As the late-life cognitive impairment and dementia are common challenges in our society, research identifying modifiable risk factors has become increasingly important. One such risk factor is low physical fitness. The present thesis focuses on cardiorespiratory fitness and its connections with brain volume and various domains of cognitive function in at-risk older people from the general population. The thesis also explores the associations between muscle strength and cognitive function in a population-based sample of older adults with a focus on the methodology of measuring muscle strength.
CARDIORESPIRATORY FITNESS, MUSCLE STRENGTH, BRAIN VOLUMES AND COGNITION IN AGEING MEN AND WOMEN

A POPULATION-BASED STUDY
Heikki Pentikäinen

CARDIORESPIRATORY FITNESS, MUSCLE STRENGTH, BRAIN VOLUMES AND COGNITION IN AGEING MEN AND WOMEN

A POPULATION-BASED STUDY

To be presented by permission of the Faculty of Health Sciences, University of Eastern Finland for public examination in Medistudia Auditorium MS301, Kuopio on Friday, January 31st 2020, at 12 o’clock noon

Publications of the University of Eastern Finland
Dissertations in Health Sciences
No 545

Foundation for Research in Health Exercise and Nutrition,
Kuopio Research Institute of Exercise Medicine
Institute of Biomedicine, School of Medicine, Faculty of Health Sciences,
University of Eastern Finland
Kuopio
2020
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ABSTRACT

As the number of older adults increases in Finland and worldwide, late-life cognitive impairment and dementia due to neurodegenerative and vascular disorders are common challenges in our society. All actions that could delay the onset of cognitive impairment are of major significance from both a humane and economic point of view. The aim of this doctoral thesis was to investigate the association between cardiorespiratory fitness (CRF) and brain volume and the associations of CRF and muscle strength with cognitive function in older men and women.

CRF was assessed as peak oxygen uptake ($V_{O2}\text{peak} \, \text{ml/kg/min or L/min}$) by respiratory gas analysis in a maximal symptom-limited exercise stress test on a cycle ergometer. Handgrip strength and strength of the main muscle groups of lower and upper body was extensively tested with three (knee extension, knee flexion, leg press) and two (chest press, seated row) exercises, respectively. Cognitive functions were assessed using an extensive neuropsychological test battery (NTB) and using the Consortium to Establish a Registry for Alzheimer's Disease (CERAD) neuropsychological test battery from which the CERAD total score was calculated. Brain magnetic resonance imaging was conducted and brain measure was performed using automatic segmentation methods.

Higher CRF was associated with larger cortical and total grey matter volumes in ageing men at increased risk for Alzheimer’s disease, but not in women. Extensively measured lower and upper body muscle strength was positively associated with global cognition but no association was observed between handgrip strength and global cognition. Over two years, CRF was positively associated with global cognition, executive functions and processing speed but not with memory.

This doctoral thesis suggests that higher CRF is associated with larger total grey matter volumes in ageing men at increased risk for cognitive impairment, and that higher CRF and higher muscle strength are both independently associated with better cognitive functions in ageing men and women.
TIIVISTELMÄ

Ikääntyvien määrän kasvaessa voimakkaasti sekä Suomessa että maailmalla myöhäisiän kognitiiviseen heikentymiseen ja dementiaan johtavat hermostoa rappuuttavat sairaudet sekä erisuonerakeräiset häiriöt ovat nykyään merkittäviä yhteiskunnallisia haasteita. Kaikki keinot joilla kognitiivista heikentymistä voitaisiin viivästyttää, ovat ensiarvoisen tärkeitä sekä inhimillisestä että taloudellisesta näkökulmasta katsottuna. Tämän väitöskirjan tarkoituksena oli tutkia kestävyyskunnon ja aivojen tilavuuksien välisiä yhteyksiä sekä kestävyyskunnon ja lihasvoiman yhteyttä kognitiivisiin toimintoihin ikääntyvillä miehillä ja naisilla.

Kestävyyskuntoa kuvattiin maksimaalisella hapenottokyvyllä (VO\textsubscript{2peak}, ml/kg/min tai L/min), joka mitattiin oirerajoitteisessa maksimaalisessa pyöräergometristestissä. Lihasvoimaa mitattiin kädien puristusvoimalla, kolmella alaraajan liikkeellä (polven ojennus, polven koukistus ja jalkaprässi) sekä kahdella ylävartalon liikkeellä (penkkipunnerrus ja kulmasoutu istuen). Kognitiivisia toimintoja arvioitiin kattavalla neuropsykologisella testisarjalla (NTB) sekä The Consortium to Establish a Registry for Alzheimer’s Disease (CERAD) -testisarjalla, josta laskettiin CERAD-kokonaispistemäärä kuvaamaan yleistä kognitiivista toimintakykyä. Aivot tutkittiin magneettikuvauksella ja kuvat segmentoitiin automaattisia menetelmiä käyttäen.

Parempi kestävyyskunto oli yhteydessä suurempaan aivoksi oikouheen harmaan aineen tilavuuteen sekä harmaan aineen kognitiiviseen toimintakykyyn. Kestävyyskunto oli kahden vuoden seurannassa positiivisesti yhteydessä yleiseen kognitiiviseen toimintakykyyn, mutta käden puristusvoiman ja yleisen kognitiivisen toimintakyvyn välillä ei havaittu yhteyttä väestöotoksessa. Parempi kestävyyskunto oli kahden vuoden seurannassa positiivisesti yhteydessä yleiseen kognitiiviseen toimintakykyyn, toiminnanohjaukseen ja prosessointinopeuteen, mutta ei muistiin.

Parempi kestävyyskunto on yhteydessä suurempaan aivojen harmaan aineen kokonaistilavuuteen, ja parempi kestävyyskunto sekä lihasvoima ovat itsenäisesti
yhteydessä parempaan kognitiiviseen toimintakykyyn ikääntyvillä miehillä ja naisilla.

Luokitus: QT 256, WE 504, WL 300, WT 104

Yleinen suomalainen ontologia: maksimaalinen hapenotto; fyysinen kunto; kestävyys; lihasvoima; aivokuori; valkea aine; hippocampus; kognitio; ikääntyminen; testit; kuntotestit; magneettikuvaus
This doctoral thesis was carried out in the Kuopio Research Institute of Exercise Medicine. It has been a privilege to be part of two world class research teams, the DR’s EXTRA and FINGER study groups.

Many individuals have supported and helped me during this laborious journey. Especially I want to thank:

My main supervisor, Professor Rainer Rauramaa for including me in your research team already when I was finishing my master’s degree. You encouraged me to undertake this thesis and gave me an opportunity to use data from the unique DR’s EXTRA Study.

My co-supervisor, Professor Hilkka Soininen for showing me the path to the world of science. I was an undergraduate student without a topic for the master’s thesis, and I contacted you without knowing anything about you or your merits in the field of neuroscience. You told me about the FINGER study which could possibly provide some opportunities for me too…I have always admired how fast someone in your position can reply to e-mails. It is amazing.

Professor Olli J. Heinonen and Docent Minna Raivio, the official reviewers of my thesis. Your comments and criticism really improved this work.

Docent Kai Savonen for your time and guidance with the various challenges that encountered me on an everyday basis. Your rock solid knowledge about the research methodology, especially about statistics, is far above the average clinical researcher.

Dr. Pirjo Komulainen for all the help with the manuscripts, grant applications and practically everything what came on my way. You kept me on track during this journey and always had time and patience to answer my many questions.

Docent David Laaksonen for the comprehensive linguistic revision of this thesis.

Professor Miia Kivipelto, you welcomed me to the FINGER team and got things going forward at a very early stage. I’m truly amazed for everything you have accomplished in science.

Teemu Paajanen and Ilona Hallikainen, for teaching me the basics of extremely complicated concept of cognition, Yawu Liu, for all the help with the interpretation of the MRI stuff. Vesa Kiviniemi, for the valuable tips with the statistical issues. All the co-authors, for your important comments with manuscript preparations.

The warm-hearted personnel of the Kuopio Research Institute of Exercise Medicine. Working with you has been a pleasure from the beginning.

Äiti ja Isä, kiitos kaikesta siitä tuesta, jota olen saanut opintojen aikana ja elämässä lapsuudesta aina tämän väitöskirjan valmistumiseen saakka. Hyviä ja ansaittuja eläkепäivien jatkoa Kuopiossa. My sister Sari, just being a great sister!

Sanna, for your love, support and being the mom of my beautiful daughter, Sofie. I couldn’t hope for a better partner beside me.
The Juho Vainio Foundation, Finnish Brain Foundation, Antti and Tyyne Soininen Foundation and Kuopio University Foundation, for financial support during this project.

Kuopio, December 2019

Heikki Pentikäinen
LIST OF ORIGINAL PUBLICATIONS

This dissertation is based on the following original publications:


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## ABBREVIATIONS

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
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<tbody>
<tr>
<td>AD</td>
<td>Alzheimer’s Disease</td>
</tr>
<tr>
<td>ADAS-Cog</td>
<td>The Alzheimer's Disease Assessment Scale - Cognitive Subscale</td>
</tr>
<tr>
<td>BDNF</td>
<td>Brain-derived neurotrophic factor</td>
</tr>
<tr>
<td>BIC</td>
<td>Bayesian information criterion</td>
</tr>
<tr>
<td>CAIDE</td>
<td>Cardiovascular Risk Factors, Aging and Dementia</td>
</tr>
<tr>
<td>CERAD</td>
<td>Consortium to Establish a Registry for Alzheimer's Disease</td>
</tr>
<tr>
<td>CERAD-TS</td>
<td>CERAD total score</td>
</tr>
<tr>
<td>CRF</td>
<td>Cardiorespiratory fitness</td>
</tr>
<tr>
<td>DR’s EXTRA</td>
<td>Dose-Responses to Exercise Training</td>
</tr>
<tr>
<td>eVO$_{2\text{max}}$</td>
<td>Estimated maximal oxygen uptake</td>
</tr>
<tr>
<td>FINGER</td>
<td>The Finnish Geriatric Intervention Study to Prevent Cognitive Impairment and Disability</td>
</tr>
<tr>
<td>GM</td>
<td>Grey matter</td>
</tr>
<tr>
<td>HIIT</td>
<td>High-intensity interval training</td>
</tr>
<tr>
<td>HR</td>
<td>Heart rate</td>
</tr>
<tr>
<td>HS</td>
<td>Handgrip strength</td>
</tr>
<tr>
<td>ICV</td>
<td>Intracranial volume</td>
</tr>
<tr>
<td>LB</td>
<td>Lower body</td>
</tr>
<tr>
<td>Max$_{dur}$</td>
<td>Maximal exercise test duration</td>
</tr>
<tr>
<td>MCI</td>
<td>Mild cognitive impairment</td>
</tr>
<tr>
<td>MCI</td>
<td>Mini-Mental State Examination</td>
</tr>
<tr>
<td>MRI</td>
<td>Magnetic resonance imaging</td>
</tr>
<tr>
<td>MTL</td>
<td>Medial temporal lobe</td>
</tr>
<tr>
<td>NTB</td>
<td>Neuropsychological test battery</td>
</tr>
<tr>
<td>SD</td>
<td>Standard deviation</td>
</tr>
<tr>
<td>SV</td>
<td>Stroke volume</td>
</tr>
<tr>
<td>UB</td>
<td>Upper body</td>
</tr>
<tr>
<td>WM</td>
<td>White matter</td>
</tr>
<tr>
<td>WMH</td>
<td>White matter hyperintensities</td>
</tr>
<tr>
<td>Symbol</td>
<td>Description</td>
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<tr>
<td>------------</td>
<td>------------------------------------</td>
</tr>
<tr>
<td>$\text{VO}_2\text{max}$</td>
<td>Maximal oxygen uptake</td>
</tr>
<tr>
<td>$\text{VO}_2\text{peak}$</td>
<td>Peak oxygen uptake</td>
</tr>
<tr>
<td>$\text{VO}_2\text{VAT}$</td>
<td>Ventilatory anaerobic threshold</td>
</tr>
</tbody>
</table>
1 INTRODUCTION

As the number of older adults increases in Finland and worldwide, late-life cognitive impairment and dementia due to neurodegenerative and vascular disorders are common challenges in our society. Estimates from the World Alzheimer Report 2015 indicate that 46.8 million people worldwide have dementia, and this number is expected to increase to 74.7 million by 2030 and 131.5 million by 2050 (1). The medical, psychosocial, and economic consequences of cognitive impairment combined with the growing population of people over 65 years of age necessitates multidimensional solutions (2) and research identifying modifiable risk factors has become increasingly important (3). One such risk factor is low cardiorespiratory fitness (CRF). CRF can be improved by aerobic training such as brisk walking, jogging, skiing, swimming, and biking, which increase breathing and heart rate. According to recently published physical activity guidelines (4), adults and older adults should do at least 150 minutes to 300 minutes a week of moderate-intensity, or 75 minutes to 150 minutes a week of vigorous-intensity aerobic physical activity, or an equivalent combination of moderate- and vigorous-intensity aerobic activity.

Previous studies in healthy older adults have reported an association of high CRF with better global cognitive function and several cognitive domains (see e.g. 5-8). While most of the evidence in older adults suggests that high CRF relates to better cognitive function, not all studies have found an association. Previous studies are based mainly on cross-sectional data, and only a few studies have investigated the longitudinal associations between CRF and cognitive function. Furthermore, these studies have methodological limitations restricting conclusions regarding cognitive change over time. For example, CRF and cognition have been assessed only at one time point or only one specific cognitive domain has been considered.

Alzheimer’s disease (AD) is a progressive neurodegenerative disorder that is characterised by the loss of brain volume. The decrease in brain volume is often visible on magnetic resonance imaging (MRI) already years before the clinical onset of mild cognitive impairment (MCI), which represents an intermediate state of cognitive function between the changes seen in ageing and those fulfilling the criteria for AD (9). High CRF is associated with larger brain volume (10,11). Interestingly, the larger volume associated with CRF is primarily in the same regions that are most seriously affected by ageing (10). However, there is a lack of studies investigating whether the association between CRF and brain volume is similar between men and women.

In addition to aerobic physical activity, new guidelines (4) recommend muscle-strengthening activities on two or more days a week. Such activity, generally performed as resistance training or by lifting weights, increases skeletal muscle strength, power, endurance, and mass. Indeed, evidence is emerging...
about the health benefits of muscle strength *per se*. A recent meta-analysis of almost two million subjects indicated that regardless of age, higher levels of upper- and lower-body muscular strength are associated with a lower risk of mortality in the adult population (12). In a study of non-demented older female volunteer twins, researchers found consistent association between leg power at baseline and cognitive function after 10 years (13). Handgrip strength (HS) has been widely used as a measure of muscle strength in various study settings, and studies examining the association between muscle strength and cognitive function are no exception. It is still unclear, however, whether the association between muscle strength and cognitive function is adequately characterised by HS as a measure of overall muscle strength, or whether extensively measured muscle strength reflects global muscle strength more appropriately.

A recent meta-analysis (14) concluded that a combination of aerobic and strength training according to current physical activity guidelines is beneficial for cognitive function. Importantly, by increasing the volume or intensity of an appropriate type of exercise, practically everyone can improve their health-related fitness. The concept of health-related fitness consists of five categories: CRF, muscular strength, motor fitness, body composition and metabolism (15). The present study focuses on CRF and its connections with brain structure and various domains of cognitive function in at risk older people from the general population. The thesis also explores the associations between muscle strength and cognitive function among a representative population-based sample of older Finnish men and women with a special focus on the methodology of measuring muscle strength.
2 REVIEW OF THE LITERATURE

2.1 AGEING OF THE CARDIORESPIRATORY SYSTEM

Cardiorespiratory fitness refers to the ability of circulatory and respiratory systems to supply oxygen to skeletal muscles, and the ability of the muscles to utilise the oxygen during prolonged exercise (16). The gold standard measure of CRF is maximal oxygen uptake, $\text{VO}_2\text{max}$ — the highest rate at which a person is able to consume oxygen during sustained, exhaustive exercise (17,18). $\text{VO}_2\text{max}$ is the product of cardiac output and arteriovenous oxygen ($a - \text{VO}_2$) difference at physical exhaustion, as shown in the following equation: $\text{VO}_2\text{max} = (\text{HR X SV}) \times a - \text{VO}_2\text{diff}$, where HR indicates heart rate and SV indicates stroke volume (19). According to a common perception, $\text{VO}_2\text{max}$ is mainly limited by maximal cardiac output rather than peripheral factors (20). True attainment of physiological $\text{VO}_2\text{max}$ is typically defined by a plateau in $\text{VO}_2$ which indicates that maximal effort is achieved and sustained for a specified period (19). In this work, however, I will use the term $\text{VO}_2\text{peak}$ which is easier to define and determine. It is straightforward term for the highest value of $\text{VO}_2$ achieved on the particular incremental exercise test.

In addition to the traditional fatigue hypothesis, that cardiac output is the most important regulator of human exercise performance, it has been proposed that fatigue is a brain-derived emotion that regulates the exercise behavior to ensure the protection of whole body homeostasis (21). This “Central Governor Model” suggests that the brain regulates exercise performance by continuously modifying the number of motor units that are recruited in the exercising limbs.

Ageing results in an annual decline of approximately 1% in $\text{VO}_2\text{peak}$ in men and women regardless of activity level from age 20-60 years (22). Studies with older individuals tend to show higher relative loss rates over time of approximately 1.5% annually in sedentary individuals (22). However, participation in relatively high frequency and intensity aerobic training may roughly halve these age-related loss rates in $\text{VO}_2\text{peak}$ (23). Maximal heart rate decreases of 8 to 10 beats per minute or roughly 7% of the heart rate reserve per decade (24). Lower heart rate together with lower stroke volume and smaller arteriovenous oxygen difference at maximal exercise all contribute to the age-related decline in $\text{VO}_2\text{peak}$ (25). Reduced skeletal muscle mass is also associated with the decline in $\text{VO}_2\text{peak}$ with ageing (26). Furthermore, there is a reduction in skeletal muscle oxidative capacity and capillary density in the elderly compared to younger subjects (27), but this is likely due to the decreased physical activity rather than ageing per se.

There are structural and functional changes in the heart and blood vessels with advancing age. Left ventricular diastolic function changes resulting in impaired diastolic filling (28). Left ventricular systolic function remains
unchanged at rest but is diminished and manifested by an inadequate rise in the
ejection fraction during exercise (29). Sensitivity to catecholamines decreases,
which diminishes myocardial contractility (30,31) and heart rate response (32,33).
Walls of the large arteries become thicker, leading to reduction of their elastic
properties and increased arterial stiffness (34) which further leads to increase in
pulse wave velocity and elevates systolic and pulse pressure (35). A significant
increase in systolic blood pressure with no change or even a decrease in diastolic
blood pressure, namely isolated systolic hypertension, typically characterises
ageing process (35).

The structural changes in the respiratory system with ageing include changes
in chest wall and thoracic spine, which impairs the total respiratory system
compliance and leads to increased work of breathing. The lung tissue loses its
supporting structure causing increment in airspace size. The alveolar dead space
increases which negatively affects to the arterial oxygen delivery without
impairing the carbon dioxide elimination. Respiratory muscle strength also
decreases with age, particularly in men (36). Despite these changes the
respiratory system is capable of maintaining adequate oxygenation and
ventilation during the entire life span and, in general, the capacity of the
pulmonary system far exceeds the demands required for ventilation and gas
exchange during exercise (37).

2.2 AGEING OF THE SKELETAL MUSCLE

Sarcopenia (38,39) is a hallmark of ageing skeletal muscle, a process
characterised by a substantial loss of muscle mass and strength which often leads
to compromised physical performance. A decline in skeletal muscle mass begins
as early as 25 years of age, and approximately 10% of muscle can be lost by the
age of 50 years (40). The rate of muscle loss then accelerates substantially, and by
the seventh decade of life about 0.7–0.8% of lower limb muscles is reduced per
year in both men and women (41). The main cause for the reduction in whole
muscle mass is the reduced number of myofibres, and to a lesser degree a
decrease in myofibre area (40). Muscle strength significantly decreases after 50–
60 years of age (42,43). The annual rates of decline are approximately 1.5%–4%
(44-46), and are greater in lower limbs than in upper limbs (47,48). The loss of
muscle strength is about three times greater than the loss of muscle mass which
suggests a decline in muscle quality, i.e. strength per unit of muscle (46).
Interestingly, high fat mass has been associated with lower muscle quality, and
high fat mass also predicts accelerated loss of lean mass (41). Impairments of
muscle strength are likely due not only to decreases in muscle lean mass, but also
a combination of other factors such as a decline in voluntary neural drive (49),
impaired neuromuscular control (50,51), increases in muscle fat accumulation
(52), and excitation-contraction uncoupling (53).
A lack of physical activity is the most important secondary factor affecting muscle ageing. Physical inactivity leads to reductions in muscle volume and power, which is more pronounced in older than younger subjects (54). Strength training promotes muscle hypertrophy and improves muscle strength. The achieved increments in muscle mass and strength in response to strength training may be blunted in older age groups (55). Ageing has also been associated with a reduced muscle protein synthetic response to protein intake (56,57). This “anabolic resistance” can be, however, counteracted by performing physical activity before protein intake which increases the use of protein-derived amino acids in the muscle (56). Sufficient physical activity may be essential to maintain the anabolic responsiveness to protein intake with ageing. However, the conclusion of a recent study (58) was that anabolic resistance to amino acids may not be a problem in healthy older adults so further studies are warranted.

Skeletal muscle has a key role in insulin-mediated glucose uptake and studies have reported a decline in insulin sensitivity with ageing (59,60). However, several later findings support the theory that changes in insulin sensitivity with physical activity and body fat are likely primary to changes in chronological ageing (61). Mitochondria have a crucial role in skeletal muscle bioenergetics. Mitochondria have been extensively examined, and studies have reported declines in mitochondrial content (62,63) and function (see e.g. 64-66) with chronological ageing. Despite the number of studies describing age-related changes in mitochondrial capacity, the results are contradictory. This may be partly because of differences in study methodology. The regenerative capacity of the muscle decreases with ageing, and muscle undergoes several morphological changes. These changes are in turn linked to age-related changes in central and peripheral nervous systems, including a loss of motoneurons and degeneration of neuromuscular junctions (67). Furthermore, the vascular system is often impaired in older people, which may further compromise skeletal muscle function by affecting delivery of oxygen, hormones, growth factors and nutrients (61).

2.3 AGEING OF THE BRAIN

The brain parenchyma of healthy older adults shrinks in volume with annual decrease on the order of 0.2–0.5% (68-70) and brains of people over 60 years of age show annual volume loss of more than 0.5% (70). The magnitude of the tissue loss is approximately similar between grey matter (GM) and white matter (WM) although there is a trend for greater longitudinal tissue loss in WM than in GM (71). Cortical volume reductions are comparable to whole brain volume losses, displaying annual decline rates of around 0.5% in most regions (72).

The atrophy of the frontal lobes has usually been regarded as a normal age-related change, but evidence is accumulating, that temporal areas go through reductions comparable to the frontal changes in healthy older adults (73-75).
Annual atrophy rates of 0.3%–2.4% for the entorhinal cortex (74,76-79) and of 0.8–2.0% for the hippocampus (68,77,80) have been observed. While total cortical volume shows an almost linear decline from third decade of life, hippocampus is relatively stable until about the age of 60 years, after which there is prominent loss (72). The amygdala, putamen, pallidum (74) and caudate nucleus (81) also show longitudinal decline. The volume of the ventricular system increases at an average rate of 2.9% annually, and the rate may also accelerate with age (81).

Levels of dopamine, serotonin and brain-derived neurotrophic factor levels also fall with advancing age and may be involved with the regulation of synaptic plasticity and neurogenesis (82). Increasing age is also one of the most important risk factors for white matter hyperintensities (WMH) (83), which are also associated with cognitive decline.

Some of the atrophy in normal ageing occurs in the areas vulnerable to AD and some in areas less characteristic of the disease (thalamus, accumbens and cerebral and cerebellar WM) in the early stages, which suggests that many of the changes observed in healthy ageing are caused by processes unrelated to degenerative disease (74). It has been hypothesised that the temporal changes seen in the older people are related to preclinical AD, while the frontal changes are less associated with developing disease (74). Atrophy rates in normal ageing described above are several times higher even at the stage of MCI (73), and they will further increase with progression to a full AD diagnosis.

2.3.1 Cardiorespiratory fitness and brain volumes

Several prospective observational studies have explored the effect of cardiorespiratory fitness on brain structures (Table 1). In the first study to explore the associations between CRF and brain volumes in healthy older adults (10), researchers found that high CRF levels attenuated the GM losses with increasing age in the frontal, temporal and parietal cortices, the same regions most affected by ageing. Another study in healthy older adults (84) reported associations between CRF and GM volumes in brain regions similar to a previous study (10), but the effect of CRF on GM volume was independent of age (84). This finding has been further replicated in sedentary older adults (8) as well as postmenopausal women with ongoing hormone replacement therapy (85).

While higher CRF levels are associated with GM volumes in the brain, the relationship between CRF and WM is not so clearly established. Colcombe et al. (10) also found an association between high CRF and larger WM volume in tracts running between the frontal and the posterior parietal lobes. A later study (11) examined the correlation of CRF with brain atrophy in nondemented older adults and older adults with early-stage AD, i.e. clinical dementia rating scale 0.5–1. There was no relationship between fitness and brain atrophy in nondemented participants. In early-stage AD, CRF was associated with whole brain volume and WM volume after adjustment for age (11).
The occurrence of WMH increases with age but high CRF may restrain the adverse effect of age on WMH (86). A study in sedentary, low-fit older adults found no independent association between CRF and WMH volume, but “CRF and physical activity” as a factor variable was positively associated with WM health, represented by the factor variable “fractional anisotropy and WMH volume” (87). A recent study found that high CRF was associated with better WM fibre integrity, measured by diffusion tensor imaging, in older adults who have normal cognitive function or MCI (88). Diffusion tensor imaging can detect impaired WM at a very early stage, much earlier than WMH occurs.

High CRF levels have also been associated with larger hippocampal volumes in healthy older adults (89-91) and also in obese older adults (92). One study (93) reported a differing relationship between CRF and hippocampal volume when researchers compared cognitively intact older adults to older adults with early-stage (clinical dementia rating scale 0.5–1) AD. CRF was not associated with hippocampal volumes in cognitively intact older adults, but positive relationship between high CRF and larger hippocampal volumes was found in older adults with early-stage AD (93). A later study (94) used a non-exercise estimate of CRF and examined whether it is related to grey matter volume in specific regions of interest and to WMH volume in a middle-aged cohort (mean age 59 years) at risk for AD [a positive family history for AD (72.4 %) and apolipoprotein E4 positive (38.7 %)]. High CRF was associated with lower WMH volume and larger volumes in the hippocampus, amygdala, and several cortical regions of interest (94). Finally, two randomised controlled trials have demonstrated the effect of aerobic training on hippocampal volume in healthy older adults (95) and older adults with MCI (96).

Few studies have examined the association between CRF levels and the size of the basal ganglia, including the caudate nucleus, putamen and globus pallidus, in healthy older adults. The first such study (97) found that higher CRF levels were associated with larger volume of the caudate nucleus but not with the volumes of the putamen or globus pallidus. Results of the later work (98) indicated that motor fitness (described as movement speed, reaction speed, balance and fine motor control) but not CRF was positively related with the volume of the putamen and the globus pallidus. Neither motor fitness nor CRF was associated with the volume of the caudate nucleus (98).
Table 1. Prospective observational studies of cardiorespiratory fitness and brain structures

<table>
<thead>
<tr>
<th>Authors</th>
<th>Study population and design</th>
<th>Exposure</th>
<th>Outcome</th>
<th>Main results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Erickson et al.</td>
<td>n=120 (men and women) healthy older adults without dementia randomly assigned to an aerobic exercise group (n = 60) or to a stretching control group (n = 60)</td>
<td>VO_{2\text{max}} (ml/kg/min) measured by treadmill exercise test</td>
<td>Hippocampal volume</td>
<td>Aerobic exercise increased hippocampal volume by 2%. Greater improvements in VO_{2\text{max}} over the 1 year were associated with greater increases in hippocampal volume for the left and right hemispheres. Higher VO_{2\text{max}} levels at baseline were associated with less hippocampal volume loss over the 1-y interval in the right hippocampus, but not the left. Correlations between changes in VO_{2\text{max}} and change in caudate nucleus and thalamic volumes were non-significant.</td>
</tr>
<tr>
<td>Vidoni et al.</td>
<td>n=90 (men and women) early-stage AD (n=37), nondemented (n=53)</td>
<td>VO_{2\text{peak}} (ml/kg/min) measured by treadmill exercise test</td>
<td>Regional brain atrophy</td>
<td>In early-stage AD, the decline in VO_{2\text{peak}} was associated with greater medial temporal atrophy, especially in the parahippocampus. In nondemented older adults, declining VO_{2\text{peak}} was associated with atrophy in the left frontal cortex and putamen and right caudate nucleus, but not in medial temporal region.</td>
</tr>
<tr>
<td>Maass et al.</td>
<td>n=40 (men and women) sedentary healthy older participants</td>
<td>Ventilatory anaerobic threshold (VO_{2\text{VAT}}) measured by recumbent cycle ergometer test. (To avoid any cardiovascular risk, VO_{2\text{VAT}} was calculated instead of VO_{2\text{max}}.)</td>
<td>Regional cerebral blood flow and volume and hippocampal volume</td>
<td>Intervention yielded non-significant results. VO_{2\text{VAT}} improvement correlated with changes in hippocampal perfusion and hippocampal head volume.</td>
</tr>
<tr>
<td>Authors</td>
<td>Study population and design</td>
<td>Exposure</td>
<td>Outcome</td>
<td>Main results</td>
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<tr>
<td>Zhu et al. 2015 (101)</td>
<td>n=565 (men and women) healthy middle-aged participants</td>
<td>Maximal treadmill test duration (Maxdur)</td>
<td>Whole brain volume, white matter volume and integrity</td>
<td>Higher Maxdur was associated with more brain volume and greater white matter integrity measured 5 years later.</td>
</tr>
<tr>
<td></td>
<td>Mean age: 46 y</td>
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<td></td>
<td>Follow-up: 5 y</td>
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<tr>
<td>Kleemeyer et al. 2016 (102)*</td>
<td>n=52 (men and women) healthy older adults</td>
<td>VO_{peak} (ml/kg/min) measured by cycle ergometer exercise test</td>
<td>Hippocampal volume</td>
<td>The change in VO_{peak} was not directly associated with the change in hippocampal volume.</td>
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<tr>
<td></td>
<td>Age: 59-74 y (mean 66 y)</td>
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<td>Follow-up: 6 months</td>
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<tr>
<td>Tian et al. 2016 (103)</td>
<td>n=146 (men and women) healthy older adults</td>
<td>Estimated midlife VO_{peak} (ml/kg/min) at age 50, and VO_{peak} measured</td>
<td>Regional brain volumes</td>
<td>Higher midlife VO_{peak} was associated with greater middle temporal gyrus, perirhinal cortex, and temporal and parietal white matter, but was not associated with atrophy progression.</td>
</tr>
<tr>
<td></td>
<td>Mean age: 70 y</td>
<td>objectively by treadmill exercise test at age 83</td>
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<td></td>
<td>Follow-up: 33 y</td>
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<tr>
<td>Ritchie et al. 2017 (104)</td>
<td>n=731 (men and women) population based sample</td>
<td>Physical fitness measured by grip strength in the dominant hand, forced</td>
<td>Grey matter, white matter, and white matter hyperintensity volumes</td>
<td>Physical fitness at baseline was significantly associated with the 3-year change in white matter volume.</td>
</tr>
<tr>
<td></td>
<td>Mean age: 73 y</td>
<td>expiratory volume in 1s, and 6-metre walk time</td>
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<td></td>
<td>Follow up: 3 y</td>
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</table>

* Study is a clinical trial also reporting prospective observational results and is thus included here.


2.4 COGNITIVE FUNCTION

Cognition denotes brain functions that are involved when we receive, store, process and utilise information. The Oxford dictionary defines cognitive function as “the mental action or process of acquiring knowledge and understanding through thought, experience, and the senses” (105). Cognitive function encompasses domains such as learning and memory, executive functions, complex attention, language, perceptual-motor function and social cognition (Figure 1). Each domain has several subdomains that overlap with each other. Hence successful performance in cognitive tasks is a result of various brain regions and functions working simultaneously together. Diminution in particular cognitive functions like episodic memory, executive functions and mental speed are commonly experienced in ageing, while verbal abilities and world knowledge are better maintained (72). Thus, in this work the focus is on global cognitive function and three specific cognitive domains: memory, executive functions and processing speed.

A long-term memory can be categorised as “declarative”, which refers to conscious recollection of facts, and “non-declarative” which refers to memory of skills and procedures (106). Declarative memory is further subdivided into semantic memory (107), which is the knowledge about the world, and episodic memory (108), which enables human beings to remember past experiences. Episodic memory involves both verbal and visuospatial (non-verbal) aspects. The process of episodic memory can be divided into three phases: 1) encoding/learning, *i.e.* capacity to take in novel information; 2) retention, *i.e.* capacity to hold information over time; and 3) retrieval, *i.e.* capacity to bring back information after a delay (106). Executive function comprises different cognitive skills, including the ability to abstract, switch tasks, plan, organise and adapt behavior to contemporary circumstances (106). Executive functions are required for controlled, goal-directed behavior and can be compromised in a variety of psychiatric and neurological disorders (109). Processing speed (110) is typically defined as speed of finishing a mental task with reasonable accuracy. It is connected to the speed in which a person can understand and react to the information they receive.
2.4.1 Muscle strength and cognitive function

Prospective observational studies have consistently shown the positive association between muscle strength and cognitive function (Table 2). Higher lower and upper body muscle strength are both found to be independently associated with better overall cognitive performance (112). The upper body muscle strength measured by handgrip strength has been widely used as a measure of muscle strength in prospective studies examining the association between strength and cognitive impairment in diverse populations including cognitively intact and impaired participants (113-123). Lower or declining HS has been independently associated with deeper decline in cognition over time (113-119), but contradictory results also exist (120-123). There is substantial variation in current methods of assessing HS, which hinders the comparison between studies (124). HS is simple and quick to measure, but caution is required when HS is generalised to predict global muscle strength (125). Higher baseline leg extensor muscle strength has also been positively associated with global cognitive function cross-sectionally (112), and over a 10-year follow-up (13) in healthy older adults.

The association between extensively (i.e. from multiple muscle groups) measured muscle strength and cognitive function has previously been explored in
only two studies (118,126). The first study (118) investigated whether extensively measured muscle strength at baseline is associated with incident AD and MCI during a mean follow-up of 3.6 years in more than 900 older individuals. Three central findings were: 1) greater muscle strength was associated with a decreased risk of developing AD; 2) greater muscle strength was associated with a slower rate of decline in global cognitive function; 3) After excluding participants with evidence of cognitive impairment at baseline, greater muscle strength was associated with a decreased risk of MCI (118). A later study (126) examined whether improvements in extensively measured muscle strength after progressive strength training would associate with improvements in cognitive function in older adults with MCI. Increased lower body muscle strength was significantly associated with improvements in global cognitive function and executive functions but not with memory. Changes in upper body and whole body strength were not associated with changes in cognitive outcomes (126). Both of these studies included participants with impaired cognitive function. Apparently, the association between extensively measured muscle strength and cognitive function has not been studied before in healthy older adults.

The positive effect of strength training on cognitive functions in healthy older adults was first time demonstrated two decades ago in a randomised controlled trial (127) which showed that an 8-week programme of resistance training lessens anxiety and self-attentiveness. Interestingly, the experimental group and control group showed no differences in memory after the intervention, but significant long-term effects were found in the training group for muscular strength and memory 1 year later (127). A later study (128) concluded that 6-month moderate- and high-intensity strength training programs had equally beneficial effects on executive functions and episodic memory. Another study (129) reported similar benefits over twelve months of once-weekly or twice-weekly strength training for the executive cognitive functions among senior women. Gains in muscular strength per se seem to mediate the benefits of strength training for executive functions (130). Finally, strength training especially with high intensities may specifically benefit memory of older adults with MCI (131).

Several biomarkers that have been associated with decreased cognitive performance, have been proven to alter favourably with strength training. Lower circulating levels of insulin-like growth factor-1 are associated with an increased risk of developing AD (132) and decreased cognitive performance in older adults (133). Moderate- and high-intensity strength training can improve cognitive functions with concomitant increase in insulin-like growth factor-1 levels (128). High cortisol levels have been associated with worse cognitive function in older adults (134). Strength training procuces either no change or somewhat reduced cortisol secretion (135). Higher homocysteine levels have been associated with worse cognitive function. Strength training can significantly reduce serum levels of homocysteine in older adults (136). Metabolic syndrome has been associated with cognitive impairment (137), and the association seems to be pronounced in
those with high level of inflammation (138). A recent meta-analysis (138) suggests that strength training is an effective strategy to ameliorate low-grade inflammation, but the reduction in inflammatory markers could be dependent on increases in muscle mass and higher training volume (139).
<table>
<thead>
<tr>
<th>Authors</th>
<th>Study population and design</th>
<th>Exposure</th>
<th>Outcome</th>
<th>Main results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alfaro-Acha et al. 2006 (113) ¶</td>
<td>n=2160 (men and women) population-based sample of community-dwelling Mexican-American older adults Age: ≥65 y (mean 72 y) Follow-up: 7 y</td>
<td>Handgrip strength</td>
<td>MMSE</td>
<td>Reduced handgrip strength at baseline predicted a greater cognitive decline over a 7-year period.</td>
</tr>
<tr>
<td>Boyle et al. 2009 (118) ¶</td>
<td>n=970 (men and women) community-based older adults without dementia at baseline Age: 54-100 y (mean 80 y) Mean follow-up: 3.6 y (range 1-6 y)</td>
<td>A composite measure of muscle strength derived from testing in 11 muscle groups in the arms, legs and axial muscles</td>
<td>Incident AD, MCI and rate of change in global cognitive function assessed by a composite score of 19 tests</td>
<td>Higher muscle strength at baseline was associated with decreased risk of developing AD and MCI, and slower rate of decline in global cognitive function.</td>
</tr>
<tr>
<td>Atkinson et al. 2010 (120)</td>
<td>n=1793 (women) high-functioning postmenopausal women Age: 65-80 y (mean 70 y) Follow-up: 6 y</td>
<td>Handgrip strength</td>
<td>Modified MMSE</td>
<td>Baseline global cognitive function and change in global cognitive function were associated with the change in handgrip strength, but baseline handgrip strength was not associated with cognitive change.</td>
</tr>
<tr>
<td>Au Yeung et al. 2011 (115)</td>
<td>n=2737 (men and women) cognitively normal older adults Age: ≥65 y (mean 72 y) Follow-up: 4 y</td>
<td>Handgrip strength</td>
<td>MMSE</td>
<td>Weaker handgrip strength at baseline was associated with a lower MMSE score 4 years afterwards.</td>
</tr>
</tbody>
</table>
Table 2. Prospective observational studies of muscle strength and cognitive function (continued)

<table>
<thead>
<tr>
<th>Authors</th>
<th>Study population and design</th>
<th>Exposure</th>
<th>Outcome</th>
<th>Main results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nakamoto et al. 2012 (140)*</td>
<td>n=39 (men) cognitively normal older adults 27 subjects also participated in a body mass-based home exercise program Age: 61-79 y (mean 69 y) Follow-up: 3 months</td>
<td>Isometric torques during maximal voluntary knee extension, plantar flexion, and elbow flexion</td>
<td>MMSE</td>
<td>The change in knee extensor torque, but not in plantar flexor or elbow flexor torque, was significantly associated with the change in MMSE scores.</td>
</tr>
<tr>
<td>Taekema et al. 2012 (119) ¶</td>
<td>n=555 (men and women) no selection criteria on health, functioning or demographic characteristics Age: 85 y at baseline for all participants Follow-up: 4 y</td>
<td>Handgrip strength</td>
<td>Global cognitive performance, attention, processing speed and memory</td>
<td>Higher handgrip strength at baseline, was associated with better global cognitive performance, but not with attention, processing speed and memory. Better performance on all cognitive tests at baseline was associated with a slower decline in handgrip strength.</td>
</tr>
<tr>
<td>Gallucci et al. 2013 (116)</td>
<td>n=309 (men and women) population-based sample Age: ≥77 y (mean 80 y) Follow-up: 7 y</td>
<td>Handgrip strength</td>
<td>MMSE</td>
<td>Higher handgrip strength at baseline was associated with slower cognitive decline.</td>
</tr>
<tr>
<td>Steves et al. 2016 (13)</td>
<td>n=324 (women) healthy female twins Age: 43-73 y (mean 55 y) Follow-up: 10 y</td>
<td>Leg extension muscle power</td>
<td>Seven computerised tests, five for memory and two for processing speed</td>
<td>Higher leg power at baseline was associated with better cognitive function over 10 years.</td>
</tr>
</tbody>
</table>
Table 2. Prospective observational studies of muscle strength and cognitive function (continued)

<table>
<thead>
<tr>
<th>Authors</th>
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<th>Main results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Veronese et al. 2016</td>
<td>n=1249 (men and women) community-dwelling older adults without cognitive impairment</td>
<td>Handgrip strength, knee extension and hip flexion</td>
<td>MMSE</td>
<td>After full adjustments, lower limbs strength and handgrip strength were not associated with cognitive status at the follow-up.</td>
</tr>
<tr>
<td></td>
<td>Age: ≥65 y (mean 72 y)</td>
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<tr>
<td></td>
<td>Follow-up: 4 y</td>
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<tr>
<td>Mavros et al. 2017</td>
<td>n=100 (men and women) subjects with MCI randomised into resistance training (n=27) or (n=24) and cognitive training (n=27) or sham cognitive training (n=22)</td>
<td>Leg press, knee extension, hip abduction, chest press, and seated row</td>
<td>Alzheimer’s Disease Assessment Scale–cognitive subscale (ADAS-Cog); global, executive, and memory domains</td>
<td>No difference observed between groups for any cognitive outcome.</td>
</tr>
<tr>
<td></td>
<td>Age: ≥55 y</td>
<td>Strength test data were combined into lower, upper, and whole-body domains</td>
<td></td>
<td>Increases in lower body strength were associated with improvements in ADAS-Cog and executive domain. Changes in upper body and whole body strength were not associated with changes in cognitive outcomes.</td>
</tr>
<tr>
<td></td>
<td>Follow-up: 6 months</td>
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</table>

¶ Study likely includes participants with cognitive impairment at baseline.

*Study is a clinical trial also reporting prospective observational results and is thus included here.
2.4.2 Cardiorespiratory fitness, physical activity and cognitive function

Prospective observational studies have revealed the positive association between CRF and cognitive function (Table 3). Objectively measured CRF levels at midlife seem to associate with the development of dementia at later life (141,142). A recent study with almost 5000 participants found that higher CRF was significantly associated with a decreased risk of cognitive impairment in healthy men and women with a mean age of 61 years. Specifically, each 1-MET increment in CRF was associated with a 7% decreased risk of cognitive impairment (5). Several cross-sectional and longitudinal studies in healthy older adults have explored the association between CRF and particular cognitive domains. They have reported associations between higher CRF and better global cognitive function (7,143-145), memory (6,7,95,143,146-150), processing speed (7,148,151) and particularly executive functions (7,8,143,145,147,148,151,152). One interesting study (151) with almost 2500 older adults observed that high CRF and low sedentary behavior were jointly associated with the highest cognitive function measured by Digit Symbol Substitution Test score, a test of visuospatial and motor speed-of-processing with a considerable executive function component. Both variables were independently associated with cognition but, in a fully adjusted model, only CRF remained significantly associated with cognitive function (151).

In addition to CRF, self-reported vigorous leisure-time physical activity have also been linked to lower risk of cognitive impairment (153) and dementia (154). However, in a recent meta-analysis (155) of over 400 000 people with a mean age of 45.5 years physical inactivity was not associated with dementia or Alzheimer’s disease when physical activity was assessed over 10 years before dementia onset. The longitudinal association was found between physical inactivity and dementia only when physical activity was measured less than 10 years before the diagnosis of dementia. The preclinical or prodromal stage of dementia is often characterised by a decline in physical activity, and researchers suggest that physical activity assessment at that time may cause reverse causation bias (155).

Kramer et al. (156) studied 124 previously sedentary older adults, 60 to 75 years old, and found that those who received 6 months aerobic training showed substantial improvements in executive functions compared with controls. In a study including men and women with MCI, 6 months of high-intensity aerobic training had superior effects on executive functions in women compared to men (157). Indeed, aerobic training probably has robust but selective benefits for cognitive functions with greatest advantage for executive-control processes (158,159), and these benefits may be larger for women than for men (158).

While most of the evidence from epidemiological studies in healthy older adults suggests that high CRF relates to better cognitive function, not all studies have found the association after controlling for age (11,126). In a recent study of 500 participants with mild to moderate dementia, a moderate to high-intensity aerobic and strength exercise training programme did not slow cognitive impairment, but rather worsened cognition (160). Moreover, a Cochrane review (161) found no cognitive
benefits from aerobic training even when only the subgroup of trials in which CRF improved in the aerobic training group was analysed. However, it is important to note that the Cochrane review did not evaluate the direct effect of CRF on cognitive functions. CRF has been observed to be a better predictor of cognitive gains than, for example, exercise duration. Thus maximising an individual’s CRF may be an important target for achieving cognitive benefits (162).
Table 3. Prospective observational studies of cardiorespiratory fitness and cognitive function

<table>
<thead>
<tr>
<th>Authors</th>
<th>Study population and design</th>
<th>Exposure</th>
<th>Outcome</th>
<th>Main results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Barnes et al.</td>
<td>n=349 (men and women) healthy older adults</td>
<td>$VO_{2\text{peak}}$ (ml/kg/min) measured by treadmill exercise test</td>
<td>modified MMSE at baseline</td>
<td>Participants with worse baseline $VO_{2\text{peak}}$ performed worse on all cognitive tests conducted 6 years later.</td>
</tr>
<tr>
<td>2003 (143)</td>
<td>Age: 55-88 y (mean 69 y)</td>
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<td></td>
<td>$VO_{2\text{peak}}$ was most strongly associated with measures of global cognitive function and attention/executive function.</td>
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<tr>
<td></td>
<td>Follow-up: 6 y</td>
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<tr>
<td>Komulainen et al.</td>
<td>n=1335 (men and women) population-based sample randomised into aerobic exercise, resistance exercise, diet, combined aerobic exercise and diet, combined resistance exercise and diet or reference group</td>
<td>$VO_{2\text{max}}$ (ml/kg/min) measured by cycle ergometer exercise test</td>
<td>Immediate memory, delayed memory, verbal performance, visual performance and MMSE</td>
<td>Improved $VO_{2\text{max}}$ was associated with improved immediate memory in aerobic, resistance, diet and combined aerobic and diet groups, with improved delayed memory in diet group and with verbal performance in aerobic group.</td>
</tr>
<tr>
<td>2010 (150)*</td>
<td>Age: 57-78 y</td>
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<tr>
<td></td>
<td>Follow-up: 2 y</td>
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<tr>
<td>Erickson et al.</td>
<td>n=120 (men and women) healthy older adults without dementia randomly assigned to an aerobic exercise group (n = 60) or to a stretching control group (n = 60)</td>
<td>$VO_{2\text{max}}$ (ml/kg/min) measured by treadmill exercise test</td>
<td>A computerised spatial memory task</td>
<td>Change in $VO_{2\text{max}}$ was not related to improvements in memory for either the entire sample or when considering each group separately.</td>
</tr>
<tr>
<td>2011 (95)*</td>
<td>Mean age: 68 in the aerobic exercise and 66 in stretching control group</td>
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<td></td>
<td>Follow-up: 1 y</td>
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</table>
Table 3. Prospective observational studies of cardiorespiratory fitness and cognitive function (continued)

<table>
<thead>
<tr>
<th>Authors</th>
<th>Study population and design</th>
<th>Exposure</th>
<th>Outcome</th>
<th>Main results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vidoni et al. 2012 (99)</td>
<td>n=90 (men and women) early-stage AD (n=37) nondemented (n=53) Age: ≥60 (mean 73 y in nondemented and 74 y in early-stage AD group) Follow-up: 2 y</td>
<td>VO2peak (ml/kg/min) measured by treadmill exercise test</td>
<td>Memory, language, working memory, executive function, visuospatial ability → a composite measure of cognitive performance</td>
<td>Baseline VO2peak and change in VO2peak were not associated with decline in global cognition in the AD group. In the nondemented group a trend was found for lower baseline VO2peak to be associated with a faster decline in global cognition. The 2-year change in VO2peak was not associated with decline in global cognition.</td>
</tr>
<tr>
<td>Wendell et al. 2014 (6)</td>
<td>n=1400 (men and women) community-dwelling healthy volunteers Age: 19-94 y Follow-up: Up to 18 y (mean 7 y)</td>
<td>VO2max (ml/kg/min) measured by treadmill exercise test</td>
<td>Memory, attention, perceptuomotor speed, language, executive function and global cognition</td>
<td>Lower VO2max at baseline was associated with greater decline in global cognitive function as well as in visual and verbal memory.</td>
</tr>
<tr>
<td>Zhu et al. 2014 (163)</td>
<td>n=2747 (men and women) healthy young adults Age: 18-30 y (mean 25 y) at baseline Follow-up: 25 y</td>
<td>Maximal treadmill test duration (Maxdur) at baseline and after 20 y (n=1957)</td>
<td>Verbal memory, psychomotor speed and executive function at 25 y</td>
<td>Longer Maxdur at baseline was associated with better verbal memory and faster psychomotor speed 25 y later. The 20-year change in Maxdur was positively associated with psychomotor speed.</td>
</tr>
<tr>
<td>Belsky et al. 2015 (164)</td>
<td>n=1037 (men and women) Age: 7-13 y at baseline and 38 y at midlife Follow-up: ~30 y</td>
<td>Submaximal exercise test to estimate VO2max (eVO2max) at age 38 y</td>
<td>Intelligence quotient score averaged across ages 7, 9, 11, and 13 y to produce childhood cognitive test performance measures. Memory, executive, and motor functioning at ages 13 y and 38 y</td>
<td>Participants with higher eVO2max scored higher on cognitive tests at age 38 y. eVO2max was not associated with midlife cognitive performance after adjustment for childhood baseline cognitive performance. Children with better cognitive functioning select healthier lifestyles, supporting neuroselection but not neuroprotection.</td>
</tr>
</tbody>
</table>
Table 3. Prospective observational studies of cardiorespiratory fitness and cognitive function (continued)

<table>
<thead>
<tr>
<th>Authors</th>
<th>Study population and design</th>
<th>Exposure</th>
<th>Outcome</th>
<th>Main results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maass et al. 2015 (100)*</td>
<td>n=40 (men and women) sedentary healthy older participants Age: 60-77 (mean 68) Follow-up: proof-of-concept intervention over 3 months</td>
<td>Ventilatory anaerobic threshold (VO₂ VAT) measured by a recumbent cycle ergometer test (to avoid cardiovascular risk, VO₂ VAT was measured instead of VO₂ max)</td>
<td>Early recall, late recall and recognition memory</td>
<td>The change in VO₂ VAT was positively related to the change in recognition memory and early recall.</td>
</tr>
</tbody>
</table>

* Study is a clinical trial also reporting prospective observational results and is thus included here.
2.4.3 Other factors and cognitive function

Advancing age is the greatest non-modifiable risk factor for cognitive impairment; 3 percent of people age 65-74, 17 percent of people age 75-84, and 32 percent of people age 85 or older have Alzheimer's dementia in the United States (165). The corresponding figures are slightly lower in Europe: 1 percent, 8 percent, and 23 percent for people aged 65-74 years, 75-84 years, and 85 or older, respectively (166). The prevalence of MCI also increases with age: from 4.5% among 60-69 year-olds to 5.8% among 70-79 year-olds, and to 7.1% among 80-89 year-olds (167). Another non-modifiable risk factor for cognitive impairment are genes. In particular, the apolipoprotein E $\varepsilon_4$ allele is associated with faster rate of cognitive decline in large population-based samples (168,169) as well as older adults with MCI (170). However, it is difficult to separate whether cognitive deficits in apolipoprotein E $\varepsilon_4$ carriers reflect the contribution of apolipoprotein E genotype to individual differences or a direct influence of prodromal dementia pathology (171). Genes are known to be involved not only in predisposition to chronic diseases but also contribute to cardiorespiratory fitness and the ageing process. This means that underlying genetic factors may have a favourable effect on many kind of traits (genetic pleiotropy), and the observed association between an explanatory variable and health outcome should not be interpreted entirely as causal (172,173).

Education is robustly associated with level of cognitive function but not with the rate of cognitive decline. This suggests that education affects the risk of late-life dementia because of its association with the level of cognition rather than the rate of cognitive decline (174). This was further confirmed in a recent study, which observed that late-life further education improved crystallised knowledge but not fluid cognitive functions (175). High socioeconomic status (176) and participation in leisure activities (176,177) like reading books and newspapers, solving crossword puzzles, playing board games, and dancing are associated with a reduced risk of MCI and dementia. Epidemiological studies suggest that lifelong experiences that include educational and occupational attainment and leisure activities in later life can increase cognitive reserve. Cognitive reserve might enable some people to be more resilient to undesirable brain changes than others (178).

A Mediterranean diet in participants at high vascular risk (179) as well as Nordic diet in a population-based sample (180) have been shown to be beneficial for cognitive functions of older adults but single nutrient interventions have generally been ineffective in MCI and AD (181).

Hypertension (182) and hypercholesterolaemia (183) in midlife have deleterious influence on cognitive function later in life. In both cases, however, there is evidence of an inverse relation in late life; higher blood pressure and cholesterol are associated with stable or better cognitive function (182,183) which, in turn, highlights the difficulties in making recommendations of optimal levels over the life course. Obesity is associated with cognitive impairment and accelerated cognitive decline in mid-life but the potentially protective effects of obesity against cognitive decline in older age require further examination (184).
Metabolic syndrome negatively impacts cognitive performance in older adults, but it is not clear whether the impact of metabolic syndrome on cognitive functions is similar between men and women (185).

Depressive symptoms are related to worse cognitive function cross-sectionally. According to interesting follow-up data, however, consistently higher-grade depressive symptoms are less strongly associated with poor cognitive functioning than with either moderate or low-grade increasing depressive symptom trajectories (186).

Smoking accelerates cognitive decline in older adults (187,188) but at least a 10-year cessation may reverse adverse effects of smoking on cognitive decline (188). Evidence suggests that low to moderate alcohol consumption in the elderly may protect against cognitive decline (189,190), but excessive alcohol consumption is detrimental to cognitive performance (191,192). Recent evidence does not support a protective effect of light drinking over abstinence on brain structure or cognitive function (193). The combined impact of cigarette smoking and heavy alcohol consumption seems to be especially harmful to cognitive functions (194).

While most commonly prescribed medications are not associated with cognitive performance, certain drugs used for nervous system disorders (e.g. antiepileptics and antipsychotics) and non-nervous system conditions (e.g. antihypertensives, antidiabetics, proton pump inhibitors and laxatives) have been associated with poorer cognitive performance (195). Available drug treatments for AD mainly decrease symptoms of the disease rather than alter the course of the disease (196).

2.5 SUMMARY OF THE REVIEW OF THE LITERATURE

Reduced cardiac output (heart rate x stroke volume) at maximal exercise is mainly responsible for the decline in VO$_{2peak}$ in the elderly. Sarcopenia and unfavourable changes in muscle metabolism like mitochondrial dysfunction and insulin resistance characterise the ageing skeletal muscle and accompanying lose in muscle strength. In early AD, episodic memory is one of the first affected cognitive functions and it also declines on a population basis after age 60 (197). Brain atrophy in healthy ageing is not restricted to the frontal lobes but rather follow the fronto-temporal pattern.

Higher CRF levels are generally associated with greater grey matter volume in the prefrontal cortex and hippocampus and less consistently in other regions (198). Brain structure and function differ between men and women, and studies are needed to clarify whether the association between CRF and brain volumes is also different between men and women. Several studies have explored the relationship between CRF and cognitive function in healthy older adults, but few studies have investigated the longitudinal associations between CRF and several measures of cognitive functions using a data set in which both CRF and cognition have been assessed at two time points. Handgrip strength has been commonly used as a
measure of muscle strength in studies examining the association between strength and cognitive functions, and the usability of extensively measured muscle strength is not clear in this context.
3 AIMS OF THE STUDY

The general aim of this thesis was to investigate the association between cardiorespiratory fitness and brain volume and the associations of cardiorespiratory fitness and muscle strength with cognitive function in older men and women.

The specific aims were to study:

1. The sex-dependent associations of cardiorespiratory fitness with total grey and white matter volumes and regional brain volumes in older men and women at increased risk for cognitive impairment (Study I).

2. The associations of handgrip strength and extensively measured muscle strength (lower and upper body) with global cognitive function in a population-based sample of men and women (Study II).

3. The longitudinal associations of cardiorespiratory fitness with memory, executive functions, processing speed, and global cognitive function in older men and women at increased risk for cognitive impairment (Study III).
4 SUBJECTS AND METHODS

4.1 STUDY POPULATION AND INTERVENTION PROTOCOL

4.1.1 The FINGER study

The Finnish Geriatric Intervention Study to Prevent Cognitive Impairment and Disability (FINGER) was a 2-year population-based multidomain randomised controlled trial (ClinicalTrials.gov Identifier: NCT01041989) done in six centres in Finland (Helsinki, Vantaa, Kuopio, Oulu, Seinäjoki, and Turku). The main objective of the FINGER trial is to investigate the extent to which a multidomain intervention can prevent/delay cognitive impairment in elderly at increased risk of cognitive decline.

Participants were recruited from previous population-based non-interventional surveys (199,200) To be eligible for participating in the trial, individuals were required to be 60-77 years old, and have a CAIDE (Cardiovascular Risk Factors, Aging and Dementia) Dementia Risk Score (201) of 6 points or higher (score based on age, sex, education, systolic blood pressure, body-mass index, total cholesterol, and physical activity; range 0–15 points). Cognitive screening was done with the Consortium to Establish a Registry for Alzheimer’s Disease (CERAD) neuropsychological battery (202) and participants had to meet at least one of the following criteria: word list memory task (ten words three times) results of 19 words or fewer; word list recall of 75% or less; or Mini-Mental State Examination (MMSE) of 26 points or less out of 30 points. These criteria selected individuals with cognitive performance at the mean level or slightly lower than expected for age according to Finnish population norms (203) Exclusion criteria were previously diagnosed dementia; suspected dementia after clinical assessment by study physician at screening visit (individuals recommended for further investigations); mini mental state examination less than 20 points; disorders affecting safe engagement in the intervention (eg, malignant disease, major depression, symptomatic cardiovascular disease, revascularisation within 1 year previously); severe loss of vision, hearing, or communicative ability; disorders preventing cooperation as judged by the study physician; and coincident participation in another intervention trial.

The control group received regular health advice. At baseline, the study nurse gave all participants oral and written information and advice on healthy diet and physical, cognitive, and social activities beneficial for management of vascular risk factors and disability prevention. The intervention group additionally received four intervention components: nutritional guidance, physical activity, cognitive training and social activity and management of metabolic and vascular risk factors. FINGER was approved by the coordinating ethics committee of the Hospital
District of Helsinki and Uusimaa. Participants gave written informed consent at screening and baseline visits. A consortium diagram of the present sub-study (Study III) is illustrated in Figure 2.

![Consortium diagram of the present sub-study (Study III).](image)

**4.1.2 The DR’s EXTRA study**

Dose-Responses to Exercise Training (DR’s EXTRA) Study was a four-year population-based randomised controlled trial on the health effects of regular physical activity and diet (ISRCTN45977199, http://isrctn.org). A random population sample of 1500 men and 1500 women, 55-74 years of age, living in the city of Kuopio in Eastern Finland was taken from the Finnish Population Register, and 1479 men and women participated in the baseline examinations in 2005-2006. Exclusion criteria were conditions that would prevent safe engagement in exercise.
training, serious medical conditions (e.g. severe cardiovascular disease, neurological disease and oncological diseases), and other conditions preventing co-operation (e.g. dementia, excess alcohol intake), as judged by the research physicians.

Altogether 1410 men and women who had no exclusion criteria were randomised into one of the 5 intervention groups (aerobic exercise, resistance exercise, diet, combined aerobic exercise+diet or combined resistance exercise+diet) or the reference group. The study protocol was approved by the Research Ethics Committee of Northern Savo Hospital District. Written informed consent was obtained from all study participants. The DR’s EXTRA study design and recruitment of the participants in the present sub-study (Study II) are presented in Figure 3.

Figure 3. DR’s EXTRA study design and recruitment of the participants in the present sub-study (Study II).
4.2 STUDY DESIGN

The designs of the original studies are summarised in Table 4. Study I was based on the baseline examinations of the FINGER study. Brain MRI was predetermined to be conducted only for a subset of the subjects. After excluding individuals with previous brain infarction (n=5) or missing data on CRF (n=4), the study included 68 participants.

Study II was based on the baseline examinations of the DR’s EXTRA study. The baseline data from resistance exercise (n=236) and resistance exercise + diet (n=234) groups were combined into a single group for analyses. After excluding individuals with missing or insufficient data on muscle strength (n=130) or cognition (n=2), the study included 338 participants.

Study III was based on the baseline and two-year-examinations of the FINGER study. As the aim was to explore the longitudinal associations between CRF and cognitive functions, the study groups were pooled in the analysis, and thus the design was a follow-up cohort study. CRF was intended to be measured for all 443 participants in two centers (Kuopio and Oulu). After excluding participants with missing CRF data at baseline (n=21) and one more participant because of a technical error during the CRF measurement at 24 months, the study included 421 participants derived equally from the intervention and control groups.

Table 4. Summary of the designs, materials and outcomes of interest in the original studies

<table>
<thead>
<tr>
<th>Study</th>
<th>Design</th>
<th>Subjects</th>
<th>Exposure</th>
<th>Outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Cross-sectional</td>
<td>n=68 (39 men, 29 women)</td>
<td>CRF</td>
<td>Total and regional brain volumes</td>
</tr>
<tr>
<td>II</td>
<td>Cross-sectional</td>
<td>n=338 (168 men, 170 women)</td>
<td>Muscle strength</td>
<td>CERAD total score</td>
</tr>
<tr>
<td>III</td>
<td>2-year follow-up</td>
<td>n=421 (226 men, 195 women)</td>
<td>CRF</td>
<td>NTB total score including memory, executive function and processing speed domains</td>
</tr>
</tbody>
</table>

Abbreviations: CRF=cardiorespiratory fitness; CERAD=The Consortium to Establish a Registry for Alzheimer’s Disease neuropsychological battery (202,204); NTB=an extended version of the neuropsychological test battery (205).

4.3 ASSESSMENT OF CARDIORESPIRATORY FITNESS

In Study I, CRF was assessed as peak oxygen uptake (VO$_{2peak}$, ml/kg/min) and in Study III VO$_{2peak}$ (L/min) measured directly by the breath-by-breath method using a paramagnetic oxygen analyzer (Vmax 29, SensorMedics Corporation, Yorba Linda, California, USA) during a maximal symptom-limited exercise test on an electrically-braked cycle ergometer (Ergoline, Bitz, Germany) at baseline and in
Study III also at 24 months. To standardise the conditions before the test, the subject received written instructions to take their medications normally, and to avoid strenuous exercise for at least 24 hours, heavy meals, coffee, tea and cola drinks for at least two hours, alcohol drinking for at least 48 hours and cigarette smoking for at least four hours before the exercise test. A nurse confirmed these prerequisites and experienced physician interviewed and examined the subjects to evaluate any contraindications. In addition, tests were supervised by a physician and a nurse.

The test started with a 6-minute sitting period in the saddle followed by a 5-minute warm-up at 0 watts. Thereafter, workload was increased 20 watts for men and 15 watts for women in every 2 minutes until exhaustion. Electrocardiography was recorded throughout the exercise test (GE Medical System, Freiburg, Germany). Self-reported ratings of perceived exertion were collected at 2-minute intervals using the 20-item Borg Scale. VO$_{2peak}$ was defined straightforwardly as the highest value of VO$_2$ achieved during the incremental exercise test. The exercise test was considered maximal if the respiratory exchange ratio was ≥1.0 or if the test had to be terminated due to cardiovascular or pulmonary reasons, muscle fatigue, or overall fatigue. Participants were verbally encouraged to reach their maximum.

4.4 ASSESSMENT OF MUSCLE STRENGTH

Muscle strength (kg) was tested with air resistance equipment (Hur Ltd., Finland) which replaces the weight plates traditionally used in weight stack machines with a pneumatic system of resistance. Strength of the main muscle groups of LB and UB was tested with three (knee extension, knee flexion, leg press) and two (chest press, seated row) exercises, respectively. For each of five exercises the aim was to find the maximum load with which a subject was able to perform 3–5 repetitions. From these loads measured separately the expected 1 repetition maximum for each exercise was estimated (206). Measurements were made by one limb at a time and a mean value was used in analyses. In the case that the value of either limb was missing, the available value was used. HS was measured two times from the dominant hand using the handheld dynamometer (Saehan, Masan, South Korea) and a mean of the two scores was used in analyses.

4.5 ASSESSMENT OF COGNITIVE FUNCTION

4.5.1 The FINGER study

A thorough cognitive assessment with standard neuropsychological tests (an extended version of the neuropsychological test battery [NTB]) (205) was done at baseline, at 12, and at 24 months by study psychologists. However, CRF data were available at baseline and at 24 months, and cognition data at the same time points were used. Outcomes were the cognitive performance measured with NTB total score, a composite score based on results from 14 tests (calculated as z-scores
standardised to the baseline mean and standard deviation [SD], with higher scores indicating better performance) (207), and NTB domain z-scores for executive functions, processing speed, and memory. The executive functions domain included Category fluency test, Digit span test, Concept shifting test (condition C), Trail making test (shifting score: time in condition B – time in condition A), and a 40-stimulus version of the Stroop test (interference score: time in condition 3 – time in condition 2). The processing speed domain included Letter digit substitution test, Concept shifting test (condition A), and Stroop test (condition 2). The memory domain included Visual paired associates test, immediate and delayed recall; Logical memory test immediate and delayed recall from the Wechsler Memory Scale III; and word list learning and delayed recall from the CERAD test battery.

4.5.2 The DR’s EXTRA study

Cognitive function was assessed using the CERAD neuropsychological test battery (199). The CERAD total score (CERAD-TS) was calculated, as previously described, including Verbal Fluency, Modified Boston Naming Test, Word List Learning, Constructional Praxis, Word List Recall and Word List Recognition Discriminability (204). The score ranged from 0 to 100 points, with higher score indicating better performance.

4.6 MAGNETIC RESONANCE IMAGING

Structural brain imaging was performed using a 1.5T Siemens Avanto scanner (Erlangen, Germany). The imaging protocol included a high resolution sagittal 3-dimensional T1-weighted magnetisation prepared rapid gradient echo (MPRAGE) volume (repetition time [TR] = 2400 ms, echo time [TE] = 3.5 ms, inversion time [TI] = 1000 ms, flip angle = 6°, and voxel size 1 x 1 x 1.2 mm³) and axial fluid-attenuated inversion recovery imaging (FLAIR) (TR = 9000 ms, TE = 109 ms, TI = 2500 ms, flip angle = 150°, and voxel size = 0.5 x 0.5 x 5 mm³).

Image processing was performed with the FreeSurfer image analysis suite version 5.0.3. FreeSurfer morphometric procedures have been demonstrated to show good test-retest reliability across scanner manufacturers and across field strengths (208,209). This highly automated processing includes motion correction and averaging (210) of multiple volumetric T1-weighted images (when more than one is available), removal of non-brain tissue using a hybrid watershed/surface deformation procedure (211), automated Talairach transformation, segmentation of the subcortical white matter and deep grey matter volumetric structures (including hippocampus, amygdala, caudate, putamen, ventricles) (212,213) intensity normalisation (214), tessellation of the grey matter white matter boundary, automated topology correction (215,216), and surface deformation following intensity gradients to optimally place the grey/white and grey/cerebrospinal fluid borders at the location where the greatest shift in intensity defines the transition to the other tissue class (217).
Before the analysis, the T1WI and FLAIR images were visually inspected by an experienced neuroradiologist. Any patients with infarction were excluded from data analysis. White matter hyperintensity was visually inspected and scored with Fazekas scale. If presence of geometric inaccuracy due to white matter hyperintensity in the automated white matter segmentation in Freesurfer analysis, manual edition was then conducted.

We used Freesurfer to obtain estimated intracranial volume (ICV) and used the estimated ICV as ICV to standardise the regional volumes. During the data processing, the automated atlas transformation generates an Atlas Scaling Factor (ASF) for each individual. Freesurfer exploits a relationship between the ICV and the linear transform to MNI305 space. The estimated ICV values are calculated as atlas mask volume/ASF. For details of this procedure, please see Buckner et al. (2004) (218). The estimated ICV has been widely used to standardise the regional volumes in the studies in where Freesurfer was used.

4.7 OTHER ASSESSMENTS

Symptoms of depression were assessed with the Zung self-rating depression scale (scores range from 25-100 with 25-49 as normal range) (219). Education was reported as years of formal education. Height was measured in a standing position without shoes and systolic blood pressure (two measurements using a validated automatic device (Microlife WatchBP Office) in a sitting position, using the right arm, after 10 min of rest) by a trained study nurse. Fasting total serum cholesterol and plasma glucose concentrations were determined enzymatically using commercial reagents from Abbott Laboratories on a clinical chemistry analyzer, Architect c8000 (Abbott Laboratories, Abbott Park, IL, USA).

4.8 STATISTICAL METHODS

Statistical analyses were performed using the IBM SPSS statistics for Windows, version 24.0 (IBM Corporation) and R version 2.11.1. Associations with a p-value of <0.05 were considered as statistically significant.

In Study I the independent samples t-test was used to compare demographic variables and neuroimaging measures between men and women. The normality of the distribution of continuous variables was determined by visual inspection. Linear regression analysis was used to explore the association between VO\(_{2\text{peak}}\) and brain volumes separately for men and women. VO\(_{2\text{peak}}\) values were standardised by calculating gender-specific z-scores, which were pooled together to enable the analysis of both sexes in the same model. Regression analyses were adjusted for age, education, Type 2 diabetes, systolic blood pressure and total cholesterol. To examine whether the regression coefficients were statistically significantly different between men and women, the product term (VO\(_{2\text{peak}}\) x sex) was entered in a regression model together with adjusting variables. An association between
VO_{2peak} and regional medial temporal lobe (MTL) volumes (hippocampus and amygdala) and striatum (caudate and putamen) volumes was analysed separately for the left and right side. All brain volumes were normalised by the total intracranial volume. The impact of multiple testing was assessed using the Benjamini and Hochberg adjustment to control the false discovery rate (220).

In Study II the independent samples t-test was used to compare muscle strength and cognition between men and women. Sum scores for lower body (LB) and upper body (UB) muscle strength were calculated as follows: knee extension + knee flexion + leg press for the LB index; chest press + seated row for the UB index. Sum scores were then standardised by calculating gender-specific z-scores, which were pooled together to enable the analysis of both sexes in the same model for each index. A similar standardising procedure was also carried out for the HS scores. Linear regression analysis was used to separately examine the association of HS, LB muscle strength and UB muscle strength with CERAD-TS adjusted for age. To explore the association between muscle strength and CERAD-TS three models were created: HS, age (Model 1), HS, age and LB index (Model 2), HS, age, LB index and UB index (Model 3). The change in global goodness-of-fit was used to explore whether the stepwise addition of new variable improved the model fit when expanding the Models from 1 to 3. The change in global goodness-of-fit was calculated using the log-likelihood ratio Chi2 (x2) statistic.

In Study III Intervention and control groups were pooled and treated as one group in the analyses. To assess the longitudinal association between VO_{2peak} and cognitive functions, linear mixed model analyses with maximum likelihood estimation were conducted according to a two-level structure, i.e. repeated (baseline and 24 months) VO_{2peak} and cognition measures were clustered within subjects. Linear mixed model is suitable for longitudinal datasets containing correlated and unbalanced data.

We used Bayesian information criterion (BIC) as a measure of model adequacy so that a lower BIC indicates a better model with a better balance between complexity and good fit. We predeterminedly chose the model with the lowest BIC as our final model for a given outcome. That is, we did not force a more complex data structure to our model if it did not improve the model fit but instead brought on unnecessary complexity to the model.

Age, sex, education, study group (intervention versus control), time and depression were included as covariates. Additional adjustments included systolic blood pressure, fasting total serum cholesterol and plasma glucose concentration. As a measure of VO_{2peak}, we used absolute L/min value instead of more commonly used weight adjusted (ml/kg/min) value to avoid the possible bias from the weight change during lifestyle intervention. Height, although not a determinant of cognitive function per se, was included to remove variation (i.e., extra noise) in VO_{2peak} resulting from differences in body size.

The standard linear mixed model pool together within-subjects (i.e., whether a change in one variable during the follow-up is associated with a change in another
one) and between-subjects (i.e. whether an overall level of one variable during the follow-up is associated with an overall level of another one) relationships in such a way that no separation can be made between the two aspects of longitudinal relationship. Because of this limitation we carried out additional analyses to sort out the within- versus between-subject aspects of the relationship between VO$_{2\text{peak}}$ and cognition by using a simple method described by van de Pol & Wright (221). With that method, the subtracting the subject’s mean value from each observation value (i.e. within-subject centering) effectively eliminates any between-subject variation thus creating a new variable that expresses only the within-subject variation component. On the other hand, a second new variable expressing only the between-subject variation component is simply the mean of baseline and two-year observations, i.e. baseline and two-year observations for the same subject are both given the same value. Whenever the parameter estimates of these two effects seem to differ, it is then possible to compare them to see whether they are statistically different from each other.
5 RESULTS

5.1 CARDIORESPIRATORY FITNESS AND BRAIN VOLUMES (STUDY I)

The mean age of the 68 participants was 70, (SD 4.0) years, and 57% were men. Men and women were similarly educated, and there were no difference in global cognitive function between men and women. Systolic blood pressure and total serum cholesterol concentrations were also similar. Men had higher VO\textsubscript{2peak} and greater total and regional brain volumes in all regions of interest (Table 5). After dividing the brain volumes by ICV, the total and cortical GM ratios and the volume of the caudate nucleus were significantly higher in women than in men.

Table 5. Characteristics of the subjects

<table>
<thead>
<tr>
<th></th>
<th>Men (n=39)</th>
<th>Women (n=29)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years</td>
<td>69.4 (4.1)</td>
<td>70.7 (3.8)</td>
<td>0.18</td>
</tr>
<tr>
<td>Education, years</td>
<td>8.9 (2.5)</td>
<td>9.2 (2.6)</td>
<td>0.56</td>
</tr>
<tr>
<td>Body mass index, kg/m\textsuperscript{2}</td>
<td>26.9 (3.1)</td>
<td>28.7 (4.2)</td>
<td>0.05</td>
</tr>
<tr>
<td>Type 2 diabetes, n (%)</td>
<td>3 (7.7)</td>
<td>5 (17.2)</td>
<td>0.29</td>
</tr>
<tr>
<td>Systolic blood pressure, mm Hg</td>
<td>140.1 (16.6)</td>
<td>143.2 (16.1)</td>
<td>0.44</td>
</tr>
<tr>
<td>Total cholesterol, mmol/L</td>
<td>4.7 (1.0)</td>
<td>5.2 (1.0)</td>
<td>0.07</td>
</tr>
<tr>
<td>VO\textsubscript{2peak}, ml/kg/min</td>
<td>26.6 (4.9)</td>
<td>19.8 (5.2)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>MMSE, points</td>
<td>27.1 (1.7)</td>
<td>26.8 (2.4)</td>
<td>0.58</td>
</tr>
<tr>
<td>Total NTB score, z-score</td>
<td>-0.16 (0.46)</td>
<td>-0.07 (0.55)</td>
<td>0.46</td>
</tr>
<tr>
<td>Total intracranial volume, cm\textsuperscript{3}</td>
<td>1658.2 (129.6)</td>
<td>1460.5 (117.6)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Cortical grey matter, cm\textsuperscript{3}</td>
<td>423.3 (33.7)</td>
<td>390.2 (32.0)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Total grey matter, cm\textsuperscript{3}</td>
<td>572.8 (40.5)</td>
<td>527.9 (40.4)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Total white matter, cm\textsuperscript{3}</td>
<td>491.2 (51.4)</td>
<td>427.9 (46.9)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Brainstem, cm\textsuperscript{2}</td>
<td>22.3 (2.0)</td>
<td>20.1 (2.1)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Hippocampus, cm\textsuperscript{3}*</td>
<td>3.7 (0.4)</td>
<td>3.5 (0.4)</td>
<td>0.01</td>
</tr>
<tr>
<td>Amygdala, cm\textsuperscript{3}*</td>
<td>1.4 (0.2)</td>
<td>1.3 (0.2)</td>
<td>0.003</td>
</tr>
<tr>
<td>Caudate, cm\textsuperscript{3}*</td>
<td>3.5 (0.5)</td>
<td>3.3 (0.4)</td>
<td>0.07</td>
</tr>
<tr>
<td>Putamen, cm\textsuperscript{3}*</td>
<td>4.8 (0.6)</td>
<td>4.5 (0.5)</td>
<td>0.008</td>
</tr>
</tbody>
</table>

Data are presented as mean (SD) or n (%). P-value derived from independent Student t-test and from χ\textsuperscript{2}-test (for type 2 diabetes) denotes difference between men and women. * Values are averaged left and right volumes.

The associations between VO\textsubscript{2peak} and brain volumes are reported in Table 6. In men, VO\textsubscript{2peak} was associated with cortical GM volume (β=0.56, p=0.001) and total GM volume (β=0.54, p=0.001). In women, no associations were found between VO\textsubscript{2peak} and brain volumes. VO\textsubscript{2peak} accounted for 23% and 1% of total variance of cortical GM volume (Figure 3a) and 25% and 4% of total variance of total GM volume in men and women (Figure 3b), respectively. VO\textsubscript{2peak} was associated with
cortical (p=0.012) and total GM volume (p=0.012) in men also after the Benjamini and Hochberg adjustment to control the false discovery rate. VO\textsubscript{2peak} did not show any association with regional volumes in MTL or striatum neither in men nor in women. VO\textsubscript{2peak} was associated with cortical (β=0.29, p=0.03) and total GM volume (β=0.34, p=0.01) also when men and women were pooled and analysed as one group (Table 6).

Finally, the interactions between VO\textsubscript{2peak} and sex on cortical and total GM volume were examined. Despite the substantial difference in the magnitude of the association of VO\textsubscript{2peak} with cortical and total GM volume between men and women, the interaction term was non-significant for cortical (p=0.16) and total GM (p=0.24) volume.

Table 6. Association of VO\textsubscript{2peak} with brain volumes separately in men and women and in the entire group (pooled data)

<table>
<thead>
<tr>
<th></th>
<th>Men (n=39)</th>
<th>Women (n=29)</th>
<th>Pooled\textsuperscript{a} (n=68)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>β</td>
<td>p-value</td>
<td>β</td>
</tr>
<tr>
<td>Cortical grey matter</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>0.56</td>
<td>0.001</td>
<td>-0.04</td>
</tr>
<tr>
<td>Total grey matter</td>
<td>0.54</td>
<td>0.001</td>
<td>0.10</td>
</tr>
<tr>
<td>White matter</td>
<td>0.08</td>
<td>0.67</td>
<td>0.34</td>
</tr>
<tr>
<td>Brainstem</td>
<td>0.03</td>
<td>0.88</td>
<td>0.42</td>
</tr>
<tr>
<td>Hippocampus</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Left</td>
<td>0.07</td>
<td>0.70</td>
<td>0.09</td>
</tr>
<tr>
<td>Right</td>
<td>-0.04</td>
<td>0.82</td>
<td>0.09</td>
</tr>
<tr>
<td>Amygdala</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Left</td>
<td>0.11</td>
<td>0.55</td>
<td>-0.09</td>
</tr>
<tr>
<td>Right</td>
<td>0.24</td>
<td>0.18</td>
<td>0.10</td>
</tr>
<tr>
<td>Caudate</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Left</td>
<td>0.18</td>
<td>0.36</td>
<td>0.25</td>
</tr>
<tr>
<td>Right</td>
<td>0.11</td>
<td>0.56</td>
<td>0.29</td>
</tr>
<tr>
<td>Putamen</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Left</td>
<td>-0.10</td>
<td>0.61</td>
<td>0.05</td>
</tr>
<tr>
<td>Right</td>
<td>0.14</td>
<td>0.47</td>
<td>0.06</td>
</tr>
</tbody>
</table>

Values are derived from linear regression analyses adjusted for age, education, type 2 diabetes, systolic blood pressure and total cholesterol. All brain volumes are normalised by total intracranial volume.

\textsuperscript{a}VO\textsubscript{2peak} values are standardised by calculating gender-specific z-scores which are pooled together to enable the analysis of both sexes in the same model.
5.2 MUSCLE STRENGTH AND COGNITIVE FUNCTION (STUDY II)

Of the 338 participants 50% were men, their mean age was 66 (SD 5.3) years, and 0.9% reported previous participation in strength training at least two times per week. Men had greater muscle strength than women in all measurements (p<0.001), but there was no difference in global cognitive function (p=0.11) between men and women (Table 7). The LB (β=0.16, p=0.007) and the UB (β=0.18, p=0.001) muscle strength was positively associated with CERAD-TS, but no association was observed between HS and CERAD-TS (β=0.04, p=0.46) adjusted for age. The association of a 1 SD increase in upper body, lower body and handgrip strength with CERAD-TS is presented in Figure 4.

The global goodness-of-fit of various models reflecting muscle strength are presented in Table 8. Model 2 fitted significantly better than model 1 (the change in log-likelihood ratio χ² statistic 6.84, df=1, p=0.009) indicating that the addition of LB index to a model improved it. Furthermore, model 3 fitted significantly better than model 2 (χ²=4.40, df=1, p=0.04), indicating that the addition of UB index to a model further improved it. Finally, HS was removed from the model to test whether the addition of the UB index to a model including the LB index and age improves the model. The addition of the UB index did not improve the fit of a model (χ²=3.61, df=1, p=0.06).
Table 7. Muscle strength and cognition of the subjects

<table>
<thead>
<tr>
<th></th>
<th>Men (n=168)</th>
<th>Women (n=170)</th>
<th>All (n=338)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leg press, kg</td>
<td>111.7 (65.9-240.0)</td>
<td>89.2 (47.8-148.9)</td>
<td>100.4 (47.8-240.0)</td>
</tr>
<tr>
<td>Knee extension, kg</td>
<td>28.1 (15.3-50.0)</td>
<td>17.0 (5.9-35.2)</td>
<td>22.6 (5.9-50.0)</td>
</tr>
<tr>
<td>Knee flexion, kg</td>
<td>32.8 (18.6-61.9)</td>
<td>19.4 (10.7-33.4)</td>
<td>26.0 (10.7-61.9)</td>
</tr>
<tr>
<td>Lower body sum score, kg</td>
<td>172.6 (112.0-323.8)</td>
<td>125.6 (74.7-193.0)</td>
<td>148.9 (74.7-323.8)</td>
</tr>
<tr>
<td>Chest press, kg</td>
<td>36.5 (17.8-84.0)</td>
<td>18.2 (6.7-32.7)</td>
<td>27.3 (6.7-84.0)</td>
</tr>
<tr>
<td>Seated row, kg</td>
<td>38.9 (20.7-50.0)</td>
<td>18.5 (7.8-29.4)</td>
<td>28.7 (7.8-50.0)</td>
</tr>
<tr>
<td>Upper body sum score, kg</td>
<td>75.5 (38.9-132.5)</td>
<td>36.7 (14.5-60.5)</td>
<td>56.0 (14.5-132.5)</td>
</tr>
<tr>
<td>Handgrip strength, kg</td>
<td>41.8 (18.5-59.0)</td>
<td>22.7 (5.0-38.5)</td>
<td>32.2 (5.0-59.0)</td>
</tr>
<tr>
<td>CERAD total score, points</td>
<td>82.1 (52-99)</td>
<td>83.6 (56-99)</td>
<td>82.9 (52-99)</td>
</tr>
</tbody>
</table>

Values are presented as the mean (range).
Lower body sum score: leg press + knee extension + knee flexion
Upper body sum score: chest press + seated row

Table 8. The association between muscle strength and the CERAD total score (n=338)

<table>
<thead>
<tr>
<th>Model</th>
<th>Log-likelihood ratio</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>-1216.9</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>-1213.5</td>
<td>0.009</td>
</tr>
<tr>
<td>3</td>
<td>-1211.3</td>
<td>0.04</td>
</tr>
</tbody>
</table>

Model 1: handgrip strength, age
Model 2: handgrip strength, age, lower body index (leg press + knee extension + knee flexion)
Model 3: handgrip strength, age, lower body index, upper body index (chest press + seated row)

The p-value denotes the change in log-likelihood ratio after adding a new variable. Lower and upper body indexes are standardised by calculating gender-specific z-scores.
5.3 CARDORESPIRATORY FITNESS AND COGNITIVE FUNCTION (STUDY III)

The mean age of the participants was 69 (SD 4.6) years, and 54% were men (Table 9). Their VO$_2$peak decreased 5.5% during two years, with no difference between study groups (study group x time, p=0.5). VO$_2$peak x group x time interactions were calculated to test whether the effect of cardiorespiratory fitness on cognitive functions varies as a function of the study group. Results were non-significant (p for interaction 0.29-0.75) for all four cognitive outcomes. Symptoms of depression were in the normal level range for a non-depressed study population both at baseline and after two years in both study groups.
Table 9. Descriptive statistics of the study participants

<table>
<thead>
<tr>
<th></th>
<th>Participants, n</th>
<th>Baseline</th>
<th>Participants, n</th>
<th>2 years</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years</td>
<td>421</td>
<td>69.0 (4.6)</td>
<td>n/a</td>
<td>n/a</td>
</tr>
<tr>
<td>Sex, number of women (%)</td>
<td>421</td>
<td>195 (46.3)</td>
<td>n/a</td>
<td>n/a</td>
</tr>
<tr>
<td>Study group, intervention, n (%)</td>
<td>421</td>
<td>212 (50.4)</td>
<td>n/a</td>
<td>n/a</td>
</tr>
<tr>
<td>Education, years</td>
<td>421</td>
<td>9.6 (3.2)</td>
<td>n/a</td>
<td>n/a</td>
</tr>
<tr>
<td>Height, cm</td>
<td>415</td>
<td>167.1 (9.1)</td>
<td>n/a</td>
<td>n/a</td>
</tr>
<tr>
<td>Systolic blood pressure, mmHg</td>
<td>415</td>
<td>141.2 (17.0)</td>
<td>380</td>
<td>139.6 (16.0)</td>
</tr>
<tr>
<td>Total serum cholesterol, mmol/L</td>
<td>420</td>
<td>5.0 (1.0)</td>
<td>392</td>
<td>5.0 (1.0)</td>
</tr>
<tr>
<td>Plasma glucose, mmol/L</td>
<td>420</td>
<td>6.2 (1.1)</td>
<td>393</td>
<td>6.3 (1.1)</td>
</tr>
<tr>
<td>Peak oxygen uptake, L/min</td>
<td>421</td>
<td>1.82 (0.5)</td>
<td>325</td>
<td>1.72 (0.5)</td>
</tr>
<tr>
<td>Peak oxygen uptake, ml/kg/min</td>
<td>415</td>
<td>23.4 (6.2)</td>
<td>322</td>
<td>22.4 (6.0)</td>
</tr>
<tr>
<td>Symptoms of depression, points</td>
<td>388</td>
<td>-</td>
<td>347</td>
<td>32.9 (7.6)</td>
</tr>
</tbody>
</table>

Cognition*

| NTB total score          | 421             | -0.15 (0.57)   | 383             | 0.14 (0.66)     |
| Executive functions      | 420             | -0.13 (0.66)   | 382             | -0.01 (0.70)    |
| Processing speed         | 421             | -0.11 (0.81)   | 382             | 0.09 (0.85)     |
| Memory                   | 421             | -0.18 (0.65)   | 384             | 0.29 (0.80)     |

Data are presented as the mean (SD) or n (%). NTB=neuropsychological test battery. *Scores on the NTB total score, and on executive functions, processing speed, and memory are mean values of the z-scores of the cognitive tests included in each cognitive outcome, with higher scores suggesting better performance.

Over two years, VO\textsubscript{2peak} was associated with executive functions (β=0.16, p=0.01) and processing speed (β=0.25, p<0.001) but not with memory (β=0.11, p=0.12) (Table 10). VO\textsubscript{2peak} was also related to the NTB total score (β=0.12, p=0.01). Results remained unchanged after adjustment for systolic blood pressure, fasting total serum cholesterol and plasma glucose concentration one at a time or simultaneously. Supplementary analyses were completed for the sub-group including only participants with complete VO\textsubscript{2peak} data available, i.e. at baseline and two years. Results remained unchanged with respect to processing speed (β=0.28, p<0.001) and memory (β=0.06, p=0.44), but became non-significant for NTB total score (β=0.10, p=0.07) and executive functions (β=0.13, p=0.07).

Next, the within-subject relationship between VO\textsubscript{2peak} and cognitive function was explored (Table 11). The within-subjects effects were not significant except for processing speed (p=0.02). Then the between-subjects relationship between VO\textsubscript{2peak} and cognitive function was explored. At the cognitive domain level, the between-subjects effect was significant for executive functions (p=0.008) and processing speed (p=0.004), but not with memory (p=0.18). The between-subjects effect was significant also for NTB total score (p=0.01). Within- versus between-subject effects were similar in all analyses (p>0.05).
Table 10. Association of cardiorespiratory fitness and covariates with cognition over two years

<table>
<thead>
<tr>
<th></th>
<th>NTB total scorea</th>
<th>Executive functionsb</th>
<th>Processing speedb</th>
<th>Memorya</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>β</td>
<td>p-value</td>
<td>β</td>
<td>p-value</td>
</tr>
<tr>
<td>Peak oxygen uptake, L/min</td>
<td>0.12</td>
<td>0.01</td>
<td>0.16</td>
<td>0.01</td>
</tr>
<tr>
<td></td>
<td>0.12</td>
<td>0.01</td>
<td>0.25</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Sexc</td>
<td>-0.03</td>
<td>&lt;0.001</td>
<td>-0.03</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>0.06</td>
<td>&lt;0.001</td>
<td>-0.04</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Baseline age, yearsc</td>
<td>-0.01</td>
<td>0.05</td>
<td>-0.01</td>
<td>0.01</td>
</tr>
<tr>
<td></td>
<td>0.01</td>
<td>0.89</td>
<td>-0.02</td>
<td>0.77</td>
</tr>
<tr>
<td>Study group (int. vs. con.)c</td>
<td>0.01</td>
<td>0.89</td>
<td>-0.02</td>
<td>0.77</td>
</tr>
</tbody>
</table>

a Values are derived from a time- and height-adjusted linear mixed model with intercept and time as random effects.
b Values are derived from a time- and height-adjusted linear mixed model with only intercept as a random effect.
c Values are from baseline.

NTB = neuropsychological test battery

Age, sex, education, study group (intervention versus control), time and depression were included as covariates in all analyses. Additional adjustments included systolic blood pressure, fasting total serum cholesterol and plasma glucose concentration both one at a time and simultaneously.

Table 11. Within- and between-subject effects between cardiorespiratory fitness and cognitive function

<table>
<thead>
<tr>
<th></th>
<th>Within-subject effect</th>
<th>Between-subject effect</th>
<th>Difference</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>β</td>
<td>95% CI</td>
<td>β</td>
</tr>
<tr>
<td>NTB total score</td>
<td>0.007</td>
<td>-0.13-0.15</td>
<td>0.21</td>
</tr>
<tr>
<td>Memory</td>
<td>-0.07</td>
<td>-0.33-0.19</td>
<td>0.13</td>
</tr>
<tr>
<td>Executive functions</td>
<td>-0.01</td>
<td>-0.21-0.18</td>
<td>0.25</td>
</tr>
<tr>
<td>Processing speed</td>
<td>0.24</td>
<td>0.04-0.44*</td>
<td>0.33</td>
</tr>
</tbody>
</table>

Difference denotes the difference between within- and between-subject effects (218).
*p-value<0.05
NTB = neuropsychological test battery
6 DISCUSSION

6.1 SUMMARY OF THE MAIN FINDINGS

- Higher cardiorespiratory fitness was associated with greater cortical and total grey matter volumes in elderly men at increased risk for cognitive impairment, but not in women. Cardiorespiratory fitness was not associated with white matter volume or regional brain volumes in the medial temporal lobe or striatum.
- An association between higher muscle strength and better global cognitive function was observed in ageing men and women. However, this association was found only between extensively measured (lower and upper body) muscle strength and global cognitive function, whereas handgrip strength was not associated with cognition.
- Over a two-years follow-up, higher cardiorespiratory fitness was associated with better executive functions, processing speed and global cognitive function but not with memory in older people at increased risk for cognitive impairment.

6.2 INTERPRETATION OF FINDINGS AND COMPARISON TO PREVIOUS STUDIES

6.2.1 Cardiorespiratory fitness and brain volumes

The observation that higher CRF levels are associated with greater cortical and total GM volume is consistent with previous findings (reviewed in reference 198). In the present study, however, CRF was associated with cortical GM and total GM volumes only in men, which suggests that CRF may be especially advantageous to the brain health of men aged 60 years and older. It has been suggested that any protective effect of higher fitness levels may only be observed after a certain age when losses in tissue volume are more prevalent (198). If so, the association between CRF and GM volume in women may not become visible until a later stage of life. Apparently, only one study (222) has previously explored the associations between CRF and brain volumes separately in men and women. In a sample of over 700 healthy older adults, they found that high CRF was associated with reduced white matter lesion load in men, but not in women (222). No associations were found between CRF and white matter volume in the present study in men or women.

Brain GM volume may decline more steeply with age in men than in women (223), and it is common, that women in their sixth decade of life have a significantly larger total GM ratio (GM volume divided by intracranial volume) than men (224). Oestrogen therapy has been argued to play a role in maintaining GM volumes in
women. The results are conflicting, however. Oestrogen therapy has been found to both preserve (225) and reduce (226) the GM ratio in women. Oestrogen therapy may also mitigate the CRF increase induced by aerobic training (227). However, another study found that women with the greatest age-related loss in VO_{2peak} did not replace oestrogen after menopause, whereas oestrogen replacement and maintenance of training volume were associated with maintained VO_{2peak} (228). Taken together the results of different studies, oestrogen therapy can induce both favourable and unfavourable effects on GM volume and CRF levels, and further studies are needed to clarify the effects of oestrogen on fitness levels and brain volumes in older women.

Collectively, there are numerous differences between men and women in brain structure, function and chemistry including differences in brain volumes, cerebral blood flow, glucose utilisation rate and serotonin transmission (229). Which of these are the key factors contributing to the different association of CRF with brain volumes in men and women remains unknown.

The lack of association of CRF with MTL structures and with deep GM structures (i.e., basal ganglia) in this study indicates that CRF may play a role in specifically maintaining peripheral GM volume (i.e., cortex), with a smaller effect on MTL or basal ganglia. However, the positive association between CRF and hippocampal volume has been observed previously (89,90,92-94). One explanation for the divergent finding may be relatively high VO_{2peak} levels observed in this study. Although the cycle ergometer was used to assess VO_{2peak}, the observed VO_{2peak} values were higher than in two previous studies in older adults (89,90) which used motor-driven treadmill to measure VO_{2peak}. Maximal VO_{2} is approximately 10% higher on a treadmill than on a cycle ergometer (230).

Two studies (93,99) also using a treadmill to assess VO_{2peak} explored the associations between CRF and hippocampal volumes separately in early AD patients and non-demented subjects. These studies found that higher CRF was associated with greater hippocampal volumes only in early AD patients. Interestingly, CRF levels in the present study were rather comparable to the CRF levels of non-demented groups in the previous studies (93,99), in which the association between CRF and hippocampal volume was not observed. Taken together, our patients were possibly “too fit” to observe any association between CRF and small brain structures like the hippocampus. Because other studies that reported the association between CRF and hippocampus, were performed on either middle-aged (94) or substantially obese (92) subjects, our results are not comparable. Given that hippocampus is a brain area with high plasticity, also factors other than CRF are likely to contribute its properties over the life course. In a recent study (231), higher educational level attenuated the progression of hippocampal atrophy during a 15-month follow-up period in healthy older men, suggesting that educational attainment may be related to preservation of hippocampal volumes during ageing.
CRF was also not related to the striatal volumes. Previously, at least two studies (97,98) have investigated the association between CRF and striatal volume in older adults. Other study (97) found that the caudate nucleus volume was a significant mediator of the relationship between CRF and cognitive flexibility, but no association was observed between CRF and the volume of the putamen. In the second study (98), motor fitness but not CRF was positively related with the volume of the putamen and the globus pallidus. In this study, however, CRF was measured submaximally, which restricts comparison of the results. Based on existing evidence, one cannot make a statement on whether there is a relationship between CRF and striatum volumes. However, as discussed earlier, benefits of CRF may relate to the GM in the cortex rather than deeper GM structures like the striatum. Of note, larger striatal and prefrontal GM volumes have been observed in physically active young twins compared with inactive co-twins (232).

Three studies (11,93,99) found differing relationships between CRF levels and brain volumes when healthy older adults and older adults with early-stage AD were compared. Based on these observations, it has been suggested that individuals with dementia may have more to gain from being physically fit than individuals without early signs of dementia (198). No evidence exists, however, that being physically fit could stop or reverse advanced degenerative processes in the brain. Rather, one theory suggests that the benefits of life-long aerobic training are limited to the expression and sustainment of cortical reserve at every age, not the age-related decline (233). When accelerated brain atrophy and cognitive decline become evident, the “therapeutic window” for being physically fit may very well be closed. Taken together, most benefits of physical fitness for brain volumes likely emerge before notable brain atrophy is evident. However, it is important to note that highly fit older adults with cognitive impairment possess notable benefits in their general ability to function compared to low fit older adults.

### 6.2.2 Muscle strength and cognitive function

An observed association between higher extensively measured (lower and upper body) muscle strength and better global cognitive function suggests that instead of measuring only HS, it may be rational to perform more extensive but relatively easy measurements of muscle strength. Boyle et al. (118) studied the association of extensively measured muscle strength with incident AD and cognitive decline in more than 900 older persons without dementia at the baseline. They found that during a mean follow-up of 3.6 years, each 1 unit increase in muscle strength (based on 11 muscle groups) at baseline was associated with about a 43% decrease in the risk of AD. Moreover, subjects belonging to the highest tenth of muscle strength had about a 48% decreased risk of developing MCI compared to subjects belonging to the lowest tenth. Of note, only axial muscle strength was individually associated with the risk of AD, whereas LB strength and UB strength were not. Unfortunately, the associations of different components of strength with global cognitive function were not reported (118), given that in the present study, LB and
UB muscle strength were both individually associated with global cognitive function. Another study (126) in older adults with MCI explored whether improvements in muscle strength after strength training intervention associated with improvements in cognitive function. Increased LB muscle strength was significantly associated with improvements in global cognitive function and executive functions but not with memory. After including only participants from the strength training group, LB, UB and whole body muscle strength were all positively associated with global cognitive function as well as with executive functions (126). Both studies (118,126) included participants with impaired cognitive function. Apparently, the present study was first to demonstrate the association between extensively measured muscle strength and global cognitive function in healthy older adults.

Handgrip strength predicts decline in cognition in older community-dwelling populations (234). However, HS was not associated with global cognitive function in the present study. This lack of association may be due to several reasons. Subjects in the previous studies were older than in the present study, and cognitive function was assessed using either MMSE, a rough measurement of global cognitive function (113,115,116,119), or incident AD (114,117,118) as an outcome. In the present study, we used the CERAD battery, which is a sensitive measure of cognitive domains that are vulnerable in early and preclinical stages of AD (202). Furthermore, the CERAD total score was previously found to be superior to the MMSE, or any single CERAD subtest in discriminating cognitively intact subjects from subjects with MCI (235). In a cohort over 2000 ageing individuals (117), poor HS was associated with increased risk of dementia over six years follow-up only among participants with possible MCI. Interestingly, among participants without cognitive impairment, other factors like gait slowing and poor balance were associated with an increased risk of dementia (117). Extensively measured muscle strength may indicate an association of muscle strength with cognitive function in participants without cognitive impairment, whereas the association between HS and cognition likely occurs with older and more cognitively impaired subjects.

6.2.3 Cardiorespiratory fitness and cognitive function

Over two years of follow-up, higher CRF was associated with better executive functions and processing speed and global cognitive function in older adults at risk for cognitive impairment. This longitudinal association between CRF and several cognitive outcomes highlights the importance of higher CRF as one of the health factors that may maintain cognitive function in late adulthood.

Our results are mostly in agreement with a previous longitudinal study (143) in older adults that reported longitudinal associations of baseline CRF with global cognitive function, executive functions and verbal memory over a six-year period. However, cognitive assessment at baseline was restricted to only global cognitive function, which limits conclusions regarding cognitive change over time (143). Wendell et al. (6) studied the longitudinal associations between CRF and cognitive
performance in a prospective observational study with a large sample (n=1400) of subjects over a wide age range (19-94 years). Greater baseline CRF was associated with less memory decline across the life span which differs from our findings in that we did not find association between CRF and memory. Although a long follow-up (18 years) is a strength of that study, CRF measurement only at baseline may be considered a limitation.

The interpretation of the results of the present study may vary depending on the presence of within-subjects or between-subjects relationships between VO$_{2peak}$ and cognitive function. The within-subjects relationship between VO$_{2peak}$ and processing speed indicates that the change in VO$_{2peak}$ was associated with the change in processing speed over time. The presence of a within-subjects relationship may reflect the potential for short-term changes, suggesting that increase in VO$_{2peak}$ may contribute the corresponding improvement in processing speed in relatively short time period (two years in this case). The presence of a between-subjects relationship between VO$_{2peak}$ and all cognitive outcomes except memory suggest that some latent, relatively fixed effects like heredity may underlie the association between CRF and cognitive function. It is also possible that the change in CRF may affect cognitive function through structural brain changes that take much longer time period than two years to emerge. Apparently, no previous studies have reported the within- and between-subject effects of the association between CRF and cognitive functions separately.

In an interesting study (164), researchers analysed a complete birth cohort (n=1037) consisting of all individuals born between April 1972 and March 1973 in Dunedin, New Zealand. They strove to examine whether children with higher CRF had better cognitive function at midlife than their less fit peers (neuroprotection), and also whether children with better cognitive function were predisposed to better adult fitness (neuroselection). Cognitive testing was conducted in childhood when study members were 7, 9, 11, and 13 years old (values averaged to produce childhood cognitive test performance measures), and at the midlife follow-up at age 38 years. Adults with higher CRF exhibited better cognitive performance at midlife, but this advantage was almost completely explained by differences in childhood cognitive function. This finding supports neuroselection rather than neuroprotection. The authors concluded that children with better cognitive functioning select healthier lives (164). This finding is consistent with a recent study (236) in which physical activity and sedentary behaviour were associated with cognition in Finnish older twins, but this was probably caused by genetic and environmental selection.

CRF is strongly and inversely associated with most cardiovascular risk factors (237), which in turn may play an important role in the aetiology of Alzheimer's disease (238). In a recent study (239) with over 6500 older adults, increased numbers of optimal cardiovascular health metrics and a higher cardiovascular health score were associated with a lower risk of dementia and lower rates of cognitive decline which further supports the promotion of cardiovascular health.
to prevent risk factors associated with cognitive decline (239). The association between CRF and cardiovascular risk factors seems the most plausible mechanism to mediate the fitness benefits for the brain health.

Interestingly, CRF can also modify the association between amyloid-β and cognitive function in late-middle-aged adults at risk for AD (240). CRF was associated with immediate memory and verbal learning among persons with high Aβ burden but not in persons with minimal amyloid-β burden (240). CRF is also related to changes in mechanisms such as cerebral blood flow, neurotrophic factors, neurotransmitter systems and neural architecture that have themselves been associated with cognitive performance (241). Lower levels of plasma brain-derived neurotrophic factor (BDNF) have been associated with higher risk for cognitive impairment (242), and animal studies have provided evidence that exercise stimulates BDNF production (243). A one-year walking intervention in humans was beneficial for both BDNF levels and executive functions, but only for individuals over the age of 71, and authors argued that moderate-intensity physical activity may be more beneficial to both BDNF and executive functions for adults over the age of 70 than for younger individuals (244). Finally, fitness-related improvements in cognitive function may be partly mediated by plasma insulin levels (245).

6.3 METHODOLOGICAL CONSIDERATIONS

This doctoral thesis was part of two large randomised controlled trials, FINGER and DR’s EXTRA. The main objective of the FINGER study was to find out whether a multi-domain intervention could prevent cognitive decline among older people. The FINGER participants were recruited from previous population-based non-interventional surveys and represent of an important part of the general Finnish older population with a number of risk factors for dementia, but without pronounced cognitive impairment. However, because of the long predementia stage of neuropathological processes, it remains possible that some participants might already have had dementia-related brain changes (246). The DR’s EXTRA study was designed to examine the effects of regular physical activity and diet on atherosclerosis, endothelial function and cognition. Participants were a representative population-based random sample of middle-aged and elderly men and women. Both original studies aimed to include participants of the general elderly population, not patients in a clinical setting. This increases the external validity of the findings in the present thesis.

The treadmill and cycle ergometer are the most commonly used exercise machines to measure VO2peak. In the present study, VO2peak was measured directly during symptom-limited incremental cycle ergometer test, which represents the “gold standard” for assessing exercise capacity (247). VO2peak values during cycle ergometer exercise average 8-15% lower than those during treadmill exercise, primarily because of the smaller volume of working muscle mass (230). The
majority of the studies examining the association of CRF with brain volumes or cognitive functions have used a treadmill to assess CRF, which hinders the comparability of their results to ours. It is also important to note that submaximal ergometer tests have advantages too. They are practical for people with restrictions such as musculoskeletal limitations, impaired balance and severe obesity (248). Improvements in submaximal measures of CRF can also be observed without a change in maximal CRF, and submaximal measures may capture peripheral adaptations not detected by maximal measures of CRF alone (249).

In study I, all brain volumes were normalised by the total intracranial volume. Studies considering the effect of sex on cross-sectional brain volumes should include a correction for head size (i.e. ICV) to reduce this potential confounding effect (68). The cross-sectional setting does not allow to conclude that higher CRF generates greater brain volumes, because the decrease in the brain volumes may precede the deterioration in CRF. However, it has been shown that aerobic training accompanied with improvement in CRF can lead to increases in brain volumes in humans (250). Thus, it seems plausible that higher CRF affects brain volumes and not the vice versa. The small sample size restricts the interpretation of the findings in the first study. This especially hindered the interaction analyses, in which the results were non-significant. Thus, results should be taken mainly as descriptive and interpreted with caution. However, the magnitude of the association observed in men was relatively high, which suggests that subsequent studies with larger sample sizes are reasonable to verify whether CRF has a larger effect on the brain grey matter volumes in men than in women.

As in the first study, the cross-sectional setting of Study II provides no evidence about the direction of causality between muscle strength and global cognitive function. The main strength of the study is a large population-based sample of ageing men and women with a wide age range. Muscle strength of several muscle groups was measured which enabled the evaluation of lower body and upper body musculature separately. A relatively high number of subjects was excluded due to musculoskeletal complaints preventing reliable measurement of muscle strength. The application of isometric strength testing instead of dynamic testing would probably have decreased the number of excluded subjects. The observed associations between muscle strength and global cognitive function are noteworthy because strength training was rare in the DR’s EXTRA study population. Less than 1% reporting that they were doing strength training according to physical activity guidelines (4), at least two times per week.

In Study III, the intervention and control groups were pooled and treated as one group in the analyses. The study design was a two-year follow-up. It is possible that the intervention yielded effects from socialisation, cognitive stimulation, and changes in lifestyle factors that could affect the observed associations. To address this, the analyses were adjusted for the randomised study group. Strengths of the study include the longitudinal study setting with a notable sample size, comprehensive cognitive assessments and a modern statistical
approach appropriate for analysing complex serial data. The change in the
independent variable between the first and second visits was compared with the
change in health outcome between visits. Results from such longitudinal designs
are used to infer that risk factor changes are related to changes in health outcomes.
However, longitudinal studies also have limitations that can influence the results.
Those who remain in the study systematically have better cognitive function than
those who drop out (selective attrition), and some of the observed change may be
attributable to effects of prior test experience (test-retest effect) (72).

The linear mixed model handles erratic measurement intervals both within and
across study participants, provides valid results despite randomly missing data,
and accounts for the correlation among recurrent measurements in the same
participants (251). Approximately one fifth of the participants did not attend CRF
measurements at 24 months, which is a limitation. However, the linear mixed
model remains unaffected by randomly missing data as described above. Previous
studies are based mainly on cross-sectional data, and only a few studies (6,95,143)
have investigated the longitudinal associations between CRF and cognitive
functions. These studies have methodological limitations restricting conclusions
regarding cognitive change over time. For example, CRF and cognition was
assessed only at one time point in two studies, (6,143) or only one specific cognitive
domain was considered in another study (95). In the present study, both CRF and
cognitive function were measured at two time points, and several cognitive
domains were studied. Analyses to sort out the within- versus between-subject
aspects of the relationship between CRF and cognitive function were also carried
out. Within- and between-subjects associations are rarely reported separately, and
much less discussed.
CONCLUSIONS

This doctoral thesis suggests that higher cardiorespiratory fitness is associated with larger cortical and total grey matter volume in ageing men at increased risk for cognitive impairment, and that higher cardiorespiratory fitness and higher muscle strength are both independently associated with better cognitive function in ageing men and women. These associations of cardiorespiratory fitness and muscle strength with several cognitive outcomes highlight the importance of regular physical activity to achieve higher fitness as one of the contributors in maintenance of cognitive function in the late adulthood. However, memory itself was not associated with higher cardiorespiratory fitness among elderly people with increased risk of dementia, which may be an indication of the unstoppable nature of neurodegenerative changes in dementive diseases (252).

An observed association between higher lower and upper body muscle strength and better global cognitive function suggests that extensive measurement of muscle strength may be more important than handgrip strength alone in studies investigating the association between muscle strength and cognition. Extensively measured muscle strength may indicate an association between strength and cognitive function in participants without cognitive impairment, whereas the association between handgrip strength and cognitive function likely occurs in older and more cognitively impaired subjects.

Possessing high physical fitness is undoubtedly beneficial for everyone. Risk profiles for cognitive impairment are, however, highly divergent in elderly populations. High fitness can protect some older adults, whereas others may benefit more from healthy diet or participating in intellectual activities. Biological brain ageing is not tied to chronological brain ageing. Physical activity to increase cardiorespiratory fitness and muscle strength may be one way to slow down biological (brain) ageing and protect against cognitive impairment.

Carefully conducted measurements of cardiorespiratory fitness and muscle strength, which then can be correlated with longitudinal changes in brain volumes and cognitive function, is the most promising avenue for understanding for their age-related associations.
FUTURE IMPLICATIONS

The findings of this doctoral thesis suggest that possessing high cardiorespiratory fitness and muscle strength may protect against cognitive decline. However, evidence is insufficient to draw conclusions whether aerobic or strength training can prevent cognitive decline or dementia in older adults (253). A multidomain intervention comprising physical activity, diet, and cognitive training seems to have potential in preventing cognitive impairment. This approach will be tested in different populations and settings across the world in the initiative called World Wide Fingers (254). Identifying persons at risk for cognitive impairment and their various risk profiles is an important future goal. Notably, the negative effect of various risk factors like LDL cholesterol, homocysteine, hypertension, history of stroke, depressive symptoms, alcohol use and smoking on cognitive decline decreases with higher age (255). Promoting exercise, healthy dietary patterns and social activities in a population level is a challenge that extends also beyond the prevention of dementia.

When referring to CRF in this work, I have repeatedly used prefix “high” or “higher” to denote the difference compared to the individuals with low CRF. However, it is not clear where the line goes between low and high CRF when evaluating the effect of CRF on health outcomes (such as cognitive function). More evidence is required to identify the cutpoints or thresholds that determine low, moderate, and high CRF across age, sex, and race (247).

Activities that include physical activity and cognitive challenges at the same time deserve to be studied more. One such activity is dancing, as it involves sensory, motor and cognitive challenges simultaneously (256). In a recent study (257), six-month dance program compared to conventional fitness training led to larger volume increases in several brain areas. Both groups improved cognitive function, without a difference between groups. Because structural brain changes precede changes in cognition, whether dancing also improves cognition more than conventional fitness training during a time period longer than six months is under investigation (257).

High-intensity interval training (HIIT) is a time-efficient and effective strategy to reduce fat mass and blood pressure and increase CRF more than moderate-intensity continuous training in cardiac patients and in patients with type 2 diabetes (258-261). Preliminary evidence suggests that HIIT can also enhance cognitive function (262). However, the optimal exercise intensity in general and with respect to possible cognitive benefits is under continuous discussion. Based on existing evidence, it is not reasonable to recommend any intensity above another to protect against cognitive impairment. While finding one “universal” exercise dose would be ideal, the response to an exercise stimulus varies individually. The same exercise prescription probably will not be optimal for everyone.
In recent years, greater technological developments have introduced new methods that can also be exploited in the field of health science. One example is a 3-D virtual reality kayak program, which can improve the cognitive function, muscle strength, and balance of community-dwelling older adults (263). It is obvious that innovations like this should increasingly be put into practice.

The human gut microbiota has been estimated to contain ten times more bacterial cells than the number of human cells present in our bodies (264). Microbiota-gut-brain research is a fast-growing field of inquiry on how microbiota may influence the way humans think, perceive, and experience the environment (265). Recently, aerobic exercise has been associated with changes in gut microbiota composition independently of diet (266, 267). The effects of physical activity on gut microbiota should be, and most likely will be, studied more in the upcoming years.

Finally, it has been argued whether a combination of aerobic and resistance training would enhance cognition to a greater extent than either training mode alone (268). Given that the combined effects of higher muscular strength and cardiorespiratory fitness on decreasing all-cause mortality have been shown to be greater than their individual effects (269), future studies should strive to answer whether this is true also for cognitive function.
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As the late-life cognitive impairment and dementia are common challenges in our society, research identifying modifiable risk factors has become increasingly important. One such risk factor is low physical fitness. The present thesis focuses on cardiorespiratory fitness and its connections with brain volume and various domains of cognitive function in at risk older people from the general population. The thesis also explores the associations between muscle strength and cognitive function in a population-based sample of older adults with a focus on the methodology of measuring muscle strength.