The Influence of Low Physical Activity Levels versus Extreme Physical Activity Levels on Brain Structure and Working Memory

By

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DECLARATION

I, Tasneem. Y. Paruk, hereby declare that the work on which this dissertation/thesis is based is my original work (except where acknowledgements indicate otherwise) and that neither the whole work nor any part of it has been, is being, or is to be submitted for another degree in this or any other university.

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Contents

Glossary of Terms

Analysis of Variance (ANOVA): is a statistical method used to determine whether there are any significant differences between the means of three or more independent groups.

Brain-derived Neurotrophic Factor (BDNF): is a protein responsible for maintaining and regenerating nerve cells in the brain.

Brodmann Area: refers to one of the 52 areas of the brain defined by German neurologist Brodmann based on its cyto-architechture.

Dopamine: is a neurotransmitter produced in the body that sends signals between nerve cells.

Global Physical Activity Questionnaire (GPAQ): is a questionnaire, based on sedentary behaviour and physical activity participation from work, travel, and recreation.

Grey matter: is the functional tissue of the brain containing neuronal cell bodies, dendrites, glial cells, synapses, and capillaries.

Executive function: is a set of cognitive processes used to control and co-ordinate cognitive abilities and behaviours.

Neurotransmission: is the transmission of nerve impulses between neurons.

Total intracranial volume: is the sum of grey matter, white matter, and cerebrospinal fluid volumes.

Voxel-based morphometry (VBM): is a quantitative neuroimaging technique used to assess the size of brain tissue using a voxel-wise approach.

White matter: is the part of the central nervous system that is mainly composed of myelinated axons and myelin-producing glial cells. White matter is the communication pathway between areas of grey matter.

White matter hyperintensities: are markers of white matter damage shown by various forms of

neuroimaging (e.g. MRI).

Working memory: Working memory is a component of short term memory of limited capacity that temporarily holds and uses information for conscious processing.

Abstract

The Influence of Low Physical Activity Levels versus Extreme Physical Activity Levels on Brain Structure and Working Memory

*Background***:** Habitual levels of moderate to vigorous levels of physical activity (MVPA) levels are associated with positive health outcomes and improvement in cognitive function. A challenge facing optimal health outcomes is determining optimal levels of physical activity levels; people can engage in extreme sedentary or high levels of physical activity. For example, levels of low physical activity and participation in ultra-endurance events are increasing. However, the impact of extreme physical activity levels on brain matter volume and working memory is not well understood. This thesis thus investigated the relationship between different levels of MVPA, brain structure, and working memory. The aim of the thesis was to understand the relationship between large volumes of physical activity with global and regional brain matter volumes using brain imaging, and the impact of physical activity on cognitive function using the N-back task. The first part of the thesis examined how habitually high or low levels of MVPA differently impacted brain matter volume in healthy individuals. Whilst, the second part of the thesis investigated the differences in working memory between the low active and high active groups.

*Methods***:** Participants aged 20-59 years, were split into a High Activity (HA) group (N = 12, 27.9 ± 26.6 years) that exercised for > 9 hours of per week (MVPA > 540 minutes) and a Low Activity (LA) group (N = 9, 28.33 ± 11.192 years) that exercised < 2 hours (MVPA < 120 minutes). Total and regional brain matter volumes were measured using Magnetic Resonance Imaging (MRI) and analysed using Voxel-Based Morphometry (VBM). Habitual levels of physical activity were measured using the Global Physical Activity Questionnaire (GPAQ). For the second part of this thesis, we tested participants aged 20-59 years. Participants were split into a High Activity (HA) group (N = 6, 24.86 ± 4.67 years) that exercised for > 9 hours of per week (MVPA > 540 minutes) and a Low Activity (LA) group (N = 6, 30.14 ± 15.31 years) that exercised < 2 hours (MVPA < 120 minutes). Working memory was compared

between the two groups using a PC-based N-back task.

Results: The HA group had greater total brain matter and total white matter volumes vs. the LA group, and brain matter volumes were positively associated with increasing physical activity levels. However, the HA group had reduced regional brain volumes in the postcentral gyrus, middle frontal lobe, and the sub-lobar thalamus. HA and LA groups did not differ in working memory performance on the N-back task.

Conclusion: Larger than normal amounts of weekly PA was associated with increased total brain matter and total white matter volumes, however, the volume of three regional brain areas were reduced.

Rationale

Research looking at the therapeutic effects of physical activity on health, and improvements in cognitive function is vast. However, the research focuses on moderate levels of physical activity. The purpose of the two components of this investigation was to find a link between physical activity levels, brain structure and cognitive function when greater than normal physical activity levels were present versus low levels of physical activity.

1 Literature Review

1.1 The Human Brain and Physical Activity Levels

1.1.1 The Life Cycle of the Human Brain

The human brain plays a sophisticated role in bodily function. It is responsible for processing stimuli, be they internal (e.g. neurotransmission) or external (e.g. environmental), and creating outputs such as behavioural changes. The brain is responsible for initiating locomotion, cognitive functioning, regulating homeostasis, and controlling emotion, etc. (McEwen, 2007). The brain is affected by all aspects of the human experience and is impacted by factors such as nutrition, trauma, disease, and activity levels that change continuously through life (Ellingson et al., 2012; Erickson et al., 2012; Gomez-Pinilla, 2011). Brain volume throughout a human's life follows a "U-shaped" curve; there are rapid periods of development in infancy that reach a peak during middle age and then decline as we age (Raz et al., 2005). The changes in volume are dynamic and are influenced by varying factors and life events such as emotional trauma (Meng et al., 2016) and positive social relationships (Kawamichi et al., 2016.) The timing of these events and the responses to the stress contribute to changes in function. Brain volume was found to be directly associated with the intrinsic brain activity that is associated with the functional ability of the brain (Qing & Gong, 2016). Thus, preserving brain volume should be prioritised in order retain physical and cognitive functions like walking and working memory (Dumurgier et al., 2012; Erickson et al., 2011; Rosano et al., 2008; Sachdev, et al., 2005).

1.1.2 Why is the Brain Important?

Health has been defined as a state characterised by physical, mental, and social well-being, not merely the absence of disease (Constitution of the World Health Organisation, 2006). Health encompasses both physiological and psychological health and is closely intertwined with cognitive health (Porges, 2007; Thayer et al., 2009). Physical activity, disease, social and emotional well-being have all been linked to health status and play a role in influencing brain structure and function (McEwen, 1998a). Factors such as

disease and negative social behaviours, showed less favourable adaptions as compared to the positive adaptations associated with physical activity. Physical activity not only improves the human condition but has also been shown to reverse the degenerative effects of poor lifestyle behaviours such as diet on health and cognition (Hillman, et al 2008). This suggests that there is a strong connection between health status, physical activity, and the brain.

1.1.3 How Does Brain Volume Impact its Functioning?

Brain tissue is composed of grey matter, white matter, and cerebrospinal fluid (CSF). Each tissue type serves a different function and contains different types of cells. Grey matter is the functional tissue containing neuronal cell bodies, dendrites, glial cells, synapses, and capillaries. White matter is the communication pathway for grey matter and is composed of myelinated axons and myelin-producing glial cells. CSF protects the brain and provides nutrients to the nervous tissue. The principal driver of brain function is governed by grey and white matter (Honey, Thivierge, & Sporns, 2010). Greater total brain matter volumes have been associated with greater collective activity in the brain, while increases in regional grey and white brain matter have been positively associated with cognitive and behavioural function (Qing and Gong, 2016).

It was previously thought that the brain could not be changed after adulthood. However, more recent research has shown that the brain continually changes in response to the environment (Raz et al., 2005). The ability of the brain to change and adapt is referred to as neuroplasticity. Maintaining its ability to change is important for its health (Cotman and Berchtold, 2002).

1.1.4 Brain Neuroplasticity

Neuroplasticity is a physiological phenomenon that allows for the brain to adapt to stimuli or stressors that are faced by the brain (Cotman and Berchtold, 2002; Feldman, 2009; McEwen, 2007; Ulrich-Lai and Herman, 2009). The brain is plastic; its ability to adapt and change with learning and growth is dependent on its plasticity. The concept of neuroplasticity was introduced in 1890 (James et al., 1890). Plasticity is

related to both positive and negative changes (McEwen, 2002), and the brains ability to re-organize and create new neural connections that are essential for adaptation to change (Zatorre et al., 2012). Neuroplasticity research focuses largely on eliciting positive changes to stimulate plasticity with interventions that strengthen the brain to cope with stressors. In contrast, research on neural degeneration focuses on reducing the rate of damage and increasing repair, such as in Alzheimer's disease (Fuchs and Flügge, 2014) or multiple sclerosis.

One approach to evaluating any neural change is to observe macroscopic grey matter and white matter changes using MRI. For example, MRI research has shown that grey matter losses occur as we age and is thought to be the basis for the overall age-related loss in physical and mental functions (Camandola & Mattson, 2017; Erickson., 2012; Fuchs and Flügge, 2014; Raz et al., 2005). On the other hand, improvements to grey matter plasticity have been positively associated with cognitive training, mental imagery and physical activity, which resulted in regional and total grey matter increases (Draganski et al., 2004; Pascual-Leone et al., 2005; West et al., 2017; Woollett and Maguire, 2011).

1.1.5 Factors That Influence Neuroplasticity and Degeneration

Plasticity and degeneration of the brain can be impacted by lifestyle, age, and gender, which cause individual variation in grey and white matter volumes (Thompson et al., 2001). For example, changes in lifestyle (e.g. physical activity and education) can alter grey and white matter at the global and regional levels (Gordon et al., 2008). Further, there are key age points during life when grey and white matter change at variable rates. Age-related matter reductions were shown to begin around 20 years old and were found to have a steady decline up until 40 years old (Svennerholm, et al., 1997). Reductions in grey matter volumes have been shown to accelerate after age 40 and significantly increased after the age of 70 (Scahill et al., 2003). It was even shown that the male brain aged faster and the decline in matter volume was more rapid (Good, et al., 2001; Király et al., 2016). Plasticity and degeneration are ever-present in the brain's structure, plasticity being the predominant force during early life and degeneration later on.

Preserving grey matter and white matter volumes are crucial to slowing the negative effects of

aging such as cognitive and physical function declines (Aribisala et al., 2013). Accelerated declines in total grey and white matter are commonly associated with degenerative diseases such as Alzheimer's and Parkinson's disease (Camandola and Mattson, 2017; Erickson et al., 2012; Fjell et al., 2013; Zhang et al., 2016). Although regional white matter was found to decrease significantly with disease and age, the total white matter was not significantly affected (Good et al., 2001). The effects of aging on total and regional brain matter volumes are not limited to unhealthy populations and even healthy individuals are subject to brain matter loss (Raz et al., 2005). Age-related matter reductions were found predominately in the caudate, cerebellum and hippocampus, cerebellar and hippocampal declines accelerated with increasing age (Raz et al., 2004; Raz et al., 2005). Age-related decreases in the volume of the parietal and temporal regions were found (Bartzokis et al., 2001). Slowing down the rates of decline in grey and white matter was found to be effective with early life and late-life implementation of physical activity to maintain or improve cognitive and physical function (Aribisala et al., 2013; Voelcker-Rehage and Niemann, 2013).

The plasticity associated with grey matter change is not entirely independent from white matter changes. For example, a study on juggling training found that regional grey matter had increased in the proximity to where white matter changes had also taken place(Scholz et al., 2009). Recently, Raichlen and Alexander (2017) proposed that with exercise and cognitive training, changes in grey matter coming from neurogenesis only take place in response to a specific demand, but that white matter strengthening is more consistent and a more efficient way to maintain brain structure and health. This illustrates the importance of assessing plasticity by measuring both grey and white matter.

1.1.6 What Is Physical Activity?

Physical activity has been defined as bodily movement which comes from skeletal movement t resulting in energy expenditure above the resting metabolic rate (Caspersen et al., 1985; World Health Organization, 2010). Energy expenditure is quantified using Metabolic Equivalents (METs). A MET is defined as the resting metabolic rate and is the amount of energy expended whilst sitting quietly in a chair (Jette et al., 1990). Regular physical activity results in adaptations that lead to improved physical fitness

(Haskell et al., 2007). Physical activity is an important component of human life and is necessary to carry out daily tasks and has been shown to impact the many systems of the human body; impacting organs such as the heart, muscle, and brain (Haskell et al., 2007; Lefèbvre et al., 2001).

All forms of physical activity engaged in by individuals are not equal. The intensity, volume, and medium of physical activity differ between individuals based on their ability and preference. Further, the mode (e.g. bike, running, swimming), frequency, and intensity of physical activity sessions result in different physical adaptations (Lefèbvre et al., 2001). The spectrum of activity ranges from sedentary behaviour to extreme levels of physical activity such as ultra-endurance activity. Sedentary behaviour is classified as an absence of voluntary physical activity and is characterised by an energy expenditure of < 1.5 METs and usually involves sitting or lying down. Moderate levels of physical activity exist in the middle of this spectrum (450-750 MET's) and people are encouraged to maintain these level of physical activity to derive benefit for healthy brain structures and overall health (Batouli and Saba, 2017; Erickson et al., 2010; Haskell et al., 2007; Warburton et al., 2006). Ultra-endurance physical activity habits lie on the other end of the spectrum, with people engaging in activities of 7-8 METs for > 300 minutes of activity during these events, not including their training load (Zaryski and Smith, 2005).

The World Health Organisations has conducted research and has created recommendations for physical activity in adults aged 18–64 years. To promote or maintain health and reduce the risk of chronic disease individuals should engage in a minimum of 150 minutes of moderate-intensity physical activity, or engage in a minimum of 75 minutes of vigorous-intensity physical activity during a week, or an equivalent combination of both moderate and vigorous physical activity. The recommendations suggest that for additional health benefits, individuals should increase their moderate-intensity physical activity to 300 minutes per week, or 150 minutes of vigorous-intensity physical activity per week (Armstrong and Bull, 2006; Haskell et al., 2007). This 150 minutes of 4-5 MET's per minute equates to 450-750 MET's weekly to achieve minimal health benefits (Haskell et al., 2007). These guidelines are applicable in South Africa and globally (Department of Health, 2000).

1.1.7 Quantifying Physical Activity

There are three primary methods used for quantifying physical activity in research settings; self-reported questionnaires, direct observation, and accelerometer devices (Sylvia et al., 2014). Accelerometers have proven to be more objective than self-reported measures such as questionnaires (Sylvia et al., 2014). However, this is costly and cannot be used to gather retrospective data on existing physical activity habits (Sylvia et al., 2014). On the other, direct observation is the most accurate and has been used to study military groups who all undergo the same training, but the feasibility of this approach for other contexts is low since it requires constant observation by a research team. A more cost-effective method that allows researchers to gather information on existing physical activity habits is the GPAQ (Sylvia et al., 2014). The GPAQ was developed by the WHO as a surveillance tool of physical activity. It is a self-reported subjective measure of physical activity. It quantifies physical activity using duration and intensity of activity using MET's. The GPAQ accounts for physical activity during working hours, transportation, and recreational physical activity. The GPAQ takes into account other sources of physical activity and not training hours alone (Armstrong and Bull, 2006). Although subjective and retrospective, it has been validated in various activity groups in several different populations from varied socio-economic and educational backgrounds globally and allows for a feasible method to gain insight on retrospective physical activity behaviours (Armstrong and Bull, 2006; Cleland et al., 2014), especially in urban populations (Mumu et al., 2017). The GPAQ is a valid measure of MVPA, but its validity in measuring sedentary behaviour is less reliable (Cleland et al., 2014).

1.1.8 The Role of Physical Activity in Health

Meeting the recommended physical activity guidelines is important because research has shown that moderate levels of physical activity can improve the conditions of psychiatric diseases, metabolic diseases, cardiovascular disease, pulmonary diseases, musculoskeletal diseases, and even cancer (Pedersen and Saltin, 2015). Physical inactivity is among the leading risk factors for global mortality (World Health Organization, 2010). The research has extensively shown that when physical activity

guidelines are met they improve disease risk factors such as insulin resistance, inflammation, hypertension, dyslipidaemia, and decrease blood pressure (Petersen and Pedersen, 2005; Pedersen and Saltin, 2015). The benefits of physical activity are important for both aspects of health; physical, and cognitive. The role physical activity plays in health is not only a preventative measure but the mere absence of regular physical activity as per the minimum recommended guidelines has also been shown to play a major role in the risk of developing chronic diseases such as cardiovascular disease, diabetes, and cancers (Booth et al., 2012). In longitudinal studies examining exercise interventions research found that cognitive decline and physical function decline rates were reduced (Erickson et al., 2011; Roberts et al., 2017).

With increasing levels of physical activity, mortality and cardiovascular risks are reduced (Biswas et al., 2015). This is however more complex as individuals can meet the physical activity guidelines and still be sedentary the rest of the time. This may counteract some of the benefits of physical activity to a certain extent and increase mortality and cardiovascular risk (Warburton et al., 2006).

1.1.9 Regular Physical Activity, Fitness, and Health

Regular physical activity plays an important role in fitness and health as it improves cardiovascular fitness by improving skeletal muscle metabolism, angiogenesis, and blood flow (Haskell et al., 2007). These physiological changes are associated with lowered resting heart rates (Jensen 2013). There are many methods to measure cardio-respiratory fitness using exercise tests (Vanhees et al., 2005), but resting heart rate is a simple, quick and cost-effective measure of cardio-respiratory fitness and health (Jensen et al., 2013; Jurca et al., 2005; Kang et al., 2017; Zhang et al., 2016). Further, resting heart rate may provide insight into the autonomic nervous system (ANS) function. The role of the ANS is to regulate physiological function by maintaining homeostasis etc. ANS balance is the interaction between sympathetic and parasympathetic activity (Thayer and Lane, 2009). With the implementation of regular physical activity, RHR usually decreases and a lower RHR was found to be associated with higher fitness levels (Jensen et al., 2013). With improvements in fitness resting sympathetic nervous system (SNS)

activity is decreased and parasympathetic nervous system (PNS) activity is increased, resulting in a more favourable ANS balance and a lowered RHR. High heart rates have been shown to reflect over activation of the SNS which is associated with autonomic nervous system imbalance and cardiovascular disease (Mueller, 2007; Böhm et al., 2015). Furthermore, decreased parasympathetic nervous system activity was shown to be an independent risk factor for all-cause mortality (Thayer and Lane, 2007; Thayer and Sternberg, 2010; Thayer et al., 2010). The enhancements in fitness and health found with regular physical activity can be attributed to a stress-adaption response.

1.1.10 Brain Neuroplasticity Induced by Stressors

Brain neuroplasticity is a phenomenon whereby brain structure is re-organised (Kramer and Erickson, 2007) and this is triggered by stressors; these can be negative such as trauma and positive stressors such as physical activity (Fuchs and Flügge, 2014). Aging, financial stress, physical, and emotional trauma have also been shown to be negative stressors that impact the brain structure negatively with reductions in grey matter volume regions such as the hypothalamus, inferior parietal lobe, frontal and temporal lobes (Cheng et al., 2015; Kokubun et al., 2018). The metabolic and CV demands of exercise have a positive affect brain structure and improve neuroplasticity over the life span (Cotman and Berchtold, 2002; Lövdén et al., 2010). Physical activity as a form of stress at musculoskeletal and cellular level is a challenge to the body that positively impacts brain health (Camandola and Mattson, 2017). Active lifestyles have been shown to offer neuroprotection that prevents degeneration and aids the regeneration of white matter (Gow et al., 2012). This is important for maintaining physiological and cognitive function. Further education may also be a positive stressor for brain structure. Gordon et al., (2008) found that not only physical activity, but a higher level of education was also associated with improved brain structure and improved white matter integrity. The ability and tolerance of the body to process stress have also been shown to impact the brain. McEwen, (1998b) found high allostatic loads to be associated with hippocampal dysfunction and atrophy. Moderate levels of physical activity, on the other hand, have been associated with positive changes in grey and white matter volume (Erickson et al., 2011, Erickson et al., 2012; McEwen, 2002), the demand it places on the body allows for physical growth and

improved neural efficiency (Dunst et al., 2014).

1.1.11 The Role of Physical Activity and Lack thereof on Brain Neuroplasticity

For neuroplasticity to occur a stimulus is required to trigger this process. Physical activity is one type of stimulus that triggers neuroplasticity, it has been associated with positive structural changes in the brain such as regional and global increases in grey and white matter (Batouli and Saba, 2017; Colcombe et al., 2006; Gow et al., 2012; Raz et al., 2005) Increases in grey matter have been attributed to the volume of physical activity individuals engaged in (Raji et al., 2016). A recent review found that physical activity-related neuroplasticity was shown to affect global grey and white matter volumes as well as specific regions (Batouli and Saba, 2017). The results of previous studies don't all concur with each other, some showed a change in white matter, some in grey matter whilst others showed no change. For example (Jochem et al., 2017) found no differences in global matter volumes which may have been due to the very wide age range in this study (25-83 years). The presence and magnitude of brain matter volume changes may be a result of various factors such as age, demographics, and even BMI (Taki et al., 2008). Age-related matter losses were shown to be reduced with exercise in elderly (Bugg and Head, 2011) Frontal white matter was also increased in an older population when meeting the guidelines performing three days of weekly exercise (Kramer and Erickson, 2007).

The exercise-induced changes do not equally affect all regions of the brain and were found predominately in certain regions such as the prefrontal and cingulate cortex (Colcombe et al., 2004; Colcombe and Kramer, 2003). Grey matter volume increases were found in the cingulate (Colcombe et al., 2004; Jochem et al., 2017) the frontal cortices (Colcombe et al., 2006; Kramer and Erickson, 2007) (Ruscheweyh et al., 2011), and the hippocampus (Erickson et al., 2011; Raichlen et al., 2019). The positive changes associated with physical activity were found in motor and cognition related areas of the brain.

When there is an absence of a stimulus, there is no demand for neuroplasticity to occur. The proposed Adaptive Capacity Model (Raichlen and Alexander, 2017) showed that in inactive populations

the brain adapts to the lack of activity and early lifestyle affects the progression in brain adaptation over the lifetime. The lack of physical activity isn't deemed as concerning as the presence of disease, however, a lifestyle lacking physical activity has been shown to have debilitating effects on health (De Rezende et al., 2014). Brain health is negatively affected by degenerative conditions, however, what is not often mentioned is that in the absence of degenerative disease a lack of physical activity can also negatively impacts brain structure (Camandola and Mattson, 2017). Exercise is often recommended as a means to improve cognition, physical health, and brain health. The impact of exercise has been shown to directly impact the volumes of total and regional grey matter. A review by Batouli and Saba (2017) showed that a large percentage of regional grey matter could be influenced by physical activity (Esteban-Cornejo et al., 2018). Larger grey matter volumes have been shown to be important because of their positive association with improved cognitive function and better physical function, whereas when grey matter atrophy was more prominent, studies showed that it was associated with degenerative conditions such as multiple sclerosis, aging, narcolepsy and even fibromyalgia (Raz et al., 2005; Ellingson et al., 2012). White matter degeneration is equally worrying. White matter has shown positive responsiveness to physical activity and it was found that white matter conditions improved with physical training and the deleterious effects of aging are slowed down (Sexton et al., 2016). White matter preservation plays a key role in disease prevention, whilst white matter lesions and degeneration were associated with Alzheimer's disease and Parkinson's disease (Erickson et al., 2012; Camandola and Mattson, 2017). In a 70-year-old population over a period of three years a linear decrease of white matter and physical function were found (Aribisala et al., 2013). Whilst physical activity is associated with improved brain function and structure, this concludes that the mere absence of physical activity could result in structural brain setbacks that are related to function. These changes in the structure of grey matter and white matter are a result of changes in the microstructure.

1.1.12 Physical Activity-Induced Brain Microstructure Plasticity

Changes in grey and white matter volumes reflect the neuroplasticity of the macroscopic structure. However, these changes are caused by processes that occur at a microscopic level (Zatorre et al.,

2012). The mechanisms by which physical activity can cause structural changes is not fully understood but can be attributed to a variety of factors. The review (Thomas et al., 2012) summarises several physical activity-related mechanisms that can be used to explain the metabolic changes which alter brain structure such as angiogenesis, neurogenesis, glial volume, and biochemical changes. In animal models, angiogenesis was upregulated with physical activity and likened to changes found with physical activity such as increases in insulin-like growth factor-1(Kramer and Erickson, 2007). Neurogenesis is the generation of new neurons that impact brain structure through increases in matter volume. Physical activity was found to be a promotor of neurogenesis (Cotman and Berchtold, 2002). The increased mitochondrial biogenesis associated with exercise brings with it benefits such as increased blood flow and vascularisation, which may be associated with the maintenance of brain structural integrity (Mattson et al., 2008; Bernardo et al., 2016). In particular, aerobic training adaptations may result from increased vascularisation, capillaries or resetting of the autonomic nervous system (Thomas et al., 2012). Biochemical changes at the molecular level include increasing gene expression and neurogenesis that play a role in the structural changes as a response to exercise (Cotman and Berchtold, 2002). Brain-derived neurotrophic factor (BDNF), expressed from the BDNF gene, which is responsible for maintaining and regenerating nerve cells in the brain was found to also be a positive contributor to brain matter changes (Ruscheweyh et al., 2009). When subjects engaged in physical activity it was shown to increase levels of BDNF, and the increase in the levels of BDNF was positively associated with increasing intensity of the exercise (Ferris et al., 2007; Kyun Jeon and Ho Ha, 2017). Glial cells which are non-neuronal cells also increased with physical activity, this increased glial volume, in turn, increased the total intracranial volume. The changes occurring in these micro-mechanisms of the brain in response to physical activity result in the changes to global and regional grey matter and white matter changes (Feldman, 2009; Sexton et al., 2016; Thomas et al., 2016).

1.1.13 The Effects of Different Modes of Physical Activity on the Brain

Physical activity is a broad term that can be categorised into different modes of activity. Different modes of activity have varying energy demands and neuromuscular demands which can impact the brain

differently (Powell et al., 2011; Voelcker-Rehage and Niemann, 2013). Literature comparing the two most common forms of physical activity, cardiovascular aerobic activity versus strength training on brain structure is limited. Previous research focuses on aerobic vs motor training. The review by Voelcker-Rehage and Niemann (2013) reviewed studies that compared cardiovascular activity and coordinative physical activity. In their review, the authors found that low metabolically challenging physical activity such as stretching and toning had no significant effects on brain structure. Co-ordinated motor tasks and cardiovascular physical activity both showed increases in hippocampal volume but at different rates (Niemann et al., 2014). (Bezzola et al., 2011) found that 40 hours of golf practice, a low intensity but more coordinative forms of physical activity resulted in grey matter increases in sensorimotor regions and that grey matter changes in the parieto-occipital junction were found to be greater with more daily hours as opposed to spreading the total training over a greater number of days. Our intensive search of the literature found that most of the studies don't discern resistance training from aerobic training and collectively have classified them as metabolic exercise. However, the majority of the studies recommend physical activity as a positive impactor on grey matter volume with a focus on aerobic activity (Hillman et al., 2008; Thomas et al., 2012; Voelcker-Rehage and Niemann, 2013). Liu-Ambrose et al., (2012) found that weekly resistance training which also places a metabolic load in an older population positively affected brain structure and found that regions affected by resistance training may be different from those affecting aerobic adaptations. A combination of aerobic and resistance training was found to have a greater effect on brain structure than aerobic training alone (Colcombe and Kramer, 2003). Motor fitness requiring co-ordination, spatial awareness, and higher-level cognitive information processing, rather than cardiovascular fitness was associated with hippocampal volume(Niemann et al., 2014). However, with 12-month interventions they both showed improvements. The literature mainly discusses the positive effects of aerobic physical activity but shows that brain structure is positively impacted by all modes of physical activity.

1.1.14 The Role of Training Load (volume, intensity, and frequency of physical activity) and Recovery on Brain Structure Changes

Training load is encompassed by duration, volume, and intensity of physical activity. Varied forms of physical activities place different demands on the body. The training load needs to fit the population that it is prescribed to (American College of Sports Medicine, 2013). This may result in differences in the magnitude or types of changes it has on physiological systems (Powell et al., 2011). The training load provides a stimulus to which the body must adapt. For physiological adaptations to occur the stimulus needs to be large enough, this is known as functional overreaching (Halson and Jeukendrup, 2004). When the stimulus from physical activity is too great in volume and intensity without adequate recovery, overtraining can occur (Fry et al., 1991; Halson and Jeukendrup, 2004). The effect of the stimulus varies based on activity levels. Gow et al., (2012) showed that the introduction of physical activity even if the activity did not meet (World Health Organization, 2010) guidelines, had a positive impact on neuroplasticity. In a study by Bugg and Head (2011) on how exercise moderated age-related grey matter losses, the authors found that the relationship between aerobic fitness was not directly impacted by the volume they had partaken in. Varying intensities of physical activity was shown to impact levels of BDNF, a factor derived from the BDNF gene, that is involved in neuroplasticity changes and was shown to be correlated with lactate, a marker of individualised intensity of exercise (Ferris et al., 2007; Flöel et al., 2010; Kyun Jeon and Ho Ha, 2017). In rodents, Rhodes et al., (2003) found that although neurogenesis increased with physical activity there was a certain point of extreme physical activity levels where it no longer increased in line with physical activity. Even minimal physical activity results in positive brain matter changes, but extreme activity may not be more beneficial than moderate levels of physical activity. Wood et al., (2016) compared master's endurance athletes to a healthy population that adhered to the minimum physical activity guidelines for at least five years and found that cortical grey matter and thickness were greater in the masters athletes, however sub-cortical grey matter peaked at the recommended guidelines. Wood et al. (2016) further found that masters athletes who were engaged in > 15 hours of weekly activity showed no significant differences in white matter hyperintensities and inflammatory markers as compared to their matched controls that performed moderate amounts

of physical activity, which is often found with excessive stress and overtraining.

Extreme levels of physical activity on the other hand have shown decreases in total intracranial and grey matter volumes (Freund et al., 2012; Freund et al., 2014). The authors (Freund et al., 2012; Freund et al., 2014) suggested that this may be from the greater than normal allostatic loads that result in overtraining (Kemp and Quintana, 2013; McEwen, 2007; Thayer and Sternberg, 2006). This in turn can lead to hypercortisolism (Bourdeau et al., 2002) and is often found in ultra-endurance athletes (Knechtle and Nikolaidis, 2018). Grey matter volumes were reduced with increases in cortisol, however when cortisol levels were reduced grey matter losses were reversed and a correlation between grey matter increases and cortisol reductions were found (Castro-Fornieles et al., 2009).

Sleep is an important aspect of recovery and plays an important role in managing the training load. Adequate sleep is important for recovery and the prevention of overtraining (Hynynen et al., 2006). Brain matter atrophy rates and white matter hyperintensities were greater in healthy individuals who slept less (Lo, Loh, et al., 2014). However, research studying sleep in ultra-endurance athletes and brain matter has not been previously conducted.

1.1.15 Brain Structure and Physical Activity in Healthy Populations

The benefits of physical activity are easily visible in debilitating disease and aging (Erickson et al., 2012; Freund et al., 2014; Pedersen and Saltin, 2015). However, the structural changes that occur with physical activity in healthy individuals who don't present with structural decline, may not be as easily detectable (Raz et al., 2005). Physical activity requires activating motor circuits; however in a study by Pascual-Leone et al. (2005) the authors showed that motor system plasticity is mostly a result of afferent feedback. This could indicate that feedback may be a driver of neuroplasticity in the motor cortex. Voxel-based morphometry studies in young and middle-aged populations are limited. When healthy young sedentary individuals engaged in physical activity, their anterior hippocampal volumes increased, however when they stopped exercising the regional grey matter returned close to baseline levels (Thomas et al., 2016). This showed that exercise could affect brain matter changes even in periods as short

as six weeks, but that the benefits were as easily reversed when they reverted to sedentary behaviour. The research on the benefits of physical activity in older populations and those with degenerative and metabolic conditions is vast (Bherer et al., 2013; Stuckey et al., 2014), however physical activity's impact on brain matter in younger healthy populations is documented only in a few studies. One of these studies (Peters et al., 2009) found that aerobic fitness was directly related to the right anterior insula volume, an area thought to be an integral part of cardiovascular fitness. However, the changes that physical activity has on brain matter structures may not be uniform across age groups. Demirakca et al. (2014) found that the posterior cingulate cortex and preceneus white matter was correlated with habitual physical activity only in subjects older than 40 years old. This may be because brain matter losses are accelerated with age. Indeed, in 2003 Colcombe and Kramer proposed that physical activity may be less correlated with brain volume in a younger population. Later, in 2006 Colcombe and co-workers found there were no significant differences in matter volumes in a younger population after a 6-month physical activity intervention, confirming their hypothesis. In a study involving a healthy young to middle-aged population (18-45 years old) Killgore et al. (2013) found a direct correlation between the minutes of weekly physical activity and right hippocampal volume. Williams et al. (2017) also found that cortical thickness in a young population was negatively associated with cardiorespiratory fitness and showed accelerated neural pruning, a physiological process where it is thought that extra neurons are reduced to enhance efficiency. This lends support to the notion that the effect of physical activity may have a slightly different mechanism in the brains of a younger population. In a young population of novice versus professional basketball players, regional grey matter was greater in the professional group in regions of the brain associated with motor and cognitive function, and was attributed to a higher level of motor skill and cognition required for elite basketball performance (Tan et al., 2016). In a judo study, greater grey matter volumes in judo athletes were found as compared to healthy sedentary controls in regions related to motor planning and execution (Jacini et al., 2009). It is clear that physical activity also has a positive impact on brain structure in healthy populations. More research in young healthy populations is needed as the literature is limited.

1.1.16 The Gap in the Literature: Ultra-endurance Training and the Brain

Moderate physical activity is well known for its benefits in the elderly and those with degenerative diseases as a preventative measure for brain matter losses (Batouli and Saba, 2017; Gow et al., 2012; Raji et al., 2016). Ultra-endurance event participation requires far greater physical exertion than moderate-intensity physical activity and in turn results in a higher caloric expenditure (Zaryski and Smith, 2005). With moderate physical activity, caloric expenditure is associated with greater grey matter volumes (Raji et al., 2016). There is limited literature on the effects of moderate levels of physical activity on structural brain changes in younger populations, but it is even more limited in ultra-endurance event participants. Ultra-marathons are a growing global trend with participation rates increasing yearly (Knechtle et al., 2011; Knechtle and Nikolaidis, 2018). Ultra-endurance events are longer and greater in distance than regular endurance events; they are cardiovascularly demanding physical activity events that exceed six hours in duration (Zaryski and Smith, 2005). Excessive physical activity in ultra-endurance participation has been found to cause excessive stress to other physiological mechanisms such as the heart, kidneys, and skeletal systems (Knechtle and Nikolaidis, 2018). The effects of a large weekly training load in preparation for an ultra-endurance event on structural brain changes have not been previously investigated. Only one research group has published data on volumetric brain differences during an ultra-endurance event (Freund et al., 2012, 2014). The study was limited and only assessed changes found with event participation in the Trans-Europe race and not pre-event training. The duration of the Trans-Europe race was unlike regular ultra-endurance events and extended over 64 days. The study imaged the brains of thirteen athletes aged 31-67 years old in the Trans-Europe race where runners ran approximately 70 kilometres daily for 64 days. The study found volumetric decreases in multiple brain regions. Changes were found in the bilateral posterior temporal, occipito-parietal brain regions, anterior cingulate cortex, and the right ventral caudate nucleus during the race. These changes were reversed in a scan 8-months post-race. The study not only found regional decreases in grey matter but the total grey matter was also decreased. The authors of the paper attributed the changes to high levels of hyponatremia, energy deficits, and hypercortisolism. Hypernatremia is associated with demyelination which in turn is associated with

white matter volume losses (Geurts and Barkhof, 2008). Energy deficits result in fat and muscle loss which are also associated with changes in brain volume (Kilgour et al., 2014). Hypercortisolism initiates metabolic processes which are related to cell injury and cell death that result in brain volume losses that were reversed when hypercortisolism was corrected (Bourdeau et al., 2002). The chronic exertion found with high levels of physical activity as seen in extreme ultra-endurance events like the Trans Europe race cannot be translated to training for single day ultra-endurance events and the effects it has on the brain. This greatly warrants further investigation into the effects that large weekly training loads required for ultra-endurance participation, have on the brain.

1.1.17 Measuring the Physical Activity- related Changes in Global and Regional Matter

Increasing global and regional matter volumes is important for maintaining and improving function. However, measuring the effectiveness of interventions and the role of degenerative conditions on grey and white matter volumes is equally important. In early brain research, animal studies and post-mortem research were used to provide insight into the brain (Andreasen, 1988). Research into the changing nature of the brain has evolved and is now powered by technology using methods such as Computer Tomography (CT), Positron Emission Tomography (PET), electroencephalography and Magnetic Resonance Imaging (MRI) to provide insight into the living brain (Andreasen, 1988). MRI imaging is a popular non-invasive technique used in medical practice and is safer than methods employing the use of radiation (e.g. CT, or PET) and offers three-dimensional insight on the structure and function of the brain (Wenzel, 2017). MRI images can be analysed using voxel-based morphometry (VBM), which is a simple quantitative technique to assess structure and size (Ashburner and Friston, 2000). VBM is used to statistically analyse structural brain anatomy differences across groups. To ensure validity, reliability, and repeatability, the data processing and analysis need to adhere to simple guidelines and methods which must be clearly defined (Ridgway et al., 2008). T1-weighted MRI images are used to assess voxel-wise differences. T1-weighted MRI images can detect tissues with high-fat content and appear bright, whilst fluid-filled compartments appear dark and are thus good for assessing structure. These images are pre-processed, segmented by tissue type (i.e., grey, white, and cerebrospinal fluid), smoothed and

normalized to a standardised brain template so that they can be compared in a standardised (common) space (Ashburner and Friston, 2000).

1.2 Cognitive Function and Physical Activity

1.2.1 Cognitive Function and the Indication it gives of Brain health

Maintaining brain health is important for keeping cognitive function optimal. A key component of cognitive function is working memory. The performance of the brain in its cognitive functions is shaped by learning, trauma, and aging. The three main components of working memory are Central Executive, Articulatory Loop, and Visuo-Spatial Scratch Pad (Baddeley and Baddeley, 2018).

1.2.2 Working memory a component of cognitive function

Working memory is a crucial component of cognitive functioning and has been defined as a temporary store of information that is important in the performance of complex cognitive tasks (Baddeley, 1992). Working memory encompasses memory, attention, and perception, and is necessary for the manipulation of information to perform cognitive tasks such as language comprehension, learning, and reasoning (Baddeley, 1992). Like the structure of the brain, working memory develops during childhood, peaks during adulthood, and declines in old age (Cowan, 2014). Impaired working memory is associated with general cognitive decline (Baddeley, 1992). Maintaining working memory is important for daily functioning and has been shown to decrease with aging. Improving working memory with interventions has been successful, but showed different results in different age groups (Bherer et al., 2013). When working memory training took place the cognitive improvements were significantly less in older populations versus younger populations due to a decreased level of neuroplasticity in the aging brain (Rhodes and Katz, 2017). In animal studies, the decline in cognitive function, in particular working memory, was related to neural structural changes in the prefrontal cortex (Wang et al., 2011). The well performing subjects performed well on divided attention task because they had the ability to perform well on each individual task and not because of their ability to divide their attention. This cognitive ability is related to working memory capacity (Conway et al., 2001).

1.2.3 Tests of Working Memory

The N-back task is a test of working memory that requires remembering a small number of figures in their correct order and is similar in principle to the running-memory task and the memory-updating task (Oberauer et al., 2000). Owen et al. (2005) concluded in their meta-analysis that the N-back task is a reliable measure of working memory. The task tests short term memory recall using letters and numbers in which the subject must respond to the stimulus only if they see that the current stimulus is the same as the one presented N trials ago. N- e.g. 0-back (stimulus matches a pre-defined target e.g. respond to all the letter 'Cs), 1-back (stimulus matches target in the immediately e.g. letter 'A' followed by the letter 'A'). The N-back task is presented on a screen and participants are provided with a keypad to respond. The N-back score is auto-generated by the software based on the correct responses, incorrect response, and no responses. A larger number of correct responses have been associated with greater working memory capacity, accuracy, and reaction time can be determined from the test as well (Meule, 2017).

1.2.4 The Link between Physical Activity, Brain Structure, and Cognitive Function

Cognitive function and brain structure are interrelated components. The structure of the brain is important in determining the level of cognitive functioning (Qing and Gong, 2016). Physical activity has been associated with positive changes in both structure and cognitive function (Verstynen et al., 2012a; Benedict et al., 2013). In a study by Flöel et al. (2010) exercise was associated with grey matter volume increases in the prefrontal area of the brain and these changes were found to correlate with memory performance, thus impacting cognitive functioning. The review by Kramer and Erickson (2007) suggested that the improved cognitive capacity associated with physical activity was due to a reduction in the risk of degenerative diseases that negatively impact cognitive function; as well as preserve the structural integrity of the brain which helps to maintain function. Previous research in obesity-related grey matter atrophy, found the atrophy to be linked to cognitive decline; and that with physical activity the grey matter volume and related cognitive decline could be improved (Jagust et al., 2005; Jagust, 2007; Bugg et al., 2012; Mueller et al., 2015). White matter microstructure atrophy was also related to

cognitive decline in individuals with elevated BMI (Alarcón et al., 2016). Raichlen and Alexander (2017) summarised that physical activity is linked to cognition because the demands of exercise, in particular at high levels of physical activity, require motor control, memory, spatial navigation, and executive function. The authors proposed a model showing how levels of physical activity improved cognition over the life span whilst simultaneously decreasing the risk of degenerative related brain matter losses and how if physical activity was implemented earlier in life the positive effects would be greater (Kramer and Erickson, 2007). It has proved to be an important link between improved brain structure and higher levels of cognitive function.

1.2.5 Impact of Physical Activity on Working Memory

The benefits of physical activity are not limited to physiological function; the positive adaptations of physical activity are also associated with positive effects on cognition (Voelcker-Rehage and Niemann, 2013). Physical activity has been shown to cause a neuromuscular stimulus that creates a demand for the brain to adapt (Lövdén et al., 2010). These exercise-induced adaptations have been associated with improved performance on working memory and cognitive tasks (Hansen et al., 2004). Greater levels of physical activity, independent of the cardiovascular benefit, were shown to improve memory function and were related to grey matter changes in the prefrontal and cingulate cortices (Flöel et al., 2010). The authors attributed the changes to the higher levels of neurotrophins they found with increased levels of physical activity (Flöel et al., 2010). In a study on the effects of physical activity and BDNF in an adolescent population, BDNF was increased with a single exercise bout. Independently research found that BDNF was also related to cognitive function (Kyun Jeon and Ho Ha, 2017). Exercise has been shown to slow down grey matter-related cognitive decline (Bherer et al., 2013; Stillman et al., 2016). In obese individuals decreases in matter volume were associated with cognitive decline (Jagust, 2007; Bocarsly et al., 2015). Jagust et al (2005) found that aerobic fitness was responsible for the differences in obese hippocampal volume and the variation in cognitive ability. When obese individuals were put on an exercise intervention, brain volume changes were found in cognition related areas (Mueller et al., 2015). There is clear evidence that physical activity positively impacts cognitive function. Aerobic

activity, in particular, has been shown to impact cognitive function greatly with changes more evident in aging populations (Hillman et al., 2008).

1.2.6 The Volume of Habitual Exercise and Cognitive Performance

Different physical activity levels have been shown to impact brain structure differently which can differently impact cognitive function (Hamer, Sharma and Batty, 2018). The benefits of physical activity on cognition were seen in a study by Ruscheweyh et al. (2009) that found a positive relationship between the volume of physical activity and performance on a memory recall task, indicating that increased activity levels lead to improved working memory (Foley and Fleshner, 2008b). In a study in an obese population physical activity was shown to increase grey matter in regions of the brain related to cognitive performance, and furthermore may reverse the maladaptive effects of obesity (Mueller et al., 2015). Cognitive performance was shown to be directly related to the level of fitness and hippocampal volume (Bugg et al., 2012). The mechanism of changes in cognitive function with physical activity has been associated with changes in dopamine neurotransmission if the stimulus was rewarding enough (Schultz, 1998).

1.2.7 Dopamine Neurotransmission and Cognitive Function

The dopamine hypothesis described by Rhodes et al. (2005) summarised that physical activity may be a natural reward in high activity mice, similar to food and drugs. Dopamine neurotransmission is regulated better in physically active populations (Foley and Fleshner, 2008). In animal studies, greater levels of dopamine were found in highly active rodents as compared to controls (Rhodes et al., 2005). Neurotransmission is the conduction of nerve impulses between neurons or between neurons and muscles. Dopamine neurotransmission is increased with physical activity levels. However, impaired dopamine neurotransmission was found to encourage sedentary behaviour making it more challenging to overcome already low levels of dopamine neurotransmission. Köhncke et al (2018) found that motivation to engage in physical activity was associated with dopamine neurotransmission in the caudate nucleus in the striatum.

The impact of physical activity on dopamine is found acutely and is further influenced by the volume of physical activity. Ruscheweyh et al. (2009) found that increases in volume directly influence increases in dopamine levels. Sedentary behaviour, on the other hand, has been shown to decrease dopamine neurotransmission. The review by Foley and Fleshner (2008a) found in animal studies that habitual activity affected plasticity in central neural circuits and with habitual physical activity dopamine synthesis was increased. Sutoo and Akiyama (2003) described how brain regulation changes in response to physical activity increased dopamine synthesis and levels, which in turn was shown to regulate the receptivity to drugs. Thus, it is clear that dopamine transmission plays a key role in brain function.

Dopamine neurotransmission has been linked to cognition, more specifically to top-down selective attention which is a factor in working memory (Noudoost and Moore, 2011). When neurotransmission of dopamine was disrupted it was shown to impact cognitive ability and cause cognitive impairment (Nieoullon, 2002). In animal studies, cognitive performance was impaired in mice bred for running, even though neurogenesis increased (Rhodes et al., 2003). This indicated that cognitive processing was disrupted in hyperactive mice and may have resulted from impaired dopamine transmission (Rinne et al., 2000).

Executive functioning is associated with the D2 receptor availability indicating that dopamine transmission plays a role in executive functioning (Volkow et al., 1998; Bäckman et al., 2000). Ruscheweyh et al. (2009) summarised the relationship between brain structure, cognitive function, and physical activity and found that in the presence of increasing physical activity there were positive associations with the prefrontal cortex cingulate grey matter structure, dopamine neurotransmission, and cognitive ability. However, the role of extreme levels of physical activity has not been previously investigated.

1.2.8 Relevance in a South African Context

The South African healthcare system is under pressure, being overburdened by non-communicable diseases like obesity, diabetes, and cardiovascular disease (Mayosi et al., 2009). Public healthcare is the fourth largest expenditure of the government (Statistics South Africa, 2017.). This burden can be eased by a simple solution like exercise. When South African employees met physical activity guidelines healthcare costs were reduced (Kolbe-Alexander et al., 2010). However, compliance to exercise as a therapeutic medium is lower than expected, and to tackle this, the mind-set toward exercise must be shifted. Research on the brain provides a further argument in favour of improving compliance to exercise Minimising cost and decreasing the burden on our healthcare system is warranted so that more serious healthcare issues can take preference. Health care practitioners are always advocating for more physical activity. Most individuals are however not meeting the recommended daily guidelines, which have been shown to prevent the development of disease and degenerative conditions (Theofilou and Saborit, 2013). Previous studies have shown moderate activity can slow the rate of brain atrophy. Exercise as a therapeutic medium is one of the few mediums shown to decrease declining rates of brain matter and to slow cognitive decline (Boutayeb and Boutayeb, 2005). Increasing the levels of physical activity participation in South Africa can help ease the burden on the healthcare system.

2 Physical Activity Level and Brain Structure

2.1 Introduction

Physical activity requires energy to be expended in a variety of biological systems (Caspersen et al., 1985) that respond through physiological adaptation. For example, the energy demands on skeletal, neuromuscular, and endocrinological during physical activity can temporarily disrupt homeostasis and lead to positive adaptation of bodily systems (McEwen, 2002). The physiological benefits of physical activity are vast and it is associated with positive effects on all aspects of the human condition when undertaken in a safe and fairly guided manner (Haskell et al., 2007; Pedersen and Saltin, 2015; Petersen and Pedersen, 2005). Habitual physical activity is often associated with being healthier; it is often advocated as a preventative and therapeutic medium for degenerative conditions such as diabetes, cardiovascular disease, and Alzheimer's (Pedersen and Saltin, 2015).

Physical activity-related adaptations are not limited to skeletal or endocrinological systems and include neuroplasticity of the brain structure (Pascual-Leone et al., 2005; Pascual-Leone and Taylor, 2011). Changes in response to physical activity have been observed in grey matter, white matter, and total intracranial volume of the brain (Batouli and Saba, 2017; Bugg and Head, 2011). These structural changes that are derived from physical activity as a stimulus which affect neuroplasticity, are not limited to structure because a resultant functional change is inevitable (Voelcker-Rehage and Niemann, 2013). Maintaining a healthy brain structure is important for keeping our physiological and cognitive function as optimal as possible throughout the life-span (Voelcker-Rehage and Niemann, 2013). Research has shown that sedentary behaviour negatively affects brain structure by accelerating grey matter and white matter atrophy (Bugg, Shah, Villareal, and Head, 2012; Kilgour et al., 2014; Torres, Strack, Fernandez, Tumey, and Hitchcock, 2015). The brain being plastic in nature allows it to adapt to stressors; but it also adapts to sedentary behaviour (Raichlen and Alexander, 2017).

Moderate physical activity as per the recommendations has not only shown to be associated with a decreased level of brain matter atrophy but research has also shown that it can improve the

volumes of grey and white matter structures over a large portion of the brain (Batouli and Saba, 2017). The response of brain structural change to physical activity is more noticeable in aging populations as the rate of decline is much larger and allows for positives changes to be easily detectable (Erickson et al., 2014; Sexton et al., 2016). Research has shown that the mechanism for these adaptations could be due to the contribution of multiple factors such as changes in gene expression, hormones, and cellular structure (Thomas et al., 2012). Structural adaptations in grey matter and white matter are crucial to everyday function as it is related to cognitive and physiological function (Honey et al., 2010). Previous literature largely focused on physical activity-related changes in elderly populations. These studies have consistently shown a positive relationship with brain structure, cardiovascular function, and cognition. Sedentary behaviour and low levels of physical activity in aging populations is not only consistent with age-related brain matter decline but concurrently affects cardiac function, and in turn, is associated with elevated resting heart rate. This indicates that some form of autonomic nervous system dysregulation exists. Autonomic nervous system imbalance has also shown improvements with physical activity (Mueller, 2007). Resting heart rate is an indicator of autonomic nervous system balance and fitness (Jensen et al., 2013). Evidently there exists an interaction between fitness and brain structure (Bugg and Head, 2011). Furthermore, associations have also been found between the structure of the brain and the autonomic nervous system (Carnevali et al., 2018; Winkelmann et al., 2017). While research has documented the effect of physical activity in older individuals and those with degenerative conditions such as Parkinson's disease, it is not as well documented in younger, healthy populations who habitually engage in ultra-endurance training and participate in ultra-endurance events.

The guidelines for physical activity to maintain health and prevent disease as recommended by the WHO are 150 minutes of weekly MVPA for adults aged 18-64 (World Health Organization, 2010). These recommendations are often not met and sedentary behaviour has become an epidemic (González et al., 2017). MVPA causes a transient physiological challenge that results in positive stress on the physiological systems of the body (McEwen, 2007; Nagai et al., 2004). Acute and habitual stimuli from MVPA create a demand for the systems to adapt to, from the increasing skeletal and metabolic demands

(McEwen, 2007). The physical activity spectrum can range from sedentary behaviour to large volumes of physical activity such as ultra-endurance participation.

The popularity of ultra-endurance athletic events has made it a global trend. Ultra-endurance events are cardiovascularly challenging events that last longer than 6 hours (Zaryski and Smith, 2005). Training for these events far exceeds the recommended guidelines for MVPA and normative moderate-vigorous physical activity participation (Knechtle et al., 2011). The training regimen for these events if not periodised correctly and without adequate recovery may have adverse health effects on the cardiovascular and skeletal systems (Knechtle and Nikolaidis, 2018). When individuals that engaged in habitual aerobic physical activity of > 15 hours weekly were compared to healthy individuals meeting the minimum guidelines, grey matter was greater in the >15 hours group with no inflammation or white matter hyperintensities present (Wood et al., 2016). Grey matter losses have been documented in (Freund et al., 2014) in an ultra-endurance event spanning weeks in duration. This study however investigated extreme activity levels that far superseded the large training loads for once-off ultra-marathon events.

The rationale for this investigation is to address a gap in the literature. Are there correlations between brain structure and habitual physical activity levels? Are the high habitual levels of physical activity in ultra-endurance athletes detrimental to their brain structure and function?

2.2 Aim and Objectives

This research chapter aimed to investigate whether brain matter, specifically total grey and total white matter, as well as regional grey matter, was increased in individuals who habitually partake in extreme levels of physical activity vs. individuals who partake in low levels of habitual physical activity.

The first objective was to measure the differences in global brain volumes of individuals who habitually partook in extreme levels of physical activity as compared to individuals who habitually partook in low levels of physical activity. The second objective was to identify significantly different regions of interest in the brain between the two groups.

2.3 Methods

2.3.1 Ethical Approval

Participants provided written informed consent before participation. This study was approved by the human research ethics committee of the University of Cape Town, South Africa (HREC 199/2017) and was carried out in accordance with the Declaration of Helsinki on the use of human participants in experiments.

This study was conducted in conjunction with another thesis examining the influence of methylphenidate on heart rate and brain connectivity. As a result, subjects had to meet other criteria, which are not relevant to this thesis.

2.3.2 Participants

We recruited 21 healthy participants without any history of cardiovascular, metabolic, neurological, and psychiatric diseases. Participants were recruited using posters, advertisements on social media, word of mouth, and from athletics clubs. Recruitment posters contained the purpose of the study, an outline of procedures, and necessary information to contact the experimenters (e.g. telephone number, email

address). Researcher's contacted interested participants by telephone and reviewed the details of the study. Participants were asked exclusion/inclusion questions (see below) such as the presence of cardiovascular-related disease, claustrophobia, or if they were taking chronic medications. They were also asked about their average weekly physical activity habits. If participants met the minimum criteria for the sedentary or ultra-endurance population (see inclusion criteria in section 2.3.2.1), a familiarisation session at the Sports Science Institute of South Africa was scheduled.

2.3.2.1 Inclusion and Exclusion Criteria

We recruited two different population groups for this study based on their physical activity habits (see consort diagram (Figure 2-2). The first group (N=11) were healthy sedentary individuals (LA). This group was defined as those who engaged in voluntary physical activity less than 2 times a week and performing less than 2 hours of total physical activity. The second group (N=14) were healthy individuals who had high activity levels and engaged in voluntary PA for at least 9 hours a week (HA). They had to be right-handed. HA participants were included if they had been training for and participated in an ultra-endurance event (e.g. Ironman) for the last year. However, participants meeting this criterion were excluded if their ironman event was within 1 month of the study.

All participants completed a neuropsychological evaluation performed by a trained psychologist using the Mini-International Neuropsychiatric Interview (Sheehan et al., 1998). From this interview, participants were excluded if they (i) had a history of drug or alcohol abuse or dependence, (ii) had past psychiatric, neurological, cardiovascular or endocrinological disease, (iii) if they had taken psychoactive drugs (e.g. Opiate analgesics, stimulants or sedatives) or prescription medications in the last month. Further, we also performed saliva tests to test for the presence of psychotropic drugs such as cocaine, phencyclidine, benzodiazepines, cannabis, opiates, barbiturates, and inhalants (Detect a Drug Saliva Drug Test, Cape Town, South Africa). Finally, participants were excluded if they had any contraindications to MRI such as the presence of any pin, prosthesis, or any other magnetic materials inside/within their bodies that had been surgically implanted before 2008) (see consort diagram Figure 2-2). All of our subjects had

at least 1 year of post-secondary education but we did not control for ethnicity or socio-economic status.

2.3.3 Familiarisation

The 1.5-hour familiarisation session took place at the University of Cape Town, Division of Exercise Science and Sports Medicine at the Sports Science Institute of South Africa. Detailed information regarding the study risks/benefits was given to participants. Participants completed a Global Physical Activity Questionnaire (GPAQ), which is a self-administered questionnaire used to measure recreational physical activity, occupational physical activity, and active transport. This questionnaire asked questions regarding the duration, intensity, and frequency of physical activity in a typical week (World Health Organization). In the GPAQ, physical activity was defined by the amount the minutes of MVPA. Moderate activity was defined to participants as moderate physical effort causing small increases in breathing or heart rate. Vigorous activity was defined as hard physical effort causing large increases in breathing or heart rate. Height and weight were also measured to obtain body mass index (BMI). One participant in each group was excluded as outliers for BMI, which was defined as a z-score of greater than 2.68 (see consort diagram (Figure 2-2). As a result, our study included N=12 in the LA group and N=9 in the HA group. Participants were then familiarised with the sensation of an MRI in a custom-built mock-MRI scanner (Figure 2-1)

Figure 2-1 Mock-MRI scanner

Thereafter we scheduled two experimental sessions within a week. Experimental sessions 1 and 2 took place at Cape Universities Body Imaging Centre (CUBIC) at Groote Schuur hospital where the MRI and Electrocardiogram (ECG) were conducted. The duration of this session was approximately an hour, with the scan itself lasting 45 minutes. Each participant was tested at the same time and of the day with 3-7 days between sessions. Participants were asked not to consume alcohol and exercise 24 hours before the scan, and asked not to consume caffeine or food three hours prior to the scan. The study was limited in that we had to trust the participants to adhere to this. If participant's clothing contained metal objects that could not be removed they were fitted with a hospital gown. The ECG electrodes were placed on participants by the trained CUBIC technicians to measure heart rate and heart rate variability.

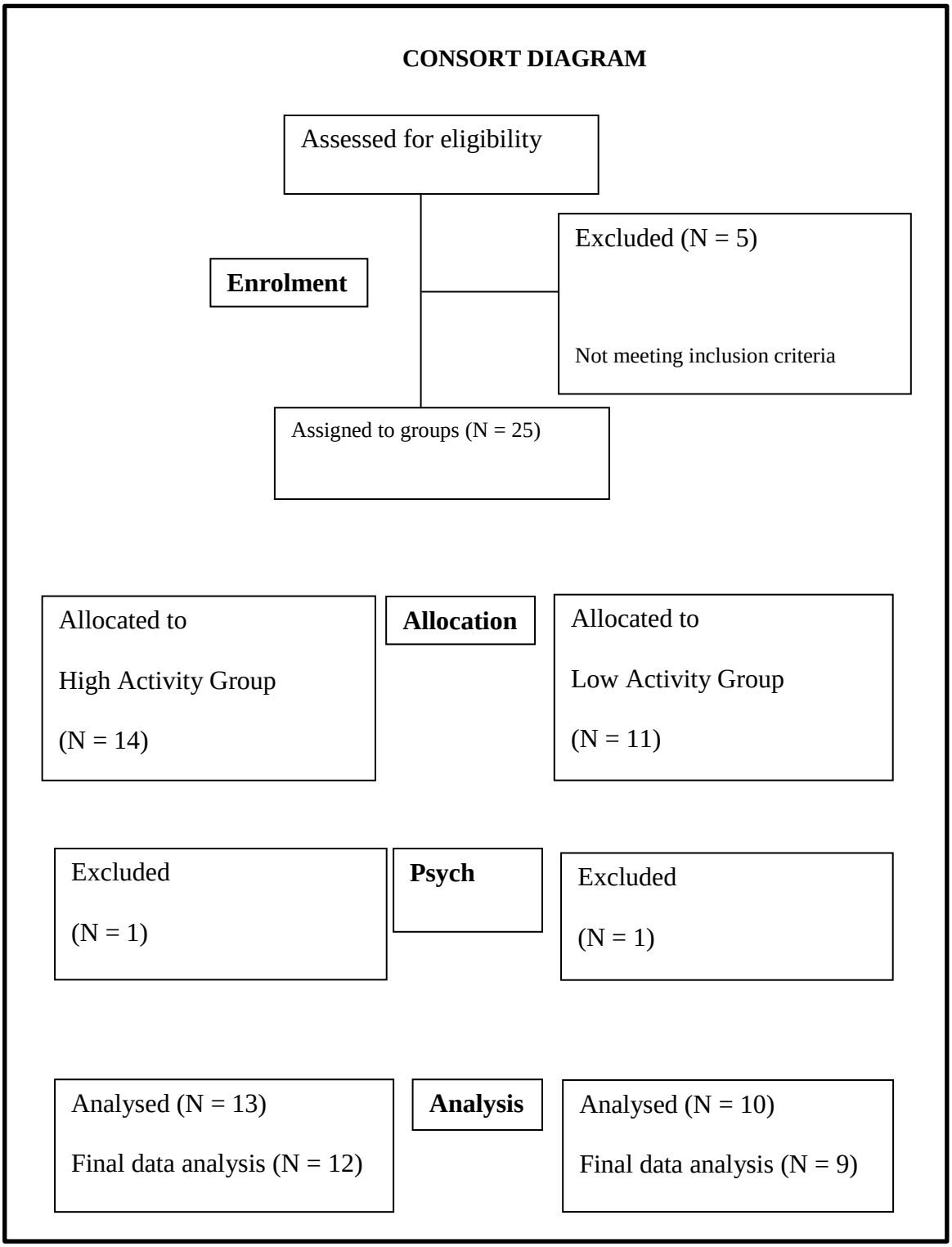

Figure 2-2 Consort diagram of the process of participant enrolment

(Figure 2-2) During the telephonic screening (enrolment phase) of participant recruitment, we excluded interested participants that did not meet the inclusion criteria (N = 5). Participants that met the criteria were allocated into the 2 groups (based on MVPA) and thereafter participants underwent a

psychological screening (N = 25). Participants who were not given approval by the psychologist to proceed in the study were excluded (N = 2). During the analysis step we had in the HA group (N = 13) and the LA group (N = 10). During this step, we excluded 1 participant from each group (N = 2) for outlying BMI's, BMI of 59.1, which was 3.65 standard deviations from the sedentary BMI mean for our recruited population.

2.3.4 MRI Acquisition, Processing, and Analyses

All participants were scanned using a 3 Tesla Siemens Skyra whole-body MRI scanner (Erlangen, Germany) at the Cape Universities Body Imaging Centre (CUBIC) at Groote Schuur Hospital in South Africa with a 32-channel head coil. For each participant a structural T1-weighted volume was acquired using a multi-echo magnetization prepared rapid gradient echo (MEMPRAGE) sequence (TR = 2530 ms, TEs = 1.59/3.4/5.21/7.02 ms, 7 degrees flip angle, voxel size = 1.14 x 1.14 x 1.50 mm, field of view 256 x 256 x 192mm^3, 128 slices). Images were analysed using the VBM 8 statistical package within SPM 8 (Welcome Dept of Cognitive Neurology, London, UK). The structural images files for each participant were pre-processed in the following manner: The file format was converted from DCM to NIFTII format using the software MRI Convert making it suitable for analysis in the next step. Using the SPM 8 package the matter probability maps were visually inspected using the display function in VBM 8. Firstly, using a cross-hair (aligning) the image to the T1-templates to correct for movement artefacts. All images were centralised to the anterior cingulate cortex and stereo-statically normalised into standard brain space template (MNI) (EPI-template provided by the Montreal Neurological Institute) and smoothed using an 8-mm Gaussian kernel toolbox which was done to reduce noise and reduce the effect of small potential errors in misregistration (http://www.fil.ion.ucl.ac.uk/~john/misc/VBMclass10.pdf).

All pre-processed smoothed images were segmented into global grey matter, global white matter, and cerebrospinal fluid using VBM 8. Total intracranial volume was also derived from this step.

Statistical analysis for demographic data was conducted using SPSS version 25 (IBM Corporation, NY, USA) and graphed using Prism (GraphPad Prism 5.0 software, La Jolla, CA, USA). T-tests were used to determine the differences between the HA and LA groups on age, BMI, and MVPA. To determine the magnitudes of significance effect size was calculated using Cohen's *d*. The incidence of gender in each group (Fisher's exact test) was used to determine whether there was a different incidence of male or female participants between group 1= (HA) and group 2= (LA).

To compare global brain volume differences, grey matter, white matter, cerebrospinal, and total intracranial volumes were extracted from the images after image processing was complete using segmentation in VBM 8. We compared grey matter, white matter, cerebrospinal and total intracranial volumes using one-tailed t-tests and effect size (Cohen's *d*, which indicates the number of standard deviation differences between the group means) using the SPSS version 25 (IBM Corporation, NY, USA) for windows.

The regional grey matter was compared between the HA and LA groups using an ANCOVA, to correct for differences total intracranial volume (Crowley et al., 2018; Hansen et al., 2015), age and gender (Luders et al., 2009) were included as covariates of interest using SPM 8 (The normalized and smoothed grey tissue probability maps were compared). We set a cluster threshold to $p < 0.05$ corrected for family-wise error (FWE). To further account for non-stationary biases associated with cluster significance in VBM analyses, all significant clusters were further corrected using the non-stationary correction toolbox (Ashburner and Friston, 2000). All voxels with a grey matter value of $p > 0.05$ were excluded and derived from the mean of grey matter and tissue probability maps, an absolute threshold masking of p= 0.15 was used to prevent the results from displaying white matter significances. T-contrasts were defined in the ANCOVA to determine the direction of significance; this is to identify which group is greater in the clusters that were significantly different between the groups. Significances were taken at the cluster level. MNI coordinates from clusters that showed significant differences between groups were used for structural identification in Pickatlas (Maldjianet al., 2003; Maldjian and Laurienti, 2004) and converted into Talairach co-ordinates using the Talairach client (Lancaster et al., 1997, 2000).

Using the Talairach co-ordinates, Marseille Bar software was used to extract the volumes of regions' interest for each participant.

We interpreted our results using the SPM Anatomy Toolbox 1.7. This toolbox is a probabilistic atlas defined by cytoarchitecture structure (Eickhoff et al., 2005) and was created to resolve the problematic use of macro-anatomical landmarks, which do not take into account microscopic architectonic organisation (Amunts and Zilles, 2001) which is aligned with function.

2.3.5 ECG Acquisition and data extraction

The ECG was conducted during the MRI Technicians at CUBIC used an alcohol swab was to clean the skin prior to the attachment of the electrodes. The ECG activity was recorded from three electrodes, two were placed sub-clavicularly bilaterally and one over the lower-left rib just above the anterior superior iliac crests; an ECG configuration known as Eindhoven's Triangle. The ECG recording began after five minutes of rest in the supine position and was recorded at a frequency of 400 hertz. ECG data was recorded and stored on the CUBIC server along with the participants' MRI data.

MATLAB 2017a (The MathWorks Inc. Natick, MA, USA) was used as the default processing software for the ECG files downloaded off the University of Cape Town's CUBIC servers. Script software was designed to filter noise caused by the scanner. The script run in MATLAB was able to detect the QRS complex of heart rate which represents ventricular depolarisation of the heart, where depolarisation triggers the heart to contract. Q waves represent depolarization of the interventricular septum and can also relate to breathing, R-waves are the largest waves as they represent mass depolarisation and the S waves represent final depolarization at the base of the heart. The R-R peaks were derived from the QRS complex of heart rate. Pre-processing of ECG data was completed to remove any noise and interference associated with the MRI scanner or other physiological parameters. The ECG sampling frequency was 400 Hz, this is sufficient for QRS analysis that can vary between 15- 30 sample points. The raw log files generated during the resting state scans were manually edited in order to remove artefact data and on/off signal triggers. The edited log files were then imported into MATLAB and the ECG waveform

was then analysed using a signal processing toolbox as well as a script that located the R peaks within the QRS complex wave. The script was adapted from the Peak detection package (https://www.mathworks.com/help/signal/examples/peak-analysis.html). The MATLAB script utilized a Savitzky-Golay FIR smoothing filter as well as a Blackman filter to remove high and low-frequency noise generated by the scanner during echo-planar acquisition with EPI sequence). Moreover, in the script, the R-peak detection algorithm was used to identify average heart rate values in beats per minute during the rest state conditions. Minimal peak distance of 0.5mm and a minimal peak height of 400 hertz were specified to filter out any false positives not associated with the QRS complex such as the P wave. Finally, ECG waveforms were visually inspected for each subject in order to ensure R-peaks were correctly identified.

2.4 Results

2.4.1 Demographics

Table 2.4.1 Participant characteristics

Variables	**High Activity Group (N=12)**	**Low Activity Group (N=9)**	**(p-value) [Cohen's *d*]**
Age (Years)	27.9 ±26.64	28.33 ±11.192	0.922 0.02
BMI (kg/m^2)	22.67 ±1.62	29.59 ±5.71	0.006** 1.86
MVPA (minutes)	1078.75 ± 407.86	18.0 ± 56.9	< 0.001** 3.56
Resting Heart Rate (BPM)	60.71 ±5.68	72.99 ±6.23	< 0.001** 2.18

Table (2.4.1) Participant information (mean and SD) of high activity and low activity populations. BMI=Body Mass Index; MVPA=Moderate-vigorous Physical activity; BPM=Beats per Minute Cohen's *d* effect size: small effect (>= 0.15 and < 0.40), medium effect (>= 0.40 and < 0.75) large effect (>= 0.75 and < 1.10) **indicates significance. Table (2.1) The T-test analysis showed that the HA and LA did not differ in age (p= 0.9). The LA group did have a greater BMI (p= 0.006; Cohen's *d* =

1.86) and lower MVPA scores ($p < 0.0001$; Cohen's $d = 3.56$). Resting heart rate was significantly in the LA group ($p < 0.001$ Cohen's $d = 2.18$).

2.4.2 Global Matter

Table 2.4.2 Mean global matter volumes for high active and low active groups

Variables	High Activity Group (N=12)	Low Activity Group (N=9)	(p-value) [Cohen's *d*]
Total intracranial volume (ml)	1549.95 ±142.12	1379.46 ±137.86	0.013** 1.28
Global Grey Matter (ml)	728.05 ±86.68	668.22 ±71.21	0.108 0.78
Global White Matter (ml)	599.62 ±59.92	497.22 ±60.57	0.001** 1.79
Global Cerebro-spinal Fluid(ml)	222.29 ±30.18	214.02 ±25.57	0.516 0.31

(Table 2.4.2) Global matter volumes (mean and SD) of high activity and low activity populations in millilitres and < 1.10) **indicates significance Cohen's *d* effect size: small effect (>= 0.15 and < 0.40), medium effect (>= 0.40 and < 0.75) large effect (>= 0.75).

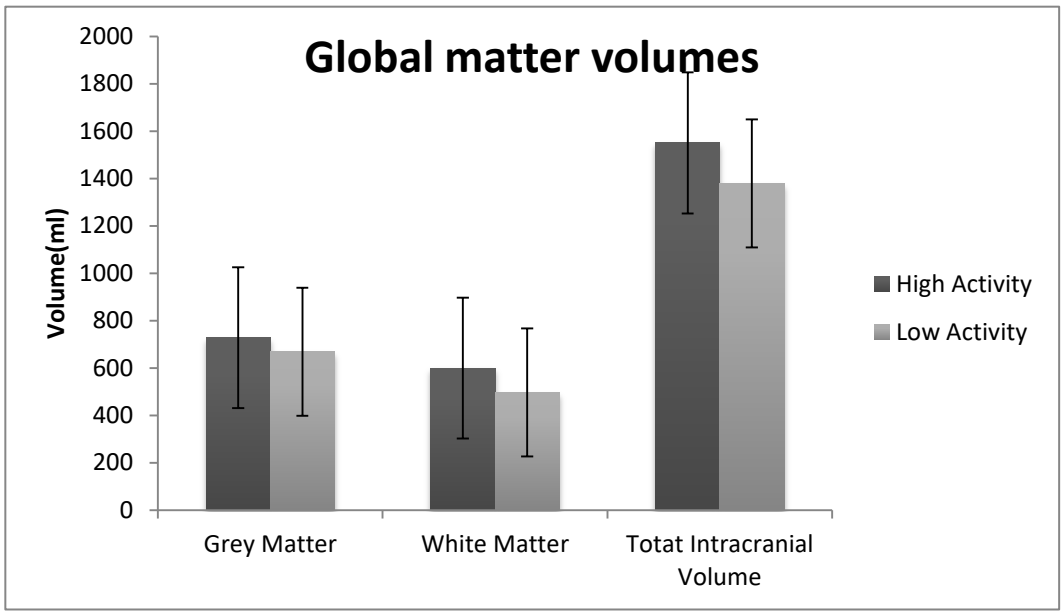

Figure 2-3 Mean global matter volumes for high activity and low activity participants: Grey matter, white matter and total intracranial volume in millilitres (ml).

Table (2.4.2) and (Figure 2-3) The T-test analysis showed the groups did not differ in global cerebrospinal fluid volumes (p=0.52) and global grey matter volumes although there was a trend towards higher global grey matter volumes in the HA vs. LA group (p=0.1; Cohen's *d* = 0.78) The HA group had significantly greater total intracranial volume (p=0.013; Cohen's *d* =1.28) and greater global white matter volumes (p=0.001; Cohen's *d* =1.79).

Table 2.4.3: Grey matter regions and coordinates in MNI space for regions, significantly different between groups (LA > HA)

Cluster #	FWE Cluster Significance	Cluster Size(s) (mm^3)	SPM Anatomy Macroscopic Label	Coordinate T - Value	Coordinate (MNI) (x,y,z)
#1	0.000	2006	R Middle Frontal Gyrus	7.09	48, 22, 42
	-		R Middle Frontal Gyrus	6.10	50, 46, 15
	-		R IFG (p. Triangularis)	5.66	52, 26, 22
#2	0.003	894	R Precentral Gyrus	5.58	45, -24, 64
	-		R Postcentral Gyrus	5.34	54, -21, 58
	-		R Precentral Gyrus	4.76	45, -12, 57
#3	0.034	532	L Thalamus	6.27	-9, -9, 18
			L Pallidum	5.79	-9, 0, 0
				3.65	-6, -9, -12

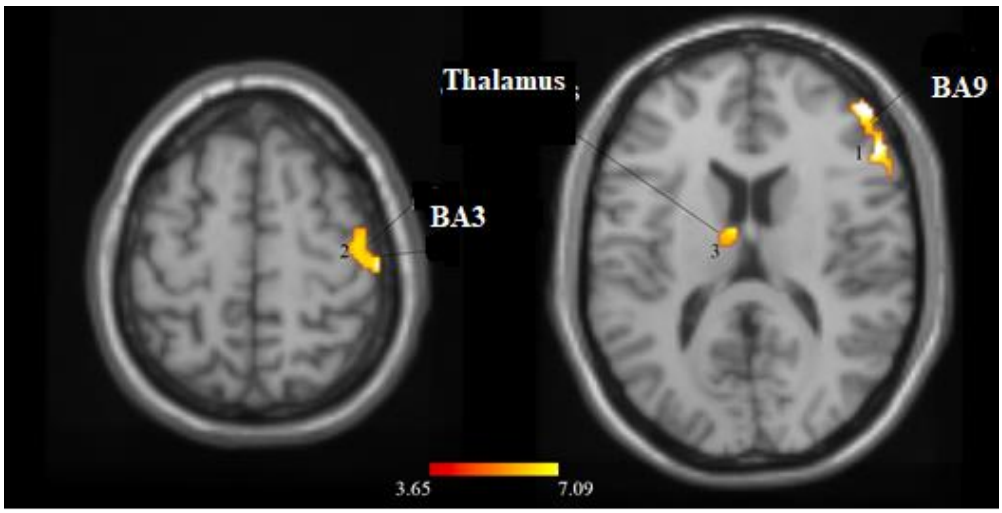

Figure 2-3 Axial brain slices showing significant regions of interest LA > HA: 3 clusters are located on a standard MNI template in an axial view of the brain 1) Right middle frontal gyrus (BA 9) Z=42; 2) Right post central gyrus (BA 3) Z=58; 3) Thalamus Z=18. The colour scale represents the significant co-ordinate T-values (FWE-corrected) p < 0.05).

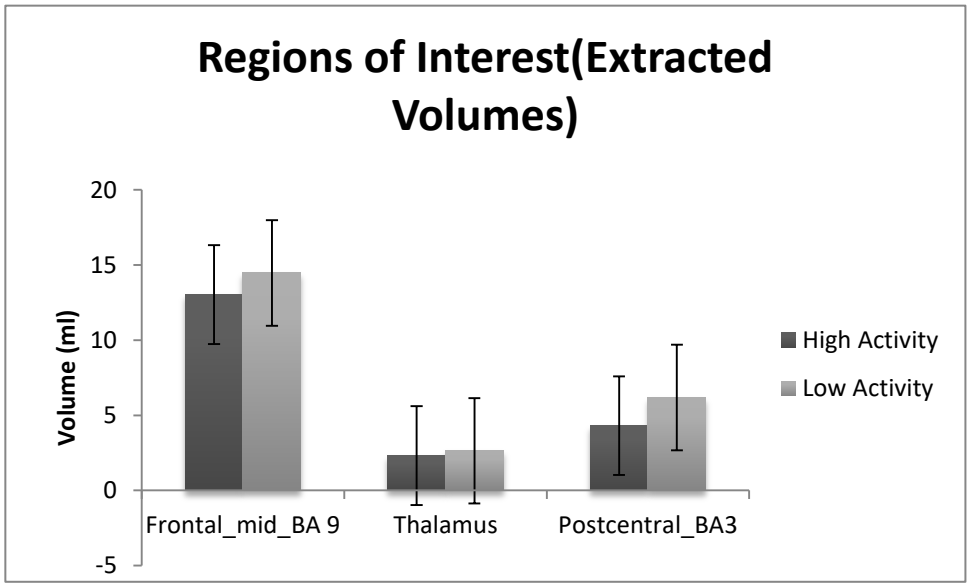

Figure 2-5 Regional grey matter volumes significantly different between HA and LA groups (LA > HA): Extracted volumes for the right middle frontal gyrus (BA 9), the left thalamus and right post-central gyrus (BA 3) in millilitres.

Table (2.4.3) presents the three significantly different clusters in the regional grey matter analysis we found as well as the sub-clusters that that form the regions of interest. The clusters were identified using their MNI co-ordinates and located on an MNI template in the axial view (Figure 2-4). The three clusters that were significantly different between the groups were the 1) right middle frontal gyrus including the inferior frontal gyrus, the 2) Right postcentral gyrus including the right precentral gyrus, and the 3) left thalamus including the left palladium. These regions were all significantly greater in the LA group. (Figure 2-5) shows the volumetric differences between the two groups in millilitres as extracted using MarsBar.

2.5 Discussion

2.5.1 Principal Findings

Our first finding was that total intracranial tissue volume and total white matter volume were significantly greater in the highly active group. We also found a statistical trend for higher total grey matter in the highly active population. This is consistent with previous findings that physical activity increases grey and white matter volumes (Batouli and Saba, 2017; Colcombe et al., 2006, 2004; Erickson et al., 2014). However, our second finding was that the high active population showed greater regional volume in the right middle frontal gyrus (BA 9), right postcentral gyrus (BA3), and the left thalamus.

2.5.2 Global Matter Findings

The role of physical activity levels and the brain structure was the cornerstone of this thesis. Brain structure maintenance and improvements are integral in maintaining cognitive and physiological function (Benedict et al., 2013; Hillman et al., 2008). Our findings showed that in a highly active and healthy population (Zaryski and Smith, 2005) higher total intracranial volume, global grey matter, and global white matter volume were achieved (Sexton et al., 2016), as was seen in previous research (Batouli and Saba, 2017; Torres et al., 2015). Importantly, these effects were large (see Cohen's *d* values in

Table 2.2) which indicates the powerful effect of high exercise levels on brain volumes.

Our findings however differed from other work (Jochem et al., 2017) that found no relation between total intracranial volumes and physical activity volumes, which may have been due to the large age range in their study. Previous research has focused largely on total matter changes in aging populations (Tseng et al., 2013), those with a degenerative disease (Camandola and Mattson, 2017; Thayer and Sternberg, 2010), obesity (Brooks et al., 2013), and recommended levels of physical activity (i.e. 150 min of MVPA/week). Our findings show that a larger than normal training load in preparation for an ultra-endurance event was associated with larger total matter volumes but without total grey and total white matter losses as were seen by Freund et al (2014). In the (Freund et al., 2012, Freund et al., 2014) study participants ran approximately 70 kms daily for 64 consecutive days. The global matter volumes in our HA group were greater than our LA group indicating that greater than MVPA recommendation levels over a long period confers a beneficial effect on global matter volumes. It is possible that excessive short term ultra-distance running has a negative effect on brain volumes whilst prolonged exercise with rest has a beneficial effect.

Our global grey matter results only showed a trend towards greater grey matter in the high active population, unlike previous studies that found the global grey matter was greater with larger volumes of physical activity (Flöel et al., 2010; Gow et al., 2012; Wood et al., 2016). Moderate levels of physical activity (Erickson et al., 2014; Gow et al., 2012) have been shown to cause increases in total grey and white matter. A limitation to our study is that we lacked a moderate activity group which would have allowed us to compare the brain structure of three different activity groups. Further research could investigate if a turn-point in grey matter volume increases exists. This could help to put the global matter losses that were found in the Trans-Europe race in perspective (Freund et al., 2012; Freund et al., 2014). The bulk of the literature on brain matter volume changes is more focused on changes in degenerative conditions and age-related declines in brain matter volume. This investigation found that the high active group had greater global white matter volume which is consistent with previous studies. It is important to note that global white matter volumes did not significantly decrease with aging (Good, et al.,

2001) and this investigation showed that even when the global white matter is not impacted by aging it physical activity can further maintain it. Deterioration of white matter associated with white matter lesions, which accelerate conditions such as dementia and Alzheimer's (Vermeer et al., 2003) have been slowed with physical activity (Burzynska et al., 2014; Torres et al., 2015). Brain matter volume loss is often researched in Alzheimer's which was further found to be associated with decreases in mitochondrial biogenesis (Bernardo et al., 2016). The increased blood flow resultant from angiogenesis may be related to the preservation of brain matter. A larger global white matter volume in the HA group ties in with previous research that showed that greater physical activity was associated with greater white matter volume and integrity (Benedict et al., 2013). Matter hyperintensities are a visual representation on MRI images that come up as areas of extra brightness and can be a marker reflecting demyelination and axonal loss in the brain. White matter hyperintensities were found to be inversely related to the volume of physical activity individuals engaged in (Torres et al., 2015).

Cardio-respiratory fitness could also be associated with the findings presented, as resting heart rate was significantly lower in the high active group and can be used as a fitness measure (Jensen et al., 2013). Most recently in the largest brain imaging and MVPA study white matter lesions were found to be inversely associated with both fitness level and MVPA and it was cardio-respiratory fitness, not MVPA that was associated with global grey matter (Raichlen, et al., 2019).

2.5.3 Regional Grey Matter Findings

A trend towards larger grey matter volumes was present in the HA group (Table 2.4.2) and (Figure 2-3). However, this investigation found clusters of significance were present in the right middle frontal gyrus (BA 9), right postcentral gyrus (BA 3), and left thalamus. These regions were significantly smaller in the HA group as compared to the LA group (Figure 2-5). This contrasted with previous findings in a review that concluded physical activity increased global and regional grey matter volumes and found no regional reductions (Batouli and Saba, 2017; Erickson et al., 2011; Erickson et al., 2014). The review by Batouli and Saba (2017) found the thalamus and postcentral gyrus increased with physical activity and that the

middle frontal gyrus was unchanged. The results in this investigation may have differed as the review focused largely on older populations. The contrasting findings in this investigation may be attributed to the larger than normal physical activity volumes, as seen in (Freund et al., 2012; Freund et al., 2014) that also found regional grey matter reductions, however; Freund and colleagues (2012, 2014) showed simultaneous global matter reductions that were not present in our findings and could be due to the extreme physical activity levels of participants in their study.

Significant differences in the postcentral gyrus (BA3) are highly relevant; this region contains the primary somatosensory cortex that is heavily engaged during physical activity. In lifelong athletes, this region was significantly greater in more active populations (Wood et al., 2016). Reductions in grey matter in the postcentral gyrus (BA 3) was found to be associated negatively with anxiety (Makovac et al., 2015). Although, previous finding that showed increasing physical activity to moderate levels of physical activity decreased anxiety disorders (Dietrich and Sparling, 2004). The MVPA in our HA group was far greater and the reductions in grey matter found with high activity levels may have psychosocial implications.

In the highly active group, a smaller right middle frontal gyrus (BA 9) was also found. This region forms part of the dorsolateral prefrontal cortex (DLFPC) and is thought to be related to co-ordinate spatial memory (Slotnick and Moo, 2006). Erickson et al (2014) found that with increased aerobic training spatial memory was improved, which could be associated with the demands of physical activity which require increasing spatial memory about the environment around them whilst being engaged in the activity. The DLFPC is functionally connected with the thalamus (Le Reste et al., 2016) and the basal ganglia. This shows an interesting connection in our study; we demonstrated that the ventral anterior part of the thalamus to be significantly smaller in the HA group. The thalamus is responsible for relaying motor and sensory signals from the basal ganglia and the cerebellum to the cortex (Hoover and Strick, 1999). With increasing motor and sensory stimulation that is found with increased levels of physical activity, there will likely be an increased need for the thalamus to be utilised. The increasing demand on the thalamus, needing improved neural efficiency could result in neural pruning which is

associated with smaller regional volume (Huttenlocher, 1990; Lowe et al., 2016).

Lastly, the regional investigation found a smaller right middle frontal gyrus (BA 9) in the highly active population, a region that was found to have reduced brain matter losses with physical activity (Colcombe et al., 2006). Erickson et al. (2014) found that the right middle frontal gyrus (BA 9) was one of the few regions to show up a consistent relationship between physical activity and increased grey matter volume. However, another study found no significant changes in a younger population with moderate physical activity (Colcombe et al., 2006). This is indicative that physical activity-related changes are not as easily detectable in young health populations as is seen in aging populations. However, some studies that looked at moderate levels of physical activity showed no changes in grey matter volume and may be explained by the populations they investigated, which were older adults (Erickson et al., 2011; Killgore et al., 2013). The physiology behind these differences needs further investigation. From previous research, we can only make speculations about the processes responsible for changes. Smaller regional grey matter and physical activity relationships may be more related to the strengthened white matter tracts than with neurogenesis related increases in grey matter (Raichlen and Alexander, 2017). This would serve as an energy-saving method while preserving grey matter volumes (Raichlen and Alexander, 2017).

Previous research has also shown the importance of physical activity in the maintenance of structure in order to maintain function. The review by Kramer and Erickson (2007) found that the risk of developing a neurologic disease and cognitive dysfunction differed amongst people and may be due to variations in their genes. Kramer and Erickson (2007) indicated that the variation in genes may also determine the effectiveness of physical activity in alleviating or reducing the negative effects of these degenerative conditions on brain structure.

This investigation found the regions that were different between the groups to be significantly smaller in the highly active group. The regions identified are involved in motor activities and as a result of our younger population, these changes may be a result of neural or synaptic pruning (Terribilli et al., 2011; Williams et al., 2017; Zatorre et al., 2012). As compared with the mass of literature particularly in aging

or degenerative populations, similar to the findings in this investigation Williams et al. (2017) showed that grey matter cortical thickness was decreased with physical activity as compared to older populations that showed an increase with physical activity.

Although the BMI between the highly active and low active groups was significantly different (p= 0.006, Cohen's *d* =1.86) this investigation found no significant relationships between BMI /gender and our regions of interest as was seen in a study by Taki et al. (2008), where the authors showed that BMI was related to greater regional volume in regions such as the frontal lobe and thalami, etc. whilst in other regions such as the postcentral gyrus inversely related to BMI in males but not in females. Obesity has been associated with atrophy of the brain, specifically grey matter regions such as the DLFPC, IFG, and left postcentral gyrus (Jagust et al., 2005; Jagust, 2007; Brooks et al., 2013). The maladaptive role of obesity on brain volume was shown to be greater with a combination of high waist-hip ratio and body mass index (BMI) rather than independently, and was associated with greater visceral fat (Hamer et al., 2018). However, when sedentary obese subjects were made to engage in physical activity it induced metabolic and neurotrophic structural changes that were associated with reduced levels of atrophy in the brain, in regions related to cognitive performance and not motor-related regions (Mueller et al., 2015).

The regional grey matter decreases may also be explained by the inflammation that is often present in HA ultra-endurance athletes (Zaryski and Smith, 2005). Inflammation can result from high volumes of training without adequate recovery (Ekdahl et al., 2003; Knechtle and Nikolaidis, 2018). In inflammatory states, even though BDNF increases were present, grey matter volumes were found to be reduced (Poletti et al., 2017). Alternatively, the reductions in the regional grey matter could be explained by the increased levels of cortisol found in HA individuals (Beat Knechtle and Nikolaidis, 2018). Increases in cortisol have been shown to reduce grey matter volumes (Castro-Fornieles et al., 2009).

Our population age (HA= 27.9 ± 26.64 years; LA= 28.33 ± 11.192 years) differed from the bulk of the research that focused on aging and populations affected by degenerative conditions (Batouli and Saba, 2017; Colcombe et al., 2006; Erickson et al., 2014; Kyun Jeon and Ho Ha, 2017). This could explain why

we found smaller regions of interest in the HA group. Regional grey matter atrophy is typically associated with degenerative conditions (Brooks et al., 2013; Rosano et al., 2008). In this investigation, it could be a result of neural pruning because of the younger healthy population (Williams et al., 2017). Although regional atrophy was present the global matter volumes were greater in the high active population which is in agreement with the literature. This is indicative that more research in young highly active populations is needed.

2.6 Conclusion

My thesis investigated the effect of high physical activity on total and regional brain volume. The main finding was that global brain matter volume and global white matter volumes were greater in the highly active group in comparison to the low active group. However, the regional grey matter volumes in three motor-related regions were found to be significantly smaller in the high active population. The regions that were found to be significantly smaller in the high active population were the right middle frontal gyrus (BA 9), the left sub-lobar thalamus and the right postcentral gyrus (BA 3). The results also indicate that total white matter had a direct relationship with MVPA scores indicating that greater volumes of physical activity impacted white matter structure positively.

2.7 Limitations

The study population size was primarily limited by the high cost of conducting MRI scans. The high cost of MRI's prevented us from testing participants over a period of time and at different points during their training status. We also did not have accelerometery data which along with multiple MRI scans could have provided more insight on how the brain's structure was directly impacted by physical activity levels. Although we measured resting heart rate, due to scanner interference with ECG recordings we were unable to obtain the heart rate variability data which has shown a strong association with cortisol (Pulopulos et al., 2018) and could have provided further insight into grey matter change. Other limitations included not having a moderate activity group that would have provided insight on optimal physical activity levels for brain structure. This was also related to the high cost of MRIS. We did

not control for sedentary time and did not have any recovery time data which is also a missing piece of the literature and could have provided further insight unfortunately it was part of a larger study and some of the data had already been collected. For this reason race, ethnicity, and socio-economic status data were also not collected. We could also not control whether participants adhered to study guidelines that fell out of the testing session's times we did however remind participants.

3 Cognitive Function and Physical Activity Levels

3.1 Introduction

Cognitive function plays an important role in everyday functioning and optimising lifestyle outcomes such as mental health, physical health, and quality of life (Diamond, 2013). Cognitive decline is an inevitable progression that is further accelerated with a lack of stimulus, aging, and degenerative diseases (Bherer et al., 2013). A key component of cognitive function is working memory which forms part of the executive functions (Diamond, 2013). Working memory is a short term store of information that is recalled shortly after storing. Its importance is not limited to daily function and it has been related to intelligence (Friedman et al., 2006) and reasoning skills (Baddeley, 1992) making it an aspect of executive function desirable to improve and maintain. The prefrontal cortex and parietal cortex are structures of the brain that were commonly found to be associated with working memory (Eriksson et al., 2015).

Physical activity is associated with a change in structure and function, executive function, in particular, has shown associated improvements (Ramnath et al., 2018). This may be attributed to changes in executive function-related brain structures such as the prefrontal cortex and parietal cortex which show increases in volume with the implementation of physical activity (Bugg and Head, 2011; Bugg et al., 2012; Ruscheweyh et al., 2009; Wood et al., 2016). Performance on a WM task like the N-back, showed improvements with physical activity (Li et al., 2014). It seems that moderate amounts of physical activity are enough to positively impact cognitive function and improve working memory (Kramer and Erickson, 2007; Verstynen et al., 2012).

The working memory adaptations resulting from physical activity have been explained by various mechanisms, one explanation is that changes in dopamine transmission are improved with physical activity and thus is better regulated with physical activity (Foley and Fleshner, 2008; Köhncke et al., 2018). Previous research focuses on the effects of physical activity on cognitive function comparing sedentary and obese populations to physically active populations (Mueller et al., 2015). Raichlen

and Alexander, 2017 likened physical activity to a form of modern-day foraging and found that with extended hours of PA this may provide a greater cognitive challenge and result in preferable adaptation such as larger grey matter volumes and stronger white matter tracts which are associated with improved working memory.

Unlike moderate volumes of physical activity, the relationship between high volume ultra-endurance training and cognitive function has not been adequately investigated. Rhodes et al., 2003 showed in a rodent model that with high levels of physical activity neurogenesis increased, however, this was without further learning enhancement indicating that a ceiling effect may be present. Physical activity improves dopamine balance and function in the basal ganglia, caudate, and hippocampus, suggesting that an association between fitness levels and volume might be apparent in this region (Eriksson et al., 2015; Köhncke et al., 2018). In a sample of 179 adults between 59 and 81 years of age without dementia (Ruscheweyh et al., 2009) showed that with an increase in the amount of physical activity, dopamine levels were increased linearly, whilst in sedentary individuals' dopamine transmission was impaired.

Another mechanism that affects cognitive function is BDNF. Physical activity-induced BDNF is associated with cognitive improvements and improved brain health (Kyun Jeon and Ho Ha, 2017). However, an over-expression of BDNF has been associated with working memory impairments (Papaleo et al., 2011). Physical activity-induced BDNF changes can continue to increase without an increase in function, as was found in high runner mice where a ceiling effect was present , beyond which cognitive function could not be further improved with physical activity (Rhodes et al., 2003). A similar phenomenon was seen in motor learning between endurance athletes and non-athletes (Seidel et al., 2017).

3.2 Aims and Hypotheses

3.2.1 Aims

The primary aim of this chapter was to examine working memory performance using an N-back task in individuals partaking in habitually high versus low levels of physical activity.

3.2.2 Hypotheses

The hypothesis in this investigation was that due to superior dopamine neurotransmission, the HA group would perform better on the N-back (Foley and Fleshner, 2008; Köhncke et al., 2018)
Although the high active group would perform better there would be a ceiling effect as was seen in animal studies (Rhodes et al., 2003).

3.3 Methods and Materials

3.3.1 Ethical Approval

Participants provided written informed consent before participation. This study was approved by the human research ethics committee of the University of Cape Town, South Africa (HREC 199/2017) and was carried out in accordance with the Declaration of Helsinki on the use of human participants in experiments.

This study was conducted in conjunction with another thesis examining the influence of methylphenidate on heart rate and brain connectivity. As a result, subjects had to meet other criteria, which are not relevant to this thesis.

3.3.2 Participants

We recruited 22 healthy participants without any history of cardiovascular, metabolic, neurological, and psychiatric diseases. Participants were recruited using posters, advertisements on social media, word of mouth, and at athletics clubs. Recruitment posters contained the purpose of the study, an outline of procedures, and necessary information to contact the experimenters (e.g. telephone number, email

address).

Researcher's contacted interested participants by telephone and reviewed the details of the study. Participants were asked exclusion/inclusion questions (see below) such as the presence of cardiovascular-related disease, claustrophobia, or if they were taking chronic medications. They were also asked about their average weekly physical activity habits. If participants met the minimum criteria for the sedentary or ultra-endurance population (see inclusion criteria (section 3.2.2.1)), a familiarisation session at the Sports Science Institute of South Africa was scheduled.

Inclusion and Exclusion Criteria

We recruited two different population groups for this study based on their physical activity habits. Group 1 (N=11) were healthy sedentary individuals. This group was defined as those who engaged in voluntary physical activity less than 2 times a week and performing less than 2 hours of total physical activity. Group 2 (N = 11) were healthy individuals who had high activity levels and engaged in voluntary PA for at least 9 hours a week. They had to be right-handed as the N-back keypad was right-handed. Participants in group 2 were included if they had been training for and participated in an ultra-endurance event (e.g. ironman) for the last year. However, participants meeting this criterion were excluded if their ironman event was within 1 month of the study.

All of our subjects had at least 1 year of post-secondary education but we did not control for ethnicity or socio-economic status.

3.3.3 Familiarization Session

Detailed information regarding the study risks/benefits was given to participants. They were then required to complete an informed consent questionnaire before any testing was conducted. All participants completed a neuropsychological evaluation performed by a trained psychologist using the Mini-International Neuropsychiatric Interview (Sheehan et al., 1998). From this interview, participants were excluded if they (i) had a history of drug or alcohol abuse or dependence, (ii) had past

psychiatric, neurological, cardiovascular or endocrinological disease, (iii) if they had taken psychoactive drugs (e.g. opiate analgesics, stimulants, or sedatives) or prescription medications in the last month. Further, we also performed a saliva test to test for the presence of psychotropic drugs such as cocaine, phencyclidine, benzodiazepines, cannabis, opiates, barbiturates, and inhalants (Detect a Drug Saliva Drug Test, Cape Town, South Africa). Finally, participants were excluded if they had any contraindications to MRI such as the presence of any pin, prosthesis, or any other magnetic materials inside/within their bodies that had been surgically implanted before 2008). The N-back protocol was explained to the participants. They were then instructed on how to perform the N-back. Our N-back task implemented 0-back and 1-back levels of difficulty. The N-back used a pseudo-random sequence of consonant letters. Each letter was presented for 500 ms, followed by a 2.5 s inter-stimulus interval during which a fixation crosshair was displayed. Subjects were asked to respond to target stimuli, consisting of a letter identical to the letter presented one or two, trials before, for the 0-back and 1-back respectively, the 1-back being the more difficult task. Each difficulty level block consisted of 50 trials (150 s total). Subjects were instructed to respond to target stimuli with their right hand using a button box. Of the 50 stimuli presented in each block, 33% were target stimuli. In both motor and cognitive tasks, (the N-back in this investigation lasted for 7 minutes. The blocks of each difficulty level were presented twice and block order was counterbalanced across subjects.

3.3.4 Experimental Session

During the experimental session, participants completed the 0-back and 1-back of the N-back task. Participants were asked to not consume alcohol and exercise 24 hours before the session and also requested not to consume caffeine or food three hours prior to the sessions. They were reminded via email to adhere to the study guidelines; the study was limited in that we had to trust the participants to adhere to this.

3.3.5 Data Analysis

During the task, the number of targets, correct responses, omission errors, and commissions

errors were recorded by the custom N-back software (Kaufman, 2005). Omissions indicate failing to respond to the stimuli and commissions were incorrect responses. A percentage score was calculated using the following equation.

$$\text{N-Back score} = \frac{\text{the number of responses-(omissions+ commissions)}}{\text{No. of Targets}} \times 100\%$$

The normality of data distribution and homogeneity was checked using SPSS using Kolmogorov-Smirnoff and Levene's test. A simple t-test was used to determine the differences in hits, omission errors, commission errors, and N-back percentage score.

Statistical analysis for demographic data was conducted using SPSS version 25 (IBM Corporation, NY, USA). Simple T-tests were used to determine the differences between the HA and LA groups on age, BMI, and MVPA. The incidence of gender for each group was determined using a Fisher's exact test.

Due to a technical setback, the data for 10 participants was corrupted. As a result, the dataset we analysed included 12 participants (Table 3.1); the ultra-endurance group were highly active participants (N = 6, 1351.33±400.56 MVPA in minutes) while the sedentary group were highly inactive (N = 6, 72.83±85.02 MVPA in minutes).

3.4 Results

Table 3.1 Participant characteristics

Variables	High Activity Group (N=6)	Low Activity Group (N=6)	(P-Value)
Age (Years)	24.86 ±4.67	30.14 ±15.31	0.411
BMI (kg/m²)	23.41 ±3.31	34.02 ±9.82	0.019**
MVPA (minutes)	1351.33 ±400.56	72.83 ±85.02	< 0.001**

Table 3.1: Demographic information (mean and SD) of high activity and low activity populations. BMI=Body Mass Index; MVPA=Moderate-vigorous Physical activity and age. **indicates significances

Table 3.2 N-back Score Parameters for High Activity and Low Activity Groups

Variables	High Activity Group (N=6)	Low Activity Group (N=6)	(P-Value)
N-back (%)	85.33 % ±17.12	86.17 % ±17.52	0.935
Commission errors	1.17 ±2.04	1.00 ±0.89	0.858
Omission errors	4.83 ±5.38	4.00 ±4.34	0.774

Table 3.2 N-back scores: N-back percentage indicates total N-back score. Commission errors are false responses. Omissions errors indicate no response. **indicate significances

3.4.1 Demographics

The two groups were not significantly different in age (p=0.411) but were different in BMI (p=0.19) and MVPA ($p < 0.001$) (Table 3.1).

3.4.2 N-back Performance

This investigation found no significant differences in N-back performance between the groups: N-back correct responses ($p = 0.935$), commission errors ($p = 0.858$), and omission errors ($p = 0.774$) were not significantly different between the two groups (Table 3.2).

3.5 Discussion

The purpose of this investigation was to determine the relationship between physical activity, working memory, and N-back scores between participants of varying activity levels. We found no significant differences present in the N-back score, omission errors, and commission errors.

The BMI between our groups was significantly different, however, no differences were found in the N-back task, which may have been due to a limited sample size. Previous research has shown BMI to be inversely related to working memory (Alarcón et al., 2016). This could be related to the impaired dopamine transmission found in overweight individuals (Wang et al., 2001).

Although we hypothesised that the HA group would perform better with the working memory task, we did not find any significant differences in total N-back score, omission error, and commission errors. Previous studies that examined the N-back task and showed significances between groups also had limited sample sizes (Redick and Lindsey, 2013). However, a major limitation in this investigation was that data for 10 subjects was unusable due to data collection errors. As a result, our study was underpowered (HA group N=6, LA group N=6) and unable detect changes between the groups. We did not account for external factors that were not controlled for and may have influenced scores e.g. education or prior exposure to the N-back. Although the sample size was very limited looking at previous research we may attribute the indistinguishability in the N-back scores due to high levels of

exercise being associated with increases in cortisol that could result in decreased BDNF and negatively affect cognitive function (Kyun Jeon and Ho Ha, 2017). Alternatively, the high levels of exercise which can result in a concurrent presence of hypercortisolism and the overexpression of BDNF may have reduced the positive effects of physical activity on working memory and subsequently resulted in cognitive impairment (Papaleo et al., 2011).

With caution, the findings of this investigation, could suggest that exercise be performed in moderation, and may be similar to the findings of (Rhodes et al., 2003; Freund et al., 2014). Although exercise in moderation improved cognition in high runner mice, the ability of the hippocampus of highly active populations to respond to learning may be impaired or overwhelmed by the excessive exercise-overactive hippocampus.

3.6 Conclusions

Our investigation found no differences in working memory on the N-back task between the high-activity and low-activity groups. Further investigation is needed to assess how high-volumes of physical activity and low volumes of physical activity may impact working memory.

3.7 Limitations

A major limitation was the small sample size due to the technical challenges we experienced with the software. Given the time constraints that came with being part of a larger study, we were unable to administer the long 2-back and 3-back which provide an increasing challenge to working memory and may have been able to find a difference between the HA and LA groups. This study also lacked a moderate physical activity level group which could compare three different activity levels on the N-back and could have provided more insight on a ceiling effect in working memory.

Concluding Remarks

The purpose of the two components of this study was to investigate the relationship between physical activity, brain structure, and cognitive function. The results of this investigation differed with the findings of Ruscheweyh et al. (2009), that summarised the relationship between brain structures, cognitive function, and physical activity, in which the authors found that in the presence of increasing physical activity there were positive associations between regional grey matter structure, dopamine transmission, and cognitive ability. The global matter volume findings agreed with Batouli and Saba, (2017) that found physical activity to be associated with increased total intracranial volume, grey matter and white matter volumes. We were unable to detect differences in working memory between the two groups. the increased global white matter volume is associated with improved working memory performance (Alarcón et al., 2016; Demirakca et al., 2014), as well as the trend to greater grey matter volume, is also associated with better cognition (Voelcker-Rehage and Niemann, 2013). The regional structural differences we found were in contrast to previous research that found larger regions of interest with MVPA (Demirakca et al., 2014; Erickson et al., 2011) and that the higher than normal levels of physical activity may be a contributing factor as was found in (Freund et al.,2012; Freund et al., 2014). This may be caused by neural pruning (Williams et al., 2017). Physical activity enhances neuroplasticity and cognition which is linked to improvements in executive function and working memory (Voelcker-Rehage and Niemann, 2013). Further research is needed to better understand the complex relationship between brain volume, MVPA, and cognition in individuals that have habitually high physical activity levels.

Summary and Future Directions

This thesis advances the current understanding of MVPA levels, brain volumes, and cognition by demonstrating that with greater than normal MVPA levels, global matter volumes were greater in healthy active populations than healthy less active populations, particularly in white matter and total intracranial volume. A relatively novel finding was that although global matter volumes were greater with a higher MVPA, decreases in regional grey matter in the right middle frontal gyrus (BA 9), the left Thalamus, and right post-central gyrus can occur (BA 3). This phenomenon may not be limited to the regions we found and requires further investigation on the role of neural pruning. This thesis did not link greater than normal MVPA levels to improved working memory performance on an N-back task. This requires furthers investigation on the ceiling effect that can be present in physical activity-related improvements in cognition.

Recommendations for future work should include a large-scale structural brain imaging study comparing three activity groups' low, moderate, and high MVPA levels in healthy younger populations. Future research should aim to scan an individual's brain multiple times over a period to account for different training time points and training status. This should be linked to accelerometery data to track MVPA and its relation to brain volumes changes over periods. Repeated structural scans and working memory tasks to better directly understand brain volume and cognition. Further, relating this back to MVPA using accelerometery.

References

Alarcón, G., Ray, S., & Nagel, B. J. (2016). Lower Working Memory Performance in Overweight and Obese Adolescents Is Mediated by White Matter Microstructure. *Journal of the International Neuropsychological Society : JINS*, *22*(3), 281–292. https://doi.org/10.1017/S1355617715001265

Amunts, K., & Zilles, K. (2001). Advances in cytoarchitectonic mapping of the human cerebral cortex. *Neuroimaging Clinics of North America*.

Aribisala, B. S., Gow, A. J., Bastin, M. E., del Carmen Valdés Hernández, M., Murray, C., Royle, N. A., Wardlaw, J. M. (2013). Associations between level and change in physical function and brain volumes. *PloS One*, *8*(11), e80386. https://doi.org/10.1371/journal.pone.0080386

Armstrong, T., & Bull, F. (2006). Development of the World Health Organization Global Physical Activity Questionnaire (GPAQ). *Journal of Public Health*, *14*(2), 66–70. https://doi.org/10.1007/s10389-006-0024-x

Ashburner, J., & Friston, K. J. (2000). Voxel-Based Morphometry—The Methods. *NeuroImage*, *11*(6), 805–821. https://doi.org/10.1006/nimg.2000.0582

Baddeley, A. (1992). Working memory. *Science*, *255*(5044), 556–559. https://doi.org/10.1126/science.1736359

Baddeley, Alan, & Baddeley, A. D. (2018). The concept of working memory. In *Exploring Working Memory* (pp. 99–106). Routledge. https://doi.org/10.4324/9781315111261-9

Batouli, S. A. H., & Saba, V. (2017). At least eighty percent of brain grey matter is modifiable by physical activity: A review study. *Behavioural Brain Research*, *332*, 204–217. https://doi.org/10.1016/J.BBR.2017.06.002

Benedict, C., Brooks, S. J., Kullberg, J., Nordenskjöld, R., Burgos, J., Le Grevès, M., … Schiöth, H. B. (2013). Association between physical activity and brain health in older adults. *Neurobiology*

of Aging, *34*(1), 83–90. https://doi.org/10.1016/J.NEUROBIOLAGING.2012.04.013

Bherer, L., Erickson, K. I., & Liu-Ambrose, T. (2013). A review of the effects of physical activity and exercise on cognitive and brain functions in older adults. *Journal of Aging Research*, *2013*, 657508. https://doi.org/10.1155/2013/657508

Biswas, A., Oh, P. I., Faulkner, G. E., Bajaj, R. R., Silver, M. A., Mitchell, M. S., & Alter, D. A. (2015). Sedentary time and its association with risk for disease incidence, mortality, and hospitalization in adults a systematic review and meta-analysis. *Annals of Internal Medicine*, *162*(2), 123–132. https://doi.org/10.7326/M14-1651

Booth, F. W., Roberts, C. K., & Laye, M. J. (2012). Lack of exercise is a major cause of chronic diseases. In *Comprehensive Physiology* (Vol. 2, pp. 1143–1211). https://doi.org/10.1002/cphy.c110025

Bourdeau, I., Bard, C., Noël, B., Leclerc, I., Cordeau, M.-P., Bélair, M., … Lacroix, A. (2002). Loss of Brain Volume in Endogenous Cushing's Syndrome and Its Reversibility after Correction of Hypercortisolism. *The Journal of Clinical Endocrinology & Metabolism*, *87*(5), 1949–1954. https://doi.org/10.1210/jcem.87.5.8493

Brooks, S. J., Benedict, C., Burgos, J., Kempton, M. J., Kullberg, J., Nordenskjöld, R., … Schiöth, H. B. (2013). Late-life obesity is associated with smaller global and regional gray matter volumes: a voxel-based morphometric study. *International Journal of Obesity*, *37*(2), 230–236. https://doi.org/10.1038/ijo.2012.13

Bugg, J. M., & Head, D. (2011). Exercise moderates age-related atrophy of the Bugg, J.M., Head, D., 2011. Exercise moderates age-related atrophy of the medial temporal lobe. Neurobiol. Aging 32, 506–14.medial temporal lobe. *Neurobiology of Aging*, *32*(3), 506–514. https://doi.org/10.1016/j.neurobiolaging.2009.03.008

Bugg, J. M., Shah, K., Villareal, D. T., & Head, D. (2012). Cognitive and neural correlates of aerobic

fitness in obese older adults. *Experimental Aging Research*, *38*(2), 131–145. https://doi.org/10.1080/0361073X.2012.659995

Camandola, S., & Mattson, M. P. (2017). Brain metabolism in health, aging, and neurodegeneration. *The EMBO Journal*, *36*(11), 1474–1492. https://doi.org/10.15252/embj.201695810

Carnevali, L., Koenig, J., Sgoifo, A., & Ottaviani, C. (2018, April 6). Autonomic and brain morphological predictors of stress resilience. *Frontiers in Neuroscience*. https://doi.org/10.3389/fnins.2018.00228

Caspersen, C. J., Powell, K. E., & Christenson, G. M. (1985). Physical activity, exercise, and physical fitness: definitions and distinctions for health-related research. *Public Health Reports (Washington, D.C. : 1974)*, *100*(2), 126–131. Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/3920711

Castro-Fornieles, J., Bargalló, N., Lázaro, L., Andrés, S., Falcon, C., Plana, M. T., & Junqué, C. (2009). A cross-sectional and follow-up voxel-based morphometric MRI study in adolescent anorexia nervosa. *Journal of Psychiatric Research*, *43*(3), 331–340. https://doi.org/10.1016/J.JPSYCHIRES.2008.03.013

Colcombe, S. J., Erickson, K. I., Scalf, P. E., Kim, J. S., Prakash, R., McAuley, E., … Kramer, A. F. (2006). Aerobic exercise training increases brain volume in aging humans. *Journals of Gerontology*, *61*(11), 1166–1170. https://doi.org/10.1093/gerona/61.11.1166

Colcombe, S. J., Kramer, A. F., Erickson, K. I., Scalf, P., McAuley, E., Cohen, N. J., … Elavsky, S. (2004). Cardiovascular fitness, cortical plasticity, and aging. *Proceedings of the National Academy of Sciences of the United States of America*, *101*(9), 3316–3321. https://doi.org/10.1073/pnas.0400266101

Colcombe, S., & Kramer, A. F. (2003). Fitness effects on the cognitive function of older adults: A meta-analytic study. *Psychological Science*, *14*(2), 125–130. https://doi.org/10.1111/1467-9280.t01-1-

01430

Cotman, C. W., & Berchtold, N. C. (2002). Exercise: a behavioral intervention to enhance brain health and plasticity. *Trends in Neurosciences*, *25*(6), 295–301. https://doi.org/10.1016/S0166-2236(02)02143-4

Cowan, N. (2014). Working Memory Underpins Cognitive Development, Learning, and Education. *Educational Psychology Review*, *26*(2), 197–223. https://doi.org/10.1007/s10648-013-9246-y

Crowley, S., Huang, H., Tanner, J., Zhao, Q., Schwab, N. A., Hizel, L., … Price, C. C. (2018). Considering total intracranial volume and other nuisance variables in brain voxel based morphometry in idiopathic PD. *Brain Imaging and Behavior*, *12*(1). https://doi.org/10.1007/s11682-016-9656-9

Demirakca, T., Brusniak, W., Tunc-Skarka, N., Wolf, I., Meier, S., Matthäus, F., … Diener, C. (2014). Does body shaping influence brain shape? Habitual physical activity is linked to brain morphology independent of age. *World Journal of Biological Psychiatry*, *15*(5), 387–396. https://doi.org/10.3109/15622975.2013.803600

Department of Health, G. C. and I. S. (2000). *Guidelines for the promotion of active ageing in older aduluts ar primary level*.

Diamond, A. (2013). Executive functions. *Annual Review of Psychology*, *64*, 135–168. https://doi.org/10.1146/annurev-psych-113011-143750

Dietrich, A., & Sparling, P. B. (2004). Endurance exercise selectively impairs prefrontal-dependent cognition. *Brain and Cognition*, *55*(3), 516–524. https://doi.org/10.1016/j.bandc.2004.03.002

Draganski, B., Gaser, C., Busch, V., Schuierer, G., Bogdahn, U., & May, A. (2004). Changes in grey matter induced by training. *Nature*, *427*(6972), 311–312. https://doi.org/10.1038/427311a

Dumurgier, J., Crivello, F., Mazoyer, B., Ahmed, I., Tavernier, B., Grabli, D., … Elbaz, A.

(2012). MRI atrophy of the caudate nucleus and slower walking speed in the elderly. *NeuroImage*, *60*(2), 871–878. https://doi.org/10.1016/J.NEUROIMAGE.2012.01.102

Dunst, B., Benedek, M., Jauk, E., Bergner, S., Koschutnig, K., Sommer, M., … Neubauer, A. C. (2014). Neural efficiency as a function of task demands. *Intelligence*, *42*(1), 22–30. https://doi.org/10.1016/j.intell.2013.09.005

Eickhoff, S. B., Stephan, K. E., Mohlberg, H., Grefkes, C., Fink, G. R., Amunts, K., & Zilles, K. (2005). A new SPM toolbox for combining probabilistic cytoarchitectonic maps and functional imaging data. *NeuroImage*, *25*(4), 1325–1335. https://doi.org/10.1016/j.neuroimage.2004.12.034

Ekdahl, C. T., Claasen, J.-H., Bonde, S., Kokaia, Z., & Lindvall, O. (2003). Inflammation is detrimental for neurogenesis in adult brain. *Proceedings of the National Academy of Sciences*, *100*(23), 13632–13637. https://doi.org/10.1073/pnas.2234031100

Erickson, K I, Raji, C. A., Lopez, O. L., Becker, J. T., Rosano, C., Newman, M. A. B., … Kuller, B. L. H. (2010). *Physical activity predicts gray matter volume in late adulthood The Cardiovascular Health Study Supplemental data at www.neurology.org*. Retrieved from www.neurology.org

Erickson, Kirk I, Voss, M. W., Prakash, R. S., Basak, C., Szabo, A., Chaddock, L., … Kramer, A. F. (2011). Exercise training increases the size of hippocampus and improves memory. *Proceedings of the National Academy of Sciences of the United States of America*, *108*(7), 3017–3022. https://doi.org/10.1073/pnas.1015950108

Erickson, Kirk I, Weinstein, A. M., & Lopez, O. L. (2012). Physical Activity, Brain Plasticity, and Alzheimer's Disease. https://doi.org/10.1016/j.arcmed.2012.09.008

Eriksson, J., Vogel, E. K., Lansner, A., Bergström, F., & Nyberg, L. (2015). Neurocognitive Architecture of Working Memory. *Neuron*, *88*(1), 33–46. https://doi.org/10.1016/j.neuron.2015.09.020

Feldman, D. E. (2009). Synaptic Mechanisms for Plasticity in Neocortex. *Annual Review of*

Neuroscience, *32*(1), 33–55. https://doi.org/10.1146/annurev.neuro.051508.135516

Flöel, A., Ruscheweyh, R., Krüger, K., Willemer, C., Winter, B., Völker, K., … Knecht, S. (2010). Physical activity and memory functions: Are neurotrophins and cerebral gray matter volume the missing link? *NeuroImage*, *49*(3), 2756–2763. https://doi.org/10.1016/J.NEUROIMAGE.2009.10.043

Foley, T. E., & Fleshner, M. (2008). Neuroplasticity of dopamine circuits after exercise: Implications for central fatigue. *NeuroMolecular Medicine*, *10*(2), 67–80. https://doi.org/10.1007/s12017-008-8032-3

Freund, W., Faust, S., Birklein, F., Gaser, C., Wunderlich, A. P., Müller, M., … Schütz, U. H. (2012). Substantial and reversible brain gray matter reduction but no acute brain lesions in ultramarathon runners: experience from the TransEurope-FootRace Project. *BMC Medicine*, *10*, 170. https://doi.org/10.1186/1741-7015-10-170

Freund, W., Faust, S., Gaser, C., Grön, G., Birklein, F., Wunderlich, A. P., … Schütz, U. H. (2014). Regionally accentuated reversible brain grey matter reduction in ultra marathon runners detected by voxel-based morphometry. *BMC Sports Science, Medicine & Rehabilitation*, *6*(1), 4. https://doi.org/10.1186/2052-1847-6-4

Fry, R. W., Morton, A. R., & Keast, D. (1991). Overtraining in Athletes: An Update. *Sports Medicine*. https://doi.org/10.2165/00007256-199112010-00004

Fuchs, E., & Flügge, G. (2014). Adult Neuroplasticity: More Than 40 Years of Research. *Neural Plasticity*, *2014*. https://doi.org/10.1155/2014/541870

Geurts, J. J., & Barkhof, F. (2008). Grey matter pathology in multiple sclerosis. *The Lancet Neurology*, *7*(9), 841–851. https://doi.org/10.1016/S1474-4422(08)70191-1

González, K., Fuentes, J., & Márquez, J. L. (2017, May 1). Physical inactivity, sedentary behavior and chronic diseases. *Korean Journal of Family Medicine*. Korean Journal of Family Medicine.

https://doi.org/10.4082/kjfm.2017.38.3.111

Good, C. D., Johnsrude, I. S., Ashburner, J., Henson, R. N. A. A., Friston, K. J., Frackowiak, R. S. J. J., … Karl J. Friston, * and Richard S. J. Frackowiak*. (2001). A Voxel-Based Morphometric Study of Ageing in 465 Normal Adult Human Brains. *NeuroImage*, *14*, 21–36. Retrieved from https://ac.els-cdn.com/S1053811901907864/1-s2.0-S1053811901907864-main.pdf?_tid=aa04ea43-f49f-4700-8bbb-be4b9d00a2ee&acdnat=1545304009_08adafbfaf5b57e763865c2b5858ecc3

Good, C. D., Johnsrude, I. S., Ashburner, J., Henson, R. N. A., Friston, K. J., & Frackowiak, R. S. J. (2001). A Voxel-Based Morphometric Study of Ageing in 465 Normal Adult Human Brains. *NeuroImage*, *14*(1), 21–36. https://doi.org/10.1006/NIMG.2001.0786

Gow, A. J., Bastin, M. E., Muñoz Maniega, S., Valdés Hernández, M. C., Morris, Z., Murray, C., … Wardlaw, J. M. (2012). Neuroprotective lifestyles and the aging brain: activity, atrophy, and white matter integrity. *Neurology*, *79*(17). https://doi.org/10.1212/WNL.0b013e3182703fd2

Halson, S. L., & Jeukendrup, A. E. (2004). Does overtraining exist? An analysis of overreaching and overtraining research. *Sports Medicine*. https://doi.org/10.2165/00007256-200434140-00003

Hansen, T. I., Brezova, V., Eikenes, L., Håberg, A., & Vangberg, X. T. R. (2015). How does the accuracy of intracranial volume measurements affect normalized brain volumes? sample size estimates based on 966 subjects from the HUNT MRI cohort. *American Journal of Neuroradiology*, *36*(8), 1450–1456. https://doi.org/10.3174/ajnr.A4299

Haskell, W. L., Lee, I.-M., Pate, R. R., Powell, K. E., Blair, S. N., Franklin, B. A., & Bauman, (2007). Physical Activity and Public Health: Updated Recommendation for Adults From the American College of Sports Medicine and the American Heart Association. *Circulation*, *116*(9), 1081–1093. https://doi.org/10.1161/CIRCULATION.107.185649

Hillman, C. H., Erickson, K. I., & Kramer, A. F. (2008). Be smart, exercise your heart: exercise effects

on brain and cognition. *Nature Reviews Neuroscience*, *9*(1), 58. https://doi.org/10.1038/nrn2298

Honey, C. J., Thivierge, J. P., & Sporns, O. (2010). Can structure predict function in the human brain? *NeuroImage*, *52*(3), 766–776. https://doi.org/10.1016/j.neuroimage.2010.01.071

Huttenlocher, P. R. (1990). Morphometric study of human cerebral cortex development. *Neuropsychologia*, *28*(6), 517–527. https://doi.org/10.1016/0028-3932(90)90031-i

Hynynen, E., Uusitalo, A., Konttinen, N., & Rusko, H. (2006). Heart rate variability during night sleep after awakening in overtrained athletes. *Med. Sci. Sports Exerc*, (8), 313–317. https://doi.org/10.1249/01.mss.0000184631.27641.b5

Jacini, W. F. S., Cannonieri, G. C., Fernandes, P. T., Bonilha, L., Cendes, F., & Li, L. M. (2009). Can exercise shape your brain? Cortical differences associated with judo practice. *Journal of Science and Medicine in Sport*, *12*(6), 688–690. https://doi.org/10.1016/j.jsams.2008.11.004

James, W., Burkhardt, F., Bowers, F., & Skrupskelis, I. (1890). *The principles of psychology*. Retrieved from https://content.taylorfrancis.com/books/download?dac=C2017-0-60287-0&isbn=9781351350938&format=googlePreviewPdf

Jensen, M. T., Suadicani, P., Hein, H. O., & Gyntelberg, F. (2013). Elevated resting heart rate, physical fitness and all-cause mortality: A 16-year follow-up in the Copenhagen Male Study. *Heart*, *99*(12), 882–887. https://doi.org/10.1136/heartjnl-2012-303375

Jetté, M., Sidney, K., & Blümchen, G. (1990). Metabolic equivalents (METS) in exercise testing, exercise prescription, and evaluation of functional capacity. *Clinical Cardiology*, *13*(8), 555–565. https://doi.org/10.1002/clc.4960130809

Jurca, R., Jackson, A. S., LaMonte, M. J., Morrow, J. R., Blair, S. N., Wareham, N. J., … Laukkanen, R. (2005). Assessing cardiorespiratory fitness without performing exercise testing. *American Journal of Preventive Medicine*, *29*(3), 185–193. https://doi.org/10.1016/j.amepre.2005.06.004

Kang, S. J., Ha, G. C., & Ko, K. J. (2017). Association between resting heart rate, metabolic syndrome and cardiorespiratory fitness in Korean male adults. *Journal of Exercise Science and Fitness*, *15*(1), 27–31. https://doi.org/10.1016/j.jesf.2017.06.001

Kaufman, G. (2005). Picture N-Back: Archives of Neurobehavioral Experiments and Stimuli. Retrieved from https://www.neurobs.com/ex_files/expt_view?id=148&tree_item_url=nback.exp&item_id=nback.exp

Kemp, A. H., & Quintana, D. S. (2013). The relationship between mental and physical health: Insights from the study of heart rate variability. *International Journal of Psychophysiology*, *89*(3), 288–296. https://doi.org/10.1016/j.ijpsycho.2013.06.018

KI, E., Leckie, R., & AM, W. (2014). Physical activity, fitness, and gray matter volume. *Neurobiol Aging*, *35*(2), 20–28. https://doi.org/10.1039/c7dt03354k

Kilgour, A. H. M., Todd, O. M., & Starr, J. M. (2014). A systematic review of the evidence that brain structure is related to muscle structure and their relationship to brain and muscle function in humans over the lifecourse. *BMC Geriatrics*, *14*, 85. https://doi.org/10.1186/1471-2318-14-85

Killgore, W. D. S., Olson, E. A., & Weber, M. (2013). Physical exercise habits correlate with gray matter volume of the hippocampus in healthy adult humans. *Scientific Reports*, *3*(1), 3457. https://doi.org/10.1038/srep03457

Király, A., Szabó, N., Tóth, E., Csete, G., Faragó, P., Kocsis, K., … Kincses, Z. T. (2016). Male brain ages faster: the age and gender dependence of subcortical volumes. *Brain Imaging and Behavior*, *10*(3), 901–910. https://doi.org/10.1007/s11682-015-9468-3

Knechtle, B., Knechtle, P., & Lepers, R. (2011). Participation and performance trends in ultra-triathlons from 1985 to 2009. *Scandinavian Journal of Medicine & Science in Sports*, *21*(6), e82–e90.

https://doi.org/10.1111/j.1600-0838.2010.01160.x

Knechtle, Beat, & Nikolaidis, P. T. (2018). Physiology and Pathophysiology in Ultra-Marathon Running. *Frontiers in Physiology*, *9*, 634. https://doi.org/10.3389/fphys.2018.00634

Köhncke, Y., Papenberg, G., Jonasson, L., Karalija, N., Wåhlin, A., Salami, A., … Lövdén, M. (2018). Self-rated intensity of habitual physical activities is positively associated with dopamine D2/3 receptor availability and cognition. *NeuroImage*, *181*, 605–616. https://doi.org/10.1016/j.neuroimage.2018.07.036

Kolbe-Alexander, T. L., Nossel, C., Conradie, J., Noach, A., Adonis, L., Patel, D., … Lambert, E. V. (2010). Meeting Physical Activity Guidelines Is Associated With Reduced Healthcare Expenditure In South African Employees. *Medicine & Science in Sports & Exercise*, *42*, 39. https://doi.org/10.1249/01.mss.0000384862.62594.fd

Kramer, A. F., & Erickson, K. I. (2007). Capitalizing on cortical plasticity: influence of physical activity on cognition and brain function. *TRENDS in Cognitive Sciences*. https://doi.org/10.1016/j.tics.2007.06.009

Kyun Jeon, Y., & Ho Ha, C. (2017). The effect of exercise intensity on brain derived neurotrophic factor and memory in adolescents. *Environmental Health and Preventive Medicine (2017)*. https://doi.org/10.1186/s12199-017-0643-6

Lefèbvre, P. J., Kesaniemi, Y. A., Danforth, E., Jensen, M. D., Kopelman, P. G., Lefebvre, P., & Reeder, B. A. (2001). Dose-response issues concerning physical activity and health: an evidence-based symposium. *Medicine & Science in Sports & Exercise*. Retrieved from https://www.researchgate.net/publication/11912476

Li, L., Men, W.-W., Chang, Y.-K., Fan, M.-X., Ji, L., & Wei, G.-X. (2014). Acute aerobic exercise increases cortical activity during working memory: a functional MRI study in female college

students. *PloS One*, *9*(6), e99222. https://doi.org/10.1371/journal.pone.0099222

Lo, J. C., Loh, K. K., Zheng, H., Sim, S. K. Y., & Chee, M. W. L. (2014). Sleep Duration and Age-Related Changes in Brain Structure and Cognitive Performance. *Sleep*, *37*(7), 821–821. https://doi.org/10.5665/sleep.3832

Lowe, C., Staines, W. R., & Hall, P. A. (2016). Effects of Moderate Exercise on Cortical Resilience: A Transcranial Magnetic Stimulation Study Targeting the Dorsolateral Prefrontal Cortex Examination of mechanisms underlying the effect of single session exercise on cortical plasticity View project Neur. https://doi.org/10.1097/PSY.0000000000000361

Makovac, E., Meeten, F., Watson, D. R., Garfinkel, S. N., Critchley, H. D., & Ottaviani, C. (2015). Neurostructural abnormalities associated with axes of emotion dysregulation in generalized anxiety. https://doi.org/10.1016/j.nicl.2015.11.022

Mayosi, B. M., Flisher, A. J., Lalloo, U. G., Sitas, F., Tollman, S. M., & Bradshaw, D. (2009). The burden of non-communicable diseases in South Africa. *The Lancet*. https://doi.org/10.1016/S0140-6736(09)61087-4

McEwen, B. S. (2002). Sex, stress and the hippocampus: allostasis, allostatic load and the aging process. *Neurobiology of Aging*, *23*(5), 921–939. https://doi.org/10.1016/S0197-4580(02)00027-1

McEwen, B. S. (2007). Physiology and Neurobiology of Stress and Adaptation: Central Role of the Brain BRUCE. *Phyisological Review*, (87), 873–904. https://doi.org/10.1152/physrev.00041.2006.

Medicine, A. C. of S. (2013). *ACSM's guidelines for exercise testing and prescription*. Retrieved from https://books.google.com/books?hl=en&lr=&id=hhosAwAAQBAJ&oi=fnd&pg=PP1&dq=acsm+guidelines+for+exercise+prescription&ots=ljHb3C0TVw&sig=DL3T0o4ibthlJJI4jatvCxKVYFo

Meule, A. (2017, March 7). Reporting and interpreting working memory performance in n-back tasks. *Frontiers in Psychology*. Frontiers Research Foundation.

https://doi.org/10.3389/fpsyg.2017.00352

Mueller, K., Möller, H. E., Horstmann, A., Busse, F., Lepsien, J., Blüher, M., … Pleger, B. (2015). Physical exercise in overweight to obese individuals induces metabolic- and neurotrophic-related structural brain plasticity. *Frontiers in Human Neuroscience*, *9*(July), 1–14. https://doi.org/10.3389/fnhum.2015.00372

Mumu, S. J., Ali, L., Barnett, A., & Merom, D. (2017). Validity of the global physical activity questionnaire (GPAQ) in Bangladesh. *BMC Public Health*, *17*(1). https://doi.org/10.1186/s12889-017-4666-0

Nagai, N., Hamada, T., Kimura, T., Moritani, T., Nagai, N., Hamada, T., … Moritani, T. (2004). Moderate physical exercise increases cardiac autonomic nervous system activity in children with low heart rate variability. *Childs Nerv Syst*, *20*, 209–214. https://doi.org/10.1007/s00381-004-0915-5

NC Andreasen. (1988). Brain imaging: applications in psychiatry. *Science*, *239*(4846), 1381–1388. https://doi.org/10.1126/science.3279509

Niemann, C., Godde, B., & Voelcker-Rehage, C. (2014). Not only cardiovascular, but also coordinative exercise increases hippocampal volume in older adults. *Frontiers in Aging Neuroscience*, *6*. https://doi.org/10.3389/fnagi.2014.00170

Noudoost, B., & Moore, T. (2011). The role of neuromodulators in selective attention. *Trends in Cognitive Sciences*, *15*(12), 585–591. https://doi.org/10.1016/J.TICS.2011.10.006

Oberauer, K., Süß, H. M., Schulze, R., Wilhelm, O., & Wittmann, W. W. (2000). Working memory capacity - Facets of a cognitive ability construct. *Personality and Individual Differences*, *29*(6), 1017–1045. https://doi.org/10.1016/S0191-8869(99)00251-2

Papaleo, F., Silverman, J. L., Aney, J., Tian, Q., Barkan, C. L., Chadman, K. K., & Crawley, J. N. (2011). Working memory deficits, increased anxiety-like traits, and seizure susceptibility in BDNF

overexpressing mice. *Learning & Memory (Cold Spring Harbor, N.Y.)*, *18*(8), 534–544. https://doi.org/10.1101/lm.2213711

Pascual-Leone, A., Amedi, A., Fregni, F., & Merabet, L. B. (2005). the Plastic Human Brain Cortex. *Annual Review of Neuroscience*, *28*(1), 377–401. https://doi.org/10.1146/annurev.neuro.27.070203.144216

Pedersen, B. K., & Saltin, B. (2015). Exercise as medicine - evidence for prescribing exercise as therapy in 26 different chronic diseases. *Scandinavian Journal of Medicine & Science in Sports*, *25*, 1–72. https://doi.org/10.1111/sms.12581

Petersen, A. M. W., & Pedersen, B. K. (2005). The anti-inflammatory effect of exercise. *Journal of Applied Physiology*, *98*(4), 1154–1162. https://doi.org/10.1152/japplphysiol.00164.2004

Poletti, S., Aggio, V., Hoogenboezem, T. A., Ambrée, O., de Wit, H., Wijkhuijs, A. J. M., … Benedetti, F. (2017). Brain-derived Neurotrophic Factor (BDNF) and gray matter volume in bipolar disorder. *European Psychiatry*, *40*, 33–37. https://doi.org/10.1016/j.eurpsy.2016.06.008

Powell, K. E., Paluch, A. E., & Blair, S. N. (2011). Physical Activity for Health: What Kind? How Much? How Intense? On Top of What? *Annual Review of Public Health*, *32*(1), 349–365. https://doi.org/10.1146/annurev-publhealth-031210-101151

Qing, Z., & Gong, G. (2016). Size matters to function: Brain volume correlates with intrinsic brain activity across healthy individuals. *NeuroImage*, *139*, 271–278. https://doi.org/10.1016/j.neuroimage.2016.06.046

Raichlen, D. A., Klimentidis, Y. C., Bharadwaj, P. K., & Alexander, G. E. (2019). Differential associations of engagement in physical activity and estimated cardiorespiratory fitness with brain volume in middle-aged to older adults. *Brain Imaging and Behavior*. https://doi.org/10.1007/s11682-019-00148-x

Raji, C. A., Merrill, D. A., Eyre, H., Mallam, S., Torosyan, N., Erickson, K. I., … Kuller, L. H. (2016). Longitudinal Relationships between Caloric Expenditure and Gray Matter in the Cardiovascular Health Study. *Journal of Alzheimer's Disease*, *52*(2), 719–729. https://doi.org/10.3233/JAD-160057

Ramnath, U., Rauch, L., Lambert, E. V., & Kolbe-Alexander, T. L. (2018). The relationship between functional status, physical fitness and cognitive performance in physically active older adults: A pilot study. *PLoS ONE*, *13*(4), 1–16. https://doi.org/10.1371/journal.pone.0194918

Raz, N., Lindenberger, U., Rodrigue, K. M., Kennedy, K. M., Head, D., Williamson, A., … Acker, J. D. (2005). Regional Brain Changes in Aging Healthy Adults: General Trends, Individual Differences and Modifiers. *Cerebral Cortex*, *15*(11), 1676–1689. https://doi.org/10.1093/cercor/bhi044

Redick, T. S., & Lindsey, D. R. B. (2013). Complex span and n-back measures of working memory: A meta-analysis. *Psychology and Aging*. https://doi.org/10.3758/s13423-013-0453-9

Rhodes, J. S., Gammie, S. C., & Garland, T. (2005). *Neurobiology of Mice Selected for High Voluntary Wheel-running Activity 1*.

Rhodes, J. S., Jeffrey, S., Girard, I., Mitchell, G. S., Garland, T., & Gage, F. H. (2003). Exercise increases Hippocampal neurogenesis to high levels but does not improve spatial learning in mice bred for increased voluntary wheel running. *Behavioral Neuroscience*. https://doi.org/10.1037/0735-7044.117.5.1006

Roberts, S., Awick, E., Fanning, J. T., Ehlers, D., Motl, R. W., & McAuley, E. (2017). Long-term maintenance of physical function in older adults following a DVD-Delivered Exercise Intervention. *Journal of Aging and Physical Activity*, *25*(1), 27–31. https://doi.org/10.1123/japa.2015-0284

Rosano, C., Aizenstein, H., Brach, J., Longenberger, A., Studenski, S., & Newman, A. B. (2008). Special article: gait measures indicate underlying focal gray matter atrophy in the brain of older adults. *The Journals of Gerontology. Series A, Biological Sciences and Medical Sciences*, *63*(12), 1380–1388.

https://doi.org/10.1093/gerona/63.12.1380

Ruscheweyh, R., Willemer, C., Krüger, K., Duning, T., Warnecke, T., Sommer, J., … Flöel, A. (2009). Physical activity and memory functions: An interventional study. *Neurobiology of Aging*, *32*(7), 1304–1319. https://doi.org/10.1016/j.neurobiolaging.2009.08.001

Sachdev, P. S., Wen, W., Christensen, H., & Jorm, A. F. (2005). White matter hyperintensities are related to physical disability and poor motor function. *Journal of Neurology, Neurosurgery, and Psychiatry*, *76*(3), 362–367. https://doi.org/10.1136/jnnp.2004.042945

Scholz, J., Klein, M. C., Behrens, T. E. J., & Johansen-Berg, H. (2009). Training induces changes in white matter architecture. *Nat Neurosci*, *12*(11), 1370–1371. https://doi.org/10.1038/nn.2412

Seidel, O., Carius, D., Kenville, R., & Ragert, P. (2017). Motor learning in a complex balance task and associated neuroplasticity: A comparison between endurance athletes and non-athletes. *Journal of Neurophysiology*, jn.00419.2017. https://doi.org/10.1152/jn.00419.2017

Sexton, C. E., Betts, J. F., Demnitz, N., Dawes, H., Ebmeier, K. P., & Johansen-Berg, H. (2016). A systematic review of MRI studies examining the relationship between physical fitness and activity and the white matter of the ageing brain. *NeuroImage*, *131*, 81–90. https://doi.org/10.1016/j.neuroimage.2015.09.071

Sheehan, D. V., Lecrubier, Y., Sheehan, K. H., Amorim, P., Janavs, J., Weiller, E., … Dunbar, G. C. (1998). The Mini-International Neuropsychiatric Interview (M.I.N.I.): The development and validation of a structured diagnostic psychiatric interview for DSM-IV and ICD-10. In *Journal of Clinical Psychiatry* (Vol. 59, pp. 22–33).

Stats SA. (2017). Public healthcare: How much per person? Retrieved October 25, 2019, from http://www.statssa.gov.za/?p=10548

Sylvia, L. G., Bernstein, E. E., Hubbard, J. L., Keating, L., & Anderson, E. J. (2014). Practical

guide to measuring physical activity. *Journal of the Academy of Nutrition and Dietetics*, *114*(2), 199–208. https://doi.org/10.1016/j.jand.2013.09.018

Taki, Y., Kinomura, S., Sato, K., Inoue, K., Goto, R., Okada, K., … Fukuda, H. (2008). Relationship Between Body Mass Index and Gray Matter Volume in 1,428 Healthy Individuals. *Obesity*, *16*, 119–124. https://doi.org/10.1038/oby.2007.4

Tan, X.-Y., Pi, Y.-L., Wang, J., Li, X.-P., Zhang, L.-L., Dai, W., … Wu, Y. (2016). Morphological and functional differences between athletes and novices in cortical neuronal networks. *Frontiers in Human Neuroscience*, *10*, 660. https://doi.org/10.3389/fnhum.2016.00660

Terribilli, D., Schaufelberger, M. S., Duran, F. L. S., Zanetti, M. V., Curiati, P. K., Menezes, P. R., … Busatto, G. F. (2011). Age-related gray matter volume changes in the brain during non-elderly adulthood. *Neurobiology of Aging*, *32*(2), 354–368. https://doi.org/10.1016/j.neurobiolaging.2009.02.008

Thayer, J. F, & Sternberg, E. (2006). Beyond Heart Rate Variability: Vagal Regulation of Allostatic Systems. *Annals of the New York Academy of Sciences*, *1088*(1), 361–372. https://doi.org/10.1196/annals.1366.014

Thayer, Julian F., & Lane, R. D. (2009). Claude Bernard and the heart–brain connection: Further elaboration of a model of neurovisceral integration. *Neuroscience & Biobehavioral Reviews*, *33*(2), 81–88. https://doi.org/10.1016/j.neubiorev.2008.08.004

Thompson, P. M., Cannon, T. D., Narr, K. L., Van Erp, T., Poutanen, V. P., Huttunen, M., … Toga, A. W. (2001). Genetic influences on brain structure. *Nature Neuroscience*, *4*(12), 1253–1258. https://doi.org/10.1038/nn758

Torres, E. R., Strack, E. F., Fernandez, C. E., Tumey, T. A., & Hitchcock, M. E. (2015). Physical Activity and White Matter Hyperintensities: A Systematic Review of Quantitative Studies.

Preventive Medicine Reports, *2*, 319–325. https://doi.org/10.1016/j.pmedr.2015.04.013

Ulrich-Lai, Y. M., & Herman, J. P. (2009). Neural regulation of endocrine and autonomic stress responses. *Nature Reviews. Neuroscience*, *10*(6), 397–409. https://doi.org/10.1038/nrn2647

Vanhees, L., Lefevre, J., Philippaerts, R., Martens, M., Huygens, W., Troosters, T., & Beunen, G. (2005). How to assess physical activity? How to assess physical fitness? *European Journal of Preventive Cardiology*. https://doi.org/10.1097/01.hjr.0000161551.73095.9c

Verstynen, T. D., Lynch, B., Miller, D. L., Voss, M. W., Prakash, R. S., Chaddock, L., … Erickson, K. I. (2012). Caudate nucleus volume mediates the link between cardiorespiratory fitness and cognitive flexibility in older adults. *Journal of Aging Research*, 1–11. https://doi.org/10.1155/2012/939285

Voelcker-Rehage, C., & Niemann, C. (2013). Structural and functional brain changes related to different types of physical activity across the life span. *Neuroscience and Biobehavioral Reviews*, *37*(9), 2268–2295. https://doi.org/10.1016/j.neubiorev.2013.01.028

Wang, G. J., Volkow, N. D., Logan, J., Pappas, N. R., Wong, C. T., Zhu, W., … Fowler, J. S. (2001). Brain dopamine and obesity. *Lancet (London, England)*, *357*(9253), 354–357. https://doi.org/10.1016/s0140-6736(00)03643-6

Warburton, D. E. R., Nicol, C. W., & Bredin, S. S. D. (2006). Health benefits of physical activity: The evidence. *CMAJ*, *174*(6), 801–809. https://doi.org/10.1503/cmaj.051351

Wenzel, A. (2017). Brain Imaging. *The SAGE Encyclopedia of Abnormal and Clinical Psychology*. https://doi.org/10.4135/9781483365817.n214

West, G. L., Zendel, B. R., Konishi, K., Benady-Chorney, J., Bohbot, V. D., Peretz, I., & Belleville, S. (2017). Playing Super Mario 64 increases hippocampal grey matter in older adults. *PLOS ONE*, *12*(12), e0187779. https://doi.org/10.1371/journal.pone.0187779

Williams, V. J., Hayes, J. P., Forman, D. E., Salat, D. H., Sperling, R. A., Verfaellie, M., &

Hayes, S. M. (2017). Cardiorespiratory fitness is differentially associated with cortical thickness in young and older adults. *NeuroImage, 146*, 1084–1092. https://doi.org/10.1016/j.neuroimage.2016.10.033

Winkelmann, T., Thayer, J. F., Pohlack, S., Nees, F., Grimm, O., & Flor, H. (2017). Structural brain correlates of heart rate variability in a healthy young adult population. *Brain Structure & Function*, *222*(2), 1061–1068. https://doi.org/10.1007/s00429-016-1185-1

Wood, K. N., Nikolov, R., & Shoemaker, J. K. (2016). Impact of Long-Term Endurance Training vs. Guideline-Based Physical Activity on Brain Structure in Healthy Aging. *Frontiers in Aging Neuroscience*, *8*, 155. https://doi.org/10.3389/fnagi.2016.00155

Woollett, K., & Maguire, E. A. (2011). Acquiring "the knowledge"; of London's layout drives structural brain changes. *Current Biology*, *21*(24), 2109–2114. https://doi.org/10.1016/j.cub.2011.11.018

World Health Organization. (2010). *Global recommendations on physical activity for health.*

Zaryski, C., & Smith, D. J. (2005). Training principles and issues for ultra-endurance athletes. *Current Sports Medicine Reports*, *4*(3), 165–170. https://doi.org/10.1007/s11932-005-0062-0

Zatorre, R. J., Fields, R. D., & Johansen-Berg, H. (2012). Plasticity in gray and white: neuroimaging changes in brain structure during learning. *Nature Neuroscience*, *15*(4), 528–536. https://doi.org/10.1038/nn.3045

Zhang, D., Shen, X., & Qi, X. (2016). Resting heart rate and all-cause and cardiovascular mortality in the general population: A meta-analysis. *CMAJ*, *188*(3), E53–E63. https://doi.org/10.1503/cmaj.150535