p=0.03. There was similar trend at 80% exercise with thalamic volume associated with strain 0.0008% lower per mm^3 (95% CI -0.00002 to 0.002, p=0.057) compared with strain achieved at 40% exercise intensity in the first interval of stress echocardiography. There were no observed associations between inferior frontal cortical thickness and peak longitudinal strain. There were no observed thalamic or cortical thickness associations with ejection fraction, or E to A ratio on exercise stress testing.

6.7 Discussion

6.7.1 Cortico-Thalamic Regulation of Cardiac Function

This study has combined multimodality MRI imaging, cardiopulmonary exercise testing and stress exercise echocardiography to identify cortical and thalamic associations with cardiovascular function at rest and during exercise. Thalamic volume was associated with resting cardiac MRI measures of left ventricular end diastolic volume, stroke volume, cardiac output, myocardial strain and ejection fraction. In analysis of cortical thickness, thalamic volume and cardiovascular indices of stroke volume and exercise function co-located to cortical areas over the precuneus and posterior cingulate areas. Cortical thickness analysis adjusted for thalamic volume identified common patterns with exercise parameters derived from cardiopulmonary exercise test. Peak oxygen uptake held the broadest associations in multivariable analysis, positively related to cortical thickness clustered over the frontal, temporal and insular areas. In multivariable vertices analysis, cortical thickness over the left inferior temporal and right frontal inferior orbital areas were independently associated with peak oxygen uptake. Furthermore, thalamic volume and inferior temporal cortical thickness were independent predictors of submaximal and peak exercise performance and cardiac output measures during stress echocardiography. This is the first study to use deep cardiovascular phenotyping with multimodal imaging to report cortical-thalamic associations in young adults. The observed patterns of cortical thickness associated with cardiac function are similar to previous reports focused on cardiovascular fitness, heart rate response and physical activity[86,88,115,272].

This is the first study to identify cortical thalamic associations with dynamic imaging measures of cardiac function. The use of stress echocardiography provides a unique

insight into the relationship between potential cortical thalamic pathways and dynamic cardiovascular function. Thalamic volume and inferior frontal cortical thickness were independently associated with submaximal cardiac output. Thalamic volume was also independently associated with myocardial strain on exercise echocardiography. The variance in cardiac output attributable to thalamic volume increased with intensity while the mean differences in strain between baseline and intervals of exercise at 40%, 60% and 80% were proportional to thalamic volume. During 40% exercise, peak longitudinal strain was higher relative to the baseline strain. Increase in myocardial strain is expected with increased exercise intensity. However, during further 3 minute intervals of exercise at 60% and 80% intensity, myocardial strain declined compared to baseline. This pattern is consistent with previous observed patterns of myocardial strain post strenuous exercise and ultra-endurance events and would suggest that during the 60% and 80% exercise intervals the participants' myocardium was still in metabolic recovery and displayed a degree of myocardial fatigue accumulated during the previous exercise bouts[292]. Activation of the thalamic-cortical pathways including the precuneus and posterior cingulate may regulate tolerance to fatiguing exercise and higher workloads[92,98,123]. The posterior cingulate and precuneus areas, have previously been observed to predict exercise adherence and tolerance to different stress environments[98,286,293]. This may explain the observation of increased cardiac output despite decline in myocardial strain, suggesting ability to go beyond a state of myocardial fatigue to achieve higher exercise performance.

Previous observational and experimental studies have demonstrated activation of thalamic nuclei during acute exercise and psychological stressor-response that has suggested a role of the thalamus in central autonomic control. Thalamic perfusion and metabolic activity have been observed to increase with acute exercise. In addition,

under direct stimulation, thalamic activation leads to an increase in heart rate and blood pressure[84,86,88,93,94,100,104,272] with similar patterns observed in clinical populations undergoing deep brain thalamic stimulation[84,86,88,93,94,100,104,272]. The patterns of cortical and thalamic associations observed in the current study may relate to excitatory and inhibitory regulation of myocardial activity via central autonomic regulation[108,128]. This study was not designed to measure thalamic or cortical autonomic function but changes in volume and thickness are proposed to reflect altered neuronal structure and integrity[259,260,262,275]. Therefore, larger thalamic volumes and the increased cortical thickness, in association with cardiac parameters, may reflect increased capacity to up-regulate and down regulate autonomic control proportional to exercise intensity.

Recent animal models suggest that central autonomic control and cardiac vagal activity are primary determinants of cardiac contractility and exercise capacity[128]. In a parallel human study, the same authors argued that heart rate recovery was a measure of vagal tone and central autonomic function that predicted exercise capacity[128]. The observation in the current study that thalamic volumes are also positively related to heart rate recovery suggest that the mechanism for thalamic regulation of cardiac function and performance may relate to central autonomic control and vagal tone. Vagal nuclei have been demonstrated in animal models to project to the thalamus[105,106]. Therefore, the observations in the current study may describe the tertiary levels of a complex heart-brain axis with multiple regulatory interfaces from local myocardial innervation and contractility, rising through the vagus to the brain stem, basal ganglia and higher functional neural networks.

A number of neural networks have been identified to be involved in cardiovascular and autonomic control[85,116,119]. The thalamus, posterior cingulate, frontal and insular areas

are neural hubs within default, somatosensory and interoceptive networks demonstrated to have distinct patterns of functional connectivity related to cardiopulmonary function[98,102,114,115]. These areas have been identified in a number of independent studies as cortical areas related to exercise and physical activity[102,294]. Evidence from functional connectivity studies in endurance-trained athletes identify patterns of functional connectivity that describe optimised anticipation of exertion and breathlessness that may build capacity and readiness to sustain effort[98]. The thalamus may provide an anatomical neural hub, which regulates functional connectivity through cortical-thalamic loops to regulate cardiovascular and exercise related pathways[108,109]. The precuneus and posterior cingulate cortex are known projections of thalamic nuclei with high structural and functional connectivity linking the thalamus and precuneus with resting default and functional somatosensory and interoceptive networks[295]. Experimental studies provide insight of how the functional neural networks related to cardiopulmonary function and exercise capacity may develop and remodel across the brainstem and cortical thalamic tracts.

In one recent study, sympathetic stimulation of locus coeruleus, a neural hub of the central autonomic network, enhanced thalamic selectivity and cortical-thalamic transmission with associated improvement in sensory processing[110]. Separate animal studies have also demonstrated distinct cortical thalamic pathways with preparatory activation to anticipate motor function[108,109]. There is also evidence that the thalamus is a circuit hub that helps to maintain persistent activity within frontal and motor thalamocortical pathways to enhance functional anticipation and predict behavioural response patterns[108,109]. The sympathetic drive associated with the workload at increased exercise intensity and persistent exposures with exercise training may provide the stimuli to drive remodelling across these pathways to improve exercise

tolerance and cardiovascular performance. Therefore, cortical thickness and higher thalamic volumes may describe anatomical remodelling associated with improved ability to anticipate and respond to exercise stimuli and provide higher tolerance of negative stimuli to facilitate increased time to exertion.

The available evidence supports the hypothesis for cortical-thalamic regulation across the heart-brain axis that may identify disease targets and explain the parallel life-course associated decline in heart and brain health. Case reports of thalamic stroke, with isolated loss of thalamic function have also been associated with acute heart failure and Takotosubo-type conditions[267]. At rest this study identifies disproportionate changes in apical circumferential strain associated with lower thalamic volumes. This is a pattern described in left ventricular disorders such as Takotosubo; thought to be linked with altered emotional inhibitory control, known functions of the thalamus and wider limbic system[268,296,297]. The current findings could reflect a degree of disordered control of the augmentation in cardiac function in association with lower thalamic volumes required to achieve exercise demands. If so, the causality in the observed associations between heart failure, brain atrophy and reduced cortical thickness in older adult populations may be bidirectional[72,74,298].

6.7.2 Interpretation of specific pattern of anatomical effects on brain imaging

The clustered cortical areas associated with measures of cardiovascular function and exercise capacity are all within the default mode network and salience networks[279]. There is further specificity in the network location when considering the cortical vertices that are significantly associated with left ventricular stroke volume, left end diastolic volume and cardiovascular performance measures. These vertices over the precuneus, cingulate cortex, lingual, calcarine, temporal and frontal cortex are all

located in the brain's "rich club" of cerebral hubs[278]. Rich club hubs are identified as the most densely interconnected regions in the cortex. Functional MRI studies exploring the rich club connectome have identified that all sensory and motor networks communicate with the default mode and salience networks through these hubs[299,300]. There is now an emerging body of evidence that supports an argument that the default mode and salience networks function as an intrinsic allostatic-interoceptive system with in the brain[279]. The proposed conceptual model presented by Kleckner and colleagues is that the allostatic-interoceptive system functions as a generative system, anticipating and predicting ascending sensory inputs from all physiological systems including the autonomic and cardiovascular systems[301]. The functional purpose of the network is anticipation and regulation of energy requirements for bodily systems and functions[302]. In the current study, the observation that higher cardiovascular function is association with increased cortical thickness in cortical hubs represented in Kleckner's allostatic-interoceptive system suggests that the cortical thickness may be an indicator of the brains efficiency to maintain energy regulation and sustain higher cardiovascular function.

The observation that the majority of cardiovascular associations with cortical thickness are mediated through thalamic volume highlights the central role of the thalamus in both the allostatic-interoceptive system and the rich club network[278,279]. Furthermore, the recent discovery that the thalamus contributes to anticipatory function and pre-excitation of cortical networks prior to initiation of energy expenditure highlights a functional role of the thalamus within the allostatic-interoceptive system[108,109,303]. An allostatic role of the thalamus is supported by the observation that thalamic volume predicted cardiac output, and higher energy expenditure during exercise in the current study.

6.7.3 Protecting future heart-brain health and relevance of cortico-thalamic function in young adult cardiovascular risk groups

Preservation of thalamic volume and thalamic-cortical connectivity may be targets to protect future heart-brain health[304,305]. Primary prevention strategies and future research are needed to understand how to maintain cardiovascular performance and explore the role of motor function as combined strategies to up-regulate cortical-thalamic loops to protect future brain health and myocardial function. Age associated decline in cardiac function are most noted with measures of left ventricular volume, with 7% or 5.5ml reduction per decade in stroke volume[306]. In the current study, controlling for age, gestational age, sex and brain size, a 7% reduction in stroke volume is seen with each 1 cm^3 reduction in thalamic volume. Age associated decline in thalamic volume increases from early adulthood and progresses at a rate approaching 1% per year and is dependent on cardiovascular risk profile[259,260,262]. Hypertension, metabolic dysfunction and adverse modifiable cardiovascular profiles can accelerate thalamic and cerebral atrophy by 2-to-3 fold, hypertensive older adults have between 15% and 20% greater reduction in thalamic volumes compared to controls[307]. There is also relevance earlier in the life-course; impaired thalamic development[141,142] may be a contributor to previously described preterm cardiovascular phenotypes and young adult cardiovascular risk[129,308]. Further work is required to investigate thalamic-cortical plasticity and cardiac haemodynamics in relation to prematurity and young adult cardiovascular risk.

The cortical thickness patterns associated with cardiovascular function in the current study may identify potential biomarkers of brain reserve and resilience to aging that may be targets for intervention in younger adults with cardiovascular risk[286]. In older

adults posterior cingulate cortex and precuneus display potential for neuroplasticity in response to exercise intervention and moderate associated improvements in executive and cognitive function[116,293]. Our results suggest these previous observations may partly be mediated through physiological differences in cardiac left ventricular performance and thalamic volume. Thalamic and cortical atrophy correlate with decline in cognitive function and a 1 cm^3 reduction in thalamic volume has been associated with 2-fold increased risk of dementia[259,260,262]. Future research is needed to investigate whether interventions that target neuroplasticity are able to modify cortico-thalamic pathways and whether this has associated benefits for cardiac performance and cortical connectivity.

6.8 Conclusions

In this study there is preliminary evidence of cortical-thalamic associations with resting MRI indices of cardiac structure and function and dynamic stress echocardiography measures of cardiac output and myocardial strain. The study supports a hypothesis that the thalamus is one of the neural hubs associated with cardiac function. The cortical thickness patterns expand on existing understanding of cortical areas related to cardiovascular function and provides evidence of cortical-thalamic regulation of peak exercise capacity. The study strengthens the evidence and understanding of central control of cardiovascular function. There is potential that the identified cardiac-cortical-thalamic pathways may be useful biomarkers to predict and track heart-brain health. Further research is needed to explore cardiac remodelling and neuroplasticity across these pathways and determine clinical importance.

7 Effectiveness of Lifestyle Intervention for cardiovascular risk reduction in young adults: Systematic Review and Meta-analysis

7.1 Abstract

Previous studies report benefits of exercise for blood pressure control in middle age and older adults, but longer-term effectiveness in younger adults is not well established. We performed a systematic review and meta-analysis of published randomized control trials with meta-regression of potential effect modifiers. Strict inclusion and exclusion criteria were applied to screen 9524 titles. Eligible studies recruited younger adults with a cardiovascular risk factor (with at least 25% of cohort aged 18 to 40 years); the intervention had a defined physical activity strategy and reported blood pressure as primary or secondary outcome. Meta-analysis included 14 studies randomizing 3614 participants, mean age 42.2+/-6.3 (sd) years. At 3 to 6 months, exercise was associated with a reduction in systolic blood pressure of –4.40 mmHg (95%CI, -5.78 to -3.01) and in diastolic blood pressure of -4.17 mmHg (95%CI, -5.42 to -2.93). Intervention effect was not significantly influenced by baseline blood pressure, body weight or subsequent weight loss. Observed intervention effect was lost after 12 months follow-up with no reported benefit over control, mean difference in systolic blood pressure -1.02 mmHg (95%CI, -2.34 to 0.29) and in diastolic blood pressure –0.91 mmHg (95%CI, -1.85 to 0.02). Current exercise guidance provided to reduce blood pressure in younger adults is unlikely to benefit long-term cardiovascular risk. There is need for continued research to improve age specific strategies and recommendations for hypertension prevention and management in young adults.

7.2 Introduction

Current National Institute of Clinical Excellence (NICE) guidelines recommend specialist review of young adults with elevated blood pressure to exclude secondary causes of hypertension and implement appropriate management[209]. In the absence of significant end organ disease or secondary causes of elevated blood pressure, young adults under 40 years old are encouraged to implement lifestyle changes, and target improvement in ideal cardiovascular risk factors as the first line for hypertension management[148]. However a major limitation of this guidance is that it is based on data from trials in older populations with mean ages greater than 50 years[309,310]. Furthermore, there is evidence that pharmacological management of mild hypertension in young adults may not have any significant health benefits and is associated with increased adverse events[154]. To reduce burden of hypertension and prevent early stroke and cardiovascular disease in young adult population guidelines need to expand to account for gaps in the current evidence base for management of young adult hypertension and cardiovascular risk.

Physical activity and exercise promotion as both a prevention and intervention strategy to protect heart and brain health is endorsed at multiple levels across society and healthcare. The sources of the evidence base to support public health guidelines for cardiovascular risk reduction are largely longitudinal population studies[311,312]. The reliance on population data creates a number of problems for translation into practical intervention. These include: limited understanding of specificity of intervention components; lack of consensus of outcomes markers; limited understanding of interaction across outcome markers and physiological mechanisms; limited understanding of heterogeneity in response patterns; and persistent disconnect between physiological and behavioural paradigms for interventions which leads to lack

of sustainable efficacious intervention. As outlined in Chapter 1 within in the current randomised control trial exercise literature there are a number of potential modifiers of intervention effect which are not consistent across age groups including baseline physical activity, barriers to participation and physiological training adaptations[313,314]. Therefore, we performed a new systematic review and meta-analysis to evaluate the quality of the evidence base and effectiveness of exercise intervention to reduce blood pressure in younger adult populations.

7.3 Methods

7.3.1 Systematic Review Methods: Search Strategy and Selection Criteria

The study protocol was prepared and published on PROSPERO by the candidate in advance of starting data extraction. Protocol registration: PROSPERO (www.crd.york.ac.uk/PROSPERO/) registration number CRD42014009604. The systematic review was completed in accordance with established methods for Cochrane reviews of physical activity interventions. Methods adhered to the Cochrane Handbook for Intervention Reviews and PRISMA statement. An information specialist (Ms Nia Roberts) from the Oxford University Bodleian Library searched the following databases: Cochrane Central Register of Controlled Trials, MEDLINE & MEDLINE In Process, EMBASE, CINAHL, AMED, PsycINFO, SPORTdiscus, OpenGrey, Science Citation Index & Conference Proceedings Citation Index-Science, ACM Digital Library and IEEE Xplore Digital Library. Cochrane highly sensitive search was used to identify randomized controlled trials. No language or date restrictions were applied. Bibliographies of review articles and selected articles were examined for relevant trials. Literature searches completed up to June 2015.

7.3.2 Systematic Review Methods: Study Selection and Data Extraction

To be included, a study needed to be a randomised control trial with the aim to examine the effectiveness of an intervention that included an exercise/physical activity/cardiovascular fitness component on a primary outcome of blood pressure. The mean population age of the study populations was required to be between 18 to 40 years old or within one standard deviation of this range to be included in the review. The age criteria were to ensure at least 25% of the study population were younger than 40 years old. To be representative of young adults who may be provided advice to manage blood pressure, included studies were required to recruit participants with one or more cardiovascular risk factor, or family history of cardio-metabolic risk. Risk factors included hypertension or pre-hypertension (systolic blood pressure ≥ 120 mmHg and/or diastolic blood pressure ≥ 80 mmHg), overweight (mean body mass index(BMI) > 25 kg/m^2) but not severely obese (BMI ≥ 35 kg/m^2), diabetes, metabolic syndrome, dyslipidaemia, smoking and alcohol consumption. The defined BMI exclusion criteria were based on the understanding that severely obese populations have higher burden of comorbidities and potential functional barriers to exercise participation that makes them a unique target audience. Studies examined the effectiveness of interventions with defined exercise, physical activity or cardiovascular fitness components. The comparator was a control group exposed to placebo, no and/or minimal intervention. Blood pressure was reported as a primary or secondary outcome after a minimum follow-up of 3 months. Studies were required to have over 80% complete follow-up data analysing the results by intention-to-treat or, if not applying intention to treat, ensuring less than 20% study attrition.

Titles and abstracts were screened independently by paired co-authors for the review (The candidate Dr Wilby Williamson (WW), Dr Hamish Reid (HR), Dr Adam Lewandowski (AL), Dr Paul Kelly (PK)). Two authors (WW, HR) independently reviewed full text articles and extracted data. Study inclusion was agreed by consensus in discussion with other authors (Dr Charlie Foster (CF), Prof Paul Leeson (PL)). Missing or ambiguous data was clarified with the corresponding author.

The risk of bias was only assessed and reported for studies that met the inclusion criteria for meta-analysis. The Cochrane Risk of Bias assessment instrument was expanded to include risk of bias assessment specific for physical activity and blood pressure interventions. Two authors (WW, HR) assessed the risk of bias. Where there was disagreement between review authors in the risk of bias assessment, a third author (CF or PL) was asked to independently appraise the study and discrepancies were resolved by consensus between authors.

Studies were assessed for the five general domains of bias: selection, performance, attrition, detection, and reporting. Risk of bias scores were allocated for:

1. Allocation sequence generation
2. Allocation concealment
3. Incomplete outcome data
4. Selective outcome reporting
5. Comparable groups at baseline
6. Contamination between groups
7. Validated blood pressure measurement protocol
8. Outcome measure applied appropriately
9. Final analysis adjusted for baseline blood pressure levels
10. Outcome assessment that was independent and blinded
11. Intention-to-treat analysis

Where sufficient information was available, each domain was identified as “high” or “low” risk of bias. Where there was a lack of information or uncertainty over the potential for bias, we described the domain as “unclear”[315]. We judged the studies overall as having a “low”, “medium”, or “high” risk of bias given consideration of the study design and size, and the potential impact of the identified weakness noted in the table for each study. The assessment of risk of bias and quality of included RCTs was then summarized using the GRADE approach[316].

7.3.3 Systematic Review Methods: Statistical Analysis

Studies were analysed using mean and standard deviation (SD) of outcomes expressed in the original papers. Clinic blood pressures, measured at rest, were reported across all studies and used as the outcome measures. The effect size was expressed as the mean difference between the post-intervention values of the randomized groups. If required imputed standard deviations were calculated from standard errors and confidence intervals using methods described in the Cochrane handbook[316]. When studies investigated multiple interventions arms, intervention arms inclusive of exercise were included as individual intervention strata. Mean values were plotted with associated error bars using forest plots. Statistically significant results were identified as confidence intervals excluding a null effect and p value<0.05. Heterogeneity was assessed through examination of the forest plots and quantified using the I^2 statistic. I^2 statistics were graded according to Cochrane interpretation (>75% considerable/large heterogeneity). In accordance with Cochrane guidance, reporting bias was assessed by plotting a funnel plot of intervention effect on blood

pressure[317,318]. To improve homogeneity, outcome data for the funnel plot was used for the earliest available follow-up, which was 3-6 months in the majority of studies.

Meta-analysis was completed according to Cochrane methods, using RevMan version 5.2 statistical software[316,319]. A random-effects model was the default to incorporate heterogeneity between studies, the inverse variance method was used to calculate the overall effect and standard error[320].

Planned subgroup analysis of included studies was completed according to the following co-variants: 1) Baseline blood pressure, 2) Baseline weight 3) Delivery method, whether exercise was self-directed or supervised 4) Estimated contact time between participants and intervention, 5) Target intensity of exercise, 6) Change in weight following intervention.

Meta-regression analysis was performed using the Wilson (2010) SPSS macro using IBM SPSS Statistics for Windows, Version 22.0. Meta-regression was performed using a random effects model to examine whether study level covariates (potential effect modifiers) predict intervention effect on systolic and diastolic blood pressure between studies at 3 to 6 months follow-up. A priori the following factors were agreed for inclusion in the meta-regression model; 1) Mean arterial blood pressure (MAP) combining systolic and diastolic blood pressure, 2) Estimated contact time between participants and intervention 3) Target exercise intensity during intervention 4) Effect of intervention on weight loss calculated as the standard mean difference between intervention and control post intervention to allow comparison between studies reporting change in weight and body mass index.

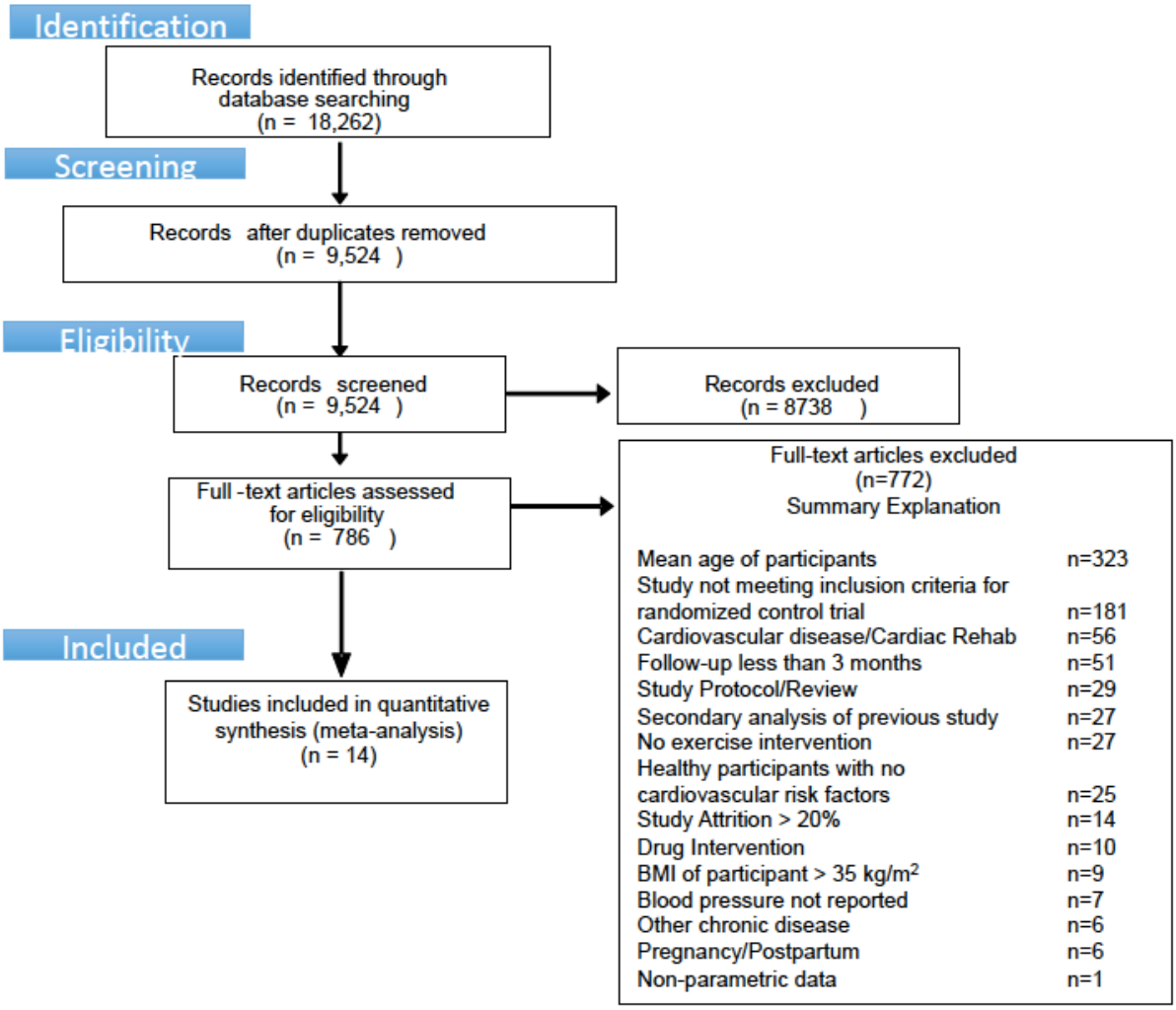


Figure 7-1 PRISMA flow diagram describing the screening and selection of studies for inclusion in meta-analysis

7.4 Results

7.4.1 Results of Search

A total of 9524 titles and abstracts were screened and 786 full text articles reviewed. 14 randomised control trials (RCTs) were identified with 20 exercise intervention arms for inclusion, published between 1985 and 2015 (Figure 7-1, Table 7.1,). The RCTs randomized 3614 participants with a recognised cardiovascular risk factor, mean age 42.2 years (SD 6.3). Over 75% of participants were Caucasian and only 35% were

female. The studies recruited participants with elevated blood pressure and hypertension. The majority of stage 1 hypertensives were not medicated at time of intervention, one study included participants that continued with antihypertensive prescription (n=15).

The major reason for study exclusion was age of population outside inclusion criteria (n=323) (Figure 7-1). 158 of these studies reported blood pressure as a primary outcome, of these, 73 studies excluded participants below 35 years old. None of the excluded cardiovascular studies performed subgroup analysis separating intervention effects by age. Other common explanations for study exclusion included non-RCT design or lack of true exercise control arms (n=181), or study objectives focused on acute or short term exercise response, primarily in healthy participants (n=51).

Table 7.1 Baseline description of study population participating in RCTs included in meta-analysis

Study	Mean Age	Age Range	Female (%)	Weight kg	BMI kg/m^2	Intervention Group mmHg		Control Group mmHg	
						SBP	DBP	SBP	DDP
Duncan 1985	30.0	21-37	0	86 (14)	.	146 (6)	94 (4)	145 (6)	93 (4)
Stamler 1989	37.0	30-44	13	84 (11)	.	122 (7)	82 (3)	123 (7)	83 (3)
Blumenthal 1991 Aerobic Exercise	45	29-59	38	82 (13)	27	141 (9)	96 (6)	142 (12)	96 (6)
Blumenthal 1991 Strength	45	29-59	42	81 (15)	27	143 (10)	95(5)	142 (12)	96 (6)
Stevens 1993 Weight Loss & Exercise	43	30-54	32	90 (13)	30	124 (8)	84 (3)	125 (8)	84 (3)
Whelton 1997 CombinedLifestyle	44	30-54	31	94 (14)	.	127 (7)	86 (2)	127 (6)	86 (2)
Whelton 1997 Weight Loss and Exercise	43	30-54	37	93 (14)	.	128 (6)	86 (2)	127 (6)	86 (2)
Blumenthal 2000 Aerobic exercise	47	>29	54	95 (15)	31	138 (15)	94 (7)	144 (7)	94 (3)
Blumenthal 2000 Exercise and Weight Loss	49	>29	62	93 (18)	32	143 (10)	93 (5)	144 (7)	94 (3)
Tsai 2002	41.0	20-60	45	.	24	134 (12)	85 (10)	138 (8)	92 (8)
Esposito 2003	35	20-46	100	95 (9)	34	124 (9)	85 (5)	123 (8)	85 (5)
Olson 2006	38	24-44	30	78 (6)	28	119 (8)	67 (8)	119 (12)	68 (12)
Kinmonth 2008 In Person	41	20-50	62	79 (16)	28	123 (13)	78 (9)	123 (13)	78 (9)
Kinmonth 2008 Telephone	41	20-50	62	80 (18)	28	124 (13)	79 (11)	123 (13)	78 (9)
Marquez-Celedonio 2009	43	30-55	.	78 (15)	31	133 (4)	88 (3)	133 (4)	86(4)
Knoepfli-Lenzin 2010 Football	37	20-45	0	82 (9)	26	134 (7)	87 (4)	134 (4)	86 (3)
Knoepfli-Lenzin 2010 Running	36	20-45	0	87 (9)	26	136 (9)	87 (5)	134 (4)	86 (3)
Edwards 2011 Aerobic Exercise	46	25-65	50	.	30	141(10)	90 (11)	138(12)	88 (9)
Edwards 2011 Aerobic Exercise and Weight Loss	46	25-65	50	.	31	140(11)	85 (10)	138 (12)	88 (9)
Krustrup 2012	46	31-54	0	98 (14)	30	151 (10)	92 (7)	153 (8)	96 (6)

Mean values presented with standard deviations (sd). Gender distribution presented as percentage of females included. BMI: Body Mass Index (kg/m^2). Missing or unreported values represented as (.).

7.4.2 Description of Included Studies

The majority of participants received a combined behavioural intervention with a defined physical activity strategy. 18 intervention exercise arms targeted increase in moderate to vigorous physical activity, 12 intervention arms delivered structured, supervised aerobic exercise programmes in gym and group environments with intensity defined by baseline exercise testing. Seven trial intervention arms promoted self-directed increase in physical activity supported by regular group and individual counselling sessions. Multi-disciplinary professional groups including psychologists, exercise physiologists, and nutritionists delivered behavioural counselling. Contact time with the intervention in the first 3 months ranged from 5 to 48 hours, average 25 hours. The average in the first 3 months for studies reporting 3 to 6 month outcome was 30 hours. The minimum follow-up for inclusion in the present systematic review was 3 months, 10 studies report 3 to 6 months follow-up data (15 intervention arms, n=2716). Five studies complete follow-up at 3 to 4 months and 6 studies report follow-up after 12 months (8 intervention arms, n=3023).

7.2 Description of Interventions delivered

Study	N	Intervention; intensity (heart rate), frequency (days) x time (minutes).	Strategies & Delivery	Contact (hours)	Follow up
Duncan 1985[321]	66	Aerobic exercise. Intensity 70-80% max heart rate determined from peak testing. 3 x 75 min session per week	Supervised exercise. Walk-Jog programme. Graduated increase in running	72	4
Stamler 1989[322]	201	Multifactorial intervention, biweekly counselling. Nutritional hygiene. Diaries and lifestyle advice. Moderate activity (70%), 3x30mins heart rate 70% of max.	Self-directed exercise. Individualised delivery. Physician and nutritionist lead. Friends & Family members for planning	40	60
Blumenthal 1991[323]	92	1. Aerobic Exercise intervention: 3 x 35 minute walk-jog 70% HR. 2. Strength and flexibility: 20 minutes flexibility, 3x 30 minutes circuit training	Supervised exercises, personal trainer	40	4
Stevens TOHP Phase 1 1993[324]	564	Lifestyle behavioural intervention, diet and exercise. 14 weekly 90-minutes group sessions, followed by month meeting for 18 months. Graduated walking (40-55% HR), 5x45mins session per week.	Self-directed and supervised group exercise. Facilitators: Nutritionists, psychologists and exercise physiologists. Self-management plans, self-monitoring and self-efficacy training	43	6 & 18
Whelton TOHP Phase 2 1997[325]	1788	1. Weight Loss and Exercise, 2. Weight Loss, Salt Restriction and Exercise: Graduated brisk walking Moderate walking (40-55% HR), 5x45mins session per week. 14 weekly 90 minute groups sessions followed by month meeting for 18 months].	Self-directed and supervised exercise. Facilitators: Nutritionists, psychologists and exercise physiologists. Weekly progress review and feedback. Self-management plans with activity self-monitoring and self-efficacy training to prevent relapse.	60	6 & 36
Blumenthal 2000[326]	133	1. Supervised Aerobic Exercise; Intensity 70-85% HR, 4 x 35 mins sessions,10 min warm-up/cool-down. 2. Supervised aerobic exercise plus weight management: as above plus 26 weekly group sessions discussing weight management	Supervised exercise. Trained Exercise Physiologist. Heart rate monitoring	95-120	6
Tsai 2002[327]	42	Supervised Aerobic Exercise, Treadmill Walk-Jog programme: Moderate Intensity (60-70%), 3 x 30 mins. 10 min warm-up/cool-down	Supervised exercise. Facilitated by trained exercise providers.	30	3
Esposito 2003[328]	120	Lifestyle behavioural intervention targeting diet and exercise. Month session for 12 months followed by bimonthly session for further 12 months. Moderate Daily walking	Self-directed exercise. Facilitated by nutritionists and exercise trainer Goals 10% weight loss.	24 to 48	24
Olson 2006[329]	30	Supervised and self-directed exercise and resistance training. 2 sessions per week for 12 months. First 12 weeks supervised, Moderate to high intensity	Supervised exercise. Self-monitoring with training logs. Feedback from exercise trainer twice monthly	38	12
Kinmonth 2008[330]	365	Self-directed exercise with behavioural coaching, 1. Telephone follow-up; home visit followed by 6 x 15-45 minute calls in 5 months followed by monthly letter. 2. In person intensive follow-up; 4 x 1 hour visits, 2 x 15 minute calls in 5 months followed by monthly 30 minute calls. No defined intensity, individualised graduated increase in activity	Self-directed. Facilitators: behaviour counsellors. Goal setting, action planning, self-monitoring, goal review, using prompts, family support, prevention plans.	5 to 8	24
Marquez-Celedonio 2009[331]	81	Multifactorial intervention. Diet and low sodium advice with supervised exercise advice. Moderate intensity (60-80% HR), 3 x 45 mins supervised aerobic exercise plus weekly group exercise (basketball, volleyball, soccer).	Supervised exercise. Heart rate monitoring.	100	6
Knoepfli-Lenzin 2010[332]	47	1. Aerobic Running; 3 x 60 minute training session per week, 65-85% HR. 2. Football; 10 minute warm up, 50 minutes 5 aside football, Heart rate above 65%	Supervised exercise. Coaching support Heart rate monitoring	36	3
Edwards 2011[333]	52	1. Aerobic Exercise; Two supervised sessions per week and encouragement to exercise for 3 more sessions. 2. Diet plus Aerobic Exercise: Target 500-1000 kcal decline in energy intake. Moderate Intensity (60-75% HR), 3 x 60 mins	Supervised exercise. Heart rate monitoring Food diaries Weekly telephone call.	48	3
Krustrup 2012[334]	33	Supervised football. Exercise trainer, 5 minutes warm-up/cool down. Moderate to vigorous intensity (75-85% HR), 2 x 60 mins per week	Supervised exercise Heart rate monitoring	48	6

7.4.3 Risk of Bias and Quality Assessment

The funnel plot of intervention effect on systolic blood pressure was symmetrical about the mean effect size line suggesting there was no particular study publication bias (Figure 7-2).

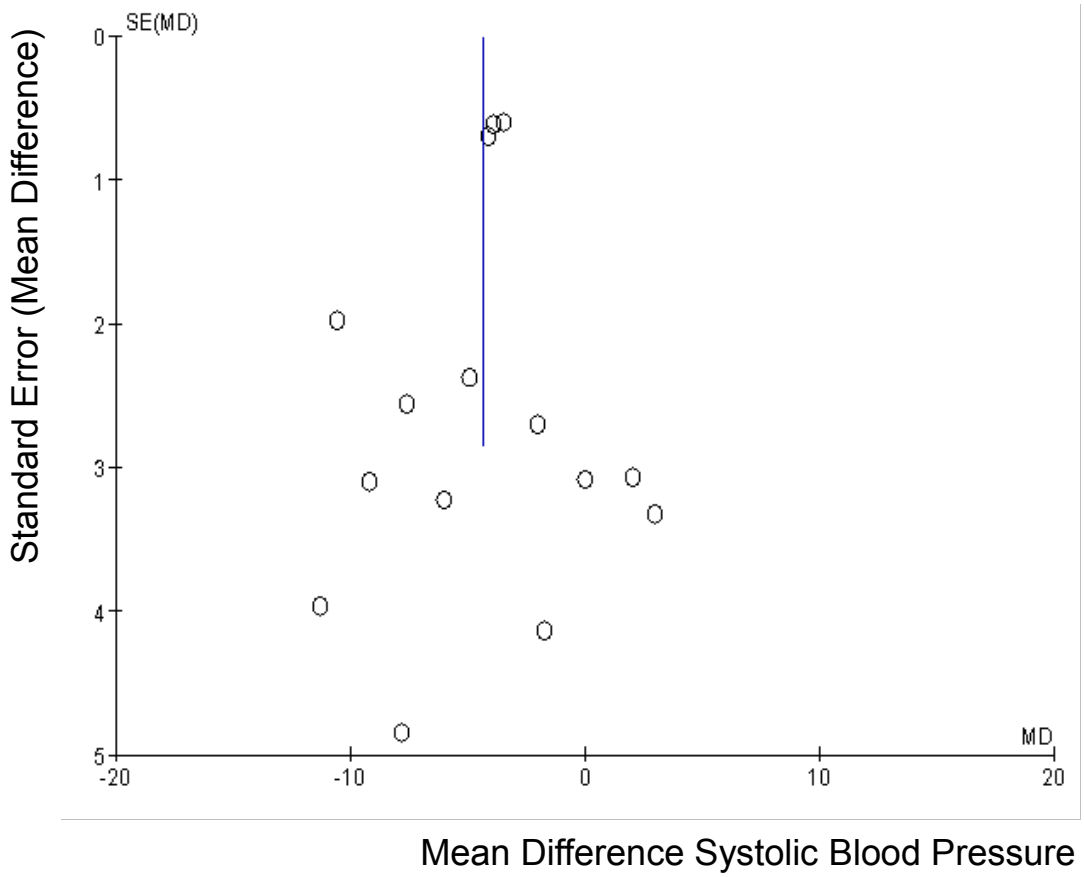


Figure 7-2 Funnel plot of intervention effects on systolic blood pressure exploring publication bias with reported trial intervention arms at 3 to 6 months follow-up

Overall quality of the included RCTs using the GRADE approach suggests moderate quality data (Figure 7-3). In total, nine studies are downgraded secondary to study design and outcome reporting being unclear or at risk of bias with limited reporting of participant allocation methods and lack of clarification of blinding during outcome assessment. A significant limitation of the included studies was the lack of reference to published study protocols that adhere to the TIDieR template[335]. Only the Trials of Hypertension Prevention (TOHP) and ProActive UK studies consistently reference published study protocols[324,325,330] (Table 7.1).

Study	Allocation Sequence Generation	Allocation Concealment	Incomplete Outcome Data	Selective Outcome Reporting	Comparable Groups at Base-Line	Contamination Between Groups	Outcome Measures Applied appropriately	Outcome Measures Repeated for regression to the mean	Final Analysis Adjusted for Baseline Blood Pressure	Outcome assessment independent and Blinded	Intention To Treat	Overall Risk of Bias
Duncan 1985	Unclear	Unclear	Low	Low	Low	Unclear	Low	HIgh	Low	Unclear	Unclear	Medium
Stamler 1989	Computer assisted	Unclear	Low	Low	Low	Unclear	Low	Low	Low	Low	Low	Low
Blumenthal 1991	Unclear	Unclear	Low	Low	Low	Low	Low	Low	Low	Unclear	Unclear	Medium
Stevens 1993 TOHP Phase 1	Low	Low	Low	Medium – PA self-report not disclosed	Medium-Gender difference	Low	Low	Low	Low	Low	Low	Low
Whelton 1997 TOHP Phase 2	Low	Low	Low	Medium – PA self-report not disclosed	Low	Low	Low	Low	Low	Low	Low	Low
Blumenthal 2000	Unclear	Unclear	Low	Low	Medium	Low	Low	Low	Low	Unclear	Low	Low
Tsai 2002	Unclear	Unclear	High	HIgh	Low	Low	Low	High	Low	Unclear	High	High
Esposito 2003	Low	Low	Low	Low	Low	Low	Unclear	Unclear	Unclear	Low	Low	Low
Olson 2006	Low	Unclear	Low	Low	Low	Low	Low	Low	Low	Unclear	Unclear	Low
Kinmonth 2008	Low	Low	Low	Low	Low	Low	Low	Unclear	Low	Low	Low	Low
Marquez Celedonio 2009	Unclear	Unclear	High	Low	Low	Low	Unclear	Unclear	Unclear	Unclear	High	High
Knoepfli-Lenzin 2010	Unclear	Unclear	High	Low	Low	Low	Unclear	Unclear	Unclear	Unclear	High	High
Edwards 2011	Low	Low	Low	Low	Low	Low	Low	High	Low	Unclear	Low	Low
Krustrup 2012	Low	Low	Low	Low	Low	High	Low	Low	Low	Unclear	Low	Low

Figure 7-3 Risk of bias and quality assessment of included trials

7.4.4 Participant Compliance and Intervention Effect of Weight and Cardiovascular Fitness

The majority of studies reported over 80% participant compliance with intervention at 3-6 months, recorded as attendance at supervised classes and group meetings, or achievement of behavioural targets such as self-reported minutes of activity. However, compliance with behavioural targets dropped to an estimated 40% beyond 12 months.

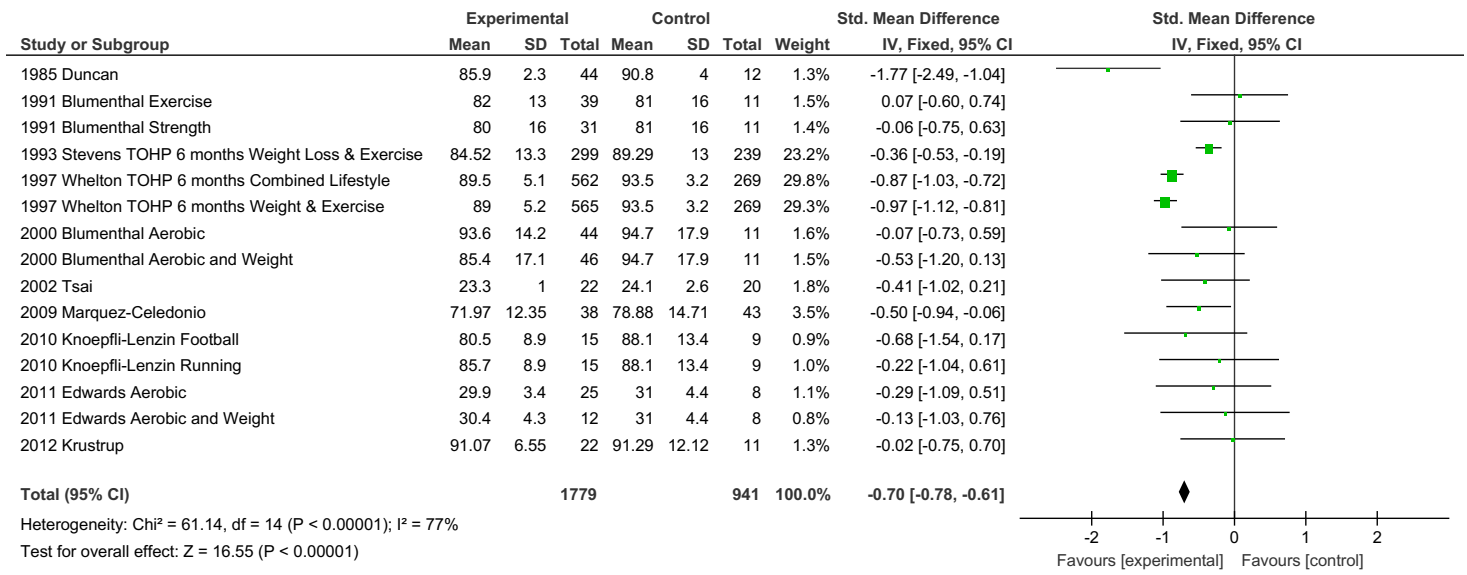


Figure 7-4 Standard mean difference intervention effect on weight loss between control and exercise group at 3 to 6 months follow-up

Dietary assessment was undertaken using self-report diaries over periods of 1 to 7 days. The majority of studies use diary cards as aids to behaviour change as opposed to assessing compliance; only three studies report the change in dietary intake. Mean cardiovascular fitness was reported from 14 intervention arms, the median increase was 12% improvement in peak exercise capacity (range 3% to 30%). Eight intervention arms deliver exercise in combination with weight management, 4 of these interventions report a significant reduction in weight loss compared to control at 3-6 months (Figure 7-4)

7.4.5 Intervention Effect on Blood Pressure

Forest plots for mean differences in systolic and diastolic blood pressure after 3-6 months of intervention are presented in Figure 7-5. Mean difference in systolic blood pressure was -4.40 mmHg (95%CI, -5.78, -3.01) and -4.17 mmHg (95%CI, -5.42, -2.93) for diastolic blood pressure when intervention was compared with control.

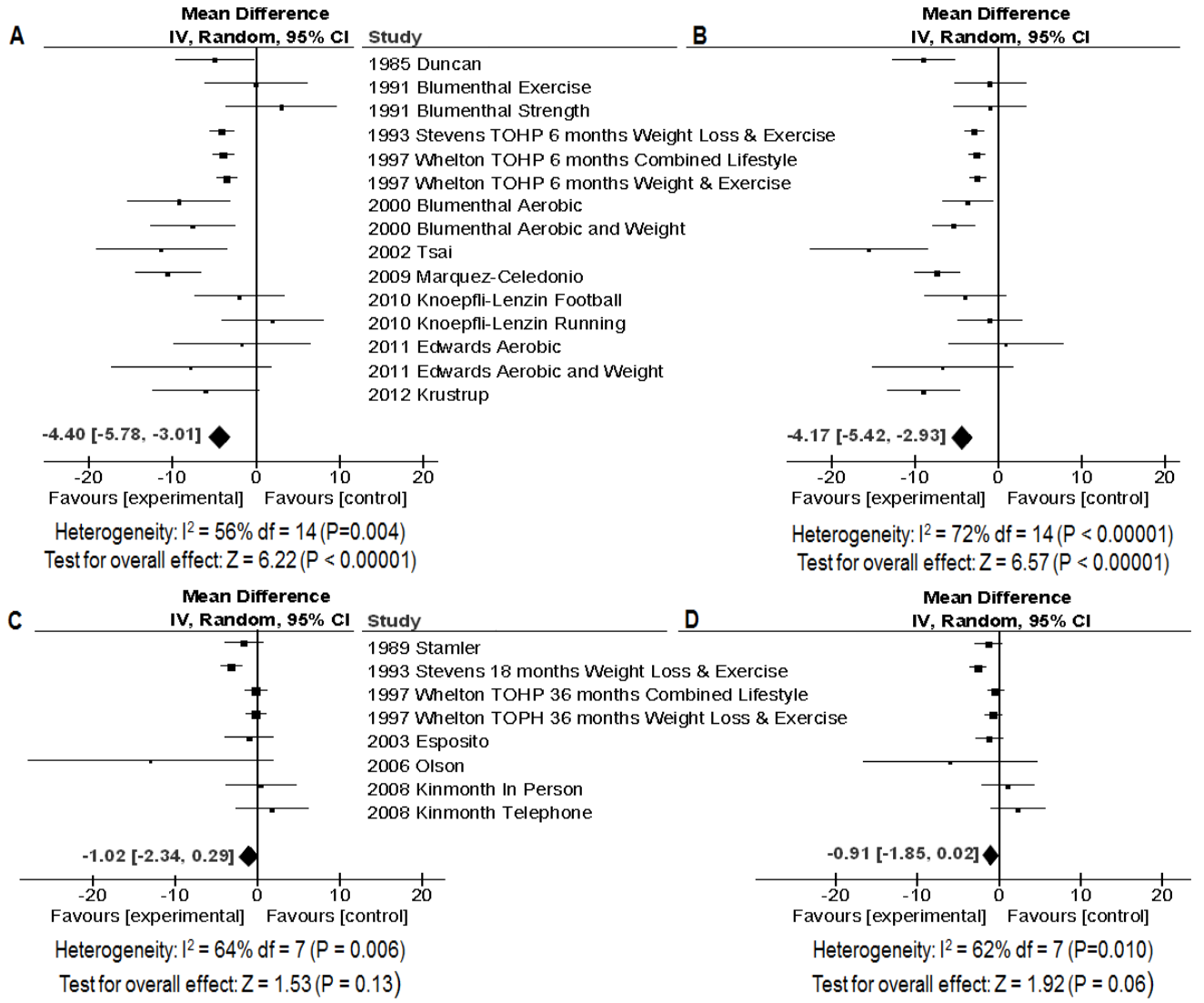


Figure 7-5 Forest plot demonstrating mean difference in systolic and diastolic blood pressure following exercise intervention.

Forest plot presents mean difference in systolic (A&C) and diastolic (B&D) blood pressure (mmHg) after 3 to 6 months and at, or beyond 12 months follow-up. Results for individual exercise intervention arms reported when available. Squares represent mean difference between intervention and control post intervention with 95% CIs, size of the square proportional to the weight of the study; pooled estimates from meta-analysis are depicted as solid black diamonds.

There are no significant differences between intervention and control group blood pressures when followed up at, or beyond, 12 months (Figure 7-5). Mean difference in systolic blood pressure -1.02 mmHg (95%CI, -2.34 to 0.29) and mean difference in diastolic blood pressure was –0.91 mmHg (95%CI, -1.85 to 0.02).

7.4.6 Subgroup Analysis of Effects of Intervention on Systolic and Diastolic Blood Pressure According to Study Level Characteristics

I^2 Statistic identified moderate to considerable heterogeneity across the studies (56% to 72%) at 3 to 6 month follow-up. Subgroup analysis did not provide a consistent explanation for heterogeneity between studies for both systolic and diastolic blood pressure. I^2 Statistic reduced to below 45% for systolic blood pressure when analysis was restricted to hypertensive groups, groups with baseline weight above 90kg and self-directed intervention (Table 7.3).

Supervised aerobic exercise, higher exercise intensity and increased contact time with intervention were associated with larger reductions in systolic and diastolic blood pressures (Table 7.3). Reduction in diastolic blood pressure was significantly greater when comparing supervised (-5.43 mmHg [95%CI, -7.58, -3.28]) with self-directed exercise (-2.64 mmHg [95%CI, -3.20, -2.08]) and when intervention targeted higher intensity compared to moderate intensity.

Table 7.3 Subgroup analysis of effects of intervention on systolic and diastolic blood pressure according to study level characteristics

Group	Arms	N	Systolic BP, mmHg	Diastolic BP, mmHg
Overall Intervention effect≤6months follow-up	15	2716	-4·40 (-5·78, -3·01) I^2=56%, Z=6·22 (p<0·00001)	-4·17 (-5·42, -2·93) $I^{2=}$72%, Z=6·57(p<0·00001)
Intervention effect ≥ 12months	8	3023	-1·02 (-2·34, 0·29) I^2=64%, Z=1·53 (p=0·13)	-0.91 (-1·85, 0·02) I^2=62%, Z=1.92 (p=0·06)
Baseline Weight <90kg	7	815	-3·0 (-6·04, 0·05) I^2 =72%, Z = 1·93 (p=0·05)	-3·88 (-6·13, -1·63) I^2 = 73%, Z=3·38 (p=0·0007)
Baseline Weight ≥90kg	5	1806	-4·23(-5·49, -2·98) I^2= 32%, Z=6·6 (p<0·00001)	-3·69 (-5·09, -2·30) I^2=69%, Z=5·2 (p<0·0001)
Baseline SBP<140mmHg and DBP <90mmHg	7	2370	-4·41 (-6·06, -2·77) I^2=69%, Z=5·25 (P<0·00001)	-3·87 (-5·33, -2·41) I^2=77%, Z=5·20 (P<0·00001)
Baseline SBP ≥140mmHg and or DBP≥ 90mmHg	8	346	-4·35 (-7·26, -1·44) I^2=42%, Z=2·93 (P=0·003)	-4·55 (-6·91, -2·19) I^2=62%, Z=3·78 (P=0·0002)
Aerobic Supervised Exercise (follow-up≤6months)	11	475	-5·40 (-8·08, -2·72) I^2=56%, Z=3·95 (p<0·0001)	-5·43 (-7·58, -3·28) I^2=67%, Z=4·95 (P<0·00001)
Self-directed physical activity (follow-up≤6months)	3	2199	-3·81 (-4·52, -3·09) I^2=0%, Z=10·39 (p<0·00001)	-2·64 (-3·20, -2·08) I^2=0%, Z=9·18 (p<0·00001)
Weight Loss >4kg	6	1586	-5·03 (-6·89, -3·17) I^2=65%, Z=5·31 (p<0·00001)	-4·77 (-6·54, -2·99) I^2=79%, Z=5·27 (p<0·00001)
Weight Loss ≤4kg	6	1236	-2·61 (-5·77, 0·55) I^2=61%, Z=2·07 (p=0·11)	-2·95 (-4·76, -1·13) I^2=53%, Z=3·18 (p=0·001)
Moderate exercise intensity ($\leq 60\%$)	5	2265	-3.40 (-4.59, -2.21) I^2 =50%, Z= 5.60 (P<0.0001)	-2.58 (-3.13, -2.03) I^2 =0%, Z= 9.14 (P<0.00001)
Moderate to Vigorous intensity (>60%)	10	451	-6.19 (-8.64, -3.73) I^2 =43%, Z= 4.93 (P<0.0001)	-5.92 (-8.09, -3.76) I^2 =64%, Z= 5.36 (P<0.00001)
Contact time less than 60 hours	9	806	-2.83 (-5.33, -0.34) I^2 =46%, Z= 2.23 (P=0.03)	-3.91 (-6.33, -1.49) I^2 =67%, Z= 3.16 (P=0.002)
Contact time 60 hours or more	6	1910	-5.61 (-7.55, -3.67) I^2 =69%, Z= 5.68 (P<0.00001)	-4.57 (-6.22, -2.92) I^2 =80%, Z= 5.42 (P<0.00001)

Results at 3 to 6 month follow-up used unless otherwise stated. Mean differences are pooled estimates from meta-analysis with 95% CIs. I^2 values reported as measure of heterogeneity

Blood pressure reductions appeared greater in association with over 4 kilogram weight loss, a weight loss threshold identified from previous systematic review[336]. However,

observed differences did not reach significance (systolic BP -5.03 mmHg [95%CI, -6.89, -3.17] vs -2.61 mmHg [95%CI, -5.77, 0.55] and diastolic BP -4.77 mmHg, [95%CI, -6.54, -2.99] vs -2.95 mmHg [95%CI,-4.76, -1.13]). There were no significant differences in intervention effect when groups were separated as hypertensive or pre-hypertensive at baseline (Table 7.3).

7.4.7 Meta-regression – Effective Intervention Components

Table 7.4 Meta-regression model comparing effect of study level covariates on post intervention systolic (model 1) and diastolic (2) blood pressure (mmHg) compared with control

	Model 1. Systolic Blood Pressure			**Model 2. Diastolic Blood Pressure**		
Variable	**$\beta_1 \pm$ SE**	**CI (95%)**	***Z(p)***	**$\beta_1 \pm$ SE**	**CI (95%)**	***Z(p)***
MABP (mmHg)	0.3161 ± 0.1625	-0.0230, 0.6346	1.95 (0.052)	-0.0255 ± 0.1966	-0.4108, 0.3599	-0.13 (0.897)
Hours of Contact	-0.0718 ± 0.0336	-0.1376, -0.0060	-2.14 (0.032)	0.0192 ± 0.0404	-0.0601, 0.0985	0.47 (0.635)
Exercise Intensity	-0.1458 ± 0.0601	-0.2636, -0.0281	-2.43 (0.015)	-0.1275 ± 0.0724	-0.2695, 0.0144	-1.76 (0.078)
Weight Loss	-0.9610 ± 1.6473	-4.1897, 2.2677	-0.58 (0.560)	2.1510 ± 1.9962	-1.7615, 6.0635	1.08 (0.281)

Model statistics for systolic blood pressure $R^2 = 0.50$, Q =16.4, df 4.0, p=0.0025.
Model statistics for diastolic blood pressure $R^2 = 0.23$ Q =4.95, df 4.0, p=0.293.
Notes: MABP Mean Arterial Blood Pressure at baseline (mmHg); $\beta_1 \pm$ Standard Error; CI (95%), 95% confidence intervals; *Z(p)*, *z*-score and alpha value.

The a priori meta-regression model explained 50% of variance in intervention effect on systolic blood pressure. Increased intensity of exercise and hours of contact with the intervention were significantly associated with reduction in systolic blood pressure (Table 7.4). Baseline mean arterial blood pressure and standard mean difference in weight loss between exercise and control groups post intervention were not significant

predictors of mean reduction in systolic blood pressure. The a priori model did not provide significant explanation for variance in diastolic blood pressure response.

7.5 Discussion

7.5.1 Effectiveness of Lifestyle intervention for Blood Pressure Reduction

This is the first systematic review to apply age criteria to evaluate the RCT evidence base to promote exercise to prevent and manage hypertension in younger adults. In the short-term (3 to 6 months), exercise and physical activity interventions are beneficial, with between 4 to 5 mmHg reductions in blood pressure. This is a larger effect than reported from previous review reporting results from older adult groups and pre-hypertension groups[310]. This may be explained by selection criteria, we excluded normotensive cohorts with no cardiovascular risk factors. Alternatively, the result may suggest that younger adults may be more responsive to exercise as an intervention to lower blood pressure. There were also comparable benefits in blood pressure reduction for both elevated blood pressure and hypertension groups which has not been observed previously in older adult groups[310].

A significant finding of this review is the observed lack of sustained effects in blood pressure reduction after 12 months. This is the first review in the blood pressure literature to explore the sustained effects of exercise intervention and the first to exclude studies with less than 3 months follow-up, which may have previously contributed to overestimation of effect[310,337]. The findings of the current review are similar to patterns observed in the general physical activity literature with a longitudinal decline in compliance with maintaining physical activity.

Current guidelines recommend review of adults under 40 years of age with elevated blood pressure for exclusion of secondary causes of hypertension[209]. The age inclusion criteria for this review were defined to align with this practice. However, a major limitation is the paucity of studies recruiting younger adults. Hypertension in younger adults is complicated by high rates of under-diagnosis and, when identified, sub-optimal treatment[337-339]. These deficiencies may reflect broader misconception that younger age is sufficiently protective against cardiovascular risk[337-339]. This pattern is reflected in this review with an observed age bias for study recruitment in favour of older adults. The majority of excluded trials recruit cohorts over 50 years of age. In addition, over 46% of studies reporting blood pressure as a primary outcome excluded participants under 35 years of age.

7.5.2 Potential Benefits with reference to findings from the YACHT study

Improved risk evaluation and interpretation of the benefits of blood pressure reduction may facilitate discussion on how to reform hypertension management for younger adults. An example is clinical interpretation of the reported 4 to 5 mmHg reduction in blood pressure, if this was sustained in a younger adult cohort with elevated blood pressure, the estimated 5 year incidence of hypertension would reduce from 1 in 5 to 1 in 10. This interpretation may be more beneficial than prediction of 10 year risk of cardiovascular events which is difficult in younger adults[340]. However, long-term benefits on cardiovascular endpoints can be estimated; a sustained 2 mmHg reduction in blood pressure could translate to 7% to 10% reduction in stroke and ischaemic cardiovascular event[341]. The major challenge is how to achieve sustained effect. In

this review intervention effect dropped to 1 mmHg by 12 months with no significant difference compared to control.

7.5.3 Future Research Requirement

To provide a platform to improve future intervention design an evaluation of study level characteristics that predict intervention effect at 3 to 6 months was completed. With regards intervention strategy and delivery, both supervised and self-directed exercise achieve reduction in blood pressure, although effect was greater with supervised exercise. This may reflect a dose effect; supervised exercise was associated with increased exercise participation in the short-term. This group also achieved higher exercise intensity and increased cardiovascular fitness. However, higher volumes of planned contact time between participant and intervention, irrespective of intensity or self-directed exercise, was also associated with greater reduction in blood pressure at 3 to 6 months. Explanation for the subgroup analyses may relate to distinctions between physiological and behavioural influences of intervention. The exposure to higher exercise intensity may drive a mechanism for change in blood pressure distinct from low-level activity. Self-directed and lower intensity exercise had relatively lower effects on diastolic blood pressure which is consistent with previous observations that blood pressure responses differ with intervention strategy[310,336].

The finding that contact time, independent of intensity is associated with a positive influence on systolic blood pressure may support a beneficial effect of increased frequency of low to moderate activity. However, interpretation is limited without objective and repeated measures of physical activity behaviour, which was not

reported in studies with 3 to 6 month outcomes. Alternatively, planned contact, inclusive of telephone and remote contact may be a stimulus for sustained behaviour. Unfortunately, the pattern across studies is that as contact is withdrawn intervention effect declines. This is despite several studies implementing recommended behaviour strategies such as promotion of participant self-efficacy, activity planning, self-monitoring and participant feedback[313,314]. Participant motivation and self-efficacy are of particular relevance as, despite the low attrition rates, the included studies report decline in scheduled attendance and compliance with intervention targets from over 80% at 3 to 6 month to 40% at 18 months. There are currently no strategies that effectively address the challenge of promoting sustained long-term physical activity behavioural change. A promising approach is personalised intervention supported by device assisted behavioural change and flexible communication strategies to better sustain effective intervention[313]. The use of wearable activity monitors and physical activity tracking applications on mobile device can provide objective measures of behaviour, facilitate self-monitoring and allow real-time feedback. However, the resource demands of maintaining high contact time and technology supported behaviour change may be a barrier to clinical translation. Evaluation of effective interventions with reference to delivery cost and process evaluation of strategies to sustain participant engagement, motivation and compliance may help to drive innovation and overcome these barriers.

Improvement in intervention design and delivery may also benefit from more transparency and disclosure of the specific methods and content of delivered communication strategies. There were often only brief summaries available, which described the professional team, if communication training was provided to the team, and broad categorical descriptors of intervention themes discussed with participants.

In the current review, a number of studies focused communication strategies around weight loss, promoting exercise as a mechanism for weight loss. However, an interesting observation from the review that may help to guide the evolution of future studies is the patterns of intervention effect associated with weight loss. Previous review identified that weight loss greater than 4kg was required to achieve significant blood pressure reduction[336]. However, in our review, short-term benefits of exercise on blood pressure were seen even in those who did not achieve this degree of weight loss. This observation is supported by weight loss not being a significant effect modifier in the regression model. The positive message is that in the short term, exercise is beneficial for blood pressure reduction independent of pre or post intervention weight. Distinguishing between the independent benefits of exercise and weight loss may facilitate effective communication and participant engagement strategies, especially when participants may be motivated by different health and well-being goals.

7.5.4 Recommendation

This review raises concern that current clinical practice to promote lifestyle and exercise intervention risks suboptimal management of young adult hypertension. Although it has been pragmatic to assume that exercise will improve blood pressure in young adults, the available evidence suggests current intervention strategies do not maintain long-term benefit. Discussion with young adult patient and public groups highlight that lifestyle intervention remains an attractive alternative to starting potential lifelong prescriptions for blood pressure. However, short-term reduction in blood pressure reported in this review involved multiple contacts over time and delivery of targeted exercise prescription. These strategies generally required supervised exercise interventions which are expensive and currently not widely supported[342]. A

major challenge for the clinical research community is to design and evaluate interventions that target sustained increase in physical activity behaviour, accommodate potential for titration of exercise prescription and deliver improvement in the cost per quality adjusted life year. Translation of research findings into clinical practice may be improved by study design incorporating comparative adaptive evaluations and exploring interactive effects with prescription medication. Going forward there appears to be a need for strategic overhaul of the approaches implemented in the prevention and management of young adult blood pressure.

7.6 Conclusions

Current exercise guidance and best practice for lifestyle intervention to reduce blood pressure in younger adults is unlikely to benefit long-term cardiovascular risk. There is need for continued research to improve age specific strategies and recommendations for hypertension prevention and management in young adults. There is need for co-ordinated and strategic engagement for across multi-disciplinary sectors to drive improvements in the prevention and management of hypertension in early and mid-life. The current population trends in prevalence and burden of hypertension and the increase in low-to-middle income counties suggested a potential crisis for global health unless action is taken. The emerging evidence of potential negative effects of pharmacological management in mild hypertension and younger adult populations highlights that more research is needed across the bio-psycho-social spectrum of medical intervention. There is an urgent need for innovation in design and delivery of lifestyle intervention and renewed pharmacological research to identify alternative pharmacological intervention for management of cardiovascular risk in young and middle age adults.

8 Future Work: Trial of Exercise for Prevention of Hypertension in Young Adults a Study Protocol

8.1 Abstract

Hypertension prevalence in young adults has increased and is associated with increased incidence of cerebrovascular and cardiovascular events in middle age. However, there is significant debate regards how to effectively manage young adult hypertension with recommendation to target lifestyle intervention. Surprisingly, no trials have investigated whether lifestyle advice developed for blood pressure control in older adults is effective in these younger populations.

TEPHRA is an open label, parallel arm, randomised controlled trial in young adults with high normal and elevated blood pressure. The study will compare a supervised physical activity intervention consisting of 16 weeks structured exercise, physical activity self-monitoring and motivational coaching with a control group receiving usual care/minimal intervention. 200 young adults aged 18-35 years, including a subgroup of preterm born participants will be recruited through open recruitment and direct invitation. Participants will be randomised in a ratio of 1:1 to either the exercise intervention group or control group. Primary outcome will be ambulatory blood pressure monitoring at 16 weeks with measure of sustained effect at 12 months. Study measures include multimodal cardiovascular assessments; peripheral vascular measures, blood sampling, microvascular assessment, echocardiography, objective physical activity monitoring and a subgroup will complete multi-organ magnetic resonance imaging.

The results of this trial will deliver a novel, randomised control trial that reports the effect of physical activity intervention on blood pressure integrated with detailed cardiovascular phenotyping in young adults. The results will support the development of future research and expand the evidence-based management of blood pressure in young adult populations.

Trial Registration: Clinicaltrials.gov registration number NCT02723552, registered on 30 March, 2016.

8.2 Introduction

Globally, modifiable risk factors have the largest attributable risk to cardiovascular events and stroke [45] and current guidelines recommend targeted lifestyle intervention, and in particular physical activity promotion, for young adults with hypertension [343]. However, the systematic review of exercise intervention trials for blood pressure, discussed in chapter 7, identified bias in recruitment towards older adults, lack of investigation of determinates of physiological and behavioural exercise remodelling and limited understanding of the cardiovascular adaptations associated with change in modifiable risk profiles [344]. Therefore, there is limited trial evidence to support physical activity intervention as a primary treatment option in younger adults [345]. In addition, at younger ages, a range of factors including birth history, familial predisposition and socio-economic exposures have influences on cardiovascular and cerebrovascular phenotypes that may make response to lifestyle interventions more complex [131,143,346]. This means there is lack of understanding regards effectiveness of intervention across subgroups of the population and potential heterogeneity in response to exercise training [130,131,143,308,347-350]. Therefore, this study will trial an

exercise intervention in a young adult population with elevated and high blood pressure to identify effectiveness for management of blood pressure and also investigate determinates of behavioural and physiological remodelling as well as differential effects across phenotypes.

8.3 Study Aims and Objectives

The primary aim of the TEPHRA study is to compare the effect of a structured aerobic exercise and physical activity intervention versus usual care/minimal intervention on ambulatory blood pressure levels in young adults with elevated or high blood pressure. Secondary aims include investigation of the broader effects of exercise on peripheral and central cardiovascular remodelling, metabolic function, physical activity behaviours, and physical function. Tertiary aims target an exploratory analysis of remodelling across the liver, heart and brain using MRI indices of structure and function to explore the direction of associations identified in chapters 4 to 6. In addition, the study will explore the potential that baseline phenotypes, such as the cortico-thalamic association discussed in chapter 6, predict exercise response.

8.4 Hypotheses

The primary hypothesis is that structured aerobic exercise training and physical activity self-monitoring will improve ambulatory blood pressure measures for young adults with elevated blood pressure. The secondary hypotheses are that: structured aerobic exercise training and physical activity self-monitoring will also improve other cardiovascular, metabolic and cerebrovascular risk profiles in young adults; that the degree of improvement will associate with changes in vascular biology and behaviour, as potential mediators of the changes; and thirdly that response to exercise will vary depending on baseline phenotypic differences between individuals.

8.5 Study Design

TEPHRA is an open label, prospective single-blinded, two-arm, parallel, randomized controlled trial in young adults with elevated blood pressure. In total, 200 participants will be randomized (ratio 1:1) following baseline measures to either exercise intervention or to a comparison control group with minimal intervention. There are two follow-up visits post-randomisation: at 16 weeks and 52 weeks. The primary endpoint will be at 16 weeks post-randomisation, immediately after the end of the intervention.

Participants who enter the trial will nominally complete 4 study visits over the course of the trial:

I. Screening assessment (Visit 0)

II. Baseline Study Visit (Visit 1)

III. 16-week follow-up Study Visit (Visit 2) at 16 weeks (post completion of structured exercise intervention for those in the intervention arm)

IV. 52-week follow-up Study Visit (Visit 3) - (52 weeks post randomisation)

Study measure collection is reduced for Visit 3 to reduce redundant data collection of selected non-primary outcome data and minimise excess burden on study participants.

8.6 Populations

8.6.1 Eligibility

Recruitment strategies will include invitation from general practice (GP) records, invitation from hospital birth registers, open recruitment, targeted online recruitment, and invitation following participation in previous studies. Participants will be verified as preterm (born before 37 weeks gestation) or full-term (born after 37 weeks gestation).

The trial inclusion criteria are listed below.

Eligible participants will:

- be willing and able to give informed consent for participation in the study
- be willing and able to complete intervention and attend all baseline and follow-up study visits at the John Radcliffe Hospital, Oxford, United Kingdom
- be able (in the investigator's opinion) and willing to comply with all study requirements
- be male or female, from 18 to 35 years old
- have verified birth history: preterm birth (<37 weeks) or full-term birth (>37 weeks)
- have the ability to access and use a computer and the internet
- have 24-hour awake ambulatory systolic and/or diastolic blood pressure >115/75 mmHg

The participant may not enter the study if any of the following apply:

- clinic blood pressure >159mmHg systolic and/or 99mmHg diastolic at initial screening
- pregnancy
- evidence of end organ damage secondary to established diagnosis of hypertension

- simultaneous participation in another human or clinical randomized trial (if there is any possibility of compromising health, safety, or well-being, or any possible compromise of study data)
- unable to walk briskly on the flat for 15 minutes
- those currently maintaining levels of cardiovascular fitness and aerobic activity at or above the levels required for the intervention arm
- unable to attend regular supervised exercise sessions
- use of beta-blockers such as atenolol or equivalent
- BMI >35 kg/m^2
- major contra-indications to exercise participation
- evidence of cardiomyopathy
- evidence of inherited cardiac conduction abnormalities
- evidence of congenital heart disease
- other significant chronic disease relevant to cardiovascular status
- a permanent pacemaker

The following are exclusion criteria for the MRI sub-study only:

- shrapnel injuries
- metal clips in blood vessels of the brain
- other metal or electronic implants adversely affected by the magnetic field
- an injury to the eye involving fragments of metal
- unsuitable for MRI based on responses on the MRI safety screening form.

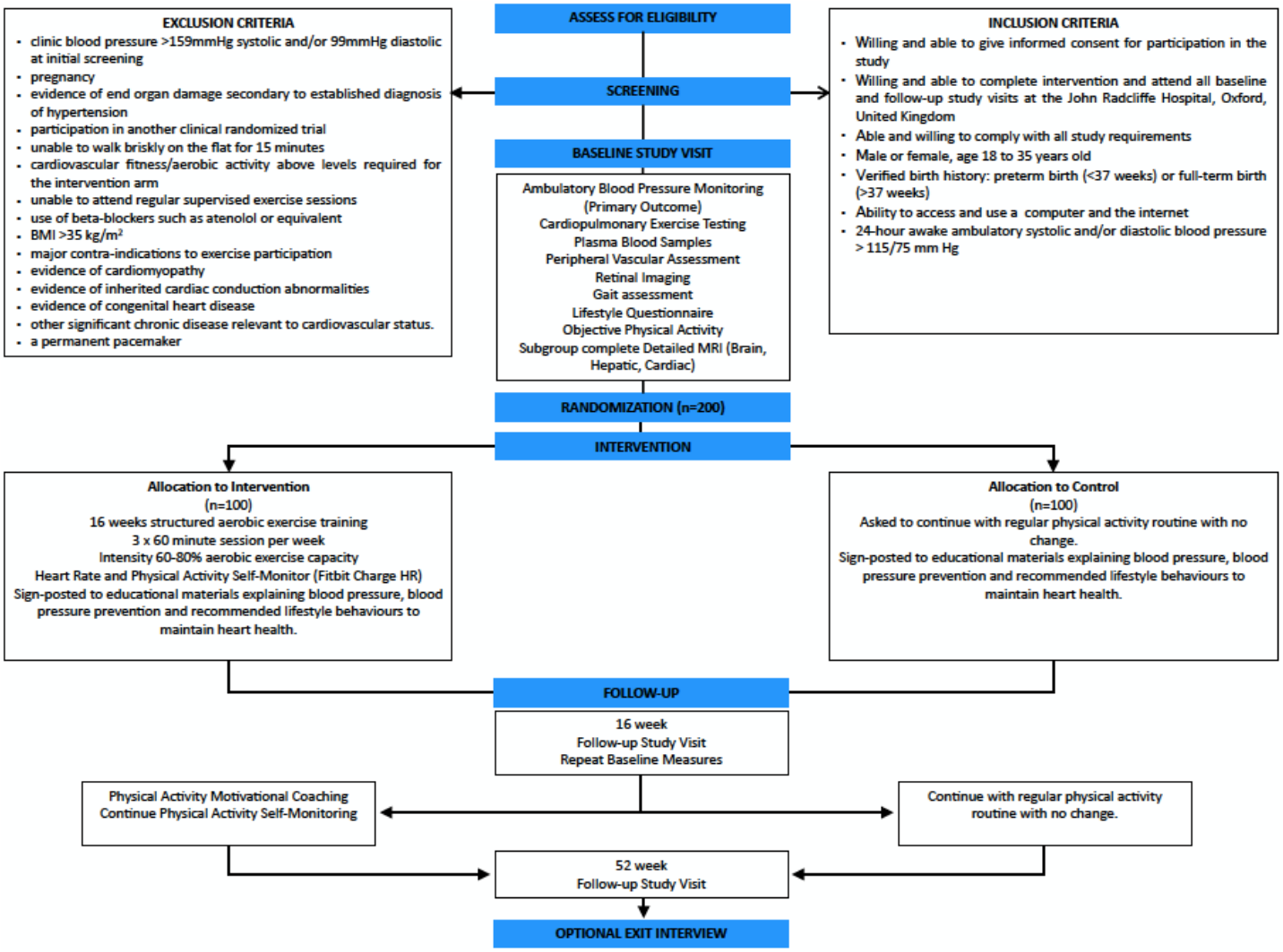


Figure 8-1 TEPHRA Study Flow Diagram

8.6.2 Setting and Participants

Participants will complete a secure, online questionnaire requesting the following information:

- Sex, age at entry to the study, birth history
- Reported pregnancy status

- Medical history to identify exclusionary medical conditions or pharmaceutical usage
- Self-reported current physical activity levels
- Willingness to change physical activity and ability to access and attend supervised exercise sessions for 16 continuous weeks

A trained clinical research investigator will review questionnaire responses and eligibility for study participation. The study investigator will clarify if there are significant contraindications, diseases or disorders, which in the opinion of the investigator might influence the individual's ability to participate in the study. Potentially eligible participants will then be invited to attend a screening visit completed at the Oxford Cardiovascular Clinical Research Facility, University of Oxford.

8.6.3 Informed Consent

Individuals willing to participate in the study will be provided with the full participant information leaflets detailing the study information in advance of the informed consent process. Only after participants have had a minimum of 24 hours to review the study information will they be invited to schedule a time for their screening visit. The informed consent process will be completed on the day of the screening study visit. The participant must personally sign and date the latest approved version of the informed consent form before any study specific procedures are performed. Written and verbal versions of the participant information leaflet and informed consent form will be presented to the participants detailing no less than: the exact nature of the study; what it will involve for the participant; the implications and constraints of the protocol; the known side effects and any risks involved in taking part. It will be clearly stated that the participant is free to withdraw from the study at any time for any reason without

prejudice to future care, and with no obligation to give the reason for withdrawal. Written informed consent will be obtained by study team members who are suitably qualified and experienced, and have been authorized to do so by the principal investigator. A copy of the Informed Consent form will be given to the participant. The original signed form will be retained at the study site.

Candidates will be asked for their consent to access their medical records including birth and neonatal records. In absence of obstetric records, self-reported birth history will be used. As appropriate, participants will be asked to confirm their birth and pregnancy details with appropriate family member(s).

8.6.4 Screening

Anthropometric measurements including body composition measures, height, weight, body mass index (BMI), waist and hip circumference, and resting blood pressure shall be collected. Participants will have their blood pressure checked after 5 minutes rest using the automated mode of a validated sphygmomanometer (Dinamap V100, GE Healthcare, Chalfont St. Giles, United Kingdom). Three blood pressure readings will be taken at intervals of 1 minute. For the outcome measure of clinic blood pressure at screening, the mean of the second and third readings will be used. Screening blood pressure will be assessed based on National Institute for Health and Care Excellence (NICE) guidelines[209] and are presented in **Table 2.**

Electrocardiogram (ECG) - A 12 lead ECG will be conducted and reviewed by a trained clinical research investigator. If the investigator determines that ECG results potentially shows evidence of exclusionary cardiac disease, the ECG (and candidate if appropriate) will be referred for further clinical review.

24 hour Ambulatory Blood Pressure (ABP) - 24-hour ambulatory blood pressure monitoring will be initiated at the end of the screening assessment session using validated, automated oscillometric, ambulatory devices (TM-2430, A&D Instruments or equivalent). Correct cuff size will be chosen based on arm circumference. Subjects will be instructed to remain still during measurements. Measurements will be automatically taken every 30 minutes during daytime and then hourly from 11:00PM to 7:00AM. Subjects will complete a diary documenting hours asleep and awake. Ambulatory blood pressure measures will be verified by a trained study investigator prior to randomisation.

8.7 Objectives and Outcome Measures

Table 8.1 TEPHRA study objectives and outcome measures exploring potential benefits of exercise.

Objectives	Measures	Time-points
Benefits of Exercise		
Short term Blood Pressure Response: The primary objective is to compare the effect of aerobic exercise intervention and physical activity self-monitoring versus usual care/minimal intervention on ambulatory blood pressure levels in young adults with high normal and elevated blood pressure.	Systolic and diastolic blood pressure measured during ambulatory blood pressure monitoring.	Measured at baseline and at 16 week follow-up
Sustained blood pressure response: To compare the sustained effect of aerobic exercise intervention with continued physical activity self-monitoring and motivational coaching versus usual care/minimal intervention on awake blood pressure levels in young adults with high normal and elevated blood pressure.	Systolic and diastolic blood pressure measured during ambulatory blood pressure monitoring.	Measured at baseline, 16 week follow-up, and 52 weeks follow-up
Cardiac Remodelling: Investigate cardiac adaptation and remodelling following intervention, with reference to gestational age and baseline cardiovascular phenotypes.	Cardiac mass. Left and right ventricular structure and function. 3D-shape and functional analysis.	Cardiac MRI Measured at baseline and16 week follow-up; echocardiography measures recorded for all participants at baseline, 16 weeks and 52 weeks.
Cerebrovascular Remodelling: Investigate cerebrovascular adaptation and remodelling following intervention, with reference to gestational age and baseline cerebrovascular phenotypes.	White and grey matter volumes. Subcortical nuclei volumes. Cortical thickness. White matter integrity. White matter hyperintensities. White matter connectivity. Brain vessel morphology. Brain vascular resistance. Brain blood flow and arrival time.	Brain MRI indices measured at baseline and 16 week follow-up
Physical Function: Compare the 16-week and sustained effect of intervention on objectively measured walking gait, walking cadence and global physical function.	Objective measure of physical activity (7 day wear of activity monitor). Objective gait analysis. Questionnaire.	Measured at baseline, 16 week follow-up, and 52 weeks follow-up
Metabolic Function: Compare the effects of intervention on the fasting metabolic profile.	Fasting glucose profile. Fasting insulin profile. Fasting lipid profile.	Measured at baseline, 16 week follow-up, and 52 weeks follow-up
Retinal microvascular structure: Compare the effect of intervention on retinal and dermal vascular structures.	Retinal imaging arteriolar and venular indices.	Measured at baseline, 16 week follow-up, and 52 weeks follow-up

8.7.1 Primary Outcome: 24 Hour Ambulatory Blood Pressure

The primary outcome is 24-hour ambulatory blood pressure. The primary objective is to compare the effect of aerobic exercise intervention and physical activity self-monitoring versus usual care/minimal intervention on ambulatory blood pressure levels after 16 weeks intervention in young adults with high normal and elevated blood pressure. Ambulatory blood pressure was selected as a more sensitive measure of young adult blood pressure than clinic blood pressure. It also provides the opportunity to measure nocturnal blood pressure profiles that had broad associations with heart-brain indices in chapters 4 to 6.

8.8 Secondary Objectives and Outcome Measures

8.8.1 Cardiovascular fitness measured via cardiopulmonary exercise testing

The secondary objectives include comparison of the effect of aerobic exercise intervention and physical activity self-monitoring versus usual care/minimal intervention on cardiovascular performance and dynamic cardiovascular stress response in young adults with high normal and elevated blood pressure, with reference to gestational age. Chapters 4 to 6 present a broad spectrum of potential benefits across the heart-brain axis of maintaining higher cardiovascular fitness. However, the analysis in chapter 6 also highlights that there may be central determinants of peak exercise capacity. There is limited understanding of the dose effects of exercise intervention across the heart-brain axis. Detailed measure of cardiopulmonary function in response to a targeted intervention will help to explore the potential for short-term benefits in peak exercise capacity. Follow-up at 52 weeks will track the sustained effects.

8.8.2 Cardiac remodelling

The detailed cardiovascular phenotyping will combine cardiac MRI imaging in a subgroup and echocardiography in the full study group to investigate cardiac adaptation and remodelling following intervention. Building on the established methods and findings presented in chapters 4 to 6 the TEPHRA study will now explore the directions of association in young adults with elevated and high blood pressure. Cardiac imaging will provide measures of cardiac mass, left ventricular shape and function. There is potential that given recruitment is targeting populations with high and elevated blood pressure that they may already display features of cardiac remodelling such as increased left ventricular mass to end diastolic volume. The age of the populations would suggest they will have the potential for positive remodelling with anticipated increase in left ventricular end diastolic volume with training. However, this required dose of exercise and remodelling potential with reference to gestational age are unknown.

8.8.3 Cerebrovascular remodelling

Brain and Cardiac MRI imaging will be complete on a subgroup of participants. Recruitment will continue to provide a minimum of 100 participants with paired baseline and 16 weeks post-intervention imaging datasets. The objective is to investigate potential for short-term cerebrovascular adaptation and remodelling across the heart brain axis. The analysis will be completed with focus on vessel morphological, vascular haemodynamics and subcortical volumes to expand on the findings presented in chapter 4 and 6. The analysis of the baseline data will provide an opportunity to validate the observed finding in chapters 4 to 6 in a new study population.

8.8.4 Physical Function

The analysis presented in chapter 6 identified cortical thickness over motor areas to be related to cardiovascular function and evidence of a potential cortico-thalamic regulations of submaximal and peak exercise capacity. The relationships between heart-brain function and motor function have not been investigated in young adult populations. The inclusion of physical activity behaviour measures and objectively measured gait and walking cadence will provide an opportunity to investigate evidence of association in young adults at baseline and potential for benefits of intervention at 16 and 52 weeks follow-up.

8.8.5 Mediators of Exercise Adaptation and Blood Pressure Response

Table 8.2 TEPHRA study objectives and outcome measures investigating mediators of exercise adaptation and blood pressure response

Objectives	Measures	Time-points
Mediators of Exercise Adaptation and Blood Pressure Response		
Physical activity behaviours: Compare the 16-week and sustained effect of intervention on objectively measured ambulatory physical activity and sedentary behaviour in young adults with high normal and elevated blood pressure.	Objective measure of physical activity (7 day wear of activity monitor)	Measured at baseline, 16 week follow-up, and 52 weeks follow-up
Molecular endothelial and angiogenic function: Compare the effect of aerobic exercise intervention versus usual care/minimal intervention on circulatory markers of angiogenesis and endothelial colony-forming cells (ECFC) function and association with change in blood pressure and cardiovascular fitness.	Circulatory markers of angiogenic function. ECFC colony growth (before or after 15 days in culture). ECFC function: ECFC proliferation rate and number of branches and closed tubes formed on matrigel.	Plasma samples for circulating biomarkers of vascular endothelial and angiogenic function will be measures at baseline and16 week follow-up.
Vascular remodelling: Compare the effect of aerobic exercise intervention and physical activity self-monitoring versus usual care/minimal intervention on central blood pressures, vascular stiffness and vascular structures.	Pulse wave velocity and augmentation index. Retinal imaging arteriolar and venular indices with retinal imaging.	Structural and functional microvascular measures will be recorded at baseline, 16 week follow-up, and 52 weeks follow-up

8.8.5.1 Physical Activity Behaviours

The TEPHRA study will compare the 16-week and sustained effect of intervention on objectively measured ambulatory physical activity and sedentary behaviour in young adults with high normal and elevated blood pressure. A major limitation identified in the systematic review of the exercise literature is the lack of sustained effect on blood pressure beyond 6 months follow-up. The study will introduce low cost components of behavior change communication and activity self-monitoring that could readily be scaled to a definitive intervention if found to effective.

8.8.5.2 Vascular Remodelling

The integrity of microvascular structure and function may be a common mediator of vascular health across the heart-brain axis that is vulnerable to modifiable risk factors. Exercise intervention may have a positive effect on the microvascular over a short-term intervention. Peripheral vascular assessments of retinal vascular structure and vascular stiffness may also be low cost and scalable assessment of vascular health that could be used as outcome measures in a larger definitive intervention. The TEPHRA study will compare the effect of aerobic exercise intervention and physical activity self-monitoring versus usual care/minimal intervention on central blood pressures, vascular stiffness and retinal vascular structures. These measures will facilitate investigation of the peripheral vascular response to exercise and association with heart-brain indices.

8.8.5.3 Metabolic and Angiogenic Profiles

The completed cross-sectional study presented a number of associations between indices of metabolic health and measures across the heart-brain axis. Young adults with metabolic dysfunction and lower indices of ideal cardiovascular health also were observed to have a higher burden of subclinical vascular impairment across the heart

brain axis. Exercise and physical activity intervention may have a positive impact on indices of metabolic health, which mediate beneficial remodelling on blood pressure and vascular health. Stimulus for favourable angiogenic response in association with exercise may also have favourable effects on the microvascular that lead to reduction in blood pressure. The TEPHRA study will study these potential changes to understand more of the benefits and mediators of exercise associated vascular remodelling. The study will compare the effects of intervention on the fasting metabolic and angiogenic profiles collected from fast blood samples.

8.8.6 Predictors of Response to Exercise and Blood Pressure Change

The systematic review presented in Chapter 7 was able to identify intervention components that were associated with significant reduction in blood pressure in association with exercise. However, there were no beneficial effects of intervention at extended follow-up. Compliance with the intervention and development of sustained behaviour change strategies based on established theoretical templates have been highlighted as required for successful intervention. The TEPHRA study will investigate the potential baseline predictors and determinants of future success or failure of intervention at the individual level.

8.8.6.1 Perceptions of the study and intervention compliance

Baseline questionnaires, self-monitoring, supervised training sessions; diary carding and objective measures of physical activity will be incorporated into a process evaluation of the study. This will include exit interviews of the study participants. These study measures will track compliance with the intervention and characterize participants' subjective and qualitative experience of the intervention, which will be

correlated with intervention effects on blood pressure and cardiovascular fitness to assess efficacy of the intervention.

8.8.6.2 Physical activity beliefs

The participants' prior experience of exercise and the interaction with the participant's acute perception of submaximal exercise may influence an individuals emotional and physical response to intervention. The study will investigate the correlation between physical activity behaviour change and the participants' cognitive and psycho-social determinants of exercise including self-efficacy to maintain physical activity and motivations to exercise. The behavioural intervention will include physical activity self-monitoring with a Fitbit and motivational coaching. It is unknown whether these intervention components may influence perceptions of exercise and physical activity behaviours.

8.8.6.3 Baseline cardiovascular phenotypes

The cross-sectional observation reported chapter 4 to 6 highlight differences in cardiovascular exercise capacity in relation to birth history. The study will now investigate the associations between baseline cardiovascular phenotypes including the preterm born phenotype and response to exercise intervention across outcomes.

8.8.6.4 Baseline cerebrovascular phenotypes

Chapter 6 identified evidence of cortico-thalamic associations with peak exercise capacity. Previous studies have suggested that cortical thickness patterns over the precuneus and posterior cingulate may predict adherence to exercise. This cross-section analysis presented in chapter 6 may suggest that this is secondary to associated subclinical cardiac dysfunction. The TEPHRA study will now investigate these associations further by relating baseline cardiac and cerebrovascular structures;

such as white matter microstructure, subcortical volumes (caudate, thalamus and hippocampus) and cortical thickness (across insular, precuneus and posterior cingualate) and response to exercise intervention.

8.8.6.5 Tertiles of blood pressure and cardiovascular risk

The cross-sectional data presented in chapters 4 and 5 identify changes across the heart-brain axis in association with indices of ideal cardiovascular health. Participants with lower indices of ideal cardiovascular health may benefit from tailored intervention and personalised assessment of outcome measures. The TEPHRA study will investigate differences in response to exercise intervention across outcomes in associations with baseline tertiles of blood pressure and ideal cardiovascular risk scores. These analyses will be hypothesis generating and will inform future research for complex intervention design and delivery.

8.9 Interventions

8.9.1.1 Exercise and Physical Activity Intervention

Participants in the intervention arm will complete 16 weeks of structured aerobic exercise training, with a target of 3 training sessions per week, duration 60 minutes, at exercise intensity of 60-80% aerobic exercise capacity. Exercise intensity of 60-80% maximal aerobic capacity defined using heart rate response during peak cardiopulmonary exercise testing at baseline. Heart rate monitors will be used to maintain training intensity. The intervention replicates similar strategies identified during systematic review of randomised control trials delivering exercise intervention for blood pressure reduction [345]. The minimum dose to start effecting positive changes in cardiovascular fitness is estimated to be 40 minutes of moderate intensity sessions three times per week. The study team will monitor participants' compliance and

progress during the structured aerobic exercise training. The study team will use motivational coaching strategies to encourage participants to maintain compliance with the training program. In addition to structured aerobic exercise training, participants will be provided with a wrist worn heart rate and accelerometer based activity monitor (Fitbit Charge HR) to facilitate physical activity self-monitoring and maintenance of heart rate targets. Participants will be consented to allow investigators access to the data collected from wear of the Fitbit. In addition, participants will be sign-posted to educational materials produced by the British Heart Foundation explaining blood pressure, blood pressure prevention and recommended lifestyle behaviours to maintain heart health.

On completion of the structured aerobic exercise training participants will be encouraged to continue to use their activity monitor and maintain physical activity goals until the final study visit at 52 weeks. The use of the activity monitors will support the three most effective behaviour change strategies to promote increased physical activity, including self-monitoring, goal setting and regular feedback [351,352]. To encourage continued regular, self-directed physical activity, participants will be provided a motivational coaching session on completion of the structured 16 week training program. A study team member trained in motivational interview techniques will provide the motivational coaching. Participants will be asked to reflect on their experience of physical activity, their experience of participating in the structured aerobic exercise training, will be encouraged to set goals and rate their confidence and motivations to achieve physical activity goals, participants will be encouraged to discuss strategies to maintaining physical activity participation.

8.9.1.2 Comparison

Participants in the control group will be sign-posted to educational materials produced by the British Heart Foundation explaining blood pressure, blood pressure prevention and recommended lifestyle behaviours to maintain heart health. Participants will be asked to continue with their regular physical activity routine with no change.

8.10 Statistics

8.10.1 Sample Size

The rationale for the number of participants required to address the primary outcome is as follows. Our systematic review and meta-analysis of randomised control trials [345] identified an average decrease of 5.4mmHg in systolic blood pressure (SBP) post-intervention in adults with elevated blood pressure (mean systolic blood pressure at baseline 126 mmHg) [345]. The post intervention standard deviation of change was 8.7 mmHg. Our available cross-sectional pilot data suggests that change will be higher in the preterm born participants and participants exposed to maternal hypertension during pregnancy. To calculate the sample size we have been conservative and used an estimated change of 5 mmHg following 16 weeks of intervention. We have used a standard deviation (SD) of 11.3 from the pooled SD for ambulatory systolic blood pressure from our cross-sectional pilot data. To observe a treatment effect on systolic blood pressure of 5mmHg, powered to 80% (p=0.05) requires a total sample size of 164 participants. The SD deviation for the pooled ambulatory diastolic blood pressure is 8.3 mmHg. To observe a treatment effect on diastolic blood pressure of 5mmHg, powered to 80% (p=0.05) requires a total sample size of 114 participants. The power calculations for systolic blood pressure are used to determine the final sample size, with adjustment to 200 participants to allow for 18% attrition. To ensure primary

objectives for the study are answered, the study team may recruit additional participants to replace those who drop out and withdraw from the study. This sample size will provide 80% power and 5% significance to analyse the proposed secondary outcomes.

8.10.2 Randomisation and Allocation

Participants meeting the study inclusion criteria will be randomly allocated 1:1 to the two intervention arms. This will be undertaken using Sealed Envelope™ (https://www.sealedenvelope.com/), a computerised randomisation program. A minimisation algorithm (with a random element of 80%) is used to ensure balanced allocation across the two groups for key prognostic factors: gender, age and gestational age of participants. The strata for each prognostic factor are as follows:

Gender: male/female;

Age: <24 years old, 24-29 years old, 30-35 years old;

Gestational age: ≤32 weeks, 32-37 weeks, > 37 weeks.

Allocation is concealed following the completion of baseline study measures and only designated trial team members monitoring potential exercise injuries and adverse events and the trial statistician will be made aware of each participant's allocation. Outcome assessors will be blinded to reduce potential bias. Due to the nature of the intervention, participants will not be blinded.

An emergency randomisation schedule has been prepared in advance and stored for use when the online system in unavailable. The randomisation list was created using simple block randomisation using variable block size.

8.10.3 Statistical Plan

The analysis will be carried out on the basis of intention-to-treat. This is, after randomisation, participants will be analysed according to their allocated intervention group irrespective of what they actually receive during the specified period.

Patient demographic characteristics and other baseline information will be summarised by intervention group. Numbers (with percentages) for binary and categorical variables and mean (standard deviation), or median (interquartile or full range) for continuous variables will be presented. Normality of variables will be assessed by visual assessment of the normality curves and the Shapiro-Wilk test. Methods such as the Student T-test will assess for differences between the groups. The analysis of the primary outcome will be assessed using analysis of covariance (ANCOVA) adjusting for baseline values and minimisation factors used in the randomisation process. Results will be presented as adjusted mean difference in change in ambulatory blood pressure between randomised groups at 16 weeks with 95% confidence intervals and associated two-sided p value. No imputation will be carried out and analysis will be performed on all available data. Sensitivity analyses will be performed on the per-protocol populations. The definitions of these will be detailed in the statistical analysis plan.

The analysis of secondary outcomes will also be done using analysis of covariance (ANCOVA) to establish a statistical model to examine the effect of cardiac structure, vascular function and lifestyle behaviours on exercise capacity. Outcomes measured on more than one occasion will be analysed using a mixed effects model. If the model assumptions are not met and evidence of departure from Normality is observed, transformations of the data will be employed or non-parametric tests will be carried

out. Statistical analysis will be carried out using STATA, SPSS or R statistical software. A detailed statistical analysis plan will be written with our statistical team and will be completed before receipt of the data.

8.11 Trial Management

A Trial Steering Committee (TSC) has been formed to oversee the conduct of the trial. The TSC meets approximately every six months, either in person or via teleconference. A data and safety monitoring committee (DSMC) has also been formed with the appropriate specialties to provide insight and oversight to the trial progression and management. The data monitoring committee meets approximately every six months, and shortly prior to TSC meetings when possible.

8.12 Trial Declaration

The protocol, informed consent form, participant information sheets, digital platforms and proposed advertising materials were approved by the University of Oxford as host institution and study Sponsor and the South Central Research Ethics Committee (REC) for the National Health Service Health Research Authority (NHS HRA) (Reference 16/SC/0016).

8.13 Discussion

The TEPHRA trial is the first study with the primary aim of determining the effects of structured aerobic exercise and physical activity self-monitoring intervention on ambulatory blood pressure in young adults [345]. Furthermore, it is the first study to explore the effects of exercise intervention with reference to novel cardiovascular risk factors such as preterm birth [345,347]. The association between maintaining high cardiovascular fitness and physical activity and the benefit for cardiovascular and

cerebrovascular health are well documented [12,50,83,162]. However, there is a paucity of trial evidence supporting exercise and physical activity intervention as an effective treatment option with sustained benefits in young adult populations [345]. Despite this lack of evidence lifestyle intervention is recommended as the primary intervention in young adults with hypertension[209]. In addition, there is limited understanding, with minimal prospective evidence in a trial setting, demonstrating the multisystem physiological adaptations to exercise which may mediate the benefits on future cardiovascular and cerebrovascular risk.

The results from the TEPHRA trial will provide preliminary evidence to support whether or not exercise and physical activity intervention should be primary treatment options for management of blood pressure. It will also provide evidence exploring potential heterogeneity in response to exercise intervention with reference to novel cardiovascular risk factors and baseline phenotypes such as those associated with preterm birth. The detailed cardiovascular and cerebrovascular phenotyping, using multimodal imaging, resting and stress measures of cardiovascular function will facilitate insights into disease mechanisms and phenotypic traits, which may demonstrate plasticity and remodelling, and may identify target mechanisms and interim outcomes for future intervention. The results will expand the available evidence-base for the management of blood pressure in young adult populations and inform the future direction of research targeting early cardiovascular risk reduction.

9 Thesis Limitations

9.1 Sample Size

A sample size calculation was completed prior to commencing recruitment and the sample was adequately powered to detect differences in vascular brain health and cardiac MRI indices between tertiles of participants with high and low ideal cardiovascular health. However, the overall sample was still small. A small sample recruited at a single site increases risk of bias and type 1 error. There were a number of limitations within the study design, study recruitment and study delivery that limited the potential size of the sample. This included the cost and time commitment required to undertake the study. The consumable spend and time for the study was limited to less than £300 000 which included staff costs. The real cost of recruitment and consumables were high, equivalent to £1500 to £2000 per participant when including recruitment costs, travel costs, imaging costs and cardiovascular phenotyping. The time commitment was also extensive with up to 5 hours of study procedures in busy and oversubscribed imaging departments and clinical research facilities. Therefore, feasibility of expanding the sample size to be able to explore more the heterogeneity within the sample and the graded effects of the different risk exposures was limited.

9.2 Recruitment

Second, purposive mixed passive and active recruitment strategies mean the sample is not population-based and could be considered similar to a convenience sample. Therefore, it is not possible to generalise expected prevalence of the observations across the heart-brain axis to the wider population. The name of the study and the

recruitment material may also have influenced decisions with regards participation. Young adult cardiovascular health, exercise, physical activity, preterm birth, blood pressure these are terms and concept that may have little meaning or resonance with some of our target audiences.

9.3 Population Bias

The location of the study site and recruitment of a population who were either living locally or born in Oxford results in a skew towards higher social economic placed populations. Socioeconomic status has a broad health effect and this is captured in the data from the Health Survey for England that demonstrate lower rates of cardiovascular disease risk exposures and incidence in Oxfordshire and surrounding catchment area. The lower health burden associated with higher SES is also reflected in the lower rates of preterm birth of approximately 5 to 6% of births. The sample recruited to this study were all in full-time employment or education and the majority had completed higher education. Therefore, the sample is not representative to the potential national picture of cardiovascular risk burden in younger adults. The study design and study measure has also influenced a trend towards recruiting healthier younger adults. This includes an exclusion criteria of body mass index above 35 kg/m^2 and the recruitment for study participants to complete a peak exercise test. Participants completing the study who were sedentary or no prior history of exercise participation expressed reservations about completing the exercise test and this may have been a disincentive from taking part.

The recruitment of young adults with history of preterm birth was targeted through invitation from local birth registers and participation in the Exercise and Blood Pressure Study Oxfordshire. There was a significant interest and enthusiasm from both

participants who were born preterm and their families to take part in the study. Many mentioned a sense of wishing to contribute and make an effort to participate as a thank you for the care and support received at the time of their birth. This may then have resulted in unintentional selection bias towards participants and families who had a favourable experience. There is also a survival bias and skew towards participants having no major physical impairments as recruitment materials informed participants of the requirement to complete a peak exercise test and to be able to walk a minimum of 10 metres unaided.

Recruitment of participants from the Oxford Hypertension Service looked to recruit participants with no secondary causes of hypertension or secondary conditions such as severe obesity, diabetes or renal impairment. These criteria were applied to reduce heterogeneity in the overall sample. However as a result the young adult participants recruited from the hypertension clinic are not representative of the young adults attending the clinic. Participants were invited to take part after receiving a letter having completed initial clinic assessments, including a specialist nurse led clinic and a consultant led clinic. As a result participants would all have received lifestyle advice or have been started on treatment. The sample is also cross-sectional and as a result participants from the hypertension clinic were all at different stages of management with potential confounding on the observed associations between risk factor and heart-brain indices.

9.4 Cross-sectional design

The study is cross-sectional and causality or even temporality of the observed relationships cannot be inferred. There is uncertainty of risk exposure and time interaction and the potential effects on remodelling of the health brain axis. It is also

not possible to interpret the direction of associations observed in the hypertension group with regards antihypertensive treatment. It also not possible to adjust for the potential effect of receiving prior lifestyle advice. The cross-sectional nature of the study also mean that it is possible that changes in cardiac function and brain structure are associated because of a separate, undefined, physiological or pathological pathway and experimental studies will be required to explore the interdependency of these findings.

9.5 Study measures

Advance in MRI acquisition and analysis is providing an opportunity to investigate the heart brain axis using non-invasive measures in young adult populations. However, the measures used may not be adequately capturing the effects of risk exposure on the heart-brain axis. Vascular endothelial dysfunction may be a plausible biological mechanism that may explain the observed changes across the heart-brain axis. The effects on the brain may include disruption in the blood brain barrier and dysfunction in auto-regulation. The use of BOLD resting state imaging, carbon dioxide reactivity and 4 D vascular flow haemodynamic might help improve the brain vascular assessments. The use of gadolinium contrast imaging may also help to assess disruption of the blood brain barrier. However, recent evidence of accumulation of gadolinium contrast in tissues may limit its use in research in health young adult populations. In addition, other types of provocation or stressor response testing such as functional MRI studies might help understand the impact of cardiovascular risk exposures on young adult brain health. Alternative study designs could look to incorporate exercise during MRI testing as a strategy to investigate the effects on the brain vascular tree and cerebral blood flow.

The assessment of cardiac output and strain patterns were all measured indirectly using medical imaging and derived indices from cardiopulmonary exercise testing. Assessment would be improved with the use of invasive monitoring such as central placed vascular catheters to measure acute haemodynamic changes. Alternative imaging strategies could also be incorporated such as cardiac 4D flow to measure vascular haemodynamic. This could include acute stress testing during cardiac MRI imaging such as using adenosine stress or incorporating exercise stress MR imaging.

The assessment of vascular risk exposures across the heart brain axis could be improved by incorporating atherosclerotic and other vascular imaging measures such as coronary calcium imaging and carotid intimal imaging. However, the use of coronary calcium imaging in low risk populations would need to be justified ethically and could increase the risk of clinically insignificant incidental findings. Other strategies might include measures of vascular reactivity such as flow mediated dilation for endothelial mediated dilation or ultrasound bubble scanning for assessment of muscular and myocardial tissue perfusion. However, these have significant limitations, with time required to measure, heterogeneity in response and reproducible. The contrasts used in ultrasound bubble imaging also have a high risk of anaphylaxis.

9.6 Observer reporting bias

MRI image acquisition was blinded to person details of the participant, specifically the imaging team did not know if the participant was preterm or hypertensive. However, the study team completing acquisition of the core cardiovascular measures including the cardiopulmonary exercise testing were not blinded from the personal details of the participant. However, all the study measures were assessed using study number and blinded to the participant personal details. However, there is a risk of the reporter being

unblinded. The complexity of the study visits and the breadth of study measures analysed mean that complete blinding with separation in roles between study measure acquisition and study measure interpretation was not possible. However, image analysis was completed offline without access to participant details some months after recruitment was completed to reduce risk of bias.

9.7 Incomplete Study Measures – Group MRI ASL and Stress Echocardiography

Following peer review of a related grant application, MRI ASL imaging was added to the YACHT study protocol. MRI ASL imaging for assessment of cerebral blood flow was completed on 52 participants who were recruited sequentially over the latter part of the study. Due to the in complete dataset it was not possible to explore interactions between perfusion and white matter hyperintensities. In addition, the echocardiography measures during exercise were only available in a subgroup so ability to understand rates of change in function and myocardial kinetics in association with cortical thickness and thalamic volume is limited.

9.8 Limitations of Review

Major limitations are the paucity of research studies recruiting younger adult or performing subgroup analysis defined by age. Included studies did not present results by age preventing analysis of effect in very early adulthood. Evaluation of the available literature base would be strengthened by meta-analysis of individual participant data but this was outside the scope of the review. Using ambulatory blood pressures would strengthen the results; only three studies reported ambulatory blood pressures in addition to clinic blood pressures. Identification of effective intervention components is limited by several study level factors including; lack of objective measurement and

tracking of physical activity behaviour, limited description of content and delivery of communication strategies and lack of disclosure of effectiveness of intermediate intervention process outputs such as strategies to maintain participant engagement and compliance. In the majority of studies there is also risk of bias in relation to participant allocation concealment and blinding of outcome assessors, with methods not discussed or unclear, which may risk overestimation of intervention effectiveness. However, overall the quality of included studies were moderate and funnel plots suggest no evidence of reporting bias, though caution in interpretation is required because of the small number of studies.

9.9 Clinical Significance of the study findings

Longitudinal follow up will be required to determine the clinical significance of the observed findings. As such, the MRI imaging finding study should be considered preliminary and exploratory but does support a need for future work. The complexity of the imaging protocol and associated financial costs may limit its widespread use but large multi-centre studies with more focused protocols, and extended follow up, may have the potential to track remodelling across the heart-brain axis.

9.10 TEPHRA Study Design

There are a number of limitations to consider with regards the TEPHRA study design.

1. Population

Strict inclusion and exclusion criteria are applied to reduce heterogeneity within the population sample. This increases the potential to target investigation of the effects of exercise on blood pressure with reference to gestational age in a young adult population. However, it reduces the real world application of the study.

2. Intervention

The intervention is multicomponent and includes supervised exercise training targeting exposure to moderate to intense exercise, technology assisted self-monitoring with use of a Fitbit and motivational interviewing. The rationale for the study components are to deliver a standardised dose of exercise training in the initial 16 weeks but also to introduce sustainable strategies to increase physical activity over the longer term. The multicomponent elements of the intervention raises the risk of not being able to identify the active components of the intervention. However, to manage these risks participant training records are maintained which record the number of weekly training session, heart rate during exercise as a measure of intensity, and duration of activity completed within the training session. In addition, participants' daily heart rate and physical activity profiles are exported from their Fitbit devices to provide an objective measure of physical activity throughout the 12 months of the intervention. These detailed measures of physical activity exposure across both the 16 week supervised intervention and self-directed period between 16-52 weeks provide objective quantitative exposures to include in assessment of the fidelity of the study and evaluation of the effects of the individual components of the study.

A comparative adaptive, multi-arm intervention study would be an alternative study design that would enable delivery of multiple intervention components which could be compared in a direct head to head comparison. A comparative adaptive trial would also increase the feasibility of personalising interventions and tailoring the intervention. However, major limitations of comparative adaptive intervention trials include the scale and expensive required to recruit large population samples to achieve adequate power, the complexity of the trial delivery, which could include a second within

participant randomisation and the skills and expertise required within the study team to maintain study fidelity and compliance.

3. Comparison

Participants entering the control group in TEPHRA are sign-posted to online educational materials produced by the British Heart Foundation explaining blood pressure, blood pressure prevention and recommended lifestyle behaviours to maintain heart health. Participants in the control group are asked to continue with their regular physical activity routine with no change. The potential risk of providing sign-posting to educational lifestyle resources is that participants may increase their physical activity participation or alternatively change other health behaviours such as nutrition, sleep or alcohol consumption. However, the available trial evidence that reports effectiveness of lifestyle education as an intervention demonstrate no positive effects[330]. Furthermore, both the intervention arm and control group are signposted to the same educational resources. An alternative design would be to deliver a trial with an active “attention” control arm. Instead of a minimal intervention control, participants would be randomised to an alternative intervention that is hypothesized to target a different physiological system. The benefits of this approach are that each individual recessives intervention and there is potential to explore alternative questions within the framework of the same study. An example of an attention control group for an exercise intervention would be randomisation to mindfulness. However, with this study design there is risk of inadvertently delivering an intervention that will influence the primary outcome and remove a true control comparison group.

4. Outcomes

The primary outcome measure in the TEPHRA study is 24 ambulatory blood pressure. Given the findings from YACHT a future study should consider a combined imaging outcome measure across the heart-brain axis. This study could potentially explore exercise effects on the integrity and function of the allostatic-interoceptive system[98,279]. The feasibility of exploring these outcomes in the future would have been strengthen by including functional MRI and measures of autonomic function in the TEPRHA MRI sub-study. Unfortunately, time and money restraints and priority to replicate MRI study measures completed in YACHT prohibited inclusion of functional MR imaging in TEPHRA. The TEPHRA MRI sub-study has now completed recruitment.

10 Thesis Summary, Interpretation, Implications and Conclusion

10.1 Ideal Cardiovascular Health and Heart-Brain Health in Young Adults

This thesis presents the first comprehensive analysis of the associations across the heart-brain axis and the interactions with indices of cardiovascular health in young adults. In Chapter 1, the established literature and understanding of the associations between ideal cardiovascular health and heart-brain health across the adult life-course was outlined. There is a strong population evidence base describing the associations between indices of ideal cardiovascular health and risk of heart failure, stroke and dementia in middle age and older adults[33,50,162,353]. Trial data and longitudinal population data suggests that the potential to change risk trajectories and remodel heart-brain health indicators is greatest in early and midlife[1,15,183]. Longitudinal evidence in young adults has identified that risk exposures in early life predict midlife risk of subclinical cardiovascular disease, cognitive impairment and even increases the risk of heart failure and stroke[11,12,16,19,36,40,183]. Some studies have reported early life associations between indices of vascular health, such as carotid intimal thickness and cardiovascular risk factors[20]. However, no studies have previously reported on the combined status of young adult brain and cardiac health and the association with cardiovascular risk factors.

10.2 Contribution to Knowledge Chapter 4 to 7

This section reflects on the value of the results presented in Chapters 4 to 7 and discusses their contribution to understanding of the heart-brain axis and cardiovascular risk in young adults. In Chapter 4, the study has provided novel evidence on association between ideal cardiovascular risk indices and vessel morphology. This is the first time that assessment of young adult brain vascular morphology and indices of cardiovascular health has been completed. Vessel density and vessel calibre are associated with evidence of vessel rarefaction and narrowing in association with lower indices of ideal cardiovascular health. The clinical implications are uncertain; however, in this study, it was found that there are associations of lower vessel density with lower cerebral blood flow and higher burden of white matter hyperintensity lesion count. There is potential that brain vessel morphology may provide a vascular brain marker to use in cardiovascular risk assessment and to track as an interim outcome marker in young adults. Further longitudinal research is required to understand the clinical significance of these associations and the utility of brain vessel morphology as an outcome marker. Chapter 8 outlines the protocol for the TEPHRA study; one objective of the study is to the test if brain vessel morphology is modifiable in the short-term with exercise intervention.

This is the first study to report the associations between ideal cardiovascular risk indices and white matter hyperintensity lesion burden and cerebral perfusion in young adults (See Chapter 4). The studied cohort were healthy and free of the clinical burden associated with white matter lesions and low cerebral hypoperfusion. However, the findings suggest that vascular pathological processes described in midlife and older adults start early in the life-course[51,52]. Current disease models and approaches to study prevention in the context of cerebrovascular disease and neuro-degeneration

largely fail to take account of the life-course aetiology of disease. There is a risk that the current dominance of applying reductionist approaches to study models of neurodegeneration and cerebrovascular disease may fail to identify biological targets to address the multifactorial disease aetiology[206,207]. This may limit the potential for intervention and ignores the complexity of behavioural and modifiable interactions that contribute to decline in heart-brain health. This current study was not designed to explore the molecular or microvascular associations across the heart-brain axis. However, a common disease mechanism may be vascular endothelial dysfunction and disruption of vascular haemodynamic function[6]. Future research will need to explore the potential for studying these disease mechanisms. Preliminary steps towards understanding microvascular health in relation to the heart-brain axis have been made by including retinal imaging and measure of circulating markers of angiogenic and endothelial function in the TEPHRA study (Chapter 8). Other directions to consider in the future are to introduce resting state heart-brain BOLD imaging and 4D vascular flow imaging across the heart-brain axis. These measures may provide further assessment of the vascular haemodynamic function and microvascular function and tissue oxygenation.

10.3 Cardiac Structure and Function and Brain Vascular Health in Young Adults

In Chapter 5, it was shown that indices of ideal cardiovascular health in young adults are associated with higher left ventricular stroke volume and left ventricular end diastolic volumes and lower left ventricular mass to end diastolic volume. The study highlights the potential benefit of reporting left ventricular mass to end diastolic volume in younger adults to accommodate for potential U shaped associations with the spectrum of cardiovascular fitness and ideal cardiovascular health. It also highlights the potential of exercise and physical activity promotion as interim steps to improve

cardiovascular fitness and provide potential for cardiovascular remodelling. The associations of cardiovascular fitness with favourable indices of cardiac structure and function were independent of gestational age. This suggests a potential universal benefit of exercise intervention independent of birth history but the specific remodelling potential of the preterm cardiovascular phenotype is uncertain. To attempt to address this question the TEPRA study will include targeted recruitment of preterm born young adults (Chapter 8).

Chapter 5 presented novel evidence on the associations between of cardiac structure and function, white matter hyperintensity lesion burden and cerebral blood flow. These are novel findings that bring a new understanding of the potential consequences of exposures to subclinical cardiac dysfunction on brain health in younger adults. It was shown that independent of brain vessel morphology and adjusted for ideal cardiovascular health, left ventricular end diastolic volume was an independent predictor of white matter lesion count and cerebral blood flow (Chapter 5). Furthermore, indexed cardiac output during exercise was independently associated with white matter hyperintensity lesion count. These findings provide new insight into the epidemiological observations of the incidence of heart failure and stroke in young adults in association with low cardiovascular fitness and musculoskeletal strength[11,12]. Future research is required to track the association between dynamic cardiovascular exercise performance and brain health. The combination of cardiopulmonary exercise testing and exercise stress echocardiography may provide a useful risk indicator to incorporation in assessment of heart-brain health in younger adults. These assessment modalities may be easier to translate into clinical practice given familiarity of health professionals with these tests and existing infrastructures within health services.

10.4 Heart-Brain Cortio-Thalamic Axis

Chapter 6 provides novel evidence on cortical-thalamic associations with cardiovascular structure and function in young adults. The multivariable models in Chapter 6 provide evidence of a functional heart-brain axis that relates to peak cardiovascular capacity. Further work is needed to understand the interactions across the identified cortico-thalamic axis to explore the potential for remodelling and the casual direction across this axis. At one end of the continuum is the question of whether functional cardiovascular brain networks determine peak exercise capacity. Then for individuals who are sedentary and have a high burden of modifiable risk factors, the question is: does lack of development across the heart-brain axis limit their capacity to engage with lifestyle intervention? There is a potential that young people with sedentary lifestyles have a negative experience and perception of exercise that is driven by the function of the corticothalamic tracts and the interaction with somatosensory and interoceptive networks. Promoting positive lifestyle engagement may need additional intervention to support individuals to overcome functional barriers relating to lower submaximal exercise performance association with heart-brain cortico-thalamic development. To explore this hypothesis, baseline cortico-thalamic structure will be investigated as a predictor of study adherence and cardiovascular remodelling in the TEPHRA study (Chapter 8).

The study supports the argument for using elevated blood pressure and hypertension as an initial screening strategy to target primary prevention intervention in young adult populations. In Chapters 4 and 5, blood pressure measures were the risk exposures most frequently associated with heart-brain MRI indices. Elevated blood pressure in young adults was associated with changes in brain vessel morphology, white matter lesion count and volume, cerebral blood flow and left ventricular mass to end diastolic

volume. The cross-sectional study also highlights the importance of measuring the spectrum of the young adult blood pressure. This includes the potential added benefit of measures of nocturnal and dynamic exercise blood pressure to provide broader profiles of young adult cardiovascular phenotypes.

Current guidance recommends the promotion of lifestyle intervention as primary intervention in young adults with elevate blood pressure[148,209]. However, the systemic review presented in Chapter 7 demonstrates that although there is evidence to suggest that physical activity intervention will be beneficial for blood pressure reduction there are no trials that specifically target the younger adult. One concern is that in the young adult with potentially more plasticity in sympathetic function and vagal tone, exercise intervention may increase blood pressure, especially in males[354]. The beneficial effects of exercise intervention may be dependent on modification of other indices of ideal cardiovascular health such as weight, lipid profile and metabolic function[354]. Therefore, tracking blood pressure in young adults identified with elevated cardiovascular risk may be too narrow an approach. Once someone is identified as having elevated blood pressure, the focus may need to broaden to encompass indices and behaviours of ideal cardiovascular health and interventions targeted at individual needs. Scoring young adults for ideal cardiovascular health may help to identify the high-risk individual who would benefit from additional intervention support. In these groups, medical imaging and exercise testing may help to additionally phenotype and segment the target audience to provide higher intensity support. The meta-analysis of intervention effect presented in Chapter 7 identifies that intensity of intervention, multimedia contact time with the team, and exercise supervision from the intervention team are key components contributing to intervention effectiveness. These intervention strategies have been incorporated into the TEPHRA study (Chapter 8).

10.5 Peak development across the heart-brain axis, potential for early decline into risk trajectories and causal relationships for future risk of disease

A life course approach provides a useful framework to interpret the observed associations and discuss the potential impact on future health across the heart-brain axis. The available longitudinal data suggests that brain vessel morphology[64,65,140], cortical thickness[260], cerebral volumes[259] and cerebral perfusion[355] reach peak values early in the life-course. Vessel morphology and cortical thickness may reach peak as early as infancy[59,65,68], cerebral volumes[259] and cerebral perfusion peaks in early to late adolescences[355]. Then functional connectivity and white matter integrity may peak in early adulthood with white matter volume peaking in midlife[356]. Peak cardiovascular function across indices such as cardiac output and maximal oxygen uptake reach peak after skeletal maturity in early adult life[357,358]. Physiologically determined cardiac mass and ventricular volume also peak in early adult life. However, cardiac volume and mass also change with age and risk exposures across the adult life-course[195]. The achieved physiological and developmental peaks across the heart-brain axis are determined by complex gene-environment interactions, with perinatal exposures, early year's nutrition, social enrichment, illness exposure and education being some of the major factors influencing peak development[15,145].

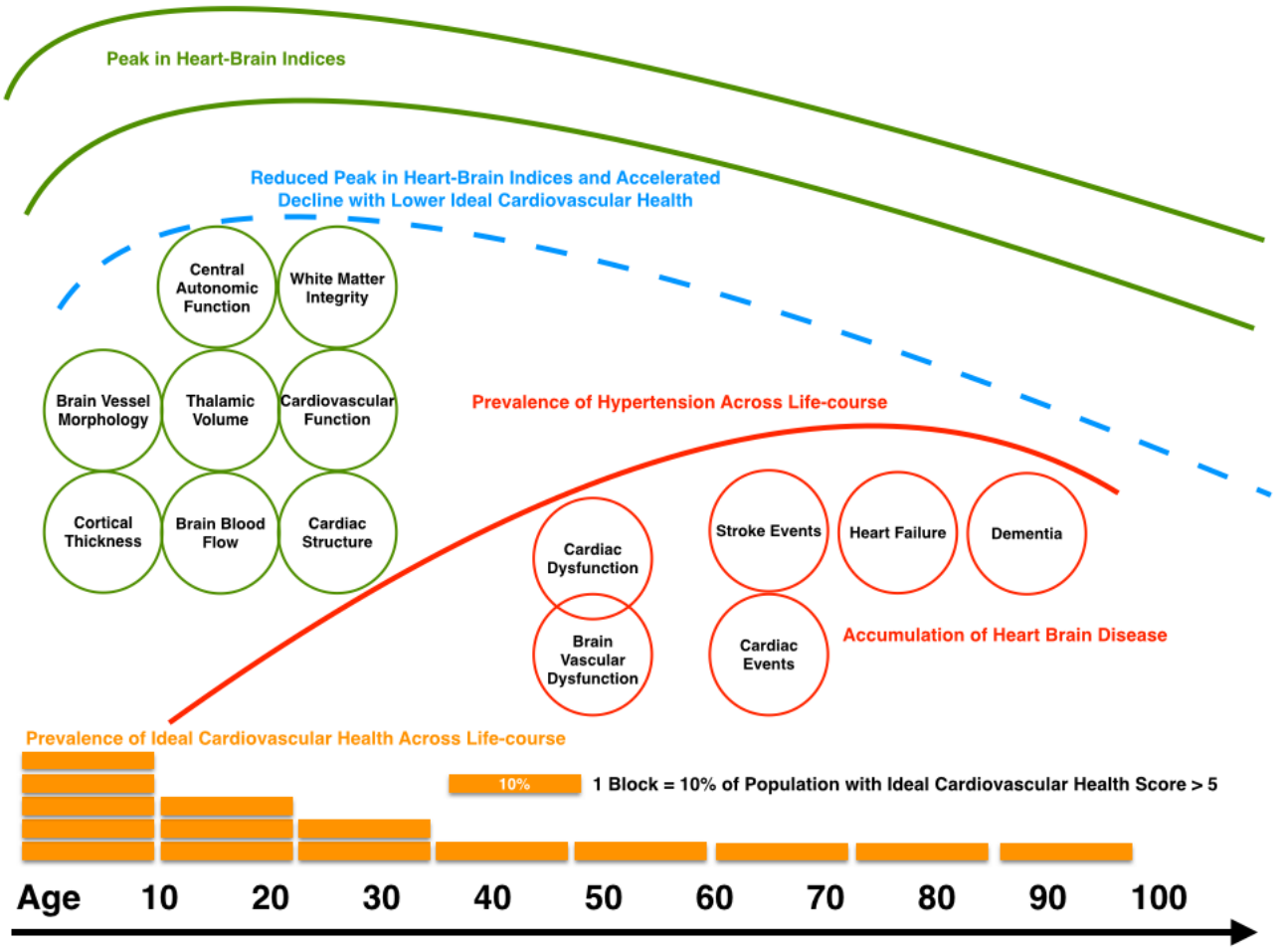


Figure 10-1 Life-Course Heart-Brain Health

This thesis provides evidence that modifiable risk factors such as elevated body mass index, low cardiovascular fitness, smoking exposure, alcohol intake, and metabolic dysfunction may lower peak heart-brain function in young adults. There is also evidence that accumulation of modifiable risk factors has added burden across the heart-brain axis. The observed cardiac-thalamic-cortical associations provide evidence to support a brain allostatic-interoceptive system. The same system may provide a mechanism for bidirectional decline across the heart-brain axis with structural and functional integrity of one organ impacting on the other. These potential

risk are particularly concerning given the population trends for modifiable risk factors across the life-course. In North American and the majority of Western Europe only 50% of children score 5 or more for ideal cardiovascular health metrics. This declines further in adolescence and early adulthood. Then across the adult lifespan less than 15% of the population have optimal health indices with no adverse modifiable cardiovascular risk factors[359].

The observed associations in the current study suggest complex interactions that may provide some insight into heart-brain resilience in young adults and known patterns of disease in midlife. Potential casual pathways across the heart-brain axis could be hypothesize to include:

1. Reduce peak thalamic-cortical development = reduced central autonomic/cardiovascular networks = reduce myocardial functional capacity= lower cardiac resilience
2. Reduced brain vessel morphology = lower cerebral haemodynamic and vascular capacity = lower brain vascular resilience = increased risk of white matter injury
3. Lower cardiac structure and performance = reduce cardiovascular stress response = potential of lower cerebral blood flow= lower brain vascular resilience
4. Lower white matter integrity/increase white matter injury = cortical-thalamic dysfunction = reduced central autonomic control = reduce cardiac capacity = lower heart-brain resilience

However, it is likely that incremental and accumulated dysfunction across the heart-brain axis leads to development of midlife disease. Across the lifespan there may be critical thresholds of resilience and potential domino effects across the heart-brain axis. Resilience determined by the underlying structure and functional integrity of the

heart, brain vessels, thalamus, white matter, cerebral cortex and associated functional connectome. Age associated decline and interaction with risk exposures may precipitate transition across a resilience threshold. The estimated age associated rates of decline in peak cardiovascular function and cerebral vascular function are 5% to 10% per decade starting in early adult life[7,8]. There are similar age associated rates of atrophy in cerebral volumes but with different critical periods of decline. Cortical volumes and cortical thickness, basal ganglia volumes and thalamic volume decline across early adult life. Thalamic volume declines at an estimated rate of 0.69% per year and maintains a strong linear trend with advancing age (r=-0.65)[260,287,360]. Decline in white matter and hippocampal volumes start later around the age of 50 years old and follows an accelerated trajectory over the age of 60 and the older life-course[261,360].

Lower cardiovascular function in young adults is a risk factor for both stroke and heart-failure in midlife[11,12]. The observed associations in the current study may provide some understanding of how these disease patterns evolve. With regards cardiac dysfunction causing decline in brain health, lower cardiovascular function may lead to impairment in vascular integrity, white matter function and cerebral perfusion which may increase risk of stroke. With regards lower cerebral function leading to cardiac dysfunction, reduced cortical-thalamic integrity and function in early adult life may be an attributable risk factor for heart failure and sudden cardiac events in middle age. In a model of parallel decline in heart and brain health, lower cardiovascular exercise capacity and modifiable risk factors may lead to lower myocardial resilience and risk of atherosclerotic changes across the heart-brain axis which increases risk of cardiac disease which lowers capacity to respond to stressors, in parallel with reduced cerebrovascular compliance and combined risk of tissue hypoperfusion which could contribute to either cardiac or cerebrovascular risk.

10.6 Biological mechanisms contributing to observed associations across the heart-brain axis

In the current study the observed associations between cardiovascular risk factors and brain vessel morphology, white matter hyperintensities and cardiac function may be explained by understanding biological effects of risk exposures on the vascular tree and impact on vascular endothelial function. Endothelial function is a primary determinant of both cerebrovascular and cardiac vascular health. Impaired endothelial and mitochondrial function leads to impaired capacity to meet oxygen and energy demands of both cardiac and cerebrovascular tissues[2,4,6,61].

In the brain the endothelial cell layer within the brain neurovascular unit is the main interface of the blood-brain-barrier and regulator of molecular exchange between the circulating blood and brain tissue. Transport channels located in the endothelial cell membrane, such as the GLUT transporters and aquaporins, regulate glucose uptake and facilitate removal of CO_2 and NH_3 to maintain metabolic haemostasis[59,60]. In addition, endothelial cells and astrocytes produce a number of growth factors such as brain-derived neurotrophic factor (BDNF), insulin-like growth factor, and vascular endothelial growth factor (VEGF), which support the sprouting of new vessels, the migration of neuronal progenitor cells and oligodendrocytes and differentiation of neuronal cell lines[64,65]. Therefore, integrity of endothelial function, the neurovascular unit, and microvascular network provide the scaffolding and essential growth factors for neurogenesis. Exposure to cardiovascular risk factors leads to dysfunction in energy metabolism, which is associated with reduced integrity of the blood brain barrier driven by reduced expression of the endothelial cell receptors. There is also associated

dysfunction in autoregulation of cerebrovascular blood flow[169-171]. Therefore, there is decline in the potential for neuroplasticity and the ability to recover following acute brain injury.

Modifiable risk factors such as elevated blood pressure cause vessel walls and the myocardium to experience significant mechanical stress stimulating hypertrophic remodelling of smooth muscle cells and cardiomyocytes[4,165]. The vascular biological response is up-regulation of extracellular metalloproteases, increased deposition of collagen and fibronectin, and extravasation of plasma proteins and macrophages around capillaries and arterioles that leads to inflammatory responses[4,165]. These inflammatory responses can then be exacerbated by the effects of exposure to smoking, alcohol and metabolic dysfunction. The vascular effects, range from luminal narrowing and atherosclerotic change in vessels, fibrotic changes in the myocardium, disruption of the blood brain barrier, and white matter damage including loss of microstructural integrity and white matter hyperintensity lesions, which are most marked around the periventricular areas as reported in the current study.[4,165,166].

The observed positive associations between cardiovascular function and increased thalamic volume and cortical thickness over the posterior cingulate and precuneus area indicate high structural integrity and functional capacity across the heart-brain axis. The thalamus, posterior cingulate and precuneus are identified as highly connected neural hubs[278,280]. The common pattern of connectedness being referred to as components of a neural brain rich club. A distinguishing feature of rich neural connection are increased thalamic volume and increased cortical thickness. Thalamic and cortical rich club nodes are also identified to have high metabolic and vascular demands to meet the functional requirements. This may therefore explain the observed association between cardiovascular capacity and thalamic volume. The increased cardiovascular capacity

required to meet the associated energy demands. However, the observed association between cardiac function, thalamic volume and patterns of cortical thickness are also consistent with a model of a brain allostatic-interoceptive system[279]. Therefore, an alternative explanation for the observed associations is that they may capture the phenotype of peak function associated with an optimal brain allostatic-interoceptive system[279]. The parallel of heightened cardiovascular function and cortical-thalamic development within the neural rich club representing an efficient energy regulation system[279,301]. With increased capacity to sense and perceive increased energy demands and functional ability to respond with increased cardiac output and workload.

The observations in the current study may help explain why decline in cortico-thalamic volume and decline in peak cardiovascular function share similar trajectories[260,261,287,360]. It also suggests potential risk for accelerated decline in cortico-thalamic volume and white matter integrity associated with higher burden of modifiable risk factors. Decline in cerebral volumes and cortical thickness in young age are associated with gene expression that regulates astrocytic metabolism and microglial signaling[361]. The observation that modifiable risk factors may disrupt the blood brain barrier and astrocyte function may then lead to increased risk of accelerated decline in cortical thickness for young adults with higher burden of modifiable risk factors[6]. The cortico-thalamic system and neural rich club have an inherently high metabolic demand and meeting these energy requirements places stress on mitochondrial function and oxidative metabolism[2,6,362]. In turn the high metabolic turn over can lead to increased production of oxidative free radicles. This can lead to dysfunctional gene expression and reduced synaptic plasticity, reduced vesicular transport and mitochondrial dysfunction[4,6,61]. This may also contribute to upregulation of stress response, antioxidants and dysfunctional DNA repair contributing to neuronal loss and

white matter damage processes exacerbated by elevated blood pressure and modifiable risk factors[67,198,363]. The high energy and densely connected cortico-thalamic white matter tracts are vulnerable to metabolic and vascular dysfunction[362]. Higher burden of white matter lesions in the periventricular areas may also be explained by the dependency of these areas on larger vessels for blood supply. Therefore these areas are more vulnerable to dysfunction in the macrovascular environment. This may explain the observation of higher white matter lesion burden in association with lower cardiac output during exercise. The lower cardiac output being an indicator of lower capacity to meet the increase haemodynamic and metabolic demands of exercise.

10.7 Cardiovascular Primary Prevention in Young Adult Populations

Completion of the cross-sectional study (Chapters 3 to 6) has shaped and informed the design and implementation of an exercise trial targeting young adults with elevated blood pressure and cardiovascular risk. The study has helped to detail the interactions across the heart-brain axis, the potential benefits of higher cardiovascular fitness and identified outcome markers. However, there remains uncertainty regarding the feasibility and effectiveness of a supervised exercise intervention in a young adult population. Major challenges include adherence to intervention allocation and compliance with intervention components. The findings presented in Chapter 6 of cortico-thalamic associations with cardiovascular function may influence adherence and compliance with intervention and this is a question of interest to explore in the context of the trial (Chapter 8). With regard to the fidelity and efficacy of exercise intervention, there is very limited understanding on the dose response and time-frame

required to achieve remodelling across the heart-brain axis. The systemic review presented in Chapter 7 and evidence to date suggests that peripheral and cardiovascular adaptation is achievable within 12 weeks of intervention. However, this has often been reported in the unique settings of military and sports and academy cohorts[42]. The TEPRHA study and the breadth of study measures will provide the investigative template to understand in more detail the heart-brain axis in young adults and the potential for remodelling. The study will also provide additional insight into the effective intervention components. It is hoped that incorporating motivational interviewing strategies, and physical activity self-monitoring with extended follow-up to 12 months will help explore the potential for more economically sustainable interventions that can be designed for clinical translation. The completion of the TEPHRA study and the associated process evaluation and analysis will provide the potential to expand to a larger definitive trial with a defined set of outcomes and a refined intervention design.

10.8 Future directions

This thesis has described the cross-sectional associations across the heart-brain axis. The study demonstrates that ideal cardiovascular health indices explain variance in the morphology and structure of the heart-brain axis. Furthermore, variance in cardiac structure and function relate to indices of brain vascular health. Cortico-thalamic development and integrity may be a determinant of cardiovascular function and explains variance in observed peak exercise capacity.

The detailed heart-brain, behavioural and dynamic cardiovascular phenotyping identifies the potential burden of cardiovascular risk in young adult populations. Younger age is not protective against the exposure to cardiovascular risk factors. The

prevalence of modifiable risk factors has increased in the adolescent and young adult populations. Population evidence has identified that risk of low ideal cardiovascular health is associated with increased risk of heart failure and stroke in midlife. Furthermore, the incidence of heart failure and stroke has increased in the young adult population. This study confirms that exposure to modifiable cardiovascular risk factors in young adults is associated with heart-brain MRI indices that are known to be on disease trajectories to heart failure and stroke in middle age.

The study highlights inherent bias in the research community to exclude young adults from participation in cardiovascular prevention research. This bias has been evident in the experience of peer review in the preparatory steps to secure funding to support this research. Some academic peers dismiss a life course approach to primary prevention and suggest a lack of relevance of young adult cardiovascular risk in the aetiology of later life stroke, heart failure and neurodegenerative disease. However, as discussed in Chapter 1 there is now clear epidemiological evidence of life-course trajectory of diseases of mid and older life. The application of the advanced medical imaging used in this current study has provided the technical innovations to describe the heart-brain axis of young adults. The development of reliable and sensitive measures of heart-brain health facilitated the observation of the associations with indices of ideal cardiovascular health.

The current study has therefore provided the platform to complete the TEPHRA study. The TEPHRA study will now investigate the potential for heart-brain remodelling and the related determinants in young adults with elevated blood pressure. The trial will provide the opportunity to expand the description of the heart-brain axis in young adults. The use of the same imaging protocol, in new cohort of young adults, provides an opportunity to validate and confirm the presented findings. It is hoped that this body

of research will help to develop a new era of primary prevention research and provide the platform for future multicentre collaborative research. An aim will be to expand the investigation of the heart-brain axis and look for opportunities to apply new multimodal investigative and analytical techniques. It is hoped that applying advances in medical imaging and cardiovascular phenotyping will get closure to understanding heart-brain vascular health in young adults and the origins of future disease. It may even offer the potential to define new biomarkers for early and mid-life heart-brain health assessment and drive advances in personalised risk assessment and tailored interventions.

10.9 Conclusion

This thesis provides preliminary findings that modifiable cardiovascular risk factors are associated with MRI indices of brain vascular and cardiac health in young adults. There is also evidence of dynamic relationships across the heart-brain axis. The observed cortical and thalamic associations with resting and exercise measures of cardiac function provides new insight on the neural-cardiac axis. The research expands the evidence and understanding of central control of cardiovascular function. Furthermore, the research challenges the perceptions of cardiovascular risk in young adults and the potential burden on the heart-brain axis. There is evidence in young adults that cardiac function is associated with white matter hyperintensity lesions and reduced or altered cerebral blood flow. These are known risk factors for future decline in heart-brain health. However, current guidance and practice to reduce cardiovascular risk in young adults is deficient of a strong evidence base. There is need for continued research to improve age specific strategies and recommendations for primary and

secondary cardiovascular risk reduction. There is also need for co-ordinated and strategic engagement from across multi-disciplinary sectors to drive improvements in the prevention and management of cardiovascular risk in early and mid-life. Until these challenges are addressed the increasing prevalence of modifiable cardiovascular risk factors in young adults will only act to accelerate burden on heart-brain health in midlife with our young adults becoming our patients of tomorrow.

11 References

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