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The Impact and Influence of Sports Related Concussion and Cardiovascular Risk Factors on Brain and Heart Health across an Ageing Population

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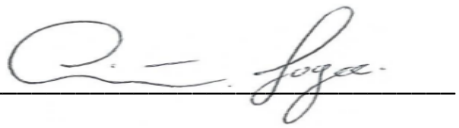
Signed 

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"*So many of our dreams seem impossible, then they seem improbable,*
and then, when we summon the will, they soon become inevitable."

- *Superman, DC Comics*

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Abstract

This study focused on the relationship between perceptual performance, brain health, and cardiovascular (CV) profiles in both recreational athletes and non-athletes, bridging the gap between sensory integration, cognitive function, and physical well-being. The study's primary aim was to explore brain health in young and middle-aged recreational athletes in diverse contact and non-contact sports via a neuropsychological measure of multisensory integration (MSI) known as the Sound-Induced Flash Illusion (SIFI) task. This task may be of use as an objective diagnostic test of sports-related concussion, thus history of concussion in all subjects was recorded. Additionally, the impact of exercise on SIFI performance and the test's reliability and overall level of agreement were examined since, if the test is to be of diagnostic use in a sports setting, performance must be resistant to exercise and the test must be robust and reliable across multiple testing sessions. Finally, the link between older community-level athletes' CV health profile and cognition in midlife was investigated alongside a systematic review of the literature investigating the influence of midlife CV risk factors on cognition across multiple domains.

Our results show that healthy young adults (aged 18-30) with or without a history of concussion showed comparable perceptual accuracy in the SIFI task across illusory conditions. While females exhibited greater susceptibility to the illusion, factors like sex and concussion history did not account for perceptual performance variance. Contact sports participants demonstrated heightened MSI, possibly due to training in open skill sports, but overall sporting cohort did not significantly affect perceptual differences. A follow-up study confirmed sex-based and sports-specific disparities in perceptual performance with open skill sport participants excelling in larger temporal asynchrony conditions, and a subgroup exhibiting a nuanced learning effect linked to cumulative SIFI exposure. The influence of moderate and high-intensity exercise on SIFI performance was negligible, supporting the hypothesis that the test may be of use in sport settings. Remarkably, the SIFI displayed strong reliability across multiple testing sessions, further supporting its utility in concussion diagnosis and prognosis.

A series of systematic reviews highlighted the complex relationship between midlife CV risk factors (hypertension, diabetes, cholesterol) and cognitive function, underscoring the need for targeted interventions at particular life stages in order to protect brain health in older age. Findings varied across different cognitive domains; notably, impairments in memory, executive function, and global cognition were correlated with poorer CV health. Longitudinal studies predominantly affirmed

an adverse link between midlife CV risk factors, specifically hypertension and type 2 diabetes (T2DM), and cognitive decline in later life. In contrast, the relationship with cholesterol was less clear, with some studies indicating positive associations while others identified negative links with cholesterol, high-density lipoproteins, low density lipoproteins, and triglycerides.

A study of SIFI performance, markers of CV health and history of concussion in middle-aged subjects with a history of recreational sporting activity revealed sex differences in BMI and health measures, with sporting engagement influencing blood pressure (BP) and mental health outcomes. A notable distinction emerged in sports participation; females engaged exclusively in non-contact sports, whereas males participated in both sporting types. Lifetime sporting involvement showed a positive correlation with systolic BP and depression, but conversely showed a negative correlation with overall physical activity and mental health metrics. Markedly, no significant correlation emerged between cognitive functioning and sports experience. Sex differences in global cognition were not significant, although females displayed an average score below the clinical cut-off, suggesting a potential increased risk of mild cognitive impairment. Susceptibility patterns to SIFI illusory conditions among older community athletes at midlife resembled those in our study of younger subjects, revealing high, moderate, and low performers. While sex did not significantly predict perceptual accuracy, males exhibited reduced susceptibility to the SIFI. Intriguingly, those who had participated in non-contact sports consistently outperformed contact sports across all SOA conditions, and despite differing outcomes from those in young participants, no significant distinctions emerged between open skill and closed skill sports.

Results included in this thesis have broad implications for athletes, coaches, healthcare professionals, policymakers, and the general public. As well as providing evidence to support the potential use of a simple but robust test of MSI in the diagnosis of concussion and monitoring of recovery, it also adds to our knowledge of the influence of CV wellbeing on brain health and function in middle age and later life. Future research should investigate the physiological mechanisms underlying the age-, sex-, disease- and exercise-related influences on cognitive function that we have observed and work towards development of tailored interventions to promote healthy aging for athletes and non-athletes alike. Ultimately, this research advances our understanding of the intricate relationships between cognitive function, brain health, and cardiovascular well-being, urging proactive measures to ensure the well-being of both athletes and the wider population.

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List of Abbreviations

AD	*Alzheimer's Disease*
AGE	*Advanced Glycation End-Products*
AHA	*American Heart Association*
ANX	*Anxiety*
APOE	*Apolipoprotein E*
AUDIT	*Alcohol Use Disorders Identification Test*
A-V	*Audio-Visual*
AXIS	*Appraisal Tool for Cross sectional Studies*
Aβ	*Amyloid-Beta*
BBB	*Blood Brain Barrier*
BDNF	*Brain-Derived Neurotrophic Factor*
BESS	*Balance-Error Scoring System*
BIC	*Bayesian Information Criterion*
BMI	*Body Mass Index*
BP	*Blood Pressure*
BPM	*Beats Per Minute*
BSI-18	*Brief Symptom Inventory*
CAC	*Coronary Artery Calcium*
CAD	*Coronary Artery Disease*
CBF	*Cerebral Blood Flow*
CC	*Corpus Callosum*
CI	*Confidence Interval*
CNS	*Central Nervous System*
CSP	*Cavum Septum Pellucidum*
CTE	*Chronic Traumatic Encephalopathy*
CTSIB	*Clinical Test of Sensory Integration and Balance*
CV	*Cardiovascular*
CVD	*Cardiovascular Disease*
CVR	*Cardiovascular Reactivity*
DAI	*Diffuse Axonal Injury*

DBP	*Diastolic Blood Pressure*
DDR	*DNA Damage Response*
DEP	*Depression*
DM	*Diabetes Mellitus*
DNA	*Deoxyribose Nucleic Acid*
DTI	*Diffuse Tensor Imaging*
EEG	*Electroencephalography*
EHCT	*Eye–Hand Coordination Test*
EICR	*Exercise-Induced Cardiac Remodeling*
ESC	*European Society of Cardiology*
FBG	*Fasting Blood Glucose*
FI	*Fatigue Index*
fMRI	*Functional Magnetic Resonance Imaging*
fNIRS	*Functional Near-Infrared Spectroscopy*
GABA	*Gamma-Aminobutyric Acid*
GAD-7	*Generalised Anxiety Disorder Assessment*
GFAP	*Glial fibrillary acidic protei*
GM	*Grey Matter*
GSI	*Global Symptom Index*
HbA1c	*Hemoglobin A1c*
HDL-C	*High-Density Lipoprotein*
HIA	*Head Injury Assessment*
HIEE	*Head Impact Exposure Estimate*
HR	*Heart Rate*
HRQoL	*Health-Related Quality of Life*
ICC	*Intra-Class Correlation Coefficient*
ICC	*and Intra-Class Correlation Coefficient*
ImPACT	*Immediate Post-Concussion Assessment and Cognitive Testing*
IPAQ	*International Physical Activity Questionnaire*
IPS	*Intraparietal Sulcus*
IQR	*Inter-Quartile Range*
IQR	*Inter-Quartile Range*

KD	*King-Devick test*
LCA	*Latent Class Analysis*
LCA	*Latent Class Analysis*
LDL-C	*Low-Density Lipoprotein*
LOC	*Loss of Consciousness*
LV	*Left Ventricular*
LVH	*Left Ventricular Hypertrophy*
MCI	*Mild Cognitive Impairment*
MCI	*Mild Cognitive Impairment*
MD	*Mean Difference*
MEG	*Magnetoencephalography*
MET	*Metabolic Equivalent*
MetS	*Metabolic Syndrome*
MoCA	*Montreal Cognitive Assessment*
MPO	*Mean Power Output*
MRI	*Magnetic Resonance Imagine*
MSI	*Multisensory Integration*
mTBI	*Mild Traumatic Brain Injury*
MTBIIM	*Michigan Traumatic Brain Injury Identification Method*
MULES	*Mobile Universal Lexicon Evaluation System*
NAA	*N-acetylaspartate*
NADPH	*Nicotinamide adenine dinucleotide phosphate*
NFL	*National Football League*
NMDA	*N-methyl-D-aspartate*
NSS	*Nike Sensory Station*
NVC	*Neurovascular Coupling*
PA	*Physical Activity*
PARQ	*Physical Activity Readiness Questionnaire*
PCS	*Post-concussion syndrome*
PHQ-9	*Patient Health Questionnaire*
POMS	*Profile of Mood States*
PPO	*Peak Power Output*

PPSC	*Pre-Participation Cardiovascular Screening*
PRISMA	*Preferred Reporting Items for Systematic Reviews and Meta-Analyses*
PSQI	*Pittsburgh Sleep Quality Index*
PSS	*Point of Subjective Simultaneity*
RAGE	*Receptors for Advanced Glycation End-Products*
RHI	*Repetitive Head Impacts*
RLD	*Representative Learning Design*
ROS	*Reactive Oxygen Species*
RPE	*Rate of Perceived Exertion*
RPM	*Revolutions Per Minute*
RTL	*Return to Learn*
RTP	*Return to Play*
RV	*Right Ventricular*
SBP	*Systolic Blood Pressure*
SC	*Superior Colliculus*
SCAT-5	*Sports Concussion Assessment Tool Version-5*
SD	*Standard Deviation*
Sdec	*Decrement Score*
SE	*Standard Error*
SF-12	*12-Item Short Form Survey*
SIFI	*Sound Induced Flash Illusion Test*
SMD	*Standardised Mean Difference*
SOA	*Stimulus Onset Asynchrony*
SOM	*Somatisation*
SRC	*Sports Related Concussion*
STC	*Superior Temporal Colliculus*
STG	*Superior Temporal Gyrus*
STROBE	*Strengthening the Reporting of Observational Studies in Epidemiology*
STS	*Superior Temporal Sulcus*
T2DM	*Type 2 Diabetes Mellitus*
TBI	*Traumatic Brain Injury*
TBW	*Temporal Binding Window*

TC	*Total Cholesterol*
TEM	*Technical Error of Measurement*
TG	*Triglyceride*
TTP	*Time to Peak Power*
UCHL-1	*Ubiquitin C-terminal hydrolase L1*
V1	*Primary Visual Cortex*
VTS	*Vienna Test System*
WHO	*World Health Organisation*
WM	*White Matter*
WMH	*White Matter Hyperintensities*

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Chapter One:

Introduction

1.1. Concussion in Sport

1.1.1. Overview

Currently, there is no standardised definition of concussion, despite its Latin roots of 'to strike together' or 'to shake violently'. The Concussion in Sport Group has classified a concussion as "a complex pathophysiological process affecting the brain, induced by biomechanical forces" (1, 2). Defining concussion has been a cause of debate since the conception of the first working definition in 2001. Different definitions exist in the literature and have led to diagnostic and classification bias (2). The interchangeable use of concussion with mild traumatic brain injury (mTBI) has contributed to the controversy and uncertainty surrounding the definition of concussion (1, 2). This lack of consensus has resulted in various definitions being used, or no definition being provided at all, in some studies (3). While traumatic brain injury (TBI) is classified as mild, moderate, or severe, concussion falls on the lower end of the spectrum and is considered a heterogeneous condition with a range of clinical profiles and phenotypes.

The threshold of impact that causes a concussion is dynamic rather than static in nature, meaning that a blow of a certain magnitude may or may not result in a concussion on different occasions (4). This highlights the potential long-term neurodegenerative effects of repetitive sub-concussive head impacts (5). Despite increased awareness of the potential consequences of a concussion, most cases go undiagnosed or are underreported due to various factors (6-8). Recently, there have been calls to change the foundation of Rugby Union to prioritise player welfare and future brain health. Dr. Stewart is a highly respected Neuropathologist at Queen Elizabeth University Hospital in Glasgow and holds Associate Professor positions at the University of Glasgow and the University of Pennsylvania. He leads a renowned research lab that investigates traumatic brain injury pathologies, focusing on their link to neurodegenerative diseases. His work is based on the comprehensive Glasgow TBI Archive and includes directing the FIELD study, examining the lifelong health and dementia risk in former soccer players, and co-leading the CONNECT-TBI research program. He has proposed ending contact training, enforcing zero tolerance for high tackles and excessive clear-outs at the breakdown, and mandating brain scans for all players during pre-season (5). Dr. Stewart advocates for changes to be made similar to the Scottish football ban on heading during training, which would provide players, coaches, and parents with the comfort of knowing that player health and welfare are at the forefront of governing sporting bodies.

Scientists and retired athletes are currently concerned for the future health and quality of life of current and former rugby players, with up to 90% of athletes lacking the knowledge to recognise concussions and receive proper care (9). A lawsuit has been filed by former players over the inability and *"failure to protect (the claimants) from the risks caused by concussion" (10)*. The potentially dangerous impact of rugby on players' health is exemplified by the case of World-Cup winner Steve Thompson, a former rugby union England hooker, who has been diagnosed with early onset dementia and probable chronic traumatic encephalopathy (CTE), likely due to repeated head injuries sustained during his career (9).

> "*It was not uncommon for me to be left dazed, seeing white spots, and not knowing where I was for a few seconds, sometimes I would pass out completely. It was just an accepted part and parcel of training."*
>
> – Steve Thompson, Former Professional Rugby Player

1.1.2. Incidence Rates

Over the past century, the incidence of concussions has significantly risen, and in the United States alone, there are approximately 1.6 to 3.8 million TBI's occurring each year. (11). Sports-related concussion (SRC) is most prevalent in rugby and American football, with overall incidence rates of 0.1 to 21.5 per 1,000 athletic exposures and 2.2 injuries per American football game (12, 13). Rugby union has been found to present with one of the highest concussion rates among all recognised contact sports with estimates reaching as high as 13.4 concussions per 1,000 contact hours (14, 15). Variations in terminology, underreporting of symptoms, and methodological discrepancies all contribute to the difficulty of obtaining reliable incidence rates of concussion across age groups and sports. Improved awareness among athletes and their support staff has contributed to increased reporting of concussions. However, many misconceptions still exist about concussion, such as the belief that loss of consciousness (LOC) is required for a diagnosis, despite LOC occurring in less than 10% of cases (16).

Various factors contribute to the clinical risk score for concussion, such as the type of sport (contact vs non-contact), age above 13 years, female sex, and prior history of concussion. Women have a higher incidence of concussion, twice that compared to men, and they also take

longer to recover, possibly due to hormonal changes occurring during different phases of the menstrual cycle (17). Weaker neck musculature in women, which reduces their ability to stabilise their head at the point of contact, can lead to more severe injuries and prolonged recovery. However, the higher reported incidence could also be because women tend to report symptoms more accurately. Women also have a higher symptom burden than men, a higher percentage report of headaches, dizziness, and confusion, and take significantly longer to fully recover cognitively (18, 19). Furthermore, athletes with a history of concussion have a x20-times increased likelihood of reporting a new concussion (20). All these factors can have a significant effect on the rates of reporting across all sports.

Retrospective self-report is frequently used to ascertain concussion history but can be inaccurate due to the subjective nature of the reporting. Low agreement exists between retrospective athlete-recall and clinically recorded concussion history. The most common explanation for discrepancies is that athletes were not aware they had a concussion, accounting for 73% of variance (21). Recently, the underreporting of concussion history has been shown to be underestimated among professional rugby players (22). The Michigan Traumatic Brain Injury Identification Method (MTBIIM) has shown fair agreement between self-reported and medically diagnosed concussion history in rugby players. Medically diagnosed concussions were 30% higher per player than self-reported concussions across 62 professional rugby players experiencing 99 rugby-related concussions (22).

1.1.3. Pathophysiology

1.1.3a Metabolic Cascade of Concussion

Concussion as a sub-category of TBI can be categorised into two types: focal and diffuse. Focal injuries are associated with moderate-to-severe TBI caused by direct impact to brain tissue, while diffuse injuries are linked to mTBI such as concussion with no direct impact to the brain. The main form of diffuse injury is diffuse axonal injury (DAI), caused by acceleration and rotational forces leading to dysfunctional contributions to neuronal axon transport systems (23, 24). DAI can cause secondary injuries including cerebral edema (25); it is considered the main pathology of concussion and historically, has been seen as pathological evidence for damage to white matter (WM) tracts. DAI is difficult to detect on standard head imaging scans and is a "stealth pathology" (25). Following a concussion, a neuro-metabolic cascade occurs, inducing ionic channel dysfunction, uncontrollable ionic flux, and a state of hyperglycolysis due to altered glucose metabolism. A state of impaired metabolism presents over the next 7-10

days due to altered glucose metabolism, coinciding with the standard symptom resolution time frame (23, 24, 26). Physiological disturbances can extend beyond the clinical recovery timeframe, leading to cellular and neuronal degeneration, and axonal separation from their cell bodies (27, 28).

1.1.3b Signs & Symptoms

Concussion and mTBI is considered a functional rather than a structural injury in the acute phase (<10 days) with no direct pathological correlate (2). It is characterised by a range of heterogeneous and short-lived symptoms, which can be categorised into four distinct groups: physical, emotional/behavioural, cognitive, and sleep disturbances, see **Table 1.1**. Headache is the most frequently reported symptom, followed by fatigue and dizziness. Symptoms typically resolve within 7-10 days, with an average duration of 3.5 days (2). When it comes to diagnosing a concussion, the most crucial factor is reporting symptoms promptly. If symptoms are reported more than 7 days after a suspected concussion, it increases the chances of a prolonged recovery five-fold, regardless of injury details and known risk factors (29). Signs and symptoms of concussion depend on the location of the injury within or outside the brain, as well as the specific brain regions affected. Certain symptoms such as visual disturbances may be useful indicators for concussion diagnosis, while others may overlap with other conditions and be non-specific to concussion alone (30).

Table 1.1. *List of Common Symptoms associated with mTBI.*

Physical/Somatic	Affective	Cognitive	Sleep Dysregulation
Headache	Mood disruption	Confusion	Drowsiness
Nausea	Irritability	Difficulty concentrating	Fatigue
Vomiting	Emotional lability	Memory deficits	Hypersomnia
Balance problems	Sadness/Depression	Feeling mentally 'foggy'	
Dizziness	Anxiety	Feeling slowed down	Hyposomnia
Decreased playing ability	Inappropriate affect	Slow reaction times	Difficulty falling asleep
Visual disturbances	Emotional outbursts	Cognitive fatigue	
Photophobia	Spontaneous crying	Memory deficits	
Phonophobia	Panic	Loss of focus	
Loss of balance		Difficulty multitasking or completing mental tasks	
Poor coordination		Forgetful of recent information and conversations	
Numbness/Tingling		Repeating questions	
Dazed		Answering questions slowly	
Stunned		Confused about recent events	

***Note:** Adapted from the work of Harmon and colleagues (31).*

Self-reported symptom scores positively correlate with the cumulative number of concussions and self-reported changes in cognition over time (32), but reliance on self-reporting may be confounded by asymptomatic athletes engaging in exercise and cognitively demanding tasks, leading to symptom re-emergence. To address this, standardised and graded exercise protocols have been established for both children and adults (26, 33), with incremental increases in physical and cognitive load below symptom exacerbation threshold before full return to play (RTP) (26). For example, the Buffalo Concussion Treadmill Test was developed to safely and reliably diagnose physiological dysfunction to differentiate from other possible diagnoses (i.e., depression, migraines, cervical injury) (34). Derived from the Balke cardiac treadmill test, the protocol begins at a speed of 3.6 mph at 0% incline gradually increasing at 1% per minute until either maximum incline is reached or the participant can longer continue exercise engagement. If patients can engage in exhaustive exercise without experiencing a recurrence or worsening of headache or other concussion symptoms, and they display a normal physiological response to exercise, then we infer that the symptoms are not caused by a physiological concussion but by another issue. However, cognitive and physical exertion testing identified decrements in cognition in recently concussed athletes eligible to RTP, and high-intensity exercise was associated with an increased symptom burden (35, 36). Recent studies have investigated the efficacy of aerobic exercise programs for acute SRC rehabilitation in athletes, showing increased attention and reduced total symptom scores in participants (37). Moreover, physical exercise assessments can help identify athletes with underlying cognitive deficits following concussion given that 10% of athletes who passed cognitive assessment at rest, failed when engaged in exercise (38).

1.1.3c Post-Concussion Syndrome

The recovery time for concussions varies widely among individuals, with most cases resolving within two weeks, although some may persist beyond this timeframe, leading to post-concussion syndrome (PCS) (25). PCS is characterised by short-lived cognitive and physiological impairments that are difficult to monitor objectively over a prolonged period with reductions in executive function and working memory (39, 40). Cognitive symptoms may persist for up to three months after injury, with deficits in attention, executive function, and processing capabilities commonly observed. PCS is characterised by persistent and underlying physiological dysfunction including altered autonomic function and diminished autoregulation and perfusion of cerebral blood flow (CBF), caused not by a single pathophysiological dysfunction but an associated conglomerate of non-specific symptoms (26, 41). While clinical

recovery may occur early post-injury, neural injury may remain in some cases in the form of damaged white and grey matter fibre tracts (42). There is evidence of significant cognitive and functional impairment in the acute stages following a concussion that will dissipate over time (43). However, it is reported that visual deficits in both central and peripheral visual reaction time can persist up to 11 months post injury (44), which may result in more player-to-player contact and incur more concussions in sports such as soccer (17, 45). It is important to acknowledge such factors and develop individualised RTP and return to learn (RTL) protocols for each athlete.

1.1.4. Concussion Across the Lifespan

TBI resulting from repetitive head impacts (RHI) during contact sports, such as American Football, has garnered significant concern due to its potential short- and long-term effects on both athletic and non-athletic populations (46). Many studies have documented that engaging in contact sports at a young age can result in substantial long-term structural and functional alterations which can negatively impact cognitive function (47-49). Specifically, disrupted myelination resulting from mTBI in childhood has been linked to delayed or interrupted cerebral development (50). Further, a correlation exists between age of first exposure to RHI and altered microstructural integrity of the corpus callosum (CC), which can impair the developmental process of the anterior CC and result in cognitive and behavioural symptoms during midlife (50, 51). In 2009, the Centre for Disease Control reported that approximately 248,418 children under the age of 19 had been treated for sports-related concussions in the emergency department (52). While most children recover from concussion within 2-4 weeks, up to 30% experience prolonged symptoms (53, 54). Given that 1 in 5 adolescents has experienced a concussion, the developing brain's susceptibility to injury is mainly due to the ongoing myelination of WM fibre tracts, which are more prone to injury and recover more slowly than myelinated fibres, accounting for differences in concussion tolerance. Various regions of the brain such as the pre-frontal cortex, known to be a primary impact centre for the effects of concussion is among the last structures for regional cerebral development, which may hinder accurate representations of saccadic eye movement, inhibitory control, and temporal integration. WM tracts and volume of the pre-frontal cortex will progressively increase through adolescence into adulthood, with completion of myelination of cortico-cortical tracts accomplished by the third decade of life (55). These deficits can have major implications in sports, as athletes may struggle to communicate and identify behaviours from teammates and opponents, leading to reduced sporting performance due to reduced

multisensory integration (MSI). No single sensory or auditory/visual function can therefore act as a universal biomarker for concussion.

Concussion was first described in Homer's Iliad as a "weakness of the knees and clouded vision" which would quickly dissipate and enable soldiers to return to battle. These warriors of old can be viewed as akin to the sporting athletes of the modern world. Concerns about the long-term effects of repeated head trauma date back to the early 20th century, with early reports suggesting a link between head injuries and long-lasting weakness in the brain (56, 57). This was later supported in 1969 by a detailed report on retired professional boxers, which provided valuable insights into the clinical neurological features associated with boxing-related brain injury (58). There is concern that RHI may exacerbate the natural cognitive decline that occurs as early as 20-years of age until an acceleration around the age of 60 (59), leading to reorganisation of functional brain networks and recruitment of additional neural resources to counteract injury-induced neurodegeneration (60-62). A growing body of literature has emerged and continues to grow regarding the neurodegenerative capacity and neuropsychological decline associated with a lifetime exposure to RHI and concussion in sport (3, 63-65).

At the turn of the 21st century the first pathological signs of a neurodegenerative disease among retired sporting athletes was reported by autopsy (66). Chronic Traumatic Encephalopathy (CTE) is a progressive neurodegenerative disease characterised by abnormal tau deposition rates in neuronal and glial cells resulting from RHI's (67, 68). CTE encompasses a clinical spectrum of motor, cognitive and psychological symptoms, with the proposed clinical features in high congruency to cognitive decline and neuropsychiatric presentation of Alzheimer's Disease (AD), with an overlap of up to 25% in some cases (69), and appearing in midlife (70). Therefore, the neuropathology of CTE may not be as unique as once thought as it has been found among those with no known history of RHI (71-74). CTE may be considered a polypathology, and that an individual may have a neurodegenerative disease with minor amounts of CTE neuropathology present as is commonly seen in those with AD (75). The supposed clinical manifestation of CTE is heterogenous and two-fold with behavioural changes appearing around the third and fourth decade in life and cognitive deficits and deterioration progressing to dementia in the sixth decade of life (70, 76, 77). One third of individuals present with symptomatic features of CTE upon their retirement from sport and half exhibiting signs of CTE within their first 4 years of their athletic retirement (78). Currently, there is no objective and diagnostic tool to diagnose CTE during life. However, recent studies used machine learning of neurophysiological indices, such as the P300, to analyse data at the individual level in retired

athletes with a history of concussion. A study on retired Canadian Football League athletes achieved an 81% accuracy rate in detecting past concussion history using this approach (79). These findings suggest that the effects of concussion can be detectable at the individual level long after the last concussion occurred, indicating potential adverse effects on the individual and the valid implementation of new assessments in sports and brain health.

1.1.5. Difficulty in Diagnosis

Diagnosis of concussion is made based on clinical jurisdiction, relying on reported mechanism of injury, signs, symptoms, and high levels of suspicion (3, 80). To address the diverse nature of SRC, international consensus statements have recommended a comprehensive, multimodal approach for management. Following the National Institute of Health (NIH) criteria, this approach encompasses evaluating symptoms, cognitive function, vestibular/oculomotor abilities, near point of convergence, and balance to enhance diagnostic accuracy. However, the on-field assessment is confounded by subjectivity, and pressure to continue playing (81). Assessments like the King-Devick test (KD), Mobile Universal Lexicon Evaluation System (MULES), Balance-Error Scoring System (BESS), SCAT5, and Immediate Post-Concussion Assessment and Cognitive Testing (ImPACT) have been developed to identify concussions early, but there are advantages and disadvantages (82). Symptom profiles have been used to group similar post-concussion symptoms into corresponding clinical subtypes, enabling clinicians to provide targeted treatment, but it lacks a definitive numerical value. With the increasing number of assessment tools devised for SRC diagnosis and prognosis, the standardisation of the procedural diagnosis becomes challenging to say the least. Identifying the most reliable tools in a multi-dimensional assessment for distinguishing athletes with sports-related concussions from healthy ones is crucial for clinicians. These tools help clinicians make informed decisions about diagnosis and the return-to-play process after a potential injury, based on functional outcomes.

A sideline and clinical assessment tool for concussion should be effective, rapid, and objective to help inform clinical decisions and protect the health and welfare of the athlete. The current RTP and RTL guidelines, based on self-reported symptoms, are unreliable due to subjectivity and incentives for athletes to continue playing. Certain athletes may underreport or forego reporting the severity of their symptoms in order to continue training and play in the tournament which is incentivised by the fact that playing sport is their livelihood (31). Performance measures possessing adequate sensitivity and sufficient specificity alongside

established markers of concussion are essential to support what remains a clinical diagnosis of concussion The introduction of blood-based biomarkers, such as Ubiquitin C-terminal hydrolase L1 (UCHL-1) and Glial fibrillary acidic protein (GFAP), as a prognostic aid has shown promise and was approved by the FDA (83, 84). These biomarkers aim to reduce unnecessary brain scans, radiation exposure rates, and economic costs. However, difficulties exist in the biomarkers themselves, such as proteolytic degradation, binding to carrier proteins, cerebral-derived protein dilution and their removal from the blood via hepatic metabolism and kidney excretion (68). An objective tool is necessary due to the heterogeneity of clinical aspects of concussion, and biomarkers can aid in diagnosis and prognosis (85).

Cognitive testing has been recognised for its ability to detect impairments or alterations following a concussion, leading to the development of several commercially available computerised test batteries that are rooted in traditional neuropsychological measures. Proponents of computerised test batteries argue that they offer numerous advantages over traditional pencil-and-paper tests, including sequential administration through alternative versions, enhanced measurement of response time, ease of administration, and minimal time demands for the testing administrator. Computer-based cognitive testing has thus become more available and standard practice in the diagnosis and prognosis of SRC at all levels of sport (86). Literature ultimately necessitates an objective tool which considers underlying physiological function.

1.1.6. Concussion Management and Physical Exercise

The management of concussion requires a graded RTL and RTP approach, as abnormalities in functional brain networks and cerebral microstructure can persist even after symptoms have resolved (87). However, simplistic cognitive tasks are not sensitive enough to detect persistent deficits beyond the acute phase of recovery (88), and early involvement in exercise may exacerbate symptomology and postpone full recovery. Aerobic exercise has the potential to decrease concussion-related physiological abnormalities such as altered autonomic function, diminished autoregulation and perfusion of CBF (89, 90), and potentially initiate neuroplasticity via BDNF leading to behavioural change (91). But the timing of exercise must be carefully managed (90, 92). In a study comparing aerobic exercise to low-impact stretching, individuals with PCS who underwent aerobic exercise showed improved brain activation patterns, while those who underwent stretching remained abnormal in several brain regions such as the posterior cingulate gyrus, lingual gyrus, and cerebellum (93). PCS is a heterogenous

condition, with each individual expressing a unique pattern of CBF and performance decrements. Concussion and PCS can give rise to an asynchrony between the autonomic nervous system and cardiovascular (CV) function with the clinical presentation of impaired CBF and altered heart rate (HR) variability. The link between concussion, cognition, and exercise is evident and an important triad to consider in future research regarding the assessment of brain health of current and retired athletes in both diagnosis and prognosis utility.

1.2. Sound-induced Flash Illusion (SIFI) Test

1.2.1. The SIFI Paradigm and Methodology

A large amount of research has studied the senses in isolation predominantly due to a lack of understanding of the mechanisms governing integration of different sense modalities, known as multisensory integration (MSI), and available assessment tools. A larger quantity of more recent research investigated multiple sensory streams simultaneously using A-V illusion paradigms to better understand environmental and sensory impressions (94-96). The SIFI occurs when a single visual flash is presented with two or more auditory tones, resulting in the perception of two or more flashes (96-98). The fission and fusion illusions have been used to measure perceptual efficiency, with the former causing an overestimation of flashes and the latter causing an under-reporting of presented flashes (99). Susceptibility to the illusion is dependent on the *stimulus onset asynchrony* (SOA) between the auditory and visual stimuli, with reduced susceptibility reported as the time delay between stimuli increases (100-102). Common metrics of performance capabilities during the SIFI task vary across studies depending on cohort and experimental design but predominantly include: proportion/mean percentage of correct responses; i.e., accuracy in the detecting the presence or absence of the target stimuli be it visual or auditory (101-104), fission and fusion illusion indices (105-107), reaction time (108, 109), signal detection theory as a measure of sensitivity *(d')* (94, 108, 110), criterion bias forming the likelihood ratio (111-113), and individual study experimental measures such as visual temporal discrimination, auditory temporal discrimination, visual gain (114), and the size of the temporal binding window (TBW) (115, 116).

The term 'prior entry' was first introduced by Titchener in 1908 and refers to the phenomenon that the visual perceptual system draws attention to visual stimuli before neglected or solitary stimuli in the individual's perceptual field (117). This processing mechanism results in a quicker processing of attended stimuli relative to unattended stimuli, as seen in the SIFI test where the visual stimulus is attended to, and the auditory beep is

ignored. The SIFI test is unique in its assessment of MSI due to the dichotomous fission and fusion illusions. Studies have varied stimulus intensity to modulate the "*point of subjective simultaneity*" (PSS), which describes the temporal asynchrony at which the unification of sensory information peaks. Decreases in stimulus intensity cause a shift in the PSS to a broader TBW where the SOA between the visual and second auditory stimulus is larger (118, 119). There are however concerns regarding the standardisation of the SIFI methodology and outcome measures used for MSI which need to be addressed “in order to select an optimal protocol and strive for harmonisation across research” (120).

The auditory stimuli used in SIFI experiments have shown variability in their sound pressure level, frequency, and duration across different studies. The original work by Shams et al. used a sound pressure level of 95 dB (96, 97) , while subsequent studies have reported levels ranging from 48-83 dB (113, 114, 121). The characteristic sine wave function at 3.5 kHz has been commonly used (94, 95, 105), but lower frequencies have also been reported (111, 122, 123). The duration of the auditory tone ranges from 7-20 ms (113, 121, 124). Over the past two decades, there has similarly been considerable variation in the visual stimuli used in the SIFI literature. The most common form of visual stimulus is a uniformly illuminated white disk presented at varying subtending visual angles and distances from the central fixation cross on a black or grey background (125-127). However, some researchers have used alternative stimuli, including squares, coloured disks, and flashes presented in rings (110, 113, 128, 129). This may be significant regarding performance in mTBI patients it has been suggested that those suffering from a head injury may present with colour vision deficits assessed using a primary colour discrimination task and event-related potentials, however, a specific colour deficit was not reported (130). Stimulus durations have also varied widely, ranging from as short as 10 ms to as long as 30 ms (103, 105, 131, 132), with some stimuli presented laterally rather than below the fixation cross (112, 133, 134). The luminance of the visual stimuli has also varied widely, with some studies using low luminance levels and others using high luminance levels (103, 135-137), with a middle-ground luminance of ~100-120 cd/m^2 (107, 125, 138). Studies have shown that the luminance of the visual stimuli can modulate the percept of the SIFI, with higher luminance values corresponding to a stronger perception of the illusion (113). LEDs of varying colours have also been used to present the visual flash (139-142). In the context of concussion and mTBI, the effect of luminance on neurosensory processing may be a crucial component as an objective biomarker of concussion, and an optimal luminance level may aid in the rapid and objective identification of a concussion (143-145).

The temporal parameters of the stimuli are crucial, especially the SOAs between the A-V pair and the secondary auditory beep in multisensory illusory conditions. The range of SOAs used in research is substantial and may account for differences in susceptibility to multisensory illusory conditions (94, 111). Studies have shown that illusion susceptibility decreases as the SOA increases. When the SOA is short, MSI occurs during the TBW and is determined based on perceptual response to perceiving the flash and beep at the same time or at different times (95, 112, 128, 146). In a study assessing the strength of the SIFI, the TBW of 31 college students was established by presenting a single flash with 0-4 beeps alongside control conditions. Two forms of TBW were established, a left TBW for audio-first presentation and a right TBW for visual-first presentation (128). It was found that the right TBW was significantly larger than the left TBW, and the strength of the multisensory illusion decreased with a narrower TBW. The authors suggest that people with narrower TBWs statistically encounter fewer occurrences of stimulus inputs perceived to be temporally synchronous, leading to greater perceptual binding of synchronous inputs and increased ability to dissociate asynchronous inputs. The "unity effect" or "assumption" is an important consideration when interpreting MSI results. Early work showed that subjects' beliefs about the relationship between different sensory cues can be manipulated by instructions given by the researcher, which can influence their perception. This effect has informed the methodology of the present doctoral work on the SIFI illusion, where verbal instructions were carefully chosen to promote MSI and eliminate observer bias. Recent work has also highlighted the potential for perceptual bias due to sound localisation, mediated by the posterior middle temporal gyrus and the superior parietal cortex. Proper control of modulatory effects and procedural commands is crucial for accurate MSI results (147, 148).

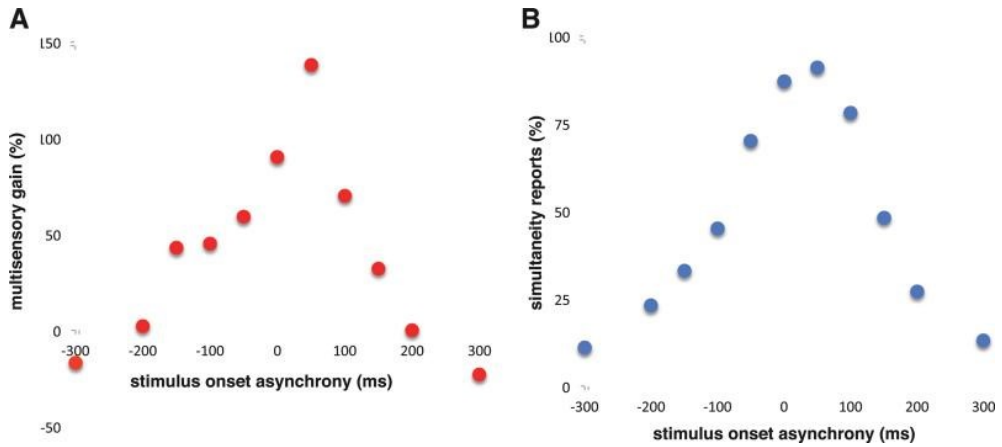

Figure 1.1. *Sensory-temporal patterns that are representative of both neural activity and perception.* **(A)** The graph displays the multisensory interactive gain of a cat superior colliculus neuron in response to audiovisual stimuli, plotted against the stimulus onset asynchrony between the two types of stimuli. This represents the temporal tuning function of the neuron. **(B)** The graph shows the results of a simultaneity judgment task performed by a human subject. The y-axis represents the percentage of responses indicating simultaneous perception, while the x-axis represents the stimulus onset asynchrony. **Adapted from the work of Wallace & Stevenson (149).*

1.2.2. SIFI Illusion and Functional Neural Mechanisms

In spite of optimal processing and integration of A-V environmental information, misleading or inaccurate representations can occur, referred to as "*environmental artefacts*" (150). The SIFI percept is only detectable above a certain threshold with SOA's up to 115 ms, with the STG and oscillatory activity playing a critical role in SIFI susceptibility and A-V integration (123, 151, 152). Recent research emphasises the importance of accounting for temporal feedback delays in MSI when applying the SIFI, due to sensory weighting where the faster sensory modality holds dominant weighting in integration (153). The temporal domains of perception in one modality are impacted by stimuli in another modality, such as the SOA associated with auditory beep presentation affecting the perception of visual flashes in the SIFI (154).

In the context of sports, optimal performance is achieved through Bayesian inference by assigning weighting to sensory stimuli through MSI and making decisions based on the integrated sensory signals (98, 155, 156). Athletes for example fine-tune their nervous system to know when and how to combine auditory and visual information to achieve statistical optimality (98). The Bayesian observer follows an inference rule based on Bayes' theorem to calculate the causes of sensory signals.

$$P(Z_A, Z_V|A, V) = \frac{P(A|Z_A)P(V|Z_V)P(Z_A, Z_V)}{P(A, V)}.$$

The ideal Bayesian observer model assumes *"that the posterior probability of events Z_A and Z_V is the normalized product of the single-modality likelihoods $P(_A|Z_A)$ and $P(_V|Z_V)$ and joint priors $P(Z_A,Z_V)$"* (98). A study of 10 participants showed a high level of consistency (r^2 = 0.92) between human perception and the ideal observer across all SIFI conditions (98). During the SIFI, the reliable auditory cue can modulate the observer's visual perception, resulting in partial combination facilitating the illusion, whereas vastly different A-V stimuli lead to reduced overlap and a lower likelihood of MSI.

1.2.3. Factors Affecting the Perceptual Illusion

1.2.3a Exercise

Exercise has been shown to modulate cognitive and perceptual abilities both in the short and long term, with varying effects on task performance. Studies have suggested that exercise can improve the sensitivity and flexibility of the human sensory systems via central nervous system (CNS) arousal, which can be useful in sporting events and performance (157). In a study of older adults (n = 58), a single bout of open skill exercise (tennis, aerobics or dance; n = 18) or closed skill exercise (swimming or gym circuits; n = 19) was assessed using the SIFI in conjunction with a working memory task (158). The open skill group and control group had higher levels of performance accuracy on congruent trials, i.e., those trials where A-V stimuli are presented in synchrony, than the closed skill group. The open skill group also significantly increased their perceptual accuracy from baseline to post exercise engagement, whereas the closed skill and control groups had worse sensitivity measures after exercise. The authors suggest that habitual physical activity (PA) is not the main factor in perceptual improvements, but that exercise intensity and modality may play a role. O'Brien and colleagues' (158) experimental design investigating the effects of exercise on SIFI performance in older adults was replicated with children aged 6-8 years (159). Fifty-one children were divided into open skill (basketball, soccer, tennis), closed skill (running, circuit training, skipping), and control (homework revision) groups with exercise intensity determined by heart rate (HR) pre- and post-exercise. The SIFI test consisted of unisensory and multisensory conditions with SOAs of 70, 110, 150, and 230ms. Perceptual sensitivity significantly increased in both exercise cohorts but not in the control cohort from pre- to post-exercise, and the authors accounted for higher

levels of performance in the open skill exercise due to alignment with the cognitive engagement hypothesis where the exercise modality would have the most cognitive reward due to WM activation (160).

1.2.3b Sensory Dominance

In sports, task-related factors such as whether participants are instructed to detect or discriminate task-relevant stimuli, or whether they have a visual or auditory bias, can influence sensory dominance and the interpretation of the SIFI illusion. Similar to professional musicians, athletes with developed expertise may exhibit enhanced spatial reasoning and working memory of temporal and spatial stimuli characteristics, which could explain differences during general cognition and sensory motor tasks (161-165). Attention and anticipation may also play a role mediating this sports-cognition relationship with a translational capacity to SIFI performance metrics with a dynamic interplay between perceptual cognitive processes that allow more efficient and apt decisions (166-168). The Dual-mode model proposes that exercise below individual ventilatory threshold sustains functional activation of the prefrontal cortex, which may improve sensory perceptual accuracy during the SIFI, whereas exercise above the ventilatory threshold may interfere with executive function and subcortical regions associated with sensory perception (169). This proposed theoretical framework may explain potential variance exhibited in sensory perceptual accuracy during the SIFI in the presence of exercise.

1.2.3c Ageing

Numerous studies have investigated MSI and susceptibility to the SIFI across different ages and cognitive maturities. In a recent investigation of children and adolescents compared to adults, perceptual accuracy was reduced overall for the fission and fusion illusions, and children showed significantly lower accuracy for fission trials than adults (121). The development of MSI across children has also been studied using the SIFI to determine whether auditory dominance persists beyond four years of age and when visual dominance starts to emerge. Results showed an overestimation of the number of flashes during the fission and fusion trials, with a decrease in overall error as age increased (170). The youngest children were found to be the most sensitive to task-irrelevant auditory tones during the A-V multisensory trials compared to older children and adults. Using short and long blocks of unimodal and cross-modal trials to measure the developmental trajectory of MSI across different age groups, younger adults were better at perceiving fewer illusions than elderly

participants, with a significant difference in performance accuracy in the +70ms condition (171). Another study enrolled 3,955 participants to investigate the effects of aging on MSI and SIFI susceptibility, finding that age was a significant factor in predicting SIFI susceptibility and governing MSI through an expanded TBW, with males having a reduced illusory susceptibility to the 2 beeps-1 flash illusion in comparison to females (95). Sensitivity to the illusion decreased as the number of SOAs increased, with younger adults exhibiting a shorter TBW than the elderly. These findings highlight the importance of considering age and cognitive development in MSI research using the SIFI.

Moreover, a recent study investigated the utility of the SIFI for assessing SRC and brain health. Retired professional rugby union players had lower correct response rates for illusory fission trials than non-contact athletes, with reduced ability to achieve correct responses as the number of reported concussions increased (172). Age-related cognitive deficits do not typically emerge until after the sixth decade of life (59), but may be exacerbated in professional athletes with a lifetime exposure to RHI. The 'selective maintenance hypothesis' and the 'compensation hypothesis' propose that certain skills will be enhanced or maintained to compensate for any decreases in other areas of skilled performance to preserve overall behavioural and cognitive performance, creating a dynamic maintenance model of ageing (173).

1.2.3d TBI and MCI

Previous studies have shown that individuals with neurological impairment, brain damage, and MCI are more susceptible to the SIFI illusion (105, 174). One study assessed patients with primary sensory deficit or impairment of visuo-spatial attention using A-V stimuli and found that patients with visual field deficit had greater accuracy and reduced sensitivity to the fission illusion compared to those with unilateral spatial neglect and healthy controls (105). Individuals with MCI were also found to have reduced accuracy across the illusory conditions compared to healthy controls (175). The SIFI has been documented to differentiate between healthy and diseased/injured populations (175-179) and may have potential utility in identifying those at risk of concussion (180). Sporting individuals may have an advantage in predicting the movement of visual stimuli due to their perceptual expertise such that saccadic eye-movement tends to increase in expert sportspeople as the stimulus velocity increases. The SIFI may help to identify any impairments in cognitive mechanisms like MSI related to coincident-timing following concussive injury (180).

The SIFI test has been used to investigate individual differences in multisensory processing and the role of gamma-aminobutyric acid (GABA) and glutamate systems (181). In the context of concussion, perturbations to neuronal membranes and axonal stretching during the neurometabolic cascade of concussion may cause an uncontrollable ionic flux and the release of glutamate, leading to diffuse axonal injury and impaired neurotransmission (23, 24). Such pathological processes may affect multisensory performance during the SIFI test long after clinical recovery (182-185). The occipital cortex is vital for the perception of the fission illusion and is known to be impacted by the biomechanical forces of concussion as visual information enters through the primary visual cortex of the occipital lobe (105, 182, 186). MSI may be impaired in individuals with a history of concussion, and therefore the SIFI may be useful for diagnosis and prediction of impaired cognitive function beyond evident morphological damage (1, 26, 187). Male athletes who had a concussion between 1-14 months prior to fMRI analysis showed significant activation peaks in the visual and auditory temporal cortices, and posterior temporal association cortex in response to verbal and visual abstract stimuli (187). These regions are involved in processing environmental stimuli and may be affected by concussion leading to cognitive function deterioration (1, 26). However, generalizing these findings requires considering internal and external factors affecting injury and recovery. The SIFI may be crucial for detecting persistent symptomatic deficits that are usually undetectable by standard clinical assessments (188-190). The SIFI paradigm is similar to sensory discrimination tasks that have shown difficulty for those with a history of concussion in A-V discrimination tasks (191).

1.2.4. Reliability of the SIFI

The SIFI is a widely used and validated psychophysical paradigm of MSI and found to be a robust percept among the literature. It may provide a new measure of neurocognitive assessment in sport provided it is resistant to feedback even during relatively easy task conditions and may also extend to the implementation of false-feedback training (when participants are given erroneous information regarding the 'correct' reaction during illusion trials) (106). This is supported by previous evidence that response bias was not inherent during the SIFI task, conferring a level of immunity to various factors such as instruction to the subject, practice, familiarity with the task, reward, etc. (192). Although the SIFI is a robust indication of auditory-dominated MSI phenomena connected with perceptual sensitivity, previous research reveals that a 7-day behavioural training programme can lessen the reaction to fission and fusion illusions by strengthening perceptual sensitivity (106). There is a plateau effect that

appears during the training stage and tends to stabilise by the fifth day. Long-term training might modify the SIFI effect by improving perceptual sensitivity, particularly in terms of the fission illusion. Furthermore, the SIFI is impacted by several elements, including unisensory and multisensory modalities, attention allocation, attention modalities, feedback factors, expectancy, and prior experience as seen across more than 100 published articles in the last two-decades (120). Although MSI can ultimately improve our overall sensory perception to become more adaptive to temporal and spatial elements of voluminous external stimuli, there is also a high possibility of maladaptation leading to perceptual illusions. Impairments among individual sensory systems can therefore have profound effects on specific populations including athletes and the ageing individual.

It is believed that our capacity for MSI tends to increase with advancing age especially in the context of congruent stimuli as a compensatory mechanism for age-related decline in vision and hearing, which may further be exacerbated in the ageing athlete (13-15). Early detection and intervention of multisensory dysfunction would allow healthcare professionals to reduce any long-term impact on cognition and brain health. Understanding the modifiable effects of susceptibility to the SIFI with prior training is crucial to evaluate its applicability in diagnostic protocols. However, to the author's knowledge, the precise within and between session reliability and overall levels of agreement of the SIFI over time has not been previously assessed. The last two decades of research has focused primarily on the perceived and recorded susceptibility to the fission and fusion illusion based on altering stimulus parameters across varying populations. Ultimately, if the SIFI is to be introduced as a clinical assessment tool in SRC for young and older athletes, not only is an optimal protocol warranted but also an establishment of the reliability and agreement levels provided detectability of the A-V stimulus can impact the likelihood that a person will experience the illusion.

1.3. Multisensory Perception & Sports Performance

1.3.1. Overview

MSI is not an innate attribute of the CNS, but instead, it is functionally created through sensory experiences and neural plasticity (193, 194). Humans react more promptly and effectively to multisensory perceptions than to unisensory perceptions (195). The 'Late Integration Approach' posits that the human sensory systems work in isolation from birth, and it is through refinement and developmental maturity over time that cognitive and neuronal reorganisation occurs to enable adaptation and the ability for MSI (196). Hearing develops in

the latter stages of gestation, while the visual system develops mainly after birth (197). The TBW, which refers to the period during which sensory stimuli can affect the processing and percept of one another, is flexible and diverse throughout development and aging (198-200). Understanding the intricacies of MSI can aid in the development of innovative techniques to improve athletic performance while also reducing the risk of injury.

An athlete's ability to make swift and informed decisions in response to diverse stimuli modalities is crucial for their performance during training and competition. MSI occurs via direct cortico-cortical pathways, allowing athletes to constantly accumulate multiple sensory systems to identify biologically significant stimuli and produce desirable reactions that may not be achievable through a singular sensory input (201-204). The posterior-parietal regions of the brain have been shown to automatically fuse multisensory inputs, while the anterior parietal regions enable a more flexible spatial representation of the event produced from statistical predictions of Bayesian causal inference (155, 156). Athletes undergo consistent training in specific sports to enhance their current ability to process multiple forms of sensory input (205-207). Understanding the neural mechanisms underlying MSI can provide a scientific basis for the development of effective training strategies to enhance athletic performance. By improving an athlete's capacity to integrate multisensory inputs, they can make better decisions, respond faster and more efficiently to external stimuli, and ultimately achieve better performance outcomes.

Over the past decade, a greater understanding of the interplay between underlying neural interactions and subsequent behavioural responses with the manipulation of space, time, and stimulus intensity has been developed, shedding light on the significant role each plays in the perception of multisensory events (135, 208-210). This knowledge may allow athletes to develop better discriminative abilities across a wider range of temporal asynchronies, ultimately leading to an enhanced TBW that enables better perceptual judgments for stimuli of reduced saliency in dynamic sporting environments. However, the question remains: how do these theoretical constructs and neuroscientific evidence translate into improved athletic performance across the sensory modalities of vision and hearing throughout an athlete's lifespan, and how does damage to sensory systems by SRC negatively impact sporting performance?

1.3.1a Neurobiological Underpinnings of MSI

The concept of MSI originated from the work of Meredith and Stein, who investigated the superior colliculus (SC) as the processing centre for sensory information from environmental cues (211-216). They studied cortical and subcortical multisensory neurons in felines to understand the neurophysiological components of MSI (211, 215). MSI is caused by various factors, such as neuronal characteristics, synaptic topography, and interconnectedness of neural populations (217). The SC receives auditory, visual, and somatosensory information from multiple brain regions and develops neuronal processes with modality-specific and spatially congruent receptive fields. The SC integrates this information to produce a behavioural response that is different from the response produced by a single sensory stimulus (211, 214, 215).

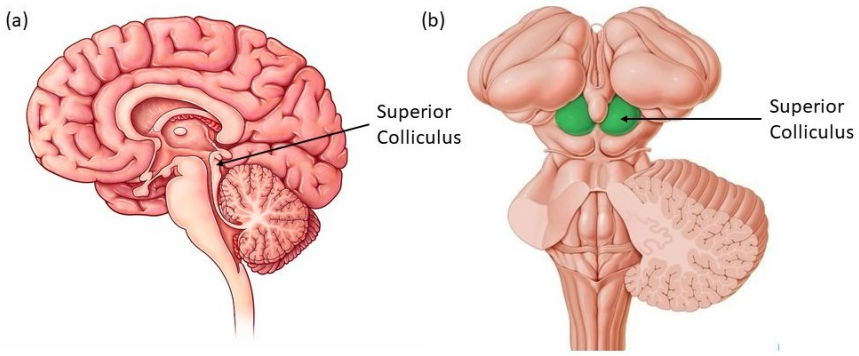

Figure 1.2. *Location of the Human Superior Colliculus.* **(a)** A mid-saggital section of the human brain depicting the location of the superior colliculus (SC). **(b)** A poster view of the mid brain with the cerebellum cut longitudinally and pushed laterally to reveal location of SC.

The initial view of MSI was that it only occurred in higher-functioning brain regions after processing in sensory-specific regions. However, it is now recognized that sensory-specific regions such as the visual cortex are inherently multisensory (218, 219). There is intrinsic inter-connectedness between the primary visual cortex and primary auditory cortex where fibre tracts from a seed region within the Heschl's gyrus connect to the occipital lobe and anterior portions of the calcarine sulcus, with previous multisensory experiences contributing to the activation of visual cortices (220, 221). There is evidence of sensory coupling between sensory-specific regions with suppressed functional activation of the primary visual cortex in response to sounds (222, 223). The suppression of visual responses by auditory input is linked to

performance and GABAergic synapses located in the infragranular layers of the primary visual cortex (224).

Although original research on MSI used single-unit electrophysiological recordings (211, 212, 214-216), contemporary MSI investigations indicate that the superior temporal sulcus (STS) and intraparietal sulcus (IPS) are important sites of MSI (225). Anatomically, the STS and superior temporal gyrus are located between visual and auditory cortices, suggesting the existence of multisensory areas at the borders of unisensory cortices (226). It has been suggested that the inferotemporal sulcus may serve as a point of crossover between the visual cortex and multisensory cortex, based on functional MRI (fMRI) data (227). There is evidence that MSI occurs early in cortical sensory processing, with neural projections from the core and parabelt auditory cortex extending into the primary visual cortex (228, 229). Beauchamp and colleagues demonstrated multisensory activations in the occipital lobe and STG, with the posterior STS and STG responding to both auditory and visual stimuli (230). However, the boundaries between unisensory and multisensory regions remain unclear.

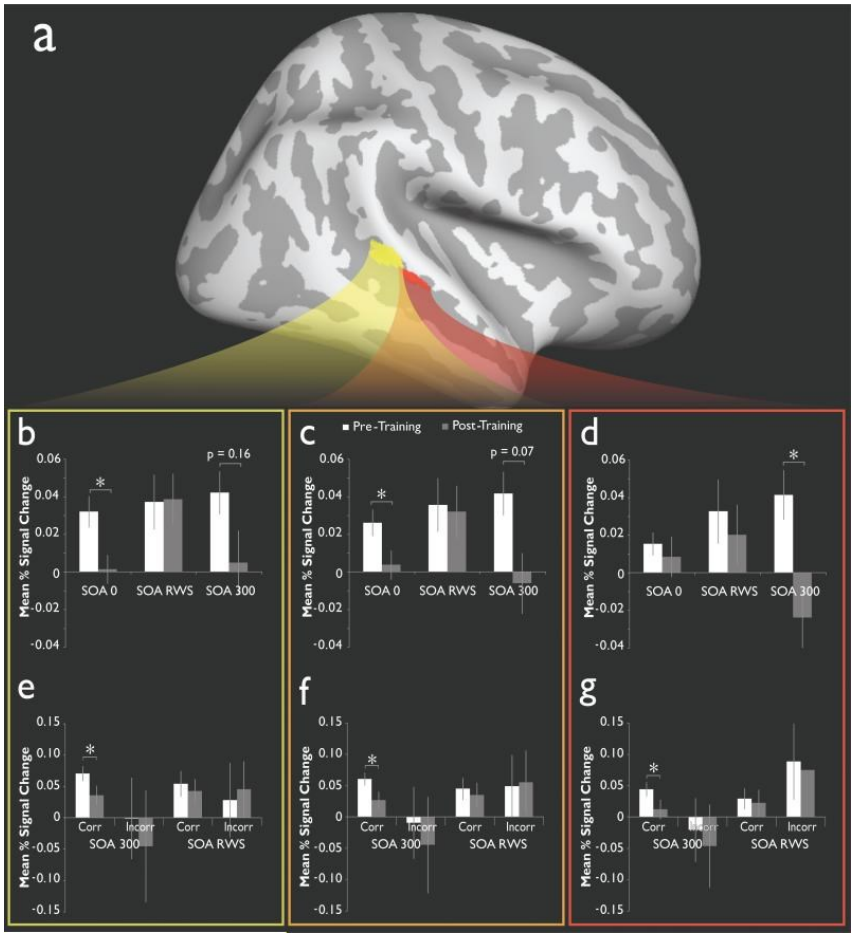

Figure 1.3. *Engaging in a multisensory temporal task leads to alterations in the activation of a network of brain regions primarily located in the pSTS area. *Adapted from the work of Powers, Hevey, and Wallace* (231).

1.3.1b How MSI transpires

MSI has been explained by various theoretical models, but the principal of sensory reliability weighting and causal inference are considered the most appropriate (232). Causal inference is essential for adaptive behaviour and is based on a person's past experiences, understanding, and new data. It follows Bayes' rule of statistical inference (233). In the human brain, sensory evidence is first accumulated in the inferior frontal sulcus, and then the anterior intraparietal sulcus combines the reliability-weighted unisensory information through Bayesian causal inference (156). This flow of sensory information has been demonstrated by electrophysiological recordings in frontoparietal neural networks based on the presented task stimuli (234). Children as young as five years old also consider the reliability of multiple sensory cues when perceiving their environment (235). While this form of Bayesian modelling provides mathematical understanding of neural and behavioural outcomes, the neurobiological process of cerebral computation remains unclear.

Selected cortical regions' inactivation can facilitate a reduced capacity for MSI, leading to a preference for unisensory stimuli interpretation and perceptual decision making in athletes (205, 236). Bayesian causal inference models the computational process underlying MSI, which can be applied to sport contexts. This involves inferring the degree to which observable variables contribute to a particular outcome, rather than considering them as separate variables (237). Recent studies using magnetoencephalography (MEG) data indicate that Bayesian models of MSI are most appropriate to explain the behavioural results, especially in the prefrontal, insular, and cingulate cortices (237). Sensory information transfer between primary sensory, multisensory, and frontal cortical regions is reflected through synchronized oscillatory activity (152, 238), and has been exhibited in the perceptual illusion of the SIFI through alpha waves (239). The temporal hierarchical approach to multiple sensory inputs guides MSI in the temporal and parietal lobes, which in turn influences later causal inference in the frontal cortex, creating a posterior-to-anterior gradient of MSI (237). Model b in **Figure 1.4.** illustrates different possibilities of causal inference when presented with two sources of information about a given variable. For instance, in Rugby Union, when an opposition player is carrying the ball (visual, S1) and calling out tactical commands to their teammate (auditory, S2), the player makes an inference as to the degree to which both variables will contribute to the opposition player's state of play, rather than considering them as separate variables.

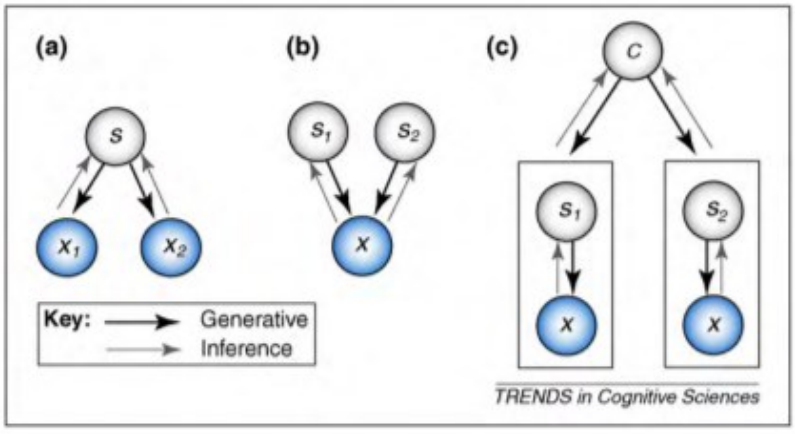

Figure 1.4. *Graphical model depictions of Causal Inference. *Adapted from the work of Shams & Beierholm* (233).

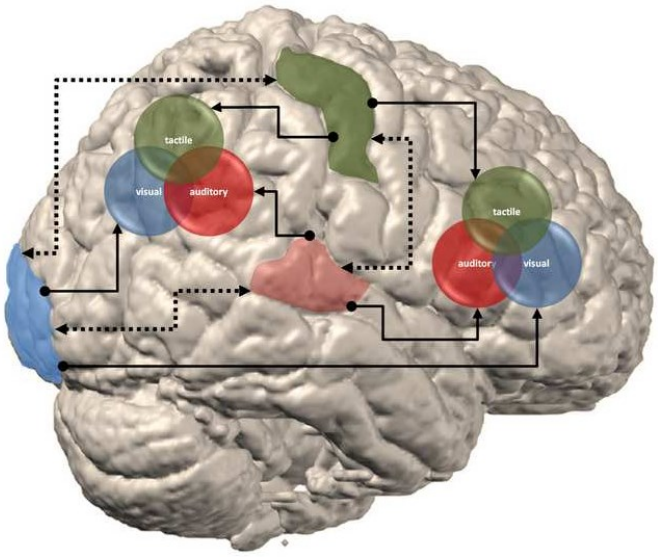

Figure 1.5. *Schematical Representation of the sites of Cortical Multisensory Processing.* The above depiction is of the right-hemisphere, with the occipital lobe on the left side of the image and the frontal lobe on the right side. Low-level visual, auditory, and somatosensory (tactile) cortices are indicated by the blue, red, and green shaded regions, respectively. The overlapping regions between coloured discs highlight the regional constrictions of higher-order processing in the prefrontal and parietal cortices, and the regional interactions are depicted through the solid black lines. The dotted lines represent direct interactions between low-level cortices. **Adapted from the work of Murray and colleagues* (240).

1.3.1c Development with Age and Sensory Weighting

The ability for MSI develops over time. It is dependent on sensory experience and will functionally alter neural networks in a scaffold representative of Bayesian statistical inference in a mathematical sense but not in a neurobiological one. Specific multisensory neurons of the subcortical and cortical regions are responsive to unisensory input in the early stages of development, and it was only with the passing of time and maturity that the responsivity adapted to multiple sensory stimuli presented together and was not an artifact of co-registration of each sensory input (211, 214, 215).

The information reliability hypothesis posits that the dominant sensory modality provides the most reliable information for MSI (120, 241, 242). Previous research has shown that the perception of the SIFI varies considerably among individuals (243), with females being more susceptible than males (95). Recent work by Zhou et al. investigated whether MSI was enhanced in older females using the SIFI (244-246). The experiment included 27 younger and 30 older females who underwent 480 trials with different delays between the visual flashes and auditory

beeps. Older females were more susceptible to the fission illusion than younger females, and this susceptibility was positively correlated with neural activation of the left middle frontal gyrus. On the other hand, the fusion illusion was positively correlated with activation of the right inferior frontal gyrus and right superior frontal gyrus. The findings support the idea that the decline of selective attention in older adults may contribute to the enhancement of the SIFI. In light of this, *'Perceptual load theory'* suggests that decreased selective attention in older adults creates a situation where they cannot ignore irrelevant stimuli, occupying a large proportion of their attentional capacity (247). In addition, age-related differences in SIFI susceptibility were observed in children, adolescents, and adults (97, 248). Brain maturation significantly affected susceptibility to the SIFI, with increased variability among aging subgroups.

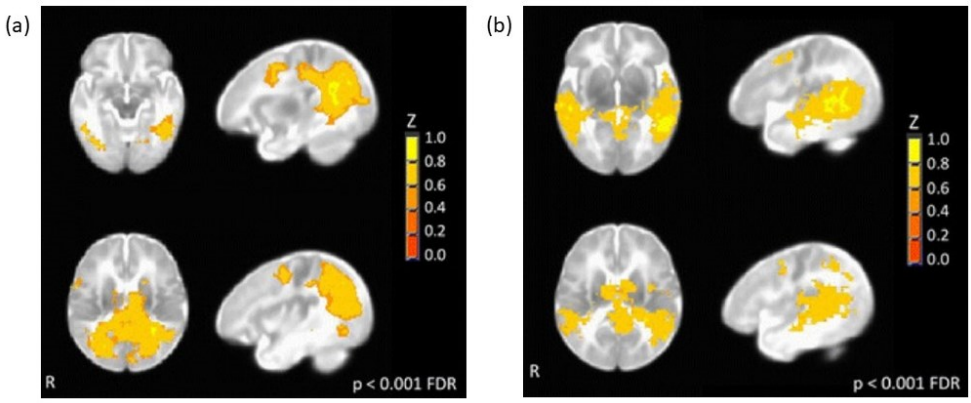

Figure 1.6. *Functional Neonatal Brain Development of MSI regions.* Demonstration of structural connectivity patterns found in the **(a)** IPS and **(b)** and STS regions of a human infant in the first few weeks of life after birth. **Adapted from the work of Sours and colleagues (249).*

Expert-level status in sports is achieved through experience, which is largely a function of age (250). MSI in developing children aged 7-16 years was measured using high-density electrical mapping (251). The study found that reaction time and percentage of correct responses to an A-V task increased with age and were higher in the multisensory conditions than in the unisensory conditions. Miller's Race Model postulates that in the presence of more than one sensory input the individual's reaction time will decline and if the model is violated during the task it is of the assumption that the individual sensory inputs have combined through MSI to facilitate an enhanced reaction time (252). Miller's Race Model was found to be violated specifically among older adults, suggesting that MSI is associated with aging. The model of perceptual narrowing, however, best represents the development of MSI in humans, with multisensory cortical areas possessing a broad level of multisensory perceptual tuning

that narrows with age (253, 254). Early participation in sports may result in better development of multisensory patterns of optimal connectivity in the IPS (249), leading to lasting effects in professional sporting careers. The STS is found to have significant functional connectivity with the visual association areas and the primary auditory cortex, and the IPS possesses robust functional connectivity with the visual association areas from birth (249). However, caution is needed as these findings only represent the presence of functional neural networks for MSI and not direct evidence of its ability to take place (249). Age-related increases in MSI can benefit sporting performance when A-V sensory inputs are congruent (255), but normal aging leads to declines in temporal and spatial perception of A-V inputs (256, 257). Aging is associated with a range of auditory and visual deficits in temporal order judgements (258, 259), as well as reductions in brain volume and cortical thinning of 0.5% annually (260-262), particularly affecting the STS and SC in the temporal lobe. These changes negatively impact multisensory brain regions and reduce neural functioning of the pre-frontal cortex and cognitive resource utilization, potentially impacting sporting performance (263-265).

One must try to consider the current understanding of sensory weighting by the brain to assess MSI accurately and objectively across all age groups in experimental design and more importantly interpretation. Sensory stimuli that are perceived in tandem, such as A-V information, will be weighted as to their relative reliability in order to achieve the most optimal multisensory depiction of the environment (232). Sensory reliability in this context refers to sensory integration and defined using the term 'estimate precision' accounting for sensory signal quality where reliability *'r'* is viewed by the brain as the inverse variance of the sensory estimate accounting for the possibility of sensory or informational noise and reducing estimate bias from each sensory source *'i'* and their weighted reliability (232):

$$r_i = 1/\sigma_i^2,$$

With increasing age, sensory re-weighting of information is however common place (266). Stimulus saliency is the key driver of a bottom-up approach to MSI, with the attention of an individual being captured by a task or object more profoundly through multiple sensory systems and dependent upon the relevance of the task and associated stimuli (267). The two most important aspects of stimulus saliency are spatial and temporal proximity. Perceptual illusions such as the SIFI are very much dependent on these two aspects of stimulus saliency,

whereby the strength of MSI is known to decrease as a function of temporal and/or spatial inconsistencies in paired stimuli. Many studies have used the SIFI as a formal assessment of MSI in older adults in hopes of detailing the contributions through which the brain will adapt to prioritise certain sensory cues over others to maintain daily activities (110, 268-270). Susceptibility across all age groups, but in particular the elderly, is heavily based on unisensory reliability to A-V illusions (99), where audition is known to affect visual perception (210).

1.3.2. Perceptual Cognitive Training

Perceptual cognitive training aims to improve an athlete's ability to integrate sensory information with prior knowledge and experience to anticipate and respond to sporting behaviour (167, 201). Field-based team sport athletes are typically assessed using video simulation models to assess perceptual decision making, which can be enhanced through training (271-273). MSI and perceptual decision making are interconnected, as visual and auditory stimuli provide complementary information (242). The goal of training is to reduce uncertainty by improving evaluation of sensory estimates in different modalities to produce an appropriate behavioural response (274). Training in a context that resembles the competitive sporting environment can improve perceptual cognitive skill acquisition (205, 275). Combining A-V cues can enhance the reliability of the statistical estimate of the event and improve 'action/performance fidelity' (275, 276). However, training to manipulate individual differences in perception is a challenging process (277).

Engaging in lifelong sports participation correlates with improved cognitive function and executive processing, especially in field-based athletes who require high levels of visuospatial processing and refined motor skills (278). The theory of deliberate practice emphasises the importance of domain-specific practice in achieving expertise, with the number of hours spent in deliberate practice being a key variable (173, 279, 280). Research has investigated whether time spent playing soccer during development discriminates against levels of expertise achieved, finding that highly skilled soccer players spend significantly more time per year engaging in soccer-specific activities during childhood, resulting in higher perceptual-cognitive performance accuracy (281). However, despite accumulating more hours in soccer activity, there were no differences between highly skilled and low-skilled athletes regarding their age of entering an elite training program or achieving semi-professional status. This would indicate that more detailed neurological mechanisms and cognitive behaviour underlie expert performance through MSI capabilities via the sensory modalities of vision and hearing.

1.3.2a Vision and Sport

Athletes rely heavily on their visual and perceptual abilities to navigate and excel in their sporting environment. Sport-derived skills such as visual scanning and reaction time in response to stimuli ranging from the movement of players and the crowd to the trajectory of the ball or terrain they are covering are key contributors to an athlete's on-field performance. The brain's remarkable ability to translate a two-dimensional image into an interactive three-dimensional space is crucial to an athlete's performance (282). Athletes must be able to inhibit irrelevant stimuli and react to ever-changing affordances in the environment to prevent injury and improve competitive performance (206, 207, 283, 284). Through extensive training, athletes can enhance various visual indices of the visuo-oculomotor system, which leads to positive changes in both neurophysiology and sporting behaviour. For example, American football players with vision training develop enhanced levels of peripheral vision and reduce the incidence rate of collisions (285). Decades of research have shown that visual information processing occurs in two distinct neural pathways known as the 'two streams hypothesis' (286, 287). The dual stream theory of vision suggests that higher order cortical processing of visual stimuli occurs in two intertwined streams, the dorsal and ventral streams, which support visuo-motor processing, control, and object recognition (288).

Sports vision training programs are gaining popularity worldwide for their ability to enhance athletic performance and reduce injury rates (289-292). These programs demand high levels of cognitive processing, placing the entire oculomotor system under strain to improve information processing efficiency. They aim to enhance visual spatial skills and target anticipation, promoting a complex interaction between sensory-motor control systems and intrinsic and environmental factors. Perceptual abilities of static and dynamic play are essential in team sports for gaining a competitive edge through opponent identification, play analysis, and environmental mapping (167, 201, 293). Such training can lead to superior levels of dynamic visual acuity and visuomotor control, accounting for almost 70% of the variability in the number of goals scored among collegiate ice hockey players (290). Athletes consistently outperform non-athletes in processing speed, attention, visual search strategies, and velocity, with these adaptations possibly due to the 'optical flow stimuli coherency' aiding in the perception of visual patterns (166, 294). In particular, radial optical flow stimuli evoke sensory processing in the fovea of an object moving through space which is determined by the middle temporal and middle superior temporal regions of the brain, improving visual performance, and reducing the risk of injury, particularly head impacts in Division One American football players (39, 40).

Elite athletes possess highly developed sensorimotor skills, which allow for rapid response to visual stimuli, even with limited sensory information, due to enhanced visual acuity, contrast sensitivity, and tracking abilities, honed through early childhood training (295). Successful performance in sports requires the integration of both visual perception and peripheral vision, the latter being the visual field of view from 60° to ~180° of horizontal diameter (290, 291, 296, 297). Sophisticated computer-based sensory tests like the Nike Sensory Station (NSS), Eye–Hand Coordination Test (EHCT), and the Vienna Test System (VTS) have been employed across a variety of sports to evaluate peripheral vision (290, 298, 299). High levels of both visual and motor skills can sometimes saturate visual reaction times, suggesting that there may be an optimal level of training for peak performance without compromising overall performance (298). Visual performance is therefore a critical aspect of athletic success and is highly dependent on both hardware and software components of the visual system. Research has shown that expert athletes possess superior visual discrimination, peripheral response time, and dynamic visual acuity compared to their less skilled counterparts, indicating the presence of visual system hardware enhancements (290, 300). On the other hand, years of deliberate and specified training of isolated sensory components have been shown to enhance visual software (167, 201, 301). However, declines in visual performance have been observed in professional baseball players in their late 30s, indicating that visual decrements may be exacerbated in contact sports where exposure to RHI and incidence rates of SRC are high, potentially leading to neural processing deficits of sensory and predictive visual information. This underscores the importance of both hardware and software components of the visual system in athletic performance and the need for targeted training programs to maintain optimal visual performance throughout an athlete's career (299).

1.3.2b Hearing and Sport

The human brain's natural inclination towards music and rhythm has been observed to synchronize with bodily movements and external acoustic stimuli. This phenomenon of auditory-motor synchronization is particularly pertinent to athletes who seek to gain a competitive advantage over their opponents by enhancing their sensory abilities and overall performance (302-306). Expert athletes demonstrate an inherent superiority in motor resonance, which allows them to better understand and predict their opponents' movements (307, 308). Accurate auditory processing is a crucial component of this ability, as athletes must be able to interpret auditory cues and use them to map various movement and behavioural patterns onto distinct auditory elements (309, 310). This will enable the individual to

systematically modulate *kinematic sonification* to enhance the quality of their movement and learn a new motor skill by integrating many congruent perceptual streams of information (311, 312). With repeated exposure over time, the motor cortex's firing rates and patterns can be enhanced by means of auditory-motor entrainment (313), leading to neuronal plasticity and improved acquisition of motor skills in athletes. These findings are supported by research on corticospinal excitability and beta brain waves, which have been linked to the development of superior and medial posterior temporal regions associated with the identification of human movement (314-316).

Expert athletes possess the ability to map physiological and physical data, i.e., rhythm, loudness, and pitch, onto psychoacoustic parameters to create a cognitive database of biomechanical information known as movement sonification (311, 317, 318). This mapping offers advantages beyond mere rhythm alteration and enhances the athlete's self-awareness of the underlying physiological processes of movement and coordination, which aids in decision-making and execution (311, 317, 318). Elite rowers have used movement sonification to perfect movement control and execution resulting in improvement in crew synchronization, faster average boat speeds, and increased distances travelled per stroke compared to when no auditory signals were present (319-321). Studies also show that sonification modelling improves perception and accuracy in novice basketball players, suggesting that auditory information facilitates predictive capabilities, response times, and inhibitory control which is necessary for acquiring and retaining new motor skills (322).

Athletes must quickly perceive and react to changes in their sporting environment with divergent states of play, relying on inhibitory control among competing sensory systems to make decisions (323). Professional basketball players can predict their opponent's movements by the sounds they make, dampening neural noise to enhance auditory perception specifically in the mid-brain (309). Athletes possess quieter neural systems enabling them to process external stimuli into a more meaningful percept and improving higher-order functions such as attention and decision-making (324). These abilities are developed through long-term training to extract and decipher natural movement sounds to anticipate behaviour through cognitive models of action-prediction involving the neuroanatomical coupling between auditory and motor systems at various levels including the spinal cord, cortex, and subcortex, such as the auditory cortex, supplementary motor area, cerebellum, and premotor cortex (325). Efferent fibres travel from the nucleus of the ventral cochlea to the sensorimotor tracts of the spinal cord through reticulospinal connections, leading to auditory-motor entrainment (325). The perception of and entrainment to auditory cues will enable the athlete to establish temporal

predictions of movement and behaviour through the oscillatory coupling of neural impulses between the auditory cortex and intertwined motor regions. While these skills are beneficial for athletic performance, concussion-induced damage can affect perception-action coupling, negatively impacting auditory and visual processing (326-330). It is unclear whether such skills protect against sporting injuries like concussion.

1.3.3. Injury-induced Sensory Deficits

1.3.3a Vision

Concussions can cause visual disturbances in up to 69% of adolescents (331), including deficits in colour vision, acuity, accommodation, and saccades, as well as photophobia and blurred vision, lasting to two years after injury (332, 333). These symptoms result from damage to both the afferent and efferent visual pathways, and are associated with delays in visual sensory information processing at the axonal level from DAI (25, 334). The diffuse nature of functional, metabolic, and structural changes associated with concussive pathology extends to visual processing streams of the extra-striate visual cortex, alongside the primary visual and visual cortices (67). While most neuro-ophthalmologic function appears normal during a clinical examination (335), more in-depth pupillary light reflex testing using digital infrared pupillometry reveals reduced pupillary indices of ocular health, such as slower dilation velocity and longer constriction latency (336, 337).

Individuals with mTBI exhibit impaired target prediction, as indicated by an increased phase lag while tracking a designated target during circular tracking paradigms (338). Diffusion tensor imaging (DTI) has revealed reduced mean fractional anisotropy indices in the WM tracts of the right anterior corona radiata, left superior cerebellar peduncle, and the genu of the CC, which are known to be affected by a concussive blow, and are associated with position error and target prediction on circular tracking paradigms. These findings are indicative of DAI and are associated with chronic concussive symptomatology (339, 340). Additionally, mTBI has been associated with loss of tissue volume, damage to the optic radiation, and deficits in perception and field of vision (334, 341). These deficits can have significant implications for athlete health and welfare, as athletes with mTBI may have a reduced capacity to protect themselves from incoming tackles and impacts in both training and match play (285). Vision-based assessments can detect concussions when other diagnostic indices fail (342), which is important as untreated concussions can result in chronic symptoms and increase the likelihood of future concussions (26, 70, 78). Chronic visual dysfunction may also occur and impact quality

of life, with photophobia and reduced depth-perception as possible symptoms of higher-level cortical impairment as a result of the diffuse nature of concussion (343, 344).

The pathophysiology underlying visual dysfunction in mTBI is not well understood, but the dorsal stream vulnerability hypothesis suggests that the magnocellular pathway and dorsal stream may be more susceptible to damage than the parvocellular pathway and ventral stream (345). It is believed that the magnocellular pathway and its reduced possession of cellular abundance predisposes the dorsal stream to more damage than the parvocellular pathway that projects to the ventral stream (144). Patients with mTBI exhibit deficits in critical flicker frequency thresholds, reduced sensitivity, and increased variability, indicative of damage to higher cortical pathways within the magnocellular retino-geniculate pathway and/or primary visual cortex (144, 145, 346). Damage to both the dorsal and ventral pathways following concussion has significant implications for balance, especially in acutely concussed athletes (347). A baseline refractive and ocular health examination should be performed before any visual assessment to rule out pre-existing visual dysfunction. A more inclusive and holistic composite measure of concussion status is required, and the SIFI may be most suitable for future study assessment protocols of vision (336, 348).

1.3.3b Hearing

Concussion can cause hearing deficits, with over 50% of individuals with a history of concussion experiencing central auditory dysfunction where it is common to experience tinnitus or *"ringing in the ears"* (349, 350). Sensorineural hearing loss, characterised by structural damage to the vestibulocochlear nerve and cochlear hair cells, is a common consequence of concussion and is associated with narrowing of the auditory radiation in both hemispheres (349, 351-353). The susceptibility of the auditory brainstem nuclei to rotational and shearing forces, as well as damage to the primary auditory cortex due to its proximity to bony ridges, can result in such injury linked to the DAI pathology of concussion (354, 355). Patients with multiple concussions are more likely to experience auditory symptoms, with hyperacusis, hearing loss, and tinnitus being the most common (356), impacting the HRQoL with 92.3% of patients having suffered a mTBI reporting one or more symptoms of auditory dysfunction.

Hyperacusis, or sensitivity to sound, is a significant subjective symptom that increases the risk of prolonged PCS by threefold at 3 months post-injury (357). This condition is characterised by a decrease in tolerance to sound, and even moderate sounds are perceived as excessively

loud (358). The Central Gain Model proposes that mechanical damage in the peripheral hearing system leads to increased sound evoked firing rates, resulting in hyperacusis and tinnitus (359, 360). This theory aligns with the Neurometabolic Cascade of Concussion, which suggests that disruptions in the excitatory-inhibitory neural balance of glutamate and GABA, and neuroinflammation contribute to the increase in neural function in the auditory cortices (24). Recent investigations have assessed athletes beyond the acute stage of concussion using psychoacoustic and psychometric measures to evaluate their sound sensitivity (361). In collegiate sport, 50% of athletes who had suffered one or more concussions reported sensitivity to sounds up to 6 weeks post-injury, despite no differences in hearing thresholds (361). Those with sound sensitivity had significantly lower loudness discomfort levels, longer symptom duration, and worsened mental health. Furthermore, most athletes reporting sound sensitivity also reported sensitivity to light, suggesting a common function between these two senses related to an increase in neuronal function or excitatory-inhibitory neuronal activity again similar to that of the central gain model. The use of auditory metrics in concussion research highlights the sensory link to the pathophysiology of concussion and MSI, which can be assessed by the SIFI. A complete understanding of how concussion affects sensory systems may identify unique and treatable problems for better concussion management protocols and sporting success. Future research should investigate the impact of age and concussion aetiology on the diagnostic and prognostic utility of SIFI which may be valuable for clinicians and researchers.

1.3.3c MSI and Associated Decrements in Sports Performance

Executive function plays a significant role in MSI and is inherently linked with an athlete's ability to compete in sporting competitions and training, processing large volumes of sensory information (362). Injury-induced executive hypofunction could result in decreased sensory abilities in a fast-paced, dynamic game situation where a reductions of relevant information processing with the aim of ignoring task irrelevant stimuli is associated with a history of concussion (188, 363). A range of cognitive networks linked to attention and working memory are commanded by auditory skills such as listening to verbal commands and that of environmental cues. These processes are centrally confined to the neural boundaries of the frontal and temporal cortices, regions of the cortex that are most susceptible to concussive injury. This can create a state of dysfunction among neural mechanisms of temporal information processing with corresponding reductions of information processing speed and reaction time which are established and predictive risk factors for concussion status (364).

These temporal sensory deficits in audition and speech processing may be more evident than those related to vision, whereby the integrated cortical-subcortical networks of the prefrontal cortex, parietal cortex, and basal ganglia are prone to WM tissue damage, particularly in the CC, and deterioration of WM cortico-cortical tracts relevant to sensory information processing as a result of functional deficits in excitatory N-methyl-D-aspartate (NMDA) receptors (23, 24).

Reduced visual integration efficiency of both visual stimulus identity and location has been observed in children having suffered a mTBI (365). This observational work precipitated the development of a novel measure of MSI measuring cognitive shifting flexibility from visual and/or A-V shift conditions of the paediatric brain referred to as *'set shifting'* (366, 367). The set-shift in the visual condition was a question mark presented above the target picture, and the A-V shift was an auditory monophonic tone set at 500 Hz. TBI severity and performance exhibited a linear effect in the A-V set-shifting condition whereby the more severe the injury the lower the performance accuracy, emphasising the effect that TBI and/or concussion has on MSI from an early age of exposure (366). Reduced performance accuracy and lower drift rate in the A-V condition were correlated with reduced general intelligence signifying diminished MSI capacity in children with TBI. The authors acknowledge that this reduction in performance accuracy can be linked back to *"reduced efficiency in integrating visual and auditory processing"* (366).

More recently, there have been advancements in clinical assessments of MSI to combat the more outdated forms of assessment that are too reliant on subjective observations and translation into clinical interpretation. The *'Clinical Test of Sensory Integration and Balance'* (CTSIB) was originally designed by Shumway-Cook and Horak in the mid-80's (368) and later modified for concussion assessment (369). The test was created to assess the sensory selection process *"by compromising available somatosensory, visual, and vestibular senses while measuring an athlete's ability to minimize postural sway",* carried out across four different testing conditions each assessed for 20 seconds to derive an composite score (370). Investigating the differences between male and female collegiate level athletes to establish preliminary normative data for the modified CTSIB, this retrospective cross-sectional study found female collegiate-level athletes had better overall MSI and balance across all four testing conditions. The amalgamation of neurobiological and multisensory perception concepts in sports performance, particularly in relation to vision and hearing, presents a vital and exciting field of study. The current evidence underscores the importance of recognizing the sensory deficits induced by SRC and their detrimental effects on athletic performance. To enhance our understanding of MSI in sports performance, future research should concentrate on exploring

the intricate neural mechanisms and examining how they are influenced by injury. This can lead to the creation of novel strategies to diagnose, treat, and prevent sports-related injuries, as well as improving athletic performance, cognitive function, and overall health among the ageing athletes.

1.4. The Brain and Cardiovascular Health of an Older Athlete

1.4.1. Risk of Ageing

Age is a significant predictor of cognitive decline accounting for 49% of cognitive function variance (371) and associated with reduced CBF (372). The levels of self-reported PA during adolescence are a significant predictor of the rate of age-related cognitive decline around the sixth decade of life (373). Those adults who were physically active when middle-aged were found to have higher total brain volume in old age in comparison to their sedentary counterparts (374). Engagement in exercise and PA in mid-life can significantly reduce the risk of dementia and AD in later life (375, 376). Arterial ageing, signified by aortic stiffness, begins as early as the second and third decades of life and is correlated with white matter hyperintensities (WMH) volume and diminished cognitive function in midlife, including executive function, processing speed, and visual memory (377). Higher aortic stiffness in midlife can lead to a diagnosis of midlife hypertension due to chronic exposure to elevated levels of BP leading to greater volume of WMH's (378). This pathological trajectory between BP and WMH load is most strongly linked to increasing age (378). WMH's and vascular WM lesions can accumulate from young adulthood and contribute towards cognitive impairment, diminished executive function, and reduced cognitive flexibility (379). Declines in cognitive function are most apparent at 58 years of age, where age and hypertension were the strongest and most predictive risk factors meeting the criteria for MCI (379). However, a single measure of longitudinal analysis may not accurately represent the cumulative burden and longitudinal variation in CV risk profiles that contribute to healthy aging and cognitive function in later life (380).

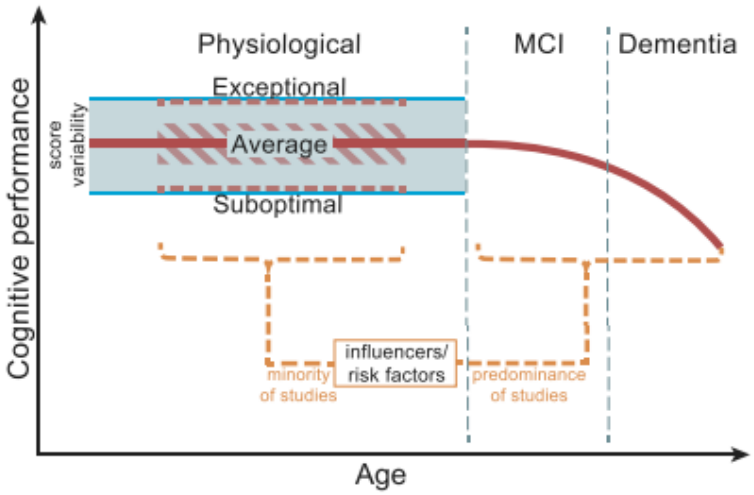

Figure 1.7. *Midlife Cognitive Performance***.** The image portrays the physiological cognitive performance during midlife, with hatching space denoting three levels of performance: exceptional, average, and suboptimal. The image also highlights mild cognitive impairment and dementia as potential pathological consequences of cognitive aging. **Adapted from the work of Novotny et al.* (381).

1.4.2. Brain Health of Older Athletes in Sporting Retirement

Early exposure to contact sports and related head impacts has been linked to decreased cognition in retired NFL athletes at midlife, particularly in areas of executive function, learning, and memory (382). Self-reported age at first concussion during childhood increases the risk of future concussions, with a decreasing associative risk of subsequent concussion annually (383). High school football players who have not sustained a concussion exhibit altered WM integrity, default mode network connectivity, decreased thalamic volume, and cortical grey matter (GM) structure, leading to poorer memory scores and early onset of neurobehavioral symptoms in retirement (382, 384-386). Persistent engagement in contact sports can cause hyperconnectivity in the default mode network, which is suggested to be a compensatory mechanism for continued executive function or compensatory regulation during high-impact exposure (387). While alterations in cerebral structure are observed during a critical period of development due to RHIs, no athletes showed any long-term clinical deficits (388), highlighting the issue of sensitivity and specificity of current assessments suitable encompassing multiple factors. The age of 12 is often chosen as the appropriate cut-off for exposure to contact sports, as it coincides with the maturation of macrostructural, microstructural, neural, and vascular networks critical for cognitive development (389). Early exposure to contact sports such as American football can have an impact on neurodevelopmental maturity and increase susceptibility to neurological deficits in adulthood (76, 382, 386). It is unclear whether the

changes in WM due to RHI early in life indicate pathological processes or altered neurodevelopment, although some studies have suggested a dose-response relationship between lifetime exposure to head impacts and cognitive dysfunction later in life (385, 390, 391).

Studies have shown that professional and community rugby union players who report experiencing one or more concussions have worse cognitive flexibility, executive function, and complex attention scores compared to those who report no concussions (392). In combat sports such as boxing and mixed martial arts, decreased processing speed is correlated with the number of professional bouts and fight exposure score (393). Thus, as concussion exposure increases there is a decrease in thalamic and caudate brain structure volume. Retired rugby league players with a self-reported average of 33.44 concussions have significant differences in GM glutathione, glutamate, N-acetyl aspartate (NAA), and creatine correlating with reduced concentration and related to depression, anxiety and stress (394). There is however a higher prevalence of previous concussion history among former elite rugby union players compared to community and non-contact control athletes (392). Regardless of sporting type, retired athletes with one or more previous concussions have worse outcome scores on measures of cognitive flexibility, executive functioning, and complex attention than those without any previous concussion (392). Brain health and function begin to decline up to 3 decades after the last concussion among retired healthy athletes across indices of motor and neuropsychological function (395-397).

1.4.3. CV Risk Factors

Originally defined by the Framingham study in the late 90's, CV risk factors included smoking, high blood pressure (BP), unhealthy cholesterol levels, diabetes, and age (398). Their presence, alongside lower cognitive function, as early as the third decade of life and during mid-life are significant and independent predictors of reduced cognition, mild-cognitive impairment, and dementia up to 20 years later (380, 399, 400). Even a modest reduction by as much as 10% of the burden of modifiable CV risk factors could see the prevention of almost 2 million dementia diagnoses globally by 2050 (401). The odds of accelerated cognitive decline significantly increase with an increasing number of CV risk factors, where ≥3 CV risk factors more than triples the odds of accelerated cognitive decline (402). Cognitive function encompasses a diverse range of cerebral processes such as memory, attention, executive function, and global cognition that are essential for daily living. Age-related alterations in

cognitive function, in the presence of CV risk factors, can lead to pathological consequences (403-405). The association between vascular and metabolic risk factors, including high BP, diabetes, and elevated serum cholesterol, with cognitive decline has been widely discussed (406-410). These risk factors may initiate cognitive decline in midlife, long before the onset of clinical dementia, thereby providing a window for suitable brain health interventions to preserve later-life cognition (411-413). Early recognition and treatment of individuals at risk of cognitive decline and dementia, with known CV risk factors, can reduce co-morbidity and mortality rates and prevent the development of symptoms (414). Although there is some controversy, epidemiological and pathological investigations suggest that modifiable CV risk factors, such as hypertension, diabetes, and high cholesterol, contribute to cognitive decline in later life (415-419).

1.4.3a Hypertension

One of the most significant risk factors for the development of neurocognitive conditions, such as vascular dementia, cognitive decline, and AD in the general and sporting populations is hypertension during mid-life. Hypertension will dismantle and degrade the form and function of cerebral vasculature and microvasculature which can lead to accelerated cognitive decline and significantly reduce cognitive reserve. It may be subtle and go unnoticed in mid-life until age reveals the true extent of the functional damage. Hypertension is a significantly important and modifiable risk factor presenting itself as a target for intervention due to its high prevalence during midlife. Elevations in BP during mid-life can negatively affect cerebral perfusion rates, where adaptive vascular changes in CBF regulation and arterial pressure can incite premature cerebral-arteriosclerotic alterations narrowing the blood vessels and increasing vascular resistance (420). Hypertension is associated with and predictive of augmented incidence rates of cognitive deficits at 50 years of age among the general population (421, 422). Although not all individuals with hypertension will develop dementia, an inverse relationship between hypertension and cognition exists (423). Higher levels of cognitive functioning in adolescents and young adults reduce the risk of developing hypertension, with females having a lower likelihood of developing hypertension than males by age 50 (424, 425). Untreated hypertension in women is associated with reduced immediate and delayed memory performance a decade later (426), although the lifetime probability risk of developing hypertension for middle-aged men and women is similar (427). Hypertension is also linked to reductions in cognitive processing speed (428), which may be subtle but detectable with assessment of CV risk factors. Hypertension contributes significantly to

cognitive decline through hypertension-induced neurovascular uncoupling, which disrupts the fine-tuning of CBF by regulation of arteriolar resistance by diminishing any reactive increases in CBF caused by neuronal activity, altering cellular homeostasis, and preventing the respiratory and metabolic demands of neural activation being met (429-431). Although efficacious treatment of hypertension can significantly reduce the incidence of cognitive impairment and dementia, it cannot eliminate it entirely (432).

1.4.3b Cholesterol

Cognitive decline is among one of the leading causes for disability (433). Ageing trajectories are tightly linked with altered paths of cognition and neural mechanics across the lifespan (265). Given that the CNS is a highly dynamic, complex, and adaptable system that changes over a lifetime, cholesterol can play an important part in synaptic plasticity due to its nature in the shape and function of synaptic drive, with the potential for cognitive impairment (434, 435). High levels of cholesterol are associated with higher incidences of hypercholesterolemia, dyslipidaemia, and worse cognition, indicating a close relationship between brain and CV health (436). Increased levels of cholesterol are associated with an accelerated risk of global cognitive decline in more than one-fifth of the middle-aged population (437). Total plasma cholesterol in midlife is a significant vascular risk factor and a predictor for early onset dementia and cognitive decline (428, 438, 439). At midlife, higher levels of total serum cholesterol is predictive of reduced cognitive function and capacity acting as a key contributor in the pathogenesis of AD and dementia (440). Beginning to decline gradually from midlife (441, 442), this advanced rate of cognitive decline is associated with structural and functional deficits, as well as an inability to recruit specific neural networks due to differentiation and weaker coupling among selective network modules associated with specific cognitive functions in later life (443-446). The impact that total cholesterol has on age-related cognitive decline and neural trajectories is therefore more pronounced in midlife (447, 448). However, the evidence of cholesterol as a significant risk factor for cognitive decline remains unsettled.

1.4.3c Diabetes

Midlife diabetes is a prevalent metabolic disorder that increases the risk of comorbidities and mortality, with a dementia risk comparable to that of the Apolipoprotein E (APOE) genotype (449). Type 2 diabetes often occurs in midlife, particularly in individuals with pre-

existing conditions such as hypertension, obesity, and sedentary behaviours. The association between diabetes and dementia is strongest in midlife with reduced association beyond 65 years of age (400), and is linked to a more severe Alzheimer's-related pathology in later life (450). Diabetes and altered glucose metabolism are linked to cerebral vasculature injury (451-456), resulting in executive function, verbal fluency, processing speed, global cognition, and memory decline (457-461). Fluctuating and intermittent states of hyperglycaemia affects not only nerve cells but also cerebral vascular tissue, leading to cerebrovascular dysfunction, cerebral infarcts, inflammation, and accelerated cognitive decline (462-464). Diabetes is now considered a modifiable risk factor for cognitive decline and mild cognitive impairment, which is a reversible and variable stage between normal cognitive aging and dementia (465), where almost 35% of diabetic patients suffer from cerebrovascular and CV diseases (466). Studies using neuroimaging have demonstrated that long-term exposure to diabetes is associated with cognitive impairment characterized by pathological features similar to those with vascular dementia and overall brain atrophy (453, 454). Structural alterations have been observed in brain regions such as the hippocampus, medial temporal lobe, and basal ganglia in midlife individuals with diabetes (467). Reduced WM integrity in regions such as the CC, cingulum, and superior and inferior longitudinal fasciculus have been consistently linked to cognitive dysfunction among diabetic patients (468-471). In vivo regional atrophy, especially in the hippocampus, has been associated with executive function and memory deficits that resemble those observed in preclinical AD (468, 472). GM volume loss, total brain volume reduction, and accelerated hippocampal volume loss have also been linked to cognitive deterioration among diabetics (428, 473). Although the underlying mechanism of structural brain changes and cognitive decline in diabetes is not fully understood, asymmetrical atrophy in the right hippocampus is associated with cognitive deterioration (474). Diabetes is closely related to other CV risk factors that may contribute to the increased risk of cognitive decline warranting further investigation (475).

1.4.4. CVD and Sports

Athletes in the NFL and Rugby Union have been increasing in size and BMI at a rate of 2.6 kg/decade and 0.4 kg/m^2/decade, respectively, putting them at an increased risk for cardiometabolic diseases (476). However, the use of BMI in this group is heavily criticised because it tends to overestimate body fat percentage due to the presence of significant muscle mass. Specific increases in body mass for athletes in particular positions can potentially subject them to long-term cardiovascular health risks. This can occur when the increase in body mass

is primarily due to excess body fat rather than an increase in lean muscle mass. In a study by Abe et al. (2018), it was observed that fat-free mass in athletes increased steadily up to 90 kg, while skeletal muscle mass increased in a curved manner, reaching a plateau at 17 kg.m2 beyond a body mass of 120 kg (477). This finding is significant as it indicates a potential upper limit for lean mass accumulation in male rugby union athletes, raising concerns considering the trend of increasing athlete size. In spite of this, these athletes have maintained a healthy CV profile, except for high BP. A cross-sectional study compared 504 active-aged, veteran NFL players to an age-matched population from the CARDIA study and found that NFL players were larger in size and had higher systolic and diastolic BP than their comparative subjects. The NFL players had a systolic BP range of 120-140 mm Hg and diastolic BP range of 80-90 mm Hg, falling into the pre-hypertensive cardiovascular disease (CVD) risk factor category (476). Cardiometabolic diseases are a concern for NFL and rugby athletes due to their increased body mass and BMI. Retired rugby players have a higher prevalence of hypertension and are more frequently prescribed anti-hypertensive medications (478). A retrospective study of American football athletes found a higher prevalence rate of hypertension during their first year of collegiate sport participation compared to non-football athletes. Collegiate male athletes were found to have pre-hypertension or hypertension, with higher BMI in football athletes accounting for some of the difference. However, the odds of developing hypertension decreased by 8.2% with each additional year of football participation (479). Long-term athletes may have mechanisms of adaptability through decades of competitive performance. The *'habituation-sensitisation hypothesis'* could explain how long-term athletes may confer long-term health benefits in the presence of exceeding long-term health risks by how they cope and manage recurrent physiological and psychological stress. Expert-level athletes may have the opportune mechanisms of adaptability through decades of competitive performance to successfully acclimate to stressful environmental and sporting parameters due to repetition and habituation. Whereas novice sporting athletes may be experiencing a competitively stressful situation for the very first time which could impact their capacity for successful coping and conditioning through repetition. Although long-term PA reduces the prevalence of CVD and cancer risk factors, the full extent of this benefit across sport levels and generations is not yet clear.

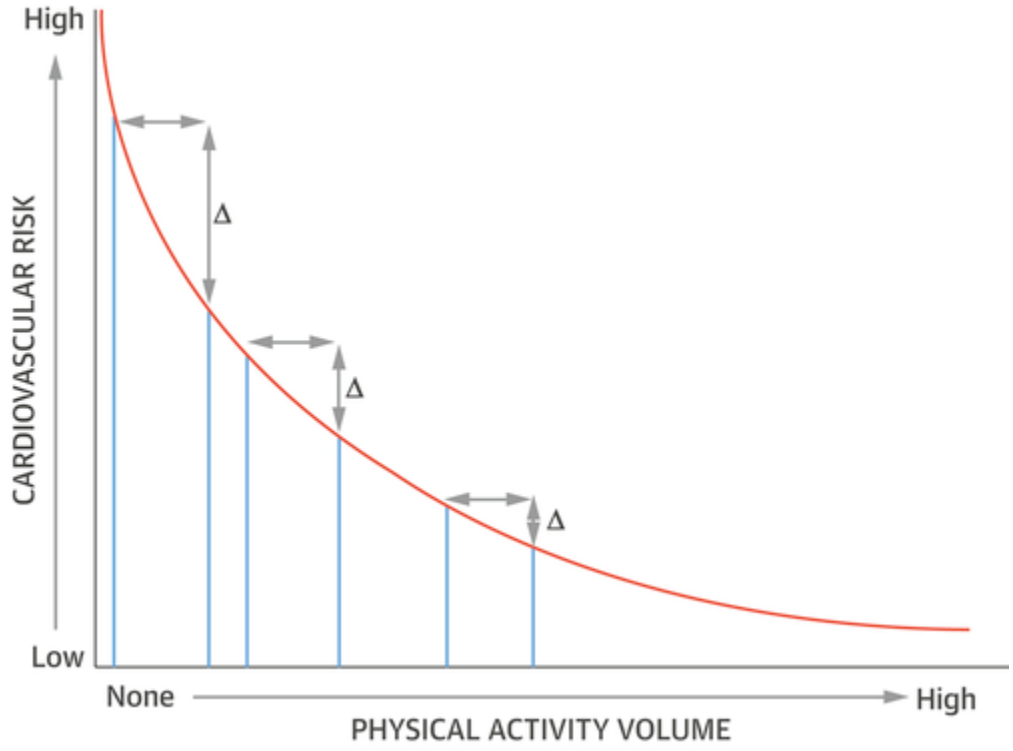

Figure 1.8. *The Curvilinear Relationship Between Physical Activity and Cardiovascular Risk. *Adapted from the work of Powell et al. (480).*

1.4.4a Contact Sports' Congruency with CV Health

Interest in contact sports and their effects on brain health and particularly CV function is growing (392, 481). Research has only recently examined health-related quality of life (HRQoL) and healthy aging among retired professional rugby players to identify modifiable risk factors and enable interventions (3, 482-484). Over a sporting career, athletes engaging in high-intensity exercise may sustain damage to their ascending aorta, leading to undiagnosed hypertension secondary to obesity, a risk for CVD and early morbidity (485, 486). In spite of a 3-fold increase in PA participation during young adulthood, after the age of 65 years, both retired athletes and sedentary individuals possess similar levels of PA (487). Regardless of possessing greater vasodilatory capacity compared to the general population, those who engage in team-based sports with isometric loading may have aortic stiffness similar to that of control subjects (488, 489). Moderate dilation of the ascending aorta may represent a previously unrecognized adaptation to sports via the Laplace law, subjected to larger wall stress, and instigate progressive modification of arterial mechanics over an athletic career (490-492). The prevalence of CVD risk factors and CV health investigated among retired male American football and soccer sporting athletes, showed two-thirds of them were classified as

obese (484). The incidence of hypertension was significantly higher among retired athletes in comparison to the control cohorts. The authors of the review suggest that retired athletes with an elevated BMI possess a similar risk for adverse CV events in the future to that of obese non-athletes in the general population. The *'Cardiovascular Reactivity hypothesis'* suggests that physiological pathways can be negatively affected by psychological stressors, and this can exacerbate the reactions of the CV system among current and retired athletes (493, 494). Individuals who experience significant increases in CV measures in response to stress are more likely to develop CVD or experience mortality up to 28 years after initial exposure (495). Long-term elite competitive participation, especially for ≥15 years, is associated with abnormal anterior aortic effacement and an increased likelihood of aortic root dilatation and ascending aortic dilatation in retired, middle-aged rugby players (496). This has been defined as a *'previously undescribed syndrome of rugby player's aorta, not previously reported in any other sport'*, and is similar to that observed in NFL players (497). Therefore, it cannot be underestimated that contact exposure rates, CV risk factors, and the risk of CVD development may in part, if not collectively, contribute to the cardiac and health status of both prospective and retired professional athletes with significant ties to future brain health.

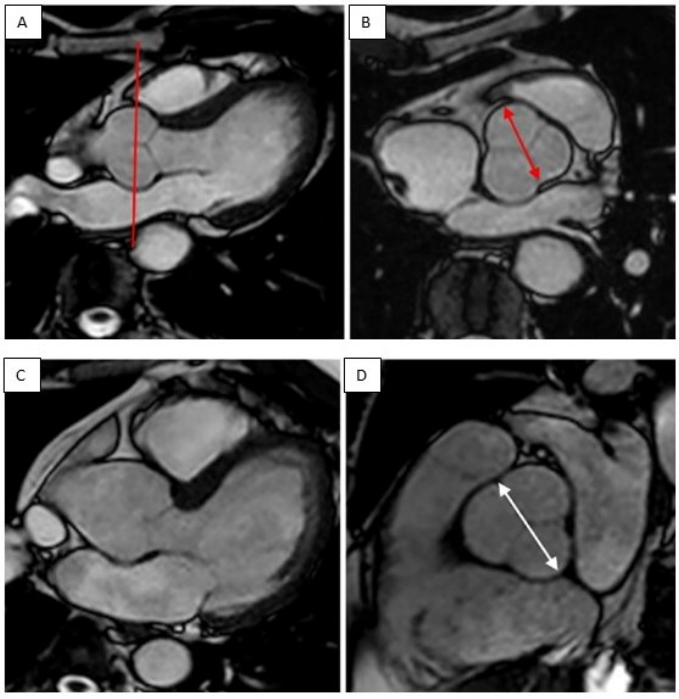

Figure 1.9. *Normal Aorta versus Aortic Dilation in Mid-life Males.* **(A & B)** Aortic root MRI of 44-year-old male with normal aortic dimensions during end diastole; **(C & D)** Magnetic Resonance Imaging (MRI) of mild dilated aortic root in 37-year-old male during end diastole. **Adapted from the work of Hoey and colleagues (498).*

1.4.4b The Intersection between CV and Brain Health in Older Athletes

Contact sports such as rugby and soccer can expose players to endure 600-800 body impacts alongside up to 100 direct head impacts over a competitive season (499), including around 2,000 headers across a 300 match sporting career (500). These may negatively affect CV health and cognitive function in later life impacting visual perception and attentional capacity as a result of concussion history (501-503). RHI's have been associated with changes in brain structure and function, leading to impaired neurovascular coupling and mitochondrial dysfunction that affects CBF velocity and autoregulation. Most importantly this can lead to endothelial cell dysfunction of the vasculature and blood brain barrier affecting cognition and brain health.

Retired rugby players may suffer from cerebral autoregulation impairments induced by inflammation that can cause neuronal cell death and synaptic dysfunction leading to cognitive decrements (504). Professional contact sports athletes, especially those with long careers, are susceptible to physical impacts that can cause vascular and aortic remodelling, affecting cerebral BP, rigidity, and peripheral resistance where the *"site of effacement at the [sinotubular junction] has a predominance of collagen type I fibres with high tensile strength, which may be uniquely susceptible"* (496). The presence of hypertension among retired sporting athletes could impact cognitive and perceptual functioning by reducing regional CBF. Hypertension can narrow the arterioles of the retina potentially reducing sensory, cognitive, and perceptual processing (505). Of significant remark, a recent study was conducted to investigate the effect of a history of SRC on neurovascular coupling (NVC) in former middle-aged rugby players (506). NVC was assessed using a modified Neary protocol with a functional near-infrared spectroscopy (fNIRS) device, and a significant difference in BMI was observed between rugby players and the control group. The results showed reduced response adequacy of O_2Hb in the caudal pre-frontal, dorsolateral pre-frontal cortices, and left middle frontal gyrus, and a higher volume of HHB in the left middle frontal gyrus among those with a history of SRC. This suggests that SRC disrupts cerebral autoregulatory mechanisms, leading to reduced cerebral perfusion pressure, impeding CBF, disturbing perivascular neuronal function and increasing susceptibility of the frontal cortices and associated functional networks through the internal carotid or vertebral arteries. Cerebral autoregulation protects against vascular damage and increases in capillary pressure, but hypertension reduces this protective role and can lead to conditions such as vascular dementia, making cerebral perfusion pressure dependent (504).

The link between CV risk factors, ageing, and cognition could be due to the strenuous and repetitive athletic experiences that athletes encounter during their sporting career. The risk of hypertension for example increases with higher levels of competitive play and doubles the risk of cognitive decline. Nonetheless, hypertension is a modifiable CV risk factor for age-related cognitive decline and dementia, although the impact of mid-life CV risk factors as a collective on later-life cognition is not yet clearly defined.

Overall Aims and Objectives

Reducing ambiguity amongst the senses to discriminate, isolate, and integrate multiple inputs can enhance response times. Modern day sporting athletes are not unlike the gladiators of days past, with enhanced MSI capabilities breeding competitive survival and sporting success, any decrement to such an ability is one to be trained, assessed, and monitored. Spatial correspondence and synchronicity between multiple senses are key factors of MSI and sensory binding (507). In sporting environments, many situations may be viewed as a single perceptual entity with multiple senses unified to enable rapid situational awareness or object detection. However, in the case of the SIFI illusory percept of flashes and beeps, any modifications in the presentation of A-V information may lead to reduced capacity for an effective response. Although such situational paradigms could be trained for in sport to be better cognitively equipped, the use of both physical exercise and neurosensory challenge may provide a new and additional platform for the assessment of concussion, making it superior to self-reports. Furthermore, with present day sporting demands increasing to professionalism, even across many community level sports such as rugby union, it is becoming more apparent that *"bigger is [not always] better"*. There is evident concern for the CV health of current and retired community level athletes; especially among those who aspire to achieve professional status. With RHI expediting the neurocognitive decline associated with the normal ageing process, it may be that cumulative bodily impacts endured in training and competitive play may take a significant and unpredictable toll on the CV health of athletes. Understanding the contributions of the CV health profile of ageing sporting athletes is pertinent among both current and retired athletes in the preservation of brain health in later life.

Overall Aim

The overall aim of the present doctoral work is to investigate the brain health of athletes across a range of contact and non-contact sports using a neuropsychological measure of MSI. The secondary aim is to determine the applicability of the SIFI as an indirect measure of brain health among university level students with and without a history of concussion. The tertiary aim is to establish if exercise influences perceptual performance during the SIFI test following the cessation of moderate and high intensity exercise reflective of a sporting environment. The quaternary aim is to assess and quantify the reliability of the SIFI test. The final aim is to determine the link between the CV health profile of older community level sporting athletes

and the general population in midlife and their relationship with cognition and brain health in later life.

Individual Study Aims and Objectives

Study 1 – The SIFI test's applicability and the assessment of MSI in a sporting context as an index of functional neuropathology related to concussion among young adults

The primary aim of this study was to determine the absolute and relative reliability of the SIFI test through a repeat test protocol. The secondary aim of this cross-sectional, exploratory study was to determine whether the SIFI test may be useful as an assessment of brain health in athletes with a history of sports concussion. The final aim of this exploratory study was to determine if varying intensities of exercise have an influence on SIFI performance in the immediate timeframe post-exercise performance.

Objectives of Primary Aim:

- Determine the Technical Error of the Measurement (TEM), 95% Limits of Agreement, and Intraclass Correlation Coefficient (ICC) for the SIFI test at baseline.
- Determine the test-retest reliability of the SIFI both across separate testing sessions.
- Determine the ICC to assess the internal consistency and between session reliability of the SIFI test from respondents measures of the SOA experimental conditions.
- Determine the level of agreement between baseline testing sessions of the SIFI using a Bayesian Hierarchical Model of analysis.

Objectives of Secondary Aim:

- Determine if there is a significant difference between those with a history of concussion to those without.
- Determine if there is a significant difference in SIFI performance between males and females across the whole population and between sporting type.
- Determine if there is a significant difference in perceptual ability through SIFI performance between those who participate in open skill versus closed skill sports versus those who do not.
- Establish a normative database for SIFI performance of young adults in third-level education and collegiate sport.

Objectives of Tertiary Aim:

- Determine whether an anaerobic exercise (high intensity) protocol will affect perceptual performance in the acute stages following the intervention.
- Determine whether the use of a submaximal, steady-state exercise (moderate intensity) protocol will affect perceptual performance in the acute stages following the intervention.
- Establish the effectiveness of both protocols to induce central and peripheral fatigue through both objective (Lactate, HR) and subjective (Rate of Perceived Exertion [RPE]) measures of assessment to simulate sporting demands.
- Investigate the validity of the SIFI in a sport setting by determining if there are any changes in perceptual performance from pre- to post-exercise.

Study 2 – Systematic Review of CV health in midlife and its influence on cognition in an ageing population

The primary aim of this study is to systematically ascertain the extent to which CV health during midlife has an impact upon cognition and brain health among middle aged adults and subsequently in later life.

Objectives:

- Investigate whether CV risk factors such as hypertension, diabetes, and cholesterol in middle-aged adults influence cognitive processes across multiple domains in later life?
- Investigate whether CV risk factors such as hypertension, diabetes, and cholesterol in middle-aged adults influence cognitive processes across multiple domains in midlife?
- Determine how the associated metrics and sub-categories of CV risk factors at midlife such as diabetes and cholesterol influence cognitive function at midlife and later life.

Study 3 – Midlife CV and Brain Health Among Older Community Sporting Athletes

The primary aim of this cross-sectional, pilot study is to objectively measure and compare the CV health profile, body composition, and neurocognitive functioning capabilities of retired community sporting athletes across a range of contact and non-contact sports.

Objectives:

- Determine the self-reported general health status of older community athletes from contact and non-contact sports.
- Determine the prevalence of CV risk factors among the older sporting population and investigate the contribution of a sporting background.
- Determine the level of neurocognitive functioning of older athletes from contact and non-contact sports, and how this correlates with health-related quality of life and mental health status.
- Determine the MSI capacity and general cognitive function of older athletes from contact and non-contact sports using the SIFI and Montreal Cognitive Assessment (MoCA) test, respectively.
- Determine whether neurocognitive functioning in older athletes is related to a history of self-reported concussion through indices of multisensory processing performance.

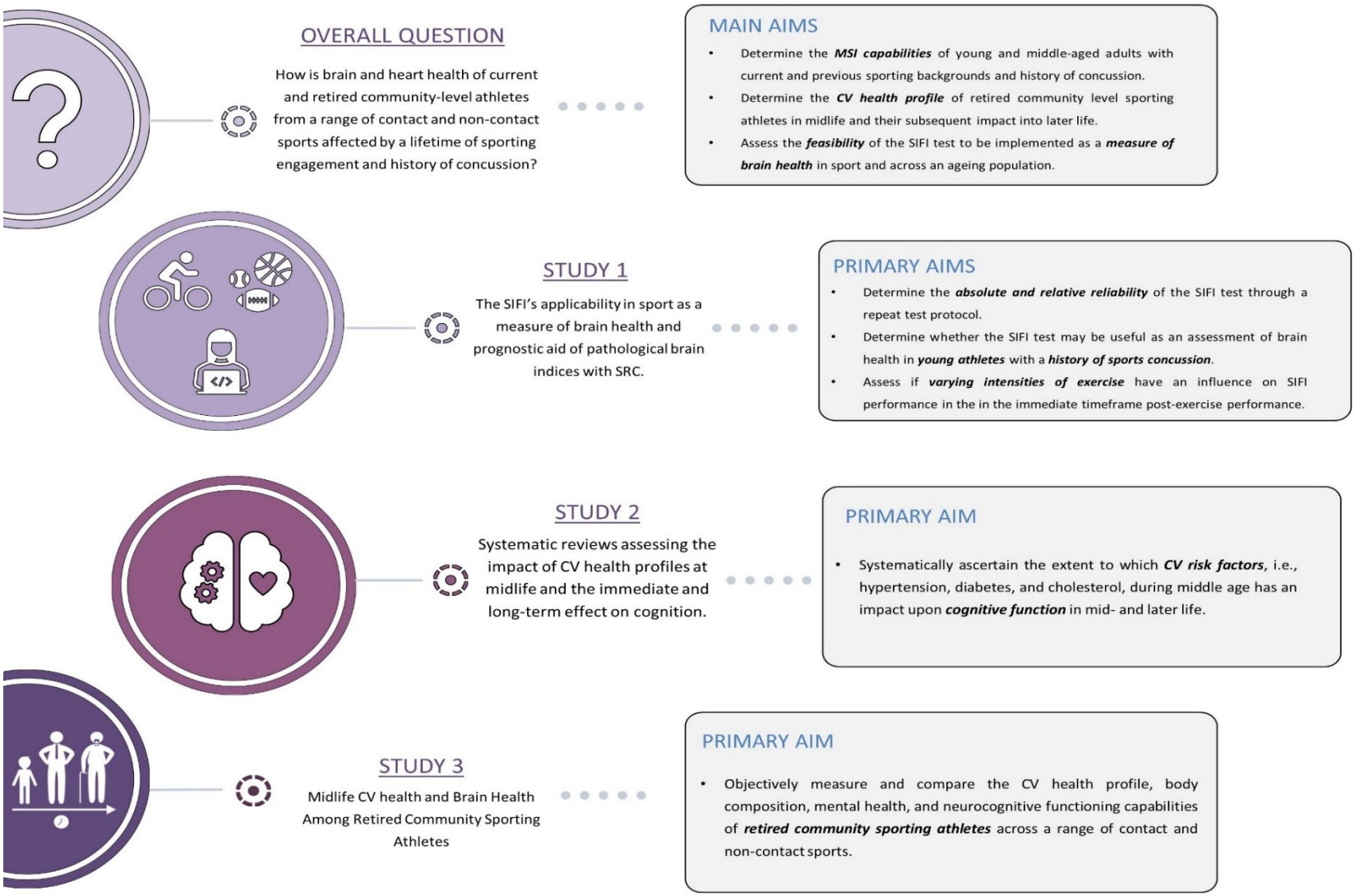

Figure 1.10. *Study Overview.*

2

Chapter Two:

Methods

2.1. Summary of overall methodological approach

This chapter outlines the methodology undertaken for all cross-sectional studies and systematic reviews conducted. The methodologies and procedures used across all investigations will be described in detail below and a brief outline of relevant methods is given in each chapter. **Study One** (**Chapters Three, Four, and Five**) aimed to assess the reliability and applicability of the Sound Induced Flash Illusion (SIFI) in sport as a measure of brain health and prognostic aid of pathological brain indices with SRC amidst the limitations of current cognitive assessment tools. To ascertain how consistent the SIFI test was before introducing it into the sporting world, the levels of agreement and reliability were determined (**Chapter Three**). Results of a pilot study (**Chapter Four**) yielded questions as to which covariables could explain any population-level differences in perceptual performance during the SIFI test. This led to a cross-sectional follow-up study (**Chapters Four and Five**) designed to test the following questions: **(1)** What additional sports-related covariables could account for and explain the variance in results from the pilot study? and **(2)** If the SIFI is to be introduced as a pitch side assessment tool integral to the immediate and long-term well-being of athletes by facilitating early recognition and appropriate management of sports-related concussions, how is test performance impacted by exercise of different intensities simulating sporting demands to minimise the risk of false negatives or positives? Based on these findings and previous evidence (3, 172, 482, 483), it was pertinent to investigate how a lifetime of sporting engagement affects perceptual performance and moreover whether cardiovascular (CV) health influences cognition with advancing age. This led to **Study Two** (**Chapters Six, Seven, and Eight**)**,** a series of systematic reviews investigating the effects of CV risk factors at midlife and their effect on cognition and brain health with age. Finally, **Study Three** (**Chapter Nine**) was conducted as an experimental investigation of any link between CV health and cognitive performance in older recreational athletes at midlife.

2.2. Study One: Protocol 1.1. - Analysing Cognitive Function by Select Covariates of Sex, Sporting Cohort, Skill Type, and Concussion History

2.2.1. Study Design and Assessment Protocol

This study was designed as a cross-sectional, observational study undertaken by two different cohorts across three separate timepoints: an initial pilot study was conducted (N = 131) prior to

the COVID-19 pandemic where soon after a follow up study was conducted (February to March 2020) which was terminated prematurely due to COVID19 pandemic; the follow-up study was subsequently continued post-pandemic (February to April 2023) and the follow-up study data sets were combined for analysis; *see **Figure 2.1.** below for illustrated timeline*. Study participants visited the designated test centre once during the pilot study and on three separate occasions during the follow-up study; *further details regarding all test visits of the follow-up study are outlined in **Protocol 1.2.** below*. Upon arrival at the test centre, all participants underwent a brief health screen including measuring height, weight, and body composition. They then completed sociodemographic and clinical questionnaires (see **Appendices 1.3. – 1.9.**, followed by cognitive testing to assess perceptual performance. Prior to each session, all equipment used was calibrated in accordance with the manufacturer's recommendations. The methods detailed for ***Protocol 1.1.*** below include all baseline measurements of cognitive assessment from both the pilot and follow-up study; *see **Figure 2.2.** below for a graphical representation of the protocol methodology*. It should be noted that additional health and sport-related questionnaires were included in the follow-up.

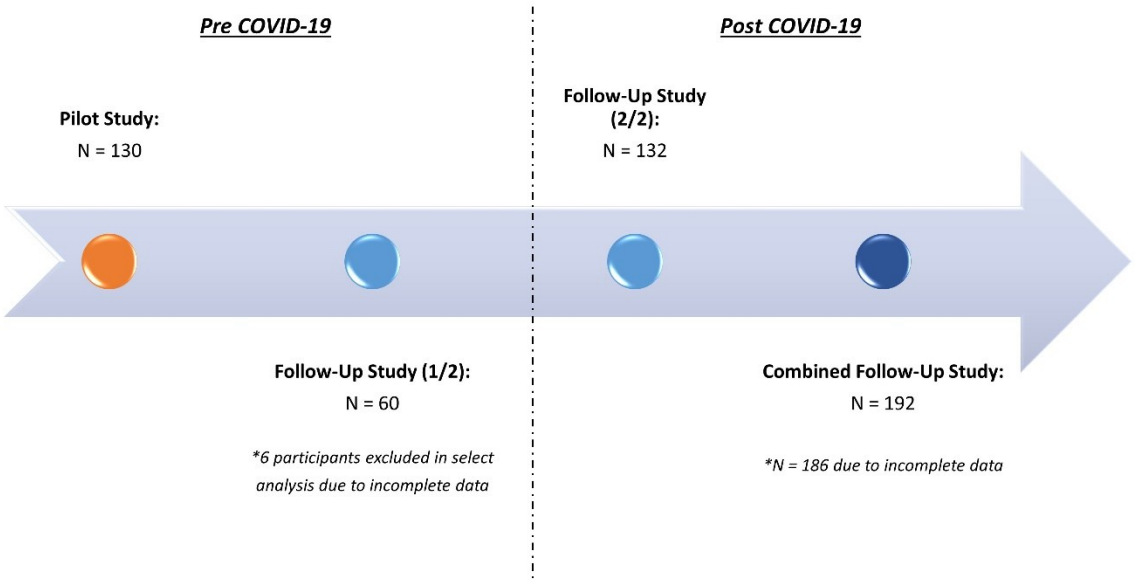

Figure 2.1. *Timeline of Study One Data Collection.*

2.2.2. Eligibility Criteria

Participants were not permitted to partake if they met any of the following exclusion criteria: outside the age range of 18-30 years old, any diagnosed metabolic or cardiovascular disease, any neurological disorder, diagnosis of dyslexia or ADHD, currently taking any prescription medications, and/or is a smoker.

A convenience sample of participants was recruited from the student population of Trinity College Dublin and collegiate sporting teams. The lead investigator obtained informed written consent from each participant. Participants were recruited through flyer advertisement, social media outreach and email contact with sports clubs. Potential participants who expressed an interest in the study were invited to examine the information sheet explaining the nature and purpose of the study with an offer from the research team to fully answer any questions they may have. Participants were given a period of seven days to consider their involvement in the study. The lead investigator individually screened participants' eligibility based on the listed exclusion criteria prior to commencing the study.

Participants were informed of all types of data to be collected via the participant information leaflet and informed consent (*see* ***Appendices 1.10. & 1.11.***). Upon initial contact and at the testing sessions all participants were fully informed of their ability to exercise their rights as a voluntary participant of the study. The participant information leaflet contains the contact information for the Data Protection Commissioner and the Data Protection Officer of Trinity College Dublin if required.

2.2.3. Ethical Approval

Ethical approval was granted by Trinity College Dublin Faculty of Health Sciences (Ref no. 191204 & 221101); see ***Appendices 1.1 & 1.2***. Written and informed consent was provided by all participants in compliance with the Declaration of Helsinki and all later amendments.

2.2.4. Study Population

Participants were recruited into three cohorts: Athletes playing contact sports (contact athletes), athletes playing non-contact sports (non-contact athletes) and controls who were age-

matched, healthy individuals not participating in organised and/or competitive sport. The contact, non-contact and control cohorts were established based on the Rice categorization tool of sporting cohorts (508). Contact included sports such as Rugby Union, Gaelic, Hurling, and Soccer. Non-Contact included Hockey, Rowing, Sailing, Swimming, and Tennis. Participants were also broken down into a sub-population for analysis based on their sporting type: open skill sport, predominantly team-based sports such as rugby or basketball involving dynamic sporting environments characterised by high cognitive load (509), or closed skill sport, determined by a more predictable environment with lower cognitive demand and including sports such as rowing, running or swimming (510). A power analysis for participant recruitment was not performed for the present study as it is exploratory in nature.

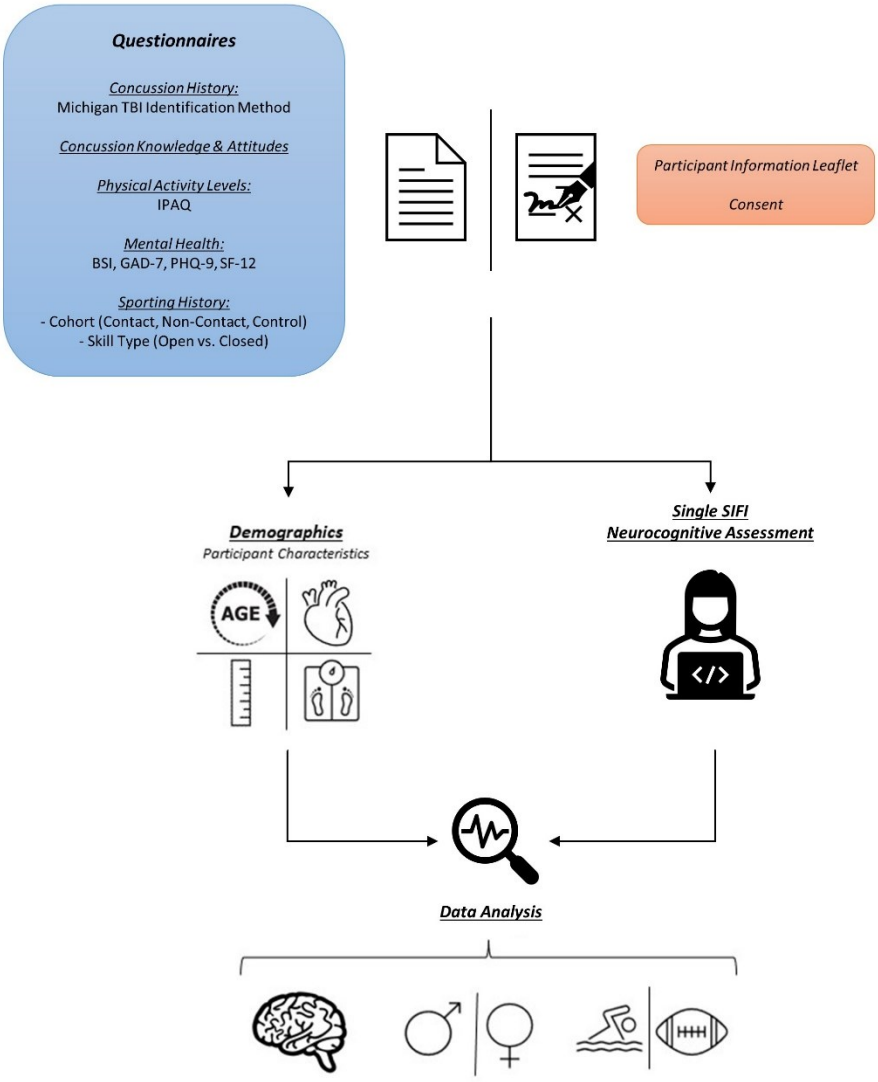

Figure 2.2. *Graphical Representation of the Methodology for Study One Protocol 1.1 of Chapter Four.*

2.3. Study One: Protocol 1.2. – Examining the impact of moderate- and high-intensity exercise on the abilities of multisensory integration.

Protocol 1.2. outlines the assessment procedure during the follow-up study. All participants engaged in both moderate and high intensity exercise on two separate occasions following initial laboratory familiarisation session where they were provided with the participant information leaflet. Participants were informed verbally of all procedures and if willing and eligible to participate, written and informed consent was obtained. This was followed by completion of baseline SIFI cognitive assessment along with several health-related questionnaires, and

acquisition of anthropometric and demographic data as detailed below. A minimum of seven days following this baseline visit, participants underwent two exercise testing sessions in the laboratory. The two exercise sessions were separated by at least 48 hours whereby all participants completed baseline and follow-up sessions within a 10-day period. The sessions took place in the same controlled testing environment and were designed to simulate physical exertion of different intensities that might be experienced during different phases of match play. The sequence of the exercise sessions was unfortunately not randomised due to time-limited access to equipment and logistical organisation, such that all individuals completed the moderate-intensity aerobic session first and the high-intensity anaerobic session second.

Participants undertook a high-intensity interval anaerobic exercise protocol adapted from the work of Pearcey and colleagues (511) and a moderate-intensity steady-state exercise protocol adapted from the work of McInnes and colleagues (512). Pearcey and colleagues' high-intensity protocol entailed a sequence of 10 maximal 10-second sprints, followed by 180 seconds of recovery. The session commenced with a 5-minute warm-up at 60–70 rpm and a workload of 50 watts. To accommodate the longer sprint duration, a 3-minute rest interval was included. Before each sprint, participants underwent a 20-second warm-up phase at 50 watts and 100 rpm, followed by verbal encouragement to exert maximal effort during the 10-second sprint. They were instructed to initiate acceleration after the electromechanical brake engagement to avoid power overestimation resulting from increased pre-brake acceleration. On the other hand, MacInnis and colleagues' moderate-intensity protocol featured a 5-minute warm-up at 25 W, with subjects maintaining a consistent cadence of approximately 80 rpm throughout each session. Upon arrival to the lab for their second and third sessions and once participants were satisfied, they completed the PAR-Q and Wellness questionnaire. This was followed by pre-exercise assessments consisting of a SIFI cognitive test, resting lactate measurement, and resting heart rate (HR). Blood lactate was taken from the earlobe and analysed using the Lactate Pro 2 analyser. Saddle height and handlebar positioning were adjusted separately for each participant. This was done to ensure that the knee was nearly fully extended, and the foot was parallel to the ground. Immediately following each exercise bout all participants retook the SIFI test, had their lactate and HR measured, and provided their level of subjective fatigue by RPE. Before any exercise testing was undertaken and ethical approval granted, all researchers completed CPR training (online and in-person).

2.3.1. Moderate-Intensity Steady State Protocol

Completion of the moderate intensity protocol was identical in delivery and equipment used for both cohorts of participants in the follow-up study. Upon arrival to the lab, participants were informed verbally of all procedures. Once participants were satisfied, they completed the PAR-Q and Wellness questionnaire. This was followed by the pre-exercise assessments consisting of a SIFI test, resting lactate measurement, and resting HR. This submaximal steady state bout consisted of aerobic exercise maintained at a HR of 70% of their predicted maximal HR in line with MacInnis et al. alongside previous investigations and meta-analytic findings (512-514). The age-adjusted maximum heart rate formula was calculated using the equation 220 minus age. Participants began with a 5-minute warm-up on the stationary ergometric bike (Monark Ergomedic 874E, Vansbro, Sweden) at 60-70 rpm and no resistance. Participants then underwent 20 minutes of steady state continuous cycling at 60 rpm with resistance appropriate to each participant to sustain 70% of their max predicted HR for the duration of the bout. Resistance was added by placing weighted loads on the bike and multiplied by the cadence of 60rpm corresponding to Load watts: i.e., 1kg @ 60 rpm = 60 watts, 1.5 Kg = 90 watts, 2 Kg = 120 watts, 2.5 Kg = 150 watts. Once completed, a five-minute cool down with no resistance was performed at a cadence of 60rpm. Upon completion of the 20 minutes continuous exercise, blood lactate and HR was taken and the SIFI test performed.

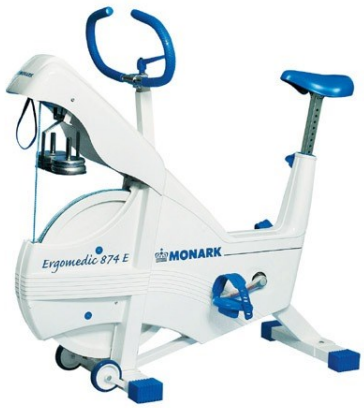

Figure 2.3. *Models of Stationary Bikes for Moderate intensity Exercise Protocol.*

2.3.5. High-Intensity Anaerobic Protocol

Participants had their lactate measured and recorded prior to commencement of exercise. Due to the COVID-19 pandemic two different bikes for this follow-up study. All participants pre-

COVID (n = 60) began with a 3-minute warm-up on a stationary wind-braked ergometric bike (Wattbike Pro, Wattbike Ltd., Nottingham, U.K.) at 60-70 rpm and a workload of an air and magnetic resistance of 1. All participants post-COVID (n = 132) completed the same workload except this was carried out on a stationary electromagnetically braked ergometer fitted with toe-clip pedals and straps (Lode Excalibur Sport 2006, model no. 20110272, Lode BV, Groningen, The Netherlands), at 50 watts post-Covid which provided measures of power (in W) and cadence (in rpm). In the follow-up cohort, all participants performed a series of 10 maximal 10-second sprints, with each sprint followed by 180 seconds (equivalent to 3 minutes) of active rest and recovery. For the first cohort of participants pre-COVID the resistance setting differed for each participant according to weight (kg) and sex as determined by the 'guide for short sprint intervals provided in the WattBike manual (*see* ***Appendix 1.15.****).* With twenty seconds remaining in the rest period, the wind and magnetic break settings were set to the required resistance. The experimenter gave the participant a five second count down preceding the sprint and on the '*go*' command the participant cycled at a maximal effort for ten seconds, maintaining a seated position throughout. Verbal encouragement was given throughout the ten second interval. Immediately after the sprint, the resistance was reset to the lowest air and magnetic resistance settings for the three-minute rest period. The participant was instructed to keep their legs moving at a cadence of 50 rpm, with minimal power to allow recovery but prevent lactate build-up.

Figure 2.4. *Models of Stationary Bikes for High intensity Exercise Protocols.*

For the post-COVID cohort, all participants completed the exercise bout on a stationary Lode ergometer bike. All data was collected and exported from the Lode Manager Software (Version 10.4.5, Serial no. 20160155) to Excel. The resistance setting for each participant during the sprints was individualised and calculated to determine the torque. The resistance (torque factor) was set at 0.70 and 0.60 $N \cdot kg^{-1}$ of bodyweight for males and females respectively. Such factory settings are preferred by practitioners and scientists because they accurately simulate overground sprinting and are unlikely to be changed since determining ideal resistive load to optimise power production is a difficult athlete- and equipment-specific technique. Although not established for this cohort, absolute muscle mass, strength, power, body weight and cadence are the most accurate predictors of performance output in repeat sprint testing allowing for large variation in results but ultimately inducing relative fatigue (515, 516). Peak and mean power outputs (PPO and MPO) were expressed as absolute measures as well as relative to body weight, and time to reach peak power (TTP, in s). Power output measurements were recorded for all completed 10 second sprints including:

- Peak, Mean and Minimum Power (Watts)
- Peak, Mean and Minimum RPM
- Anaerobic Capacity (Mean Power ÷ Body Weight [Watts/Kg])
- Anaerobic Power (Peak Power ÷ Body Weight [Watts/Kg])
- Total Work (Average Watts x Test Duration [Joules])

HR was recorded alongside RPE after each of the 10 sprint intervals. Following the 5th sprint lactate was measured again from the earlobe. Participants were instructed verbally to complete the rest period on the bike and then begin the 6th sprint interval upon command– participants did not dismount the bike during this period. Following completion of the 10th sprint, the participant then dismounted the bike to have their post-exercise blood lactate and HR assessed and complete the SIFI immediately after.

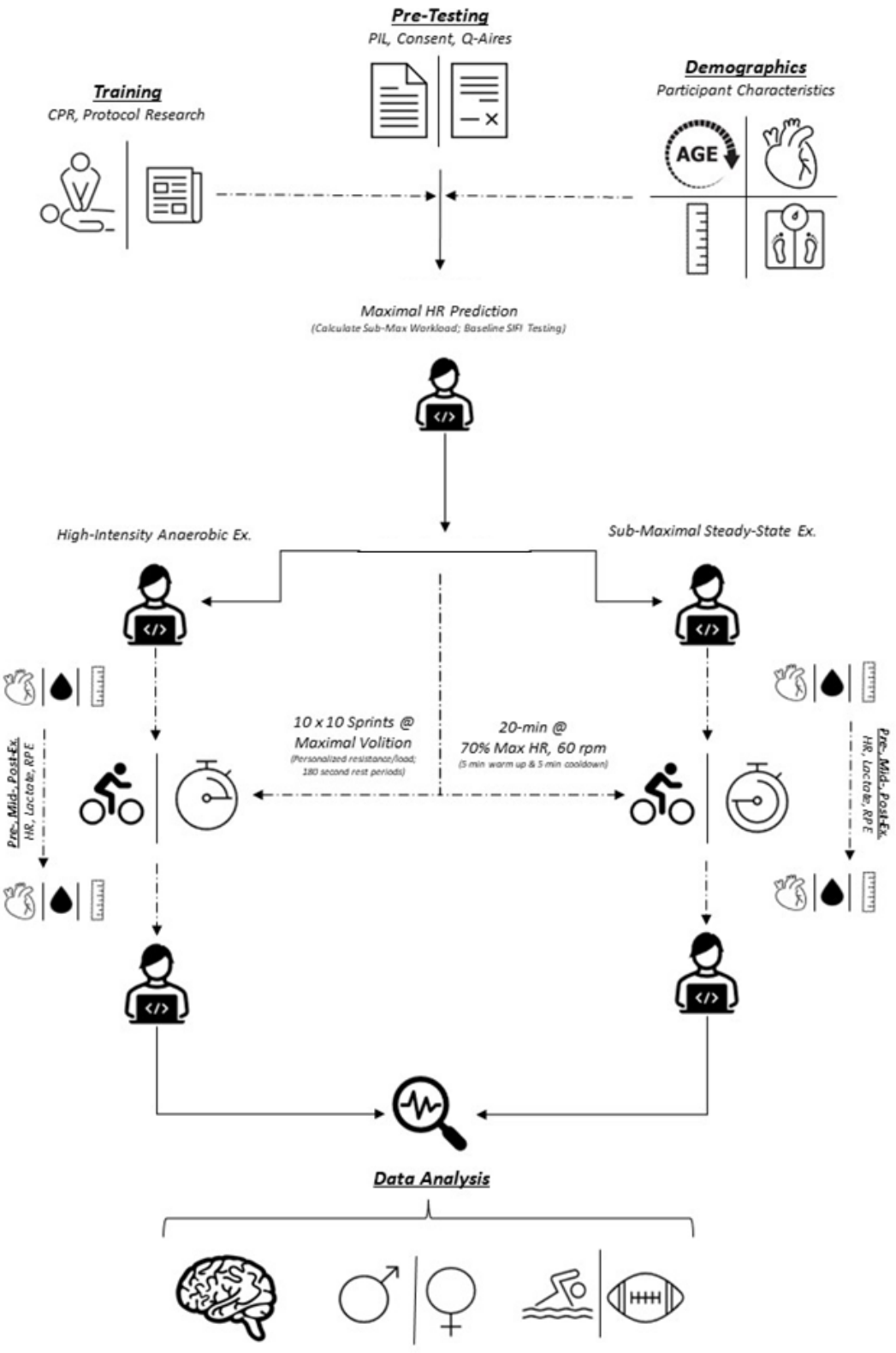

Figure 2.5. *Graphical Representation of the Methodology for Study One Protocol 1.2 of Chapter Five.*

2.4. Outcome Measures

2.4.1 Anthropometric Measurements

Body composition was determined by anthropometry, and measurements taken included height, weight, and body mass index (BMI).

Barefoot standing height was measured using the Seca 222 Mechanical telescopic measuring rod with large measuring range (Seca, Birmingham, United Kingdom). All participants height were taken to the nearest 0.1cm. Participants were instructed to stand without shoes, with their backs facing the wall, legs together, arms down by their sides, and mid-axillary line parallel to the wall. A line travelling between the tragion (front of the ear) and the lowest point of the eye socket was used to put the participant's head in the Frankfort horizontal plane, the standard plane used for accurate head alignment. The participant's legs were straight, and shoulder blades and buttocks were in contact with the uprights. The individual was asked to keep their arms relaxed at their sides. The headboard was lowered until it met the crown of the head and flattened the hair.

The weight of each participant was measured using a digital scale to the nearest 0.1 Kg. Participants were measured in one layer of light clothing and with shoes removed.

BMI uses the formula *(weight (kg) ÷ height (m) squared: Kg/m^2)* to calculate an individual's relative weight based on height as an indicator of health. The individual can be classified as: (1) underweight (BMI: < 18.5 kg/m^2), (2) normal (18.5 ≤ BMI ＜ 25 kg/m^2), overweight (25 ≤ BMI ＜ 30 kg/m^2) and obese (BMI ≥ 30 kg/m^2).

Figure 2.6. *Height Measurement Stadiometer. (Images Courtesy of Seca)*

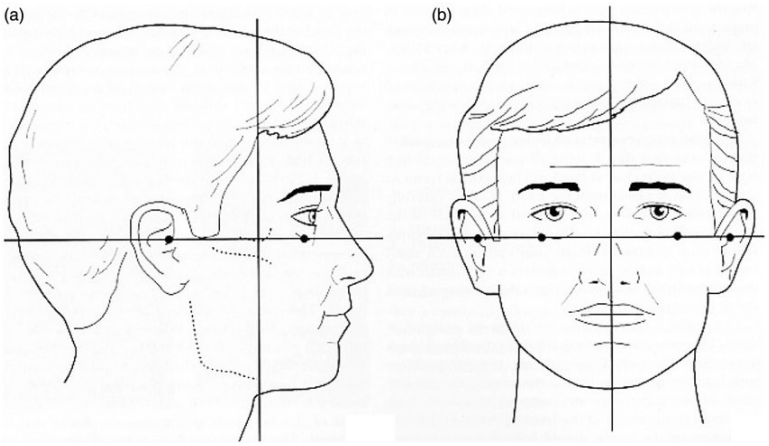

Figure 2.7. *Frankfort Horizontal Plane.*

**Image adapted from the work of Capon and colleagues (517)*

2.4.2 Clinical, Health-Related, and Sporting Questionnaires

2.4.2a Concussion History

Concussion history was self-reported and acquired from each participant using the Michigan TBI Identification Method (MTBIIM) (518). The MTBIIM has a fair level of agreement between self-reported concussion and medically diagnosed concussion (22). Prior to completion of the questionnaire, participants were given a written copy of the definition of concussion defined as a form of TBI induced by biomechanical forces in accordance with the most recent published definition by the international Concussion in Sport Group in 2017 based on the most up to date guidelines (26), see **Appendix 1.7**. The MTBI concussion questionnaire was designed to assess concussion history among athletes. It is a self-report pen and paper questionnaire that looks at a person's lifetime history of concussions, both sports-related and non-sports-related. It evaluates each concussion based on similar criteria such as the injury mechanism and timing, the presence or absence of related loss of consciousness, and the symptoms reported. A lifetime history of concussion was obtained to assist research into the links between sports-related concussion history and measures of brain health and cognition using the SIFI. The MTBI identification method is a NIH Common Data Element (518) (see **Appendix 1.7.**).

2.4.2b Concussion Knowledge and Attitudes Questionnaire

This survey was based on previously published work to investigate concussion reporting behaviour, knowledge, attitudes, and education in Irish camogie players and jockeys (519-521). The survey took approximately 5-10 minutes to complete and was aimed at better understanding the participants' understanding of concussion. The definition of concussion used for the purpose of this thesis was as a "short lived impairment of neurological function that resolves spontaneously", whereby "forces [are] transmitted to the head [that] disrupt neuronal membranes, which cause a metabolic imbalance within the brain" and "may or may not involve a loss of consciousness" in line with the most recent research (1). Data was analysed based on concussion history (number of previous suspected and medically diagnosed concussions), reporting behaviour (behaviour following most recent concussion, reasons for not reporting past concussions), concussion knowledge (correct recognition of signs and symptoms, general concussion knowledge, and concussion management), and attitude (intention to report in a variety of scenarios, reasons for not reporting concussion in the future). A concussion knowledge score was calculated by scoring and summing correct responses in the concussion knowledge section (correct = 1, incorrect = 0), with a maximum range of 0–29 and higher scores indicating greater knowledge (522); see **Appendix 1.9.** for full details.

Table 2.1. *Variables of Analysis for Concussion Knowledge and Attitudes.*

Category	Variables
Concussion History	○ Number of Suspected Concussions in the Past ○ Number of Concussions Diagnosed Previously
Reporting Behaviour	○ Reporting After Recent Possible Concussion (Yes/No) ○ Playing After Recent Possible Concussion (Yes/No) ○ Reasons for Not Reporting
Concussion Knowledge	○ Recognizing Concussion Signs and Symptoms Correctly ○ Correct Responses on Concussion Knowledge Questions
Concussion Attitude	○ Intention to Report a Possible Concussion in Various Circumstances ○ Causes for Failure to Report a Concussion in the Future ○ Response to a Teammate's Concussion ○ Recommended Sources of Information for Reporting a Possible Concussion

**Adapted from the work of Leahy and colleagues (522).*

2.4.2c The International Physical Activity Questionnaire (IPAQ) Short Form

The IPAQ is a 7-day recall questionnaire which has been validated across 12 countries (523). As the IPAQ-SF version is easier and more realistic to complete than the lengthy form, it is suggested for population prevalence investigations where time is restricted. This measure assesses the types of intensity of physical activity (PA) and sitting time that people do as part of their daily lives. The IPAQ-SF questionnaire was specifically designed for adults between the ages of 18-65 years and pertains to 4 distinct domains: **1)** Transport, **(2)** Work, **(3)** Household activity and gardening tasks, and **(4)** Leisure time activity inclusive of participation in exercise and sport (523). This questionnaire also poses questions surrounding the amount of time spent sitting being indicative of sedentary behaviour. Each of the four sections' questions record the value of moderate and vigorous activity through the metrics of the number of days per week, and the number of hours and minutes per day. Practical examples pertaining to what constitutes a specific level of PA; mild, moderate, or vigorous; is provided for each question. The IPAQ is based on the PA levels of the past 7 days, or in our case for standardisation and given the time of data acquisition coincided with college exams, it was based on an average week. The short version has been previously evaluated to maintain a good reliability of 0.76 (523), and enables a more insightful understanding of the time and pattern of PA undertaken at population level (see **Appendix 1.3.**).

For the purpose of this study activity levels were divided according to walking, moderate and vigorous activity, and the results scored according to 'Metabolic Equivalent Task' (MET) minutes per week. MET's represent the value of energy expenditure carried out depending on how long, how intense, and how often an activity is carried out. MET's can be classified as a multiple of one's estimated resting energy expenditure. Walking activity for the present study was classified as 3.3 MET's, moderate activity as 4 MET's, and vigorous activity as 8 MET's (524). All PA levels were measured in minutes. MET's were calculated using the following equation: Level of PA MET Score (3.3, 4, 8) x minutes engaged in activity x frequency of activity engagement (days per week). MET scores for all subjects across selected questions were summed to derive overall PA levels.

According to the IPAQ scoring system, participants were classified as having either high levels of PA (i.e., 7 or more days of any combination of walking, moderate intensity or vigorous intensity activities achieving a minimum total physical activity of at least 3000 MET minutes a week), moderate levels of PA (i.e., 5 or more days of any combination of walking, moderate intensity or vigorous intensity activities achieving a minimum total physical activity of at least 600 MET minutes

a week), or low levels of PA (i.e., not meeting any of the criteria for either high of moderate levels of PA).

2.4.2d Brief Symptom Inventory (BSI)

The BSI-18 is a commonly used standardised screening instrument for objectively assessing psychological discomfort and mental illnesses (see **Appendix 1.8.**). It is the most modern and streamlined version of a series of comparable instruments created in the 1980's (525). It is a 53 item self-report symptom inventory that assesses nine patterns of clinically relevant psychological symptoms and is a brief version of the Symptom Checklist List 90-R (526). The BSI-18 as implied is made up of 18 different descriptions of emotional and physical issues (527). The measure is divided into three six-item scales: somatisation, depression, and anxiety, as well as a Global Severity Index (GSI). Respondents are asked to rate how concerned they are by the complaints on a scale of zero to four (zero being not at all and four being a lot). Depression, anxiety, and somatization are assessed throughout the preceding week. Each BSI-18 subscale has six items from the three relevant BSI subscales: six items from the Somatization, Depression, and Anxiety dimensions. The BSI provides three symptom scores: somatisation (items 1, 4, 7, 10, 13, 16), depression (items 2, 8, 11, 14, 17), and anxiety (items 3, 6, 9, 12, 15, 18) (max for each = 24) as well as an overall GSI score (max score = 72), and enables cases of clinical distress to be identified using normative data proposed by Derogatis (2001). Higher scores reflect greater distress.

The GSI is a total score that measures the respondent's overall level of psychological suffering. The GSI is a brief, quantitative assessment of a respondent's present level or depth of psychological distress. In general, the GSI is the best single predictor of a respondent's overall emotional adjustment or psychopathologic condition. The remaining two global indices include Positive Symptom Distress Index, and Positive Symptom Total which measure the intensity of symptoms, and number of reported symptoms, respectively. The Somatisation (SOM) items represent discomfort induced by the impression of physical malfunction, with a focus on symptoms emerging from cardiovascular, gastrointestinal, and other physiologic systems with strong autonomic mediation. Moreover, symptoms on the Somatisation dimension are frequently reported in highly somaticized anxiety and depressive illnesses. The Depression (DEP) elements reflect the basic symptoms of several clinical depression syndromes. Disaffection and dysphoric mood symptoms are mentioned, as are self-deprecation, anhedonia, loss of hope, and suicide thoughts. The BSI-18

handbook instructs users that t-score elevations of 63 (representing the top 9% of normative respondents) indicate considerable scale elevation on each of the three BSI-18 scales, DEP, ANX, SOM, or on the overall GSI.

2.4.2e Generalised Anxiety Disorder Assessment (GAD-7)

The most prevalent type of anxiety is generalised anxiety disorder (GAD), which is characterised by excessive and persistent concern. Due to its simplicity and operability, the Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition (DSM-IV) is a frequent tool used in the screening of generalised anxiety disorders (528). The GAD-7 has been translated into multiple languages over the last two decades (529, 530), with an established reliability, validity, and diagnostic capability among the general population and university level students in previous psychometric investigations (531-533). The GAD-7 is a 7-item self-report questionnaire designed to test for the existence of general anxiety disorders in the preceding two weeks (534, 535); see **Appendix 1.5**. Seven statements concerning worry or somatic tension are scored on a four-point Likert scale, as follows: 0 (never); 1 (a few days); 2 (more than half the number of days); and 3 (almost every day) indicate the frequency of GAD symptoms. The GAD-7 summed score runs from 0 to 21, with cutoff points of 5, 10, and 15 used by researchers to categorise anxiety as none/normal (0-4), mild (5-9), moderate (10-14), or severe. (15–21).

2.4.2f Patient Health Questionnaire (PHQ-9)

The PHQ-9 is a regularly used tool for screening and monitoring depression. The nine-item survey is simple to administer and comprehend, requiring little time. The respondent is asked to think back on any symptoms they may have had in the two weeks prior and rank how bothersome they were on a scale of one to 10; (zero being not at all and three being nearly every day) (see **Appendix 1.4.**). There are pre-established cut-off thresholds to identify respondents as having serious depression, moderate depressive symptoms, or being in the normal range (536). The PHQ-9's validity and reliability have been extensively studied and evaluated, and a high level of internal validity has also been discovered (537).

2.4.2g Short Form 12 (SF-12) Health Survey

Physical and mental health was assessed using the SF-12 which is now widely employed across the literature (see **Appendix 1.6.**). The reliability and validity of the SF-12 have been established and quantified over the last two decades (538, 539). The SF-12 is a condensed form of the SF-36, a widely used HRQoL measurement (540). The survey consists of 12 questions drawn from the 36-item short-form (SF-36), asking questions on characteristics of both physical and mental health and covers the same eight categories of health outcomes. They include bodily pain (BP), general health (GH), vitality (VT), social functioning (SF), role emotional (RE), and mental health (MH). Each inquiry receives a score using a number ranking system (NRS). The PCS-12 and MCS-12 scores, each represented by six items (see **Table 2.2**. below), were calculated using published techniques in this study. For both the PCS and MCS components, the scale runs from 0 to 100, with higher scores reflecting improved physical and mental health functioning. A PCS-12 score of 50 or less has been proposed as a cut-off for determining a physical condition, but an MCS-12 score of 42 or less may be suggestive of 'clinical depression' (540).

Table 2.2. *Measurement Properties of the SF-12*

Scales	Item No.	Contents	Response Categories
Physical Component Summary (PCS-12)	1	General health	Excellent/very good/good/fair/poor
	2	Moderate activities	Limited a lot/limited a little/not limited at all
	3	Climb several flights of stairs	Limited a lot/limited a little/not limited at all
	4	Accomplished less (physical)	All of the time/most of the time/some of the time/a little of the time/none of the time
	5	Limited in kind of work	All of the time/most of the time/some of the time/a little of the time/none of the time
	8	Pain—interference	Not at all/a little bit/moderately/quite a bit/extremely
Mental Component Score (MCS-12)	6	Accomplished less (emotional)	All of the time/most of the time/some of the time/a little of the time/none of the time
	7	Did work less carefully	All of the time/most of the time/some of the time/a little of the time/none of the time
	9	Calm and peaceful	All of the time/most of the time/some of the time/a little of the time/none of the time
	10	Energy or vitality	All of the time/most of the time/some of the time/a little of the time/none of the time
	11	Downhearted and blue	All of the time/most of the time/some of the time/a little of the time/none of the time
	12	Social limitations	All of the time/most of the time/some of the time/a little of the time/none of the time

2.4.3. Objective Cognitive Assessment

2.4.3a The Sounds Induced Flash Illusion (SIFI) Task

The SIFI task was adapted from previously published studies (96, 97, 268), and employed in the most recent work of Cunningham and colleagues (172). It was deemed unnecessary to assess visual or hearing acuity in the current study; participants were presumed to have thresholds within the normal limits due to the pre-defined age range of 18-30 years old. The SIFI task involves the presentation of brief visual (flash, F) and auditory (beep, B) events, where the participant will report the perceived number of flashes or beeps. The experiment was designed to be self-paced with no importance placed on the speed of response. The fission illusion would occur when participants were presented with a single visual flash in conjunction with two auditory beeps (*i.e.,* 1F2B) and would subsequently perceive two visual flashes across a designated range of stimulus onset asynchronies (SOA's). Control conditions were classified as unimodal (1F0B, 2F0B, 0F1B, and

0F2B) and bimodal congruent conditions (1F1B and 2F2B) to establish baseline performance for each participant. These baseline measures of correct responses to unisensory stimuli were to ensure participants did not differ in the ability to perceive either visual or auditory information. The unisensory visual flashes were presented as either one or two flashes with no auditory beeps at an SOA of 70ms (*i.e.,* 1F0B and 2F0B) and instructed to report the perceived number of flashes. The unisensory visual conditions were interleaved among the multisensory conditions of the first stimulus block. The auditory only conditions were presented as part of a separate block signified by a written command appearing across the computer screen informing the participant to report how many beeps were perceived, and also interleaved among the multisensory conditions. They were presented with either a single auditory beep (0F1B) or two auditory beeps (0F2B) at SOA's of 70ms, 150ms, and 230ms.

Two different stimulus conditions were presented for the multisensory trials, either congruent or incongruent. The congruent trials consisted of the same number of visual flashes being presented with the same number of auditory beeps; either 1 flash was synchronously paired with 1 beep (1F1B) or two flashes were synchronously paired with two beeps (2F2B) at SOA's of 70ms, 150ms and 230ms, see **Figure 2.8**. The incongruent trials were representative of the illusory multisensory fission trials, whereby one visual flash was always paired with two auditory beeps (1F2B) with auditory SOA's of 70ms, 150ms or 230ms, see **Figure 2.9**. One of these auditory beeps was always presented in synchrony with the visual flash, the second auditory beep would either precede or lag behind the visual-auditory pairing to bring about six SOA pairings (*i.e.,* ±70ms, ±150ms, and ±230ms). These time delays are herein referred to as negative and positive SOA's whereby the beep is presented before or after the visual-auditory pairing, respectively. Similar to the unisensory visual trials, participants were instructed to report the perceived number of visual flashes for all multisensory conditions and ignore the auditory beeps.

There were 16 conditions each presented four times yielding a total of 64 trials per participant. The experiment was designed as a 3 x 16 mixed design, with the cohorts (Contact, Non-Contact, Control) and the unisensory and multisensory conditions, inclusive of the SOA conditions (±70, 150, 230 ms) as the factors of analysis. The inter-individual presentation of the visual and auditory stimuli was randomly generated to reduce the possibility of a future learning effect.

The SIFI task was performed in a darkened room, with the participant seated approximately 60cm from the laptop screen. The laptop and screen were adjusted with respect to height and

location to ensure all visual angles of stimuli presentation were accounted for. The SIFI task was carried out using the following devices: a Packard Bell EasyNote Notebook laptop, a Dell laptop, and a HP laptop all with a refresh rate of 60 Hz and spatial resolution of 1366 x 768 pixels. Stimuli were generated using Matlab and displayed using the Presentation software package (Neurobehavioral Systems Incorporation, Berkeley, California). The visual stimulus was a hard-edged white annulus with a diameter with a visual angle of 1.5˚ and luminance of 31.54 foot-lambert displayed for 17 ms against a black background, and 5˚ below the fixation crosshairs. The sound stimulus was a momentary auditory tone or beep at a frequency of 3.5 KHz. The auditory stimulus was presented for 10 ms at 65 decibels with a 1 ms ramp. All participants wore headphones for the duration of the task. The SiFI task was blocked into two distinct sections consisting of all visual unisensory and multisensory trials in the first bock and followed by the auditory only trials block. Participants also undertook a practice block to familiarise themselves with the testing procedure. The practice block was comprised of one trial from each of following conditions: 1F2B (SOAs of 70, 150, and 230ms), 1F1B, 2F0B, 2F2B, and auditory-only trials (0F1B; 0F2B with SOAs of 70, 150, and 230ms). This yielded a total of 11 practice trials per participant. Each participant was guided through the short practice test by the lead investigator. All participants completed the practice block just once before completing the two experimental blocks. Participants were informed to maintain their gaze at the centre white crosshairs during each trial. Each trial was initiated by pressing the spacebar on the keypad to bring about the presentation of the white fixation crosshairs in the middle of a black screen and followed by the appearance of one or two brief visual flashes. These visual flashes were accompanied by one, two or no auditory beeps, depending on the experimental condition. Participants were instructed to note how many flashes they had perceived after each trial, whilst ignoring the auditory beeps, by using the response keys on the keyboard. This process would be continued for each trial until the end of the first block. If participants did not perceive any visual flashes (i.e., during the auditory-only trials), then they were to report perceiving no visual flashes and report the number of auditory beeps instead. During the second block of auditory-only trials, participants were instructed to now report the number of auditory beeps that they heard. Similar to the first block, the end of a trial would be denoted by the absence of the white fixation cross hairs and the participants were instructed to press a numbered response key and then the spacebar to initiate the next trial. The SIFI task lasted approximately 7 minutes for each participant.

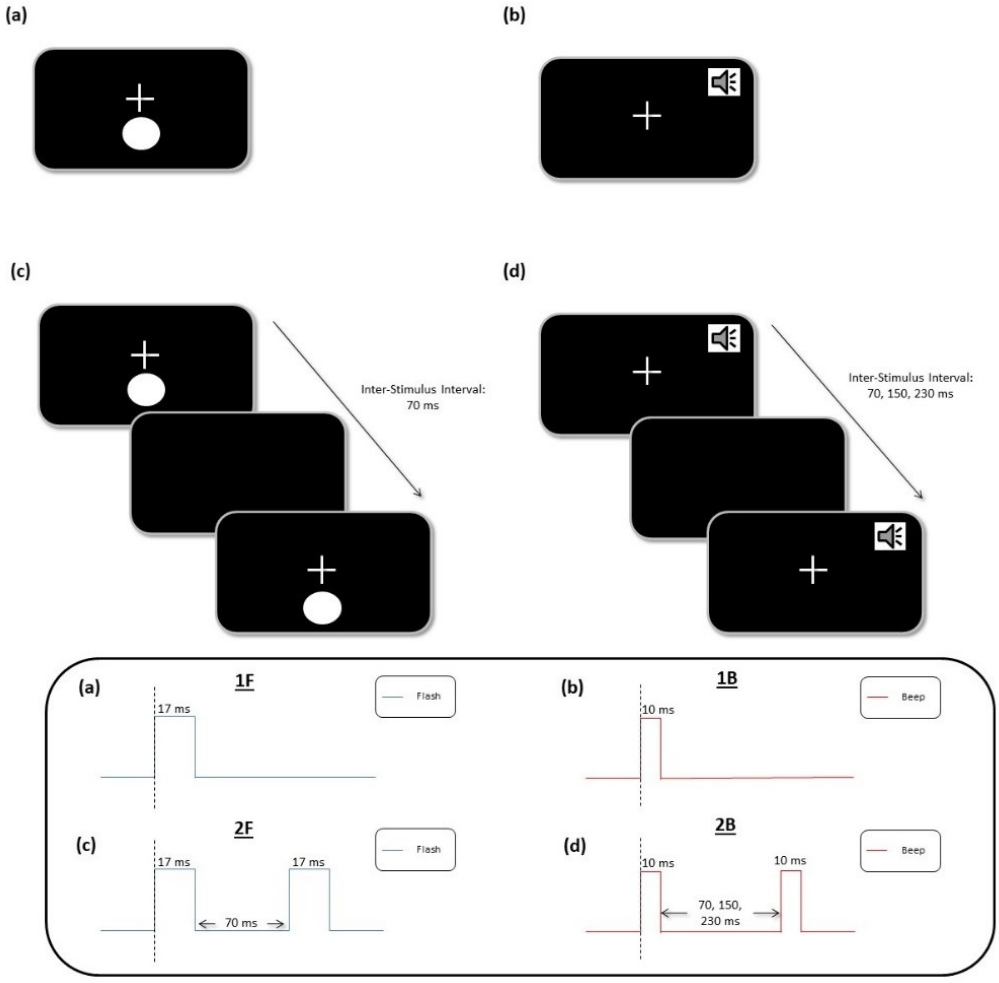

Figure 2.8. *Unisensory SIFI Conditions.* **(a)** One visual flash presented for a duration of 17 ms. **(b)** One auditory beep presented for a duration of 10 ms. **(c)** Two visual flashes with SOA of 70 ms. **(d)** Two auditory beeps presented with SOA's of 70, 150, and 230 ms.

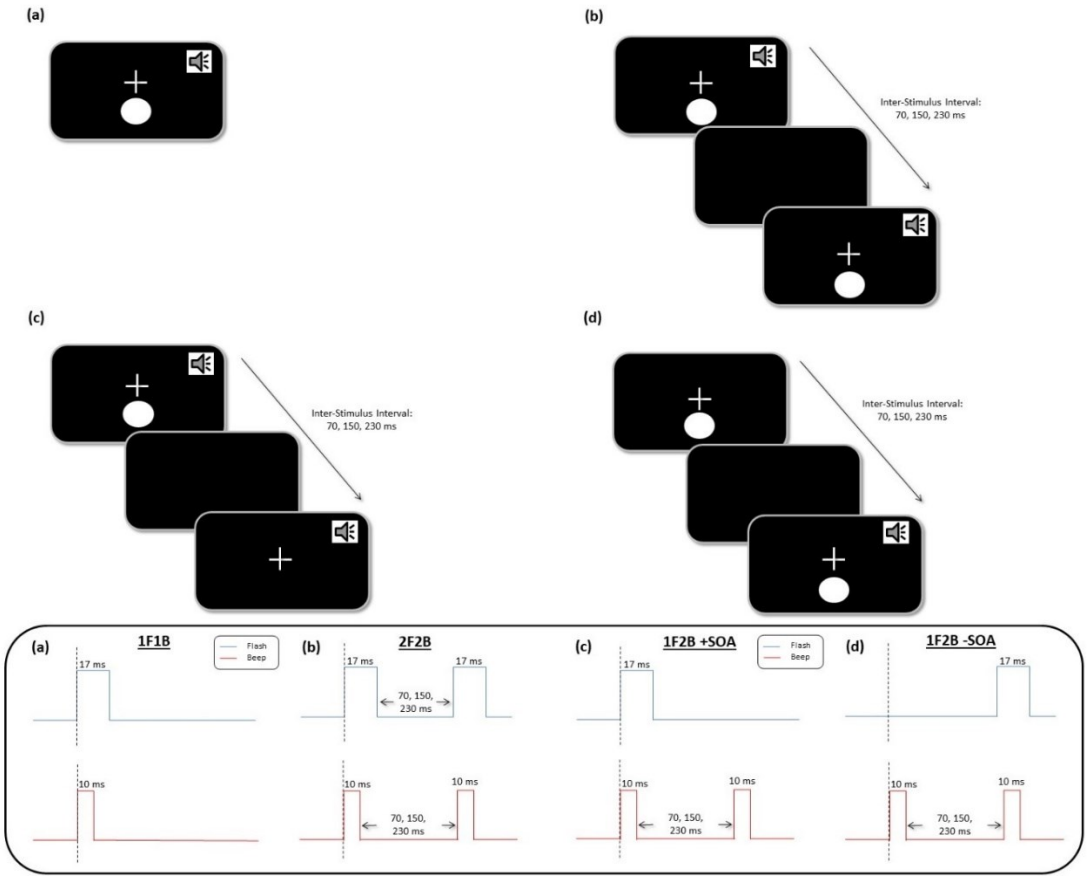

Figure 2.9. *Multisensory SIFI Conditions*. **(a)** Multisensory congruent condition of one visual and auditory beep. **(b)** Multisensory congruent condition of two flashes and two beeps with a stimulus onset asynchrony (SOA) of 70, 150 and 230 ms. **(c)** Illusory fission trial with auditory beep following flash-beep combination by 70, 150, and 230 ms. **(d)** Illusory fission trial with auditory beep preceding flash-beep combination by 70, 150, and 230 ms.

2.4.4. Exercise Metrics of Performance and Physiological Disturbance

2.4.4a Wellness Questionnaire

Prior to both exercise sessions, participants completed a subjective wellbeing questionnaire. McLean and colleagues developed the questionnaire as a brief version of the profile of mood states (POMS) questionnaire to measure the overall degree of wellbeing on that particular day (541). On a scale of 1 to 5, five distinct physiological factors (fatigue, sleep quality, general muscular soreness, stress level, and mood) were scored. The scores from each of the five components were

added together to give each participant an overall account of vitality out of a possible 25 (*for further details see* ***Appendix 1.13.****)*

2.4.4b Lactate

An alcohol swab was used to clean the earlobe before using a sterile single-use safety lancet (Safety Lancet Super 1.5 Blade, Penetration Depth 1.6 mm, Sarstedt, Germany) to puncture the earlobe and extract blood. An automated blood lactate analyser was used to test blood lactate when a lactate strip was placed in contact with the blood droplet (Lactate Pro 2 analyser & strip, Arkay, Amsterdam, The Netherlands). The site was wiped down using a tissue to dry off the area. The lancet was then be used to create a small puncture for analysis of blood lactate throughout the testing session. The test strip was inserted into the Lactate analyser and placed at the blood droplet for analysis. At each lactate measurement, the site was abraded using a tissue to remove any clotting and open the puncture wound again for blood extraction and lactate analysis. In all procedures, lactate levels were assessed before and immediately after exercise. Lactate levels were also assessed following the fifth interval of the anaerobic exercise constituting midway completion of the protocol.

Figure 2.10. *Lactate Pro 2 Analyser.* A precise portable measurement device for determining whole blood lactate levels within 15-seconds. It is ideally suited for portable usage within sports medicine and research settings storing 330 test results and requires only 0.3 µl test volume.

2.4.4c Rate of Perceived Exertion

To evaluate levels of exertion felt during and after the exercise sessions, the Borg CR-10 Scale was employed (542). This is a 10-point scale, with 0 representing '*rest*', 5 representing *'somewhat hard'*, and 10 representing *'all out'*. Before the activity, the participants were shown a chart representing rate of perceived exertion (RPE) levels. Participants in the intermittent, high-intensity sprint session were given the chart after each interval and asked to rate their RPE. The RPE rating was also measured following the completion of the 20-minute aerobic session of moderate intensity (*see* ***Appendix 1.14.****).* Participant sex, age, training status, and the RPE scale used has been previously shown to have no effect on RPE validity (543). There is a wide body of research in support of RPE as a valid metric of both exercise intensity and physiological exertion during exercise and, and competition and training in team based sports (544, 545). When HR, blood lactate and maximum rate of oxygen consumption (VO_{2max}) are used as outcome measures, RPE possesses the highest significant validity coefficients during cardiovascular exercise (545).

2.4.4d Heart Rate

HR was measured using a Kinetik wellness heart rate monitor and adjustable strap HRM4 (Kinetik medical devices Ltd, Nottingham, UK). Before being fastened on the subject, the electrodes on the strap were dampened with water. Prior to activity, participants' resting heart rates were measured while they were sitting. During all sessions, heart rate was measured throughout and immediately after activity. For the high-intensity session, the greatest heart rate achieved at a single point in time during each interval was recorded.

2.4.4d Fatigue

Fatigue was measured indirectly only for the high intensity exercise protocol using two commonly used techniques (516), the Fatigue Index (FI) and the performance decrement score (Sdec) with a higher percentage achieved indicating a higher rate of fatigue and decline in exercise performance, using the following equations:

$$\mathrm{FI}\ (\%) = 100 \times \left(\frac{[S_{best} - S_{worst}]}{S_{best}} \right) \quad (1)$$

$$S_{dec}\ (\%) = \left\{ 1 - \frac{(S_1 + S_2 + S_3 \ldots + S_{10})}{S_{best} \times n} \right\} \times 100 \quad (2)$$

where S refers to sprint performance (Peak Power Output [PPO], in Watts) and n is the number of sprints performed.

2.5. Study Two – Systematic Reviews investigating the Impact of Midlife CV Risk factors on Cognitive function with Advancing Age

Each chapter for Study Two was carried out as a *subset analysis* of a wider review investigating the influence of midlife CV risk factors on cognition at mid- and later life. The reviews detailed in **Chapters 6-8** were conducted in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses guidelines (PRISMA; www.prisma-statement.org) and was recorded in PROSPERO, a registry of systematic reviews. Registration of this review can be found at https://www.crd.york.ac.uk/prospero/ (registration number: CRD42021238293).

2.5.1. Search strategy

Online electronic databases were searched, and relevant articles retrieved from the following: EMBASE, MEDLINE, PubMed, Web of Science and CINAHL, from their inception to May 2022. All search strategies were conducted by a medical librarian with methodological experience and the full search strategy can be found in the ***Supplementary Material***. The search strategy comprised of key words, MeSH terms, common medical terms, and a combination of these including, but not limited to, middle age, midlife, cardiovascular disease, cardiovascular risk, cognition, and cognitive defect. The search strategy for **Chapter Six** included the key terminology of *hypertension* and *high blood pressure*; **Chapter Seven** included *diabetes*, diabetes mellitus, and *diabetic*; and **Chapter Eight** included *cholesterol* and *total cholesterol*. The search strategy focused on the inclusion of longitudinal, prospective and follow-up studies to ensure later life cognition was captured. No search restrictions for language or publication date were implemented. The search of electronic

databases was supplemented by a manual literature search of the reference lists of included studies and appropriate databases to ensure all relevant studies were captured.

All stages of the screening process were conducted independently by two reviewers, including title and abstract screening and subsequent full text screening. Disagreements between the two reviewers were resolved through discussion. If a consensus was not achieved, a third reviewer was consulted. Titles, abstracts, and full texts of all eligible articles were screened using Covidence (https://www.covidence.org/home).

2.5.2. Eligibility criteria

Studies were deemed eligible based on the following inclusion criteria: human participants, adults between ages of 40-65 years were classified as middle-aged [World Health Organisation (WHO) definition of middle age]; CV risk factor (hypertension, DM, and/or cholesterol) as an outcome measure at later life, midlife, or both for determination of the longitudinal association with midlife CV risk factors and cognition across domains including memory, attention, executive function, intelligence, and global cognitive functioning (see ***Appendix 2.1.*** *and* ***2.2.***). Studies not published in the English language where a translation could not be obtained were excluded. Studies were excluded if cognitive testing was undertaken by a proxy or designated respondent, such as a friend or family member, if the participant cohorts included those with midlife dementia or any form of pre-existing cognitive impairment and if studies of specific disabilities (traumatic brain injury, stroke, human immunodeficiency virus (HIV), spinal cord injury, etc.) were associated with modifiable behavioural risk factors.

2.5.3. Data extraction

Data extraction was carried out in accordance with the STROBE guidelines (546), including study aims, participant characteristics, measures of cognition and CV risk factors alongside relevant outcome data as group means, standard deviation (SD), standard error (SE) of the mean, statistical significance, and precision estimates for **Chapters 6-8**. Adults between the ages of 40-65 years were considered middle aged in line with the WHO definition and those beyond the age of 65 years were classified as later life participants. To prevent double reporting of data from prospective longitudinal cohorts, the most recent publication relating to each was selected as the reference

study for the determination of baseline data (*see* ***Appendices 6.2., 7.2.,*** *and* ***8.2.***). If uncertainties arose, the corresponding authors were contacted for further clarification. Each study was assigned a reference number and separate data collection form. To ensure accurate reporting the data extraction pro-forma was piloted against a selection of papers. Endnote version 20 was then used to create a database.

2.5.4. Risk of bias and methodological assessment

The methodological quality of included studies was evaluated using the Appraisal Tool for Cross sectional Studies (AXIS) (547). This tool employs 20 questions to determine quality of study design and risk of bias with questions being answered as 'Yes', 'No' or 'Unsure'. Using the method outlined by McHugh and Colleagues (484) answers were inserted in colour coding to reflect the impact on the text, including: green, positive impact on quality of study; red, negative impact on quality of study; and amber, unknown impact on quality of study. Two reviewers independently evaluated the included studies. Disagreements between reviewers were resolved through discussion. If a consensus was not achieved, a third reviewer was consulted. Study quality was then classified as either low, moderate, or high.

2.5.5. Meta-Analysis

A random effects meta-analysis was conducted for **Chapters Six and Seven** to compare the difference across each cognitive domain between two independent groups: hypertension vs normotension, and diabetes vs no diabetes. A meta-analysis was not possible for **Chapter Eight** and associated cholesterol metrics. Group mean differences, 95% confidence intervals (CIs) and P-values were calculated using Review Manager (RevMan) software (Version 5.4, Copenhagen: The Nordic Cochrane Centre, The Cochrane Collaboration, 2020). Sub-grouping for meta-analyses included study design and quality. The heterogeneity between studies was established using the I^2 statistic. I^2 values of 25%, 50%, and 75% ($p > 0.05$) correspond to low, moderate, and high degrees of heterogeneity respectively (548). Where high levels of heterogeneity ($I^2 > 75\%$) were detected and a sufficient availability of studies was present, sensitivity analysis were applied, and studies were removed one by one to assess their overall influence. Studies that were removed due to the sensitivity analysis are represented by a 0.0% weight in the forest plots.

2.6. Study Three – Cognitive and CV Health of Older Community Athletes

2.6.1. Study Design

This pilot study was a cross-sectional cohort design. Older community athletes classified as middle aged (40-65 years old) were invited to partake in the study from TCD Sports Alumni. Participants who enrolled in the study attended the designated test centre for a once off testing session. All participants were informed their participation in the study was voluntary. This exploratory study was carried out from May 2023 to June 2023.

2.6.2. Study Population

Participants were recruited from the alumni clubs of TCD Sport. Participants were recruited through flyer advertisement, social media outreach and email contact with sports clubs (*see **Appendix 3.9.**)*. A power analysis for participant recruitment was not performed for the present prospective study as it is exploratory in nature. All participants in this study were obtained using a non-probability convenience sampling from the chosen target population (549). This form of convenience sampling ensures availability of participants, ease of access, time efficiency and affordability for creating a large dataset for analysis. However, there are inherent concerns of bias where a high risk of self-selection and hidden biases in non-probability sampling become apparent where the researcher cannot fully ensure that the convenience sample accurately represents the target population (550). With concerns that a small population reduces the power of results, this does not apply in the present study given its exploratory nature where power calculations were not undertaken (551). Given the retrospective nature of repetitive head impact (RHI) exposure it has been deemed appropriate by the National Institutes of Health/National Institute of Neurological Disorders and Stroke (NINDS) to use contact sport career duration as a proxy measure as was the case for this chapter (552).

2.6.3. Eligibility Criteria

The inclusion criteria included Male and female older community athletes in midlife (40-70 years) and must have completed at least one season of sport. Participants must have completed at least one season of sport. Potential participants were deemed ineligible based on the following criteria: **1)** Individual had a history of a non-sports related concussion or form of brain injury, **(2)**

Individual has suffered a previous concussion (self-reported or diagnosed) within the past year, and **(3)** Individual reporting previous neurological or neurodevelopmental disorder. The rationale and background for the study was explained to all participants prior to the commencement of the study. The protocol and participant information were subsequently provided to athletes who were interested in participation.

2.6.4. Ethical Approval

The Institutional Research Ethics Committee gave ethical approval (Ref No. 221102; see **Appendix 3.1.**). As necessary, the primary investigator presented a research summary to all prospective participants through email and at the club's training facilities. Athletes who were interested in participation were given a participant information pamphlet (see **Appendix 3.8.**). Athletes who were still interested in participation signed written informed consent forms (see **Appendix 3.7.**).

2.6.5. Assessment Protocol

Enrolled participants in this study underwent a single testing session conducted at the Trinity Sports Centre in Dublin. A standardised test protocol was employed for all assessments. The evaluation process involved completing sociodemographic and clinical questionnaires, undergoing a physical assessment, and participating in cognitive functioning tests. The lead investigator administered all measures. The testing session took a total of 1-hour (30-minutes for the series of questionnaires and 30-minutes for the physical and cognitive assessment).

The testing protocol was explained to the participant prior to commencing the test. This was followed by questionnaires enquiring into sporting career, mental and general health, physical activity and lifestyle factors, and concussion history. All participants then undertook a brief health screen including resting HR and BP, height, weight, and BMI. The participants then completed the neurocognitive SIFI assessment.

2.6.6. Outcome Measures

2.6.1. CV Assessment

2.6.6.1a Resting Heart Rate & Blood Pressure Measurement

HR and BP were both measured using an electronic automated BP monitor (Omron 705IT, Omron Corporation, Kyoto, Japan). HR was obtained via sensors and algorithms integrated within the device itself. BP was measured using the oscillatory method for measuring BP (553). The participant was instructed to sit in a supported chair with uncrossed feet, resting in a relaxed position. After a five-minute period of relaxation and quiet, the lead investigator performed the BP measurement. The participant's left arm was prepared by clearing clothing and placing a cuff at heart level on the upper left arm (unless contraindicated). The cuff automatically inflated and deflated, with pressure changes monitored by a transducer that detects heartbeat-related variations in pulse volume. Subsequently, a specialized algorithm calculated systolic (SBP) and diastolic (DBP) blood pressure values. The lead investigator recorded the SBP and DBP values, reported in mmHg, as well as the HR reported in bpm. The participant was informed of the SBP/DBP measurements verbally and on paper. This measurement process was repeated three times, with a 1–2-minute interval between each repetition. The averaged readings were used to estimate the participant's BP level and average HR.

The reported results are the average of three measurements. On the day of testing, all participants were measured in the morning prior to food or fluid ingestion or participation in exercise. Participants participated in normal activities outside of these hours in accordance with the most recent ESH recommendations.

BP was classified according the European Society of Hypertension (ESH) guidelines into the following categories: optimal, defined as systolic BP < 120 mm Hg and diastolic BP < 80 mm Hg; normal, defined as systolic BP 120 – 129 mm Hg and/or diastolic BP 80 – 84 mm Hg; high-normal, defined as systolic BP 130 – 139 mm Hg and/or diastolic BP as 85 – 90 mm Hg; grade 1 hypertension, defined as systolic BP 140 – 159 mm Hg and/or diastolic BP as 90 – 99 mm Hg; grade 2 hypertension, defined as systolic BP 160 – 179 mm Hg and/or diastolic BP as 100 – 109 mm Hg and; grade 3 hypertension as systolic BP ≥ 180 mm Hg and/or diastolic BP ≥ 110 mm Hg (554).

Figure 2.9. *Blood Pressure Monitor used during participant assessment.*

2.6.6.2. Clinical Questionnaires

In addition to the clinical, health-related, and sporting questionnaires detailed in ***Protocol 1.1.*** of **Study One**, the following questionnaires were deemed appropriate to further investigate the long-term effects of a history of sporting engagement and concussion history with age.

2.6.6.2a Pain Disability Index (PDI)

The widely used Pain Disability Index was chosen to quantify pain-related impairment linked with persistent musculoskeletal pain (555) (see **Appendix 3.3.**). This test is intended to measure self-reported degrees of impairment encountered while doing important living activities on a daily basis (556). The rating scales assess the extent to which chronic pain interferes with many elements of life. The PDI measures the following activities: **(1)** family and home responsibilities: activities related to home and family, **(2)** recreation: hobbies, sports and other leisure time activities, **(3)** social activity: participation with friends and acquaintances other than family members, **(4)** occupation: activities partly or directly related to working including housework or volunteering, **(5)** sexual behaviour: frequency and quality of sex life, **(6)** self-care: personal maintenance and independent daily living (bathing dressing etc.) and **(7)** life-support activity: basic life-supporting behaviours (eating, sleeping, breathing, etc.). Higher ratings imply increased pain and impairment. Many investigations have shown the PDI to be reliable and valid (557, 558). Long-term sports participation has been associated with the prevention of and relationship with several adverse

health conditions including but not limited to osteoarthritis, back pain, and joint replacement (559-562). As a result, measurement of impairment linked with chronic pain was judged relevant in this group.

2.6.6.2b The Pittsburgh Sleep Quality Index (PSQI)

The PSQI is an effective self-reported questionnaire used to examine sleep quality and patterns over a 1-month period. It distinguishes between "*bad*" and "*excellent*" sleep by assessing seven domains: subjective sleep quality, sleep latency, sleep length, habitual sleep efficiency, sleep disruptions, use of sleep medicine, and daytime dysfunction in the previous month. Each of these seven sleep regions is self-rated by the responder. The responses are scored on a 0 to 3 scale, with 3 being the negative extreme on the Likert scale. A "*poor*" sleeper has a global total of "5" or above (see **Appendix 3.4.**). For its seven components, the PSQI has internal consistency and a reliability coefficient (Cronbach's alpha) of 0.83. Around 56 languages have been translated into the scale. Numerous investigations in a range of adult groups have shown the PSQI to have good validity and reliability (563, 564).

2.6.6.2c The Alcohol Use Disorders Identification Test (AUDIT)

The AUDIT is a 10-item screening tool developed by the World Health Organisation (WHO) (565). The purpose of this questionnaire is to examine alcohol use, drinking habits, and alcohol-related issues. Participants were urged to respond to the AUDIT using standard drinks. For reference, they looked to a chart indicating the estimated number of standard drinks in various alcoholic beverages. A score of 8 or above is considered hazardous or dangerous alcohol usage (see **Appendix 3.5.**). The AUDIT has been found as a viable and valid screening tool for identifying high-risk drinkers and people with alcohol use disorders (AUD) (566). It has been verified across genders and racial/ethnic groups and is well suited for usage in primary care settings (567). Research suggests athlete groups have greater rates of alcohol consumption or dependency than non-athlete communities (568-571). Alcohol abuse is known to be a complicating factor in cognitive performance, as well as a risk factor for psychiatric diseases and poor mental health (572-574).

2.6.6.3. Cognitive Assessment

Cognitive health was examined through the administration of the SIFI and Montreal Cognitive Assessment (MoCA). Both cognitive assessments were administered in a quiet room in order to enhance optimal testing conditions. For full details on the SIFI task please see ***Section 2.4.3a*.**

2.6.6.3a The Montreal Cognitive Assessment (MoCA)

The MoCA test is a quick 10-minute 30-point cognitive screening tool for mild cognitive impairment (MCI) and dementia that is utilised all over the world (575) (see **Appendix 3.2.**). Further clinical assessment is necessary to assess the aetiology of cognitive impairment and to make a diagnosis of dementia. Considered the gold standard in cognitive testing, it has greater sensitivity for detecting MCI (90%) compared to the mini mental state examination (MMSE) (18%) (576, 577). The test may also be used to measure multiple cognitive domains and grade the severity of cognitive impairment. Domains which are assessed include: **1)** attention and concentration, **(2)** executive function-memory, **(3)** language, **(4)** visuoconstructional skills, **(5)** conceptual thinking-calculations, and **(6)** orientation. Normative databases for middle aged adults (578) and sporting populations (579, 580) were used as reference markers to the present study population. Online training version 8.1 was completed to gain a certificate covering administration, scoring, and interpretation of the MoCA test (see **Appendix 3.10.**).

2.7. Statistical Analysis

Data were analysed using the IBM Statistical programme for the Social Sciences (SPSS) Version 26.0 statistic software programme (SPSS Inc, Chicago, IL) and R version 3.6.0 using package lme4. Preliminary analyses for all investigations included determining normality by visual inspection of histograms and Q-Q plots, as well as utilising the Kolmogorov-Smirnov test ($p>.05$). Non-normally distributed data are provided as medians and interquartile ranges (IQR), whereas regularly distributed data are presented as mean and standard deviation (SD). If the data was not normally distributed, non-parametric tests were applied. Significance was taken at $p<0.05$. Figures were generated using GraphPad Prism (GraphPad Software version 8, San Diego, CA).

In **Study One**, the intraclass correlation coefficient (ICC2*k*) was used to assess the between and within reliability of the SIFI across multiple sessions. The technical error of measurement (TEM) was used to assess the variability in experimental SOA conditions of the SIFI. Cronbach's alpha to determine the internal consistency was also assessed across the 6 experimental SOA illusory

conditions across the testing sessions. Bland-Altman plots were used to describe the differences between sessions graphically. A Bayesian hierarchical model was designed to assess the level of agreement of the SIFI across three distinct testing session: baseline, pre-moderate, and pre-high intensity exercise. In **Study One**, between-group differences for perceptual performance during the SIFI test were initially determined using mixed model ANOVA's to uncover main effects. Two-way ANOVA's were subsequently run to determine the effects of each multisensory SOA condition across between-subject factors. Logistic Regression was then deemed more appropriate to model the number of correct responses in each condition for each individual, allowing the probability of a correct response to vary according to group and condition in the *pilot study* of **Study One**. Due to the multivariate nature of the response for each individual (six SOA conditions), latent class analysis (LCA) or Bayesian classification was then used to determine whether there were any consistent patterns across the SIFI conditions. Based on further observations and the inclusion of additional covariates, the data was categorised into strata to assess whether the additional baseline variables measured in participants can better predict overall patterns in SIFI response. A multinomial logistic regression was undertaken whereby the model was fit through Markov Chain Monte Carlo using a Bayesian Pólya-Gamma sampler. This analysis was also undertaken for the cohort of participants in **Study Three**. One-way ANOVA or T-test where appropriate were used to assess for multiple comparison across groups for all exercise-related data in **Chapter Five**. Correlations between outcomes of interest such as associations between common sporting metrics (concussion history, time since sporting retirement, sporting type, etc.) and mental health indicators were explored using bivariate correlation analysis. Multiple linear regression modelling was also employed to assess the predictive nature of several sporting characteristics and their effect on mental health.

In **Study Two**, the weighted mean for demographics, cognitive measures (cognitive-specific domains and associated neuropsychological tests), and metrics of associated CV risk factors, i.e., hypertension, diabetes, and cholesterol were calculated across all studies. Cognitive outcome measures were grouped according to cognitive domain. Qualitative analysis assessed the relationship between midlife CV risk factor status and cognition at later life and midlife; positive, negative, or neutral, across studies. Meta-analysis was only deemed appropriate for **Chapters Six and Seven**. *See* ***Section 2.4.5.*** *for full details*. As all studies within this thesis were considered exploratory work a power calculation was not performed. Full details of statistical analysis for each study are presented in the relevant chapters.

3

Chapter Three:

An Investigation into the Absolute and Relative Reliability, and Level of Agreement of the Sound Induced Flash Illusion Test

3.1. Highlights

- The reliability between conditions within a given session was reported as excellent through intraclass correlation coefficient (ICC) analysis.
- The reliability across multiple sessions was found to be excellent by ICC analysis.
- The largest relative Technical Error of Measurement (TEM) was found for those conditions with the shortest stimulus onset asynchrony (SOA).
- Those with the largest SOA, i.e., -230 and +230, were seen to exhibit the least amount of measurement error.
- The within and between session reliability was found to have an excellent level of internal consistency as measured by Cronbach's Alpha.
- The measure of agreement based on our Bayesian Hierarchical Model computed for $\hat{k}$ = 13 is given by 0.8488 suggesting a reliability for the SIFI of ∼85% for the present cohort.

3.2. Introduction

Audio-visual (A-V) illusion paradigms can allow assessment of multisensory integration (MSI) (94-96). First described more than two-decades ago by Shams et al., the *'Sound-induced Flash Illusion'* (SIFI) has provided an example of perceptive ability in the midst of multisensory products where several unisensory modalities can work in unison (97, 98). The SIFI occurs when a single visual flash that is shown in conjunction with two or more auditory tones is subsequently perceived as two or more flashes (96-98). Previous studies have used susceptibility to the SIFI as a measure of perceptual efficiency, particularly using the fission and fusion illusions (96-99). Susceptibility to the illusion is however dependent upon the time delays, or stimulus onset asynchrony (SOA), that are present between the A-V stimuli. A common reported feature amongst the literature is reduced susceptibility as the time delay between the stimuli increases (100-102). Although widely accepted as a valid and reliable measure of MSI, there is no published data to be found in support of the accuracy and reliability of results, even though trust in the experimental procedure is based on previous evidence of perceiving the illusion itself.

Measurements of reliability and levels of agreement are an important aspect to any level of quantitative research to ensure reproducibility of results and is centred on the principle of consistency and credibility of published outcomes (581). Reliability is defined as the extent to which

measurements can be replicated over time reflecting not only the strength of correlative properties but also the agreement between the measurements. This core pillar forms a foundational premise alongside inherent validity for high-quality, evidence-based research to be conducted. Reliability and agreement have become an essential component in concussion assessment with many separate and serial testing policies (582). Performance measures with adequate sensitivity and sufficient specificity alongside established markers of mTBI are essential to support a clinical diagnosis of concussion. Hence, post-concussive clinical decisions backed by neurocognitive tests must have a high level of sensitivity, specificity, and most importantly reliability to a concussive injury. There is inherent variability in the reliability and levels of agreement of several neurocognitive tests due to sample size, test-retest intervals, participant age, and a narrowed validity of supporting evidence even among those most widely used (583). If the SIFI test is to have sufficient utility in SRC diagnosis and prognosis, the determination of an agreeable level of reliability is paramount.

Table 3.1. *Various forms of Reliability.*

Type	Definition
Intra-rater reliability	Indicates the difference between two or more raters measuring the same set of respondents.
Inter-rater reliability	Represents the variance in data recorded by a single rater over two or more trials.
Test-retest reliability	Signifies the variance in measurements collected by the same instrument on the same subject under the same conditions. It is often suggestive of dependability when raters are not involved, or the rater effect is negligible.

Depending on the properties of the data (categorical or continuous) and the settings of testing variables, such as percentage agreement, many statistical approaches can be employed to assess reliability ranging from Kappa statistics and the Phi approach to Pearson's correlation and Intra-Class Correlation Coefficients (ICCs). ICC is a frequently used metric for determining the test reliability of continuous variables. It is known to be generated from repeated measures of analysis of variance, which yield results that are closest to the strict definition of dependability. However, different types of ICC can provide different findings when used to the same set of data, and the methods for reporting ICC may change amongst studies. ICC comes with inherent pitfalls and limitations with a strong dependence on the variance of the assessed population. They are prone to subject variability, which

may result in different results even for the same measurement errors in identical dimensions. ICCs can be calculated using a variety of models (*one-way random, two-way random, and two-way fixed*), types (*absolute agreement or consistency; single rater/measurement or the mean of k raters/measurements*), and measures (*single or average measurements*), which can produce disparate results and perplex researchers attempting to choose an appropriate ICC model, type, or measure. In scientific literature, inadequate statistical approaches for assessing agreement and erroneous interpretations of agreement results are common. As such, the most often used methodologies for measuring agreement on continuous variables have been the ICC and Limits of Agreement, or a combination of both. Despite identical levels of agreement, higher ICC values may be produced when applied to a more diverse population as opposed to a more homogenous one. In contrast, the variance of the examined population has no effect on Limits of Agreement, but interpretation of the significance of its results is very subjective, since it is dependent on people's understanding of the clinical value of the given range. The mean difference between two measurements in the same individuals and the standard deviation of these differences are used to compute Limits of Agreement. Approximately 95% of these differences will be between the mean differences of 1.96 standard deviations. ICC values, however, are primarily used for statistical purposes, which might make clinical interpretation of ICC findings problematic. Although ICC values represent within-target variability (*measurement errors*), they are thought to be sensitive to between-target variability (*subject variability*). Other approaches for assessing the clinical significance of measurement error should thus be included in ICC-based reliability testing.

What is ultimately required is an investigation of the overall test-retest reliability of the SIFI measures - since a measure must reliably define individual variations over time to be replicable. Mathematically, reliability depicts a ratio of true variance over true variance plus error variance, such that dependability is defined as a measure's consistency or stability in producing the same findings under a variety of probable circumstances (584, 585). To guarantee validity, internal consistency should be confirmed before a test is used for study or examination. Furthermore, reliability estimates indicate the degree of measurement error and agreement in a test. Simply put, this view of reliability is the test's connection with itself. This chapter was thus designed as a cross-sectional validation study with the primary aim to explore the reliability and reproducibility of the SIFI test over time. To our knowledge, this study is the first attempt to determine the reliability and overall level of agreement of the SIFI test.

3.3. Methods

3.3.1. Study Design

This study was designed as a cross-sectional validation study including a population of young adults attending third-level education. This study was split across two time periods due to the COVID-19 pandemic, i.e., between January-March 2020 and February-April 2023 and thus involved two separate cohorts of participants that were combined to form the present study cohort. The following chapter used data collected from ***Study One*** (**Chapters Three, Four & Five**) to investigate the reliability of the SIFI neurocognitive assessment tool. The methods used to collect this data can be found in **Chapter Two**; *please refer to* **Study Protocol 1.1. and 1.2.** *for full details*. The reliability of the SIFI was assessed on three separate occasions from a cohort of university level students (n = 192) during the two previously defined testing periods due to the COVID-19 pandemic.

3.3.2. Participants

A sample of 192 university students were recruited. See **Chapters Two and Four** for full details of the study population demographics and experimental testing procedure for data collection. The primary outcome measure was the SIFI neurocognitive assessment. A complete description of the SIFI test, method of assessment, associated performance metrics, and interpretation of results can also be found in **Chapters Two, Four, and Five**. Using the data collected for **Study One**, all participants undertook 5 SIFI neurocognitive assessments over three testing sessions. In this analysis, we are only using data from 3 of the 5 assessments (Baseline, Pre-Moderate, and Pre-High). Due to the COVID-19 pandemic, there were 5 participants with missing SIFI data removed from the dataset when undertaking the reliability analysis for this chapter.

3.3.3. Test Re-Test Reliability

3.3.3a Intraclass Correlation Coefficient

The ICC, which indicates the statistical stability between two data measurements, is one of the most consistent indices used to evaluate the reliability of neuropsychological and cognitive tests. In order to derive the ICC from the SIFI, a minimum of two tests are required for each participant. In theory, a sample of N participants (i = 1, 2, 3….N) is selected randomly from a population P, and the susceptibility and proportion of correct responses from the SIFI test, S is measured k times (j = 1, 2….k)

across repeated testing sessions with all 6 SOA conditions. Hence, S_{ij} denotes the SIFI performance results j for a given participant i which can be represented as a matrix with N rows and k columns. Participants are commonly referred to as "targets," while measures are referred to as "conditions" or "raters." Thus, the ICC of a sample may be calculated using the *N x k matrix*.

ICCs (i.e., 2-way mixed-effects models with absolute agreement) were used as a reliability index for test–retest reliability to assess the internal consistency of the SIFI test from respondents measures of each SOA condition (within) and across different sessions (between). The ICC values generally range from 0 to 1 such that an ICC = 1 indicates perfect reliability whereas an ICC = 0 denotes perfect unreliability. The ICC values are typically divided into four levels of stability which are as follows: poor (ICC < 0.4), fair (0.4 < ICC < 0.59), good (0.6 < ICC < 0.74), and excellent (ICC ≥ 0.75). The ICC for the SIFI was assessed between the baseline session and both follow-up sessions involving exercise testing (pre-moderate and pre-high intensity exercise); *see* ***Study Protocol 1.2.*** *in* ***Chapter Two*** *for full details*. Data analyses were conducted using SPSS (version 26.0; SPSS Inc, Chicago, IL), and codes were written in Python 3.7 and executed on either Google Collaboratory or Jupyter Notebook using Anaconda version 1.10.0, where statistical significance was set at α = .05.

In principle, ICC is formulated as a ratio of variances (σ) based on the source of error or unwanted variances below:

$$\mathrm{ICC} = \frac{\text{variance of interest}}{\text{total variance}} = \frac{\text{variance of interest}}{\text{variance of interest} + e} \qquad (1,1)$$

The ratio of between-subject variation to within-subject variance is another way to define ICC (586-588). When the error source is equal to or larger than the variance of interest (i.e., the number of SIFI tests, testing sessions) the dependability is likely to be low (ICC 0.4). As ICC is calculated based on how *e* is stated, it can take one of three forms: when *e* is not specified, ICC(1,1) represents "agreement" in exact values; ICC(2,1) represents "absolute agreement" in exact values when *e* is specified and modelled as random effects; and ICC(3,1) represents "consistent agreement" in exact values when *e* is specified and modelled as fixed effects. The ICC(1,1), ICC(2,1) and ICC(2,3) models, for example, can be stated mathematically as follows:

$$ICC(1, 1) = \frac{\sigma_\lambda^2}{\sigma_\lambda^2 + \sigma_\varepsilon^2} = \frac{MSBS - MSWS}{MSBS + (k - 1)\ MSWS} \quad (1,2)$$

$$ICC(2, 1) = \frac{\sigma_\lambda^2}{\sigma_\lambda^2 + \sigma_\pi^2 + \sigma_\varepsilon^2} = \frac{MSBS - MSE}{MSBS + (k - 1)\ MSE + \frac{k}{N}(MSBM - MSE)} \quad (1,3)$$

$$ICC(3, 1) = \frac{\sigma_\lambda^2}{\sigma_\lambda^2 + \theta_\pi^2 + \sigma_\varepsilon^2} = \frac{MSBS - MSE}{MSBS + (k - 1)\ MSE} \quad (1,4)$$

where λ = the focus of a measurement such as participants, π = specified error source, ε = unspecified error source, $\theta_\pi^2 = \frac{\sum_j(\pi_j - \bar{\pi})^2}{k-1}$, MSBS = mean square between participants, MSWS = mean square within participants, MSE = mean square error, and MSBM = mean square between sessions (e.g., SIFI tests across time). Of note, the reliability of the proportion of correct responses can be generalised by averaging across *k* measurements and be further expressed as ICC(1, *k*), ICC(2, *k*), and ICC(3, *k*) models (589). For this chapter, the flowchart of ICC selection process below was reviewed and it was determined that a two-way mixed effects model based on the mean of *k* was appropriate to determine the absolute reliability of the SIFI test across multiple testing sessions.

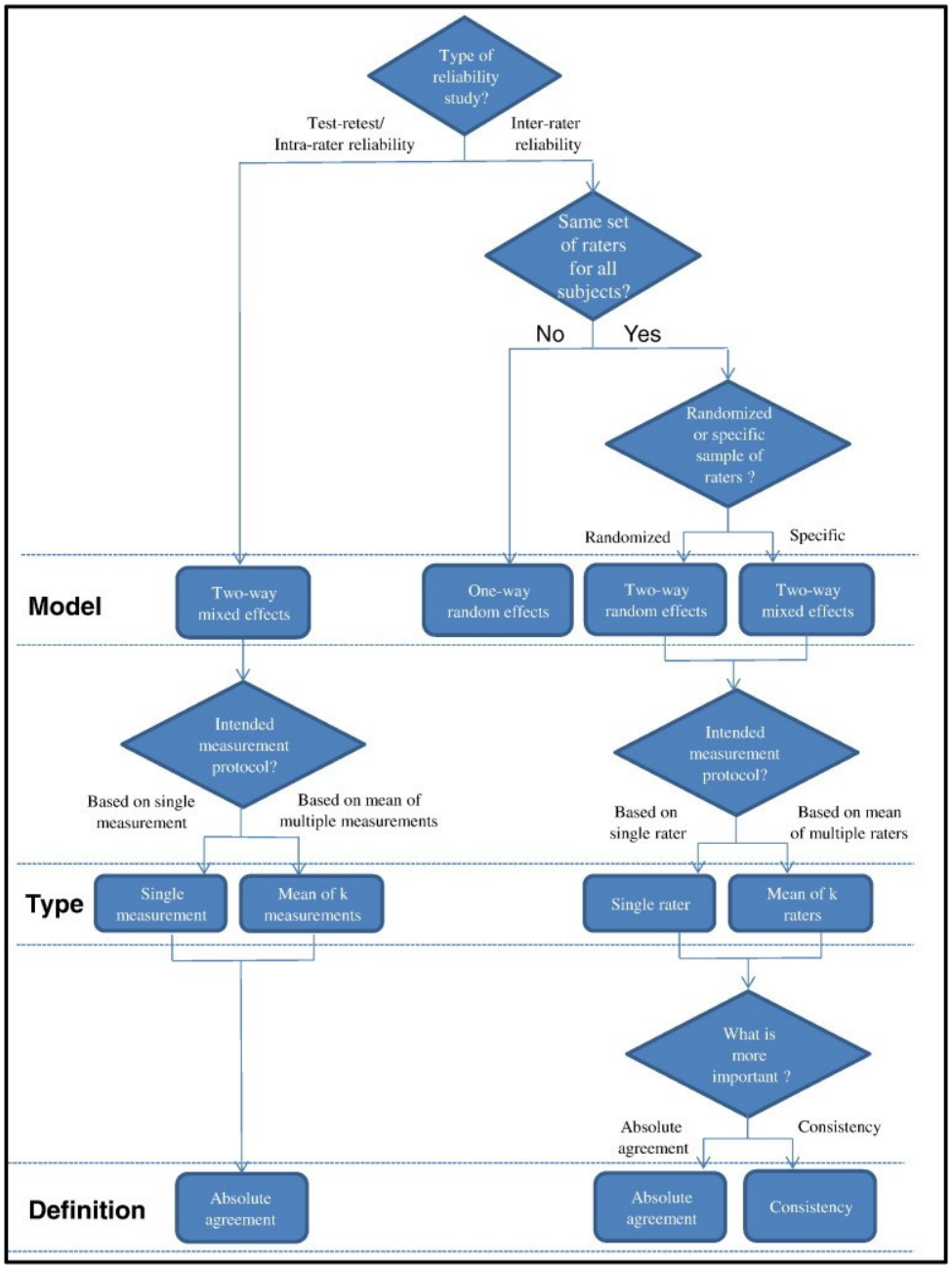

Figure 3.1. *A flowchart illustrating the process of selecting the ICC form based on the experimental design of a reliability study. *Adapted from the work of Koo et al. (586).*

3.3.3b Technical Error of Measurement (TEM) & The Coefficient of Reliability (R)

The TEM was used to assess the variability across the 6 experimental SOA conditions of the SIFI across several testing sessions (590, 591). The measurement quality and control dimension are represented by the TEM, which is an accuracy index. The TEM index is essentially a method of assessing the intra- and inter-testing reliability between the same participant or group of participants

and is the standard deviation between repeated measurements. The TEM calculation allows for the estimation of confidence intervals around the actual value of the measurement obtained. This includes non-controllable possible variations - for example, biological disparities - allowing for the verification of whether the changes detected in repeated measurements before and after a training session are a result of this training or a result of the method's relative variation. The method of differences was adopted to calculate the TEM which is found through the standard deviation between repeat assessments and is the level of deviation or dispersion of the given values in relation to the mean. A TEM with a lower value indicates greater precision of the participant undertaking the assessment. The TEM calculation can be represented using the following equation:

$$\text{AbsoluteTEM} = \sqrt{\frac{\Sigma\, d_i^2}{2n}} \tag{1,5}$$

Where Σd^2 is the summation of all deviations which are raised to the second power, *n* is the population total, and *i* is the total number of deviations. To derive the error stated as a percentage proportional to the entire average of each SOA, the absolute TEM was translated into relative TEM. The variable average value was calculated by determining the mean of the means for each participant across both sessions. This was calculated as the sum of the results from each respective SOA condition and divided by 2. The means from the total population were then summed and divided by the number of participants with data available for analysis to generate the variable average value. The following equation was used:

$$\text{relative TEM} = \frac{\text{TEM}}{\text{VAV}} \times 100 \tag{1,6}$$

Bland-Altman plots were used to describe the differences between sessions graphically. The TEM, the relative TEM (%TEM), and the coefficient of reliability (R) were the statistical tests used to further assess the reliability of the SIFI across each condition within the Limits of Agreement. The

reliability coefficient, *R* , denotes the estimated fraction of inter-subject variation that is not attributable to measurement error and was classified as:

$$R = 1 - \left(TEM^2/SD^2\right) \tag{1,7}$$

The overall inter-subject variation for the research population is denoted by SD^2. Scores range from 0 to 1, with 0 indicating that all differences between subjects are attributable to measurement error and 1 indicating that no measurement error exists. Greater precision is indicated by higher R values, with values of 0.9 and above classified as excellent, 0.80 – 0.89 deemed good, 0.70 – 0.79 regarded as adequate and values below 0.70 may have limited applicability with minimal acceptability.

3.3.3c Cronbach's Alpha

Given the ongoing debate and concern regarding reliability metrics, specifically internal consistency, Cronbach's alpha was also assessed across the 6 experimental SOA illusory conditions across the 3 testing sessions (592, 593). This measure of reliability informs us of how closely linked a set of test items are to each other as a group. Cronbach's alpha is designed to measure the reliability for latent variables or '*hidden variables*' that are thought to be otherwise not observable such as susceptibility to perceptual sensory illusions. In the present study this comes in the form of the SOA SIFI conditions where there are several unidentified variables at play which may alter performance results. This belief in the unrecognised *'hypothetical constructs'* are inherently inferred in most modelling techniques such as those used in Principal Component analysis and regression modelling. What Cronbach's alpha can potentially account for is the unobserved heterogeneity from random effects within a given sample or population for example where all additive error terms are undefined and thus latent by nature. The following formula was used:

$$\alpha = \frac{N \cdot \bar{c}}{\bar{v} + (N-1) \cdot \bar{c}} \tag{1,8}$$

where N is the number of items, $\bar{c}$ accounts for the average covariance between item-pairs, and $\bar{v}$ refers to the average variance. Cronbach's alpha can be interpreted as the following: ≥ 0.9 is excellent internal consistency, $0.9 > \alpha \geq 0.8$ is good, $0.8 > \alpha \geq 0.7$ is acceptable, $0.7 > \alpha \geq 0.6$ is questionable, $0.6 > \alpha \geq 0.5$ is poor, and $0.5 > \alpha$ is unacceptable. However, there is caution for interpretation as a high alpha level may indicate that the test items are highly connected. However, α is affected by the quantity of items in a test. A bigger number of objects can lead to a larger α, while a lesser number of items can lead to a smaller α. It should be mentioned that an instrument's reliability is directly related to its validity. A valid instrument is one that is trustworthy. However, an instrument's dependability is independent of its validity and ultimatley level of agreement.

3.3.4. Level of Agreement

We developed a hierarchical mixture model to assess the agreement of the SIFI test method. Agreement here refers to consistency of the SIFI test (*at baseline conditions*) taken by an individual on different occasions.

Suppose there are $i = 1, \ldots, n$ individuals. Each individual undergoes m experimental conditions e.g. −230, −150, −70, +70, +150, +230. These experimental conditions are each repeated r times with conditions presented in a random order to the participant. Thus for each individual i we have an m dimensional outcome, where each outcome $y_{ij} \in \{0, \ldots, r\}$ for $j = 1, \ldots, m$. We can approximate y_{ij} using a Binomial distribution with r trials and a given probability of success.

Figure 3.10. in **Section 3.4.** shows the result of a stratification with 13 strata on our data to be discussed in detail later. The row dimensions are arranged by strata labelling obtained through our proposed method. What is clear is a strong grouping of participants into groups of varying performance patterns over different conditions. Any method of assessing agreement should account for this observed heterogeneity.

3.3.4a Definition of Agreement

First, we need to define what we mean by agreement. Suppose individuals undertake the SIFI on T different occasions (assumed to be equally spaced in time). Let y_{tij} be the SIFI count result for individual i under SOA condition j at time (or occasion) t. Heuristically, agreement amounts to $y_{1ij}, \ldots, y_{Tij}$ being "*close*" together for all $j = 1, \ldots, m$. We need to provide a concept of closeness which is

native to the ordinal data that SIFI produces for each individual under varying SOA conditions. Taking the Binomial assumption then, it would correspond to condition *j* having a similar probability of success over all times *t*: $y_{tij} \sim \text{Binomal}(r, \theta_{tj})$ with:

$$\theta_{1j} = \theta_{2j} = \cdots = \theta_{Tj} \qquad j = 1, \ldots, m.$$

However, from ***Figure 3.10.*** located in **Section 3.4.** we can appreciate considerable heterogeneity in the response patterns. It is not unreasonable to expect the probability θ_{tj} to be specific to a subgroup exhibiting a particular response pattern. To account for this heterogeneity we introduce a mechanism for stratifying participants by response pattern through introduction of a class label for individual *i* and defining stratum specific probabilities θ_{tkj} , $k = 1, \ldots, K$.

$$[c_i \mid \pi, K] \quad \sim \quad \text{Categorical } (K, \pi)$$

$$[y_{tij} \mid c_i = k, \theta] \quad \sim \quad \text{Binomial } (r, \theta_{tkj})$$

where $\boldsymbol{\theta}$ denotes all the θ_{tkj}. Letting the strata labels c_i act as an individual level effect, correlating a participant's observations over time, we assume that y_{tij} are independent of $y_{t'ij}$ for $t' \neq t$ and all *i*, *j* conditional on c_i. We also assume local independence, so that conditional on c_i, y_{tij} is independent of $y_{tij'}$, $j' \neq j$. Our definition of agreement is to judge agreement within stratum *k* if:

$$\theta_{1kj} = \theta_{2kj} = \cdots = \theta_{Tkj} = \theta_{kj} \qquad j = 1, \ldots, m$$

i.e., the probability of success within the stratum is the same across all time points for all *m* SOA conditions.

3.3.4b Bayesian Hierarchical Model of Agreement

Bayesian methods can be used to effectively explore the agreement of individuals within strata over time. For a given stratum, we have two possibilities: agreement or non-agreement. If there is agreement, the stratum specific parameters are $\theta_{k1}, \ldots, \theta_{km}$. If there is not agreement, the parameters are $\theta_{tk1}, \ldots, \theta_{tkm}$, $t = 1, \ldots, T$. We introduce an indicator variable γ_k which indicates agreement across time points within stratum k. A value of $\gamma_k = 1$ indicates agreement, and 0 non-agreement. The generating model is thus:

$$[c_i \,|\boldsymbol{\pi}, K] \quad \sim \quad \text{Categorical } (K, \pi)$$

$$[y_{tij} \mid c_i = k, \boldsymbol{\theta}, \gamma_k = 0] \quad \sim \quad \text{Binomial } (r, \theta_{tkj})$$

$$[\gamma_{tij} \mid c_i = \text{k}, \boldsymbol{\theta}, \gamma_k = 1] \quad \sim \quad \text{Binomial } (\text{r}, \theta_{kj})$$

where the vector notation $\boldsymbol{\theta}$ represents whichever regime $\gamma_k = 0$ or $\gamma_k = 1$ is being used for stratum k. Taking standard conjugate priors on $\boldsymbol{\pi}$ (Dirichlet symmetric) and $\boldsymbol{\theta}$ (Beta symmetric) allows these parameters to be marginalised out of the joint Bayesian model, leaving a closed form expression for the marginal likelihood of the data for a given stratification $c_1, \ldots, c_n$ and model agreement indicators $\gamma_1, \ldots, \gamma_K$ under a given number of strata K. Using techniques similar to those described in detail in White et al. (594), we can generate samples of K, and conditional on K, then $\boldsymbol{c}$ and $\boldsymbol{\gamma} = (\gamma_1, \ldots, \gamma_K)$, through a "*collapsed*" posterior:

$$p(K, \boldsymbol{c}, \boldsymbol{\gamma}|\mathbf{y}) = p(K)p(\gamma|K)p(\boldsymbol{c}|K, \boldsymbol{\gamma}, \mathbf{y}).$$

We take a prior probability of 0.5 that any stratum does/does not agree, so that $p(\gamma|K)$ is a product of independent simple Bernoulli random variables. We take $p(K)$ to be Poisson(1) which is a well justified choice for these settings (595). The resulting Monte Carlo sample from $p(K, \boldsymbol{c}, \boldsymbol{\gamma}|\mathbf{y})$.

3.3.4c Correspondence between the Posterior and Hypothesis Tests

It is worth exploring the correspondences between this approach and what we would call more "classical" hypothesis tests. A test of consistency can be stated as:

$$H_0 : \theta_{1kj} = \theta_{2kj} = \cdots = \theta_{Tkj} = \theta_{kj} \qquad j = 1, \ldots, m$$

with the alternative being lack of agreement. The Bayesian method proposed allows flexible testing of this hypothesis, with potentially varying conclusions over different strata. Indeed, in the MCMC algorithm something similar to a Bayes factor appears to compare agreement and non-agreement models within a stratum. The advantage of this proposed approach is the flexibility and the huge model space that is searched.

3.3.4d Remarks on the Agreement Model

The introduction of *K* as a model variable, as well as the agreement indicators **γ** have several interpretational advantages.

Anomaly detection: By grouping participants into strata jointly over multiple time points, it can provide an insight into anomalous individuals whose data does not fit any common pattern.

Varying agreeability: The model allows for varying modes of agreeability over different strata, which allows for an unambiguous assessment of the full picture of agreeability (which may not be a dichotomous phenomenon for all groups).

Capturing learning and forgetting effects: If the *t* indexes represent a chronologically arranged time, then learning and forgetting effects for strata can be captured through examination of the posterior distribution of **γ**.

3.3.4e An Agreement Measure derived from the Model using Weighted Average

There is an obvious Bayesian measure of agreement which can be proposed from this model. An average of the agreement probability γ_k weighted by strata proportion was taken:

$$\mathcal{A}_K = \sum_{k=1}^{K} \pi_k \, \gamma_k$$

which we can approximate via

$$\widehat{A}_K = \sum_{k=1}^{K} \frac{n_k}{n} \widehat{\gamma}_k \tag{1}$$

where $\hat{y}_k$ denotes the MCMC estimated E { γ_k | K, **y**} and n_k = ≠ { $\hat{c}_i$ = k} gives the number of participants allocated to strata k based on some optimal **ĉ**.

3.4. Results

ICC2*k* estimates and their 95% CIs were calculated based on a mean-rating, absolute agreement, 2-way mixed effects model. The reliability between conditions within a given session was reported as excellent across baseline (ICC2*k*= 0.881; 95% CI [0.84-0.91]), pre-moderate (ICC2*k*= 0.892; 95% CI [0.85-0.92]), and pre-high (ICC2*k*= 0.888; 95% CI [0.84-0.92]) testing sessions of which all were statistically significant (p<0.000). The reliability across multiple sessions was found to be excellent between baseline-pre-moderate and pre-high (ICC2*k*= 0.922; 95% CI [0.87-0.95]), which was statistically significant (p<0.000). See **Table 3.2** and **Table 3.3.** below for all within and between session ICC values. A graphical illustration of results can be seen in **Figure 3.2.** and **Figure 3.3.** for the within and between ICC results respectively.

The relative TEM was assessed for each SOA illusory condition between testing sessions ranging from 22.45% to 64.55% across all SOA conditions. The largest relative TEM was found for those conditions with the shortest SOA, i.e., -70 and +70. Those with the largest SOA, i.e., -230 and +230, were seen to exhibit the least amount of measurement error. The coefficient of reliability, *R* , across all conditions was found to be minimally acceptable (<0.7) to good (>0.7). The conditions with the highest level of temporal delay between stimulus presentation produced the highest level of reliability between sessions. i.e., these conditions represent those with the highest level of possible discriminatory ability between stimulus presentation. See **Table 3.4.** below for further details and **Figure 3.8.** for a graphical depiction of results. Bland Altman plots were also created to better represent the reliability of the SIFI and can been seen below in **Figures 3.4. and 3.5**.

The within session reliability across all six SOA conditions was also defined using Cronbach's Alpha; this analysis revealed statistically significant levels of reliability across all three testing sessions

(p<0.000). Although the number of cases varied between sessions due to participant attrition rates in the wake of the COVID-19 pandemic, the within session reliability was found to have excellent internal consistency (i.e., >0.9). Full details of the results can be found in **Table 3.5.** below. The between session reliability across all testing sessions was found to produce an excellent level of internal consistency between baseline-pre moderate-pre high (α = 0.962), which was statistically significant (p<0.000). However, upon further inspection of each individual SOA condition between the testing sessions, only the SOA-150, +70, and +230 conditions showed significant levels of consistency (p<0.05 and <0.01). These individual SOA condition values can be interpreted as having good ($0.9 > \alpha \geq 0.8$) and acceptable ($0.8 > \alpha \geq 0.7$) levels of reliability. Full details of the results can be found in **Tables 3.5.** and **Table 3.6.** below.

We assessed the agreement of the baseline, pre-moderate intensity, and pre-high intensity exercise SIFI sessions. After removal of participants with missing values, we were left with n = 186 participants observed at T = 3 timepoints at m = 6 SOA conditions. ***Figure 3.9.*** shows the posterior probability mass function of K. We select $\hat{k}$ = 13 to present the remaining results. ***Figure 3.8.*** shows the observed SIFI values over the three time points, arranged by cluster (roughly in order of $\hat{y}_k$). ***Figure 3.10***. gives a similar plot, but within cluster variable sample means indicated through heat colours. ***Table 3.7.*** shows the estimated posterior agreement for each of the strata. The measure of agreement suggested in (1) computed for $\hat{k}$ = 13 is given by 0.8488 suggesting a reliability for SIFI around 85% for this group of participants. ***Figure 3.11.*** shows the observed means for each variable for the top six strata (for clarity) shown in ***Figure 3.8.***

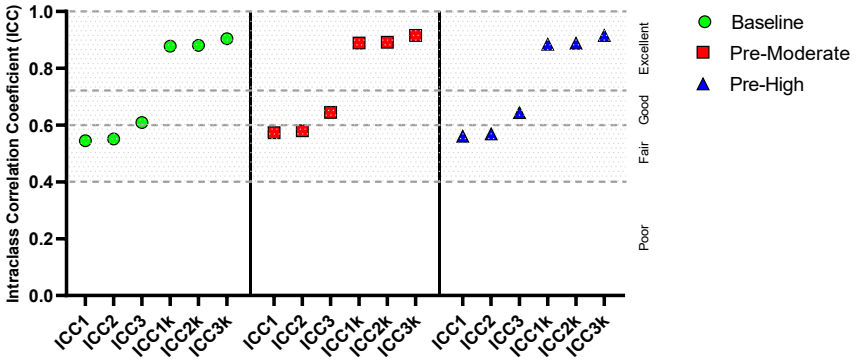

Figure 3.2. *ICC results of SIFI Neurocognitive Assessment among Young Adults – Within Session Reliability.* ICC(1):"agreement" in exact values; ICC(2):"absolute agreement" in exact values when *e* is specified and modelled as random effects; and ICC(3): "consistent agreement" in exact values when *e* is specified and modelled as fixed effects; Reliability of the proportion of correct responses can be generalised by averaging across *k* measurements and be further expressed as ICC(1, *k*), ICC(2, *k*), and ICC(3, *k*). Higher ICC values closer to 1.0 represent perfect reliability.

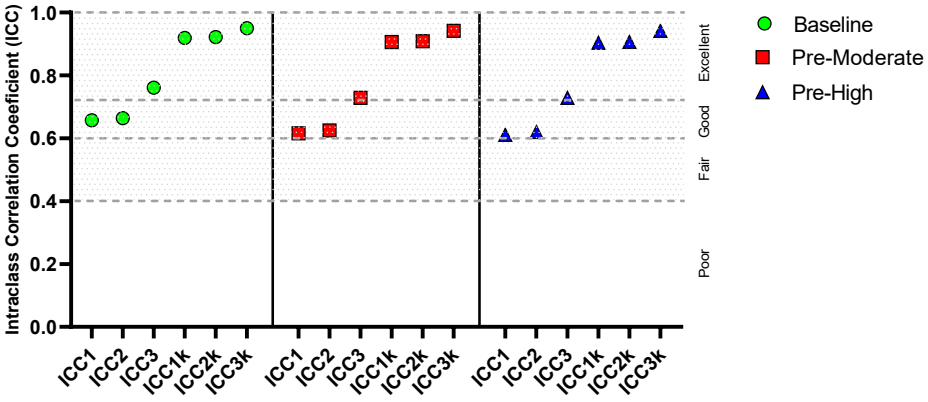

Figure 3.3. *ICC results of SIFI Neurocognitive Assessment among Young Adults – Between Session Reliability.* ICC(1):"agreement" in exact values; ICC(2):"absolute agreement" in exact values when *e* is specified and modelled as random effects; and ICC(3): "consistent agreement" in exact values when *e* is specified and modelled as fixed effects; Reliability of the proportion of correct responses can be generalised by averaging across *k* measurements and be further expressed as ICC(1, *k*), ICC(2, *k*), and ICC(3, *k*). Higher ICC values closer to 1.0 represent perfect reliability.

Table 3.2. *Within Session ICC Results.*

	Type	Factor of Analysis	ICC	F	df1	df2	p-value	95% CI
Baseline	ICC1	Single Raters Absolute	0.545	8.177	190	955	0.000***	[0.48, 0.61]
	ICC2	Single Random Raters	0.551	10.376	190	950	0.000***	[0.46, 0.64]
	ICC3	Single Fixed Raters	0.609	10.376	190	950	0.000***	[0.55, 0.67]
	ICC1*k*	Average Raters Absolute	0.878	8.177	190	955	0.000***	[0.85, 0.9]
	ICC2k	***Average Random Raters***	***0.881***	***10.376***	***190***	***950***	***0.000******	***[0.84, 0.91]***
	ICC3*k*	Average Fixed Raters	0.904	10.376	190	950	0.000***	[0.88, 0.92]
Pre-Moderate							0.000***	
	ICC1	Single Raters Absolute	0.573	9.046	191	960	0.000***	[0.51, 0.63]
	ICC2	Single Random Raters	0.580	11.904	191	955	0.000***	[0.48, 0.67]
	ICC3	Single Fixed Raters	0.645	11.904	191	955	0.000***	[0.59, 0.7]
	ICC1*k*	Average Raters Absolute	0.889	9.046	191	960	0.000***	[0.86, 0.91]
	ICC2k	***Average Random Raters***	***0.892***	***11.904***	***191***	***955***	***0.000******	***[0.85, 0.92]***
	ICC3*k*	Average Fixed Raters	0.916	11.904	191	955	0.000***	[0.9, 0.93]
Pre-High							0.000***	
	ICC1	Single Raters Absolute	0.561	8.682	186	935	0.000***	[0.5, 0.62]
	ICC2	Single Random Raters	0.569	11.838	186	930	0.000***	[0.46, 0.66]
	ICC3	Single Fixed Raters	0.644	11.838	186	930	0.000***	[0.59, 0.7]
	ICC1*k*	Average Raters Absolute	0.885	8.682	186	935	0.000***	[0.86, 0.91]
	ICC2k	***Average Random Raters***	***0.888***	***11.838***	***186***	***930***	***0.000******	***[0.84, 0.92]***
	ICC3*k*	Average Fixed Raters	0.916	11.838	186	930	0.000***	[0.9, 0.93]

*ICC(1):"agreement" in exact values; ICC(2):"absolute agreement" in exact values when *e* is specified and modelled as random effects; and ICC(3): "consistent agreement" in exact values when *e* is specified and modelled as fixed effects; Reliability of the proportion of correct responses can be generalised by averaging across *k* measurements and be further expressed as ICC(1, *k*), ICC(2, *k*), and ICC(3, *k*). Higher ICC values closer to 1.0 represent perfect reliability.

**Significance was set at p<0.05 (two-tailed).

ICC2*k values were selected as a two-way mixed effects model based on the mean of *k* average random raters was appropriate to determine the absolute reliability of the SIFI test across multiple testing sessions.

Table 3.3. *Between Session ICC Results.*

	Type	Factor of Analysis	ICC	F	df1	df2	p-value	95% CI
	ICC1	Single Raters Absolute	0.657	12.493	185	930	0.000***	[0.6, 0.71]
	ICC2	Single Random Raters	0.664	20.094	185	925	0.000***	[0.52, 0.76]
Baseline – Pre	ICC3	Single Fixed Raters	0.761	20.094	185	925	0.000***	[0.72, 0.8]
Moderate – Pre High	ICC1*k*	Average Raters Absolute	0.919	12.493	185	930	0.000***	[0.9, 0.94]
	ICC2k	***Average Random Raters***	***0.922***	***20.094***	***185***	***925***	***0.000******	***[0.87, 0.95]***
	ICC3*k*	Average Fixed Raters	0.950	20.094	185	925	0.000***	[0.94, 0.96]

*ICC(1):"agreement" in exact values; ICC(2):"absolute agreement" in exact values when *e* is specified and modelled as random effects; and ICC(3): "consistent agreement" in exact values when *e* is specified and modelled as fixed effects; Reliability of the proportion of correct responses can be generalised by averaging across *k* measurements and be further expressed as ICC(1, *k*), ICC(2, *k*), and ICC(3, *k*). Higher ICC values closer to 1.0 represent perfect reliability.

**Significance was set at p<0.05 (two-tailed).

***ICC2*k* values were selected as a two-way mixed effects model based on the mean of *k* average random raters was appropriate to determine the absolute reliability of the SIFI test across multiple testing sessions.

Table 3.4. *Between-session TEM reliability across all SOA conditions.*

	N=	Condition	Mean ± SD	Variance	Limits of Agreement		Absolute TEM	VAV	Relative TEM (%)	Coefficient of Reliability (R)
					Lower	Upper				
Baseline-Pre Moderate	191	SOA-230	0.65 ± 0.35	0.123	-0.04	1.34	0.199	0.651	30.714	0.675
		SOA-150	0.63 ± 0.35	0.126	-0.07	1.32	0.209	0.626	33.469	0.652
		SOA-70	0.47 ± 0.34	0.116	-0.19	1.14	0.230	0.471	48.862	0.543
		SOA+70	0.41 ± 0.34	0.114	-0.26	1.07	0.219	0.406	54.052	0.578
		SOA+150	0.60 ± 0.34	0.112	-0.04	1.24	0.232	0.602	38.475	0.521
		SOA+230	0.74 ± 0.31	0.094	0.26	1.21	0.174	0.738	23.588	0.678
Pre Moderate – Pre High	188	SOA-230	0.68 ± 0.35	0.123	-0.01	1.36	0.219	0.680	32.217	0.609
		SOA-150	0.65 ± 0.35	0.119	-0.03	1.33	0.207	0.645	32.209	0.637
		SOA-70	0.48 ± 0.35	0.122	-0.21	1.16	0.224	0.483	46.516	0.587
		SOA+70	0.42 ± 0.34	0.116	-0.25	1.08	0.248	0.419	59.201	0.469
		SOA+150	0.63 ± 0.34	0.114	-0.04	1.29	0.218	0.632	64.553	0.582
		SOA+230	0.75 ± 0.29	0.090	0.17	1.34	0.179	0.759	23.519	0.645

*ICC(1):"agreement" in exact values; ICC(2):"absolute agreement" in exact values when *e* is specified and modelled as random effects; and ICC(3): "consistent agreement" in exact values when *e* is specified and modelled as fixed effects; Reliability of the proportion of correct responses can be generalised by averaging across *k* measurements and be further expressed as ICC(1, *k*), ICC(2, *k*), and ICC(3, *k*). Higher ICC values closer to 1.0 represent perfect reliability.

**Significance was set at p<0.05 (two-tailed).

***ICC2*k* values were selected as a two-way mixed effects model based on the mean of *k* average random raters was appropriate to determine the absolute reliability of the SIFI test across multiple testing sessions.

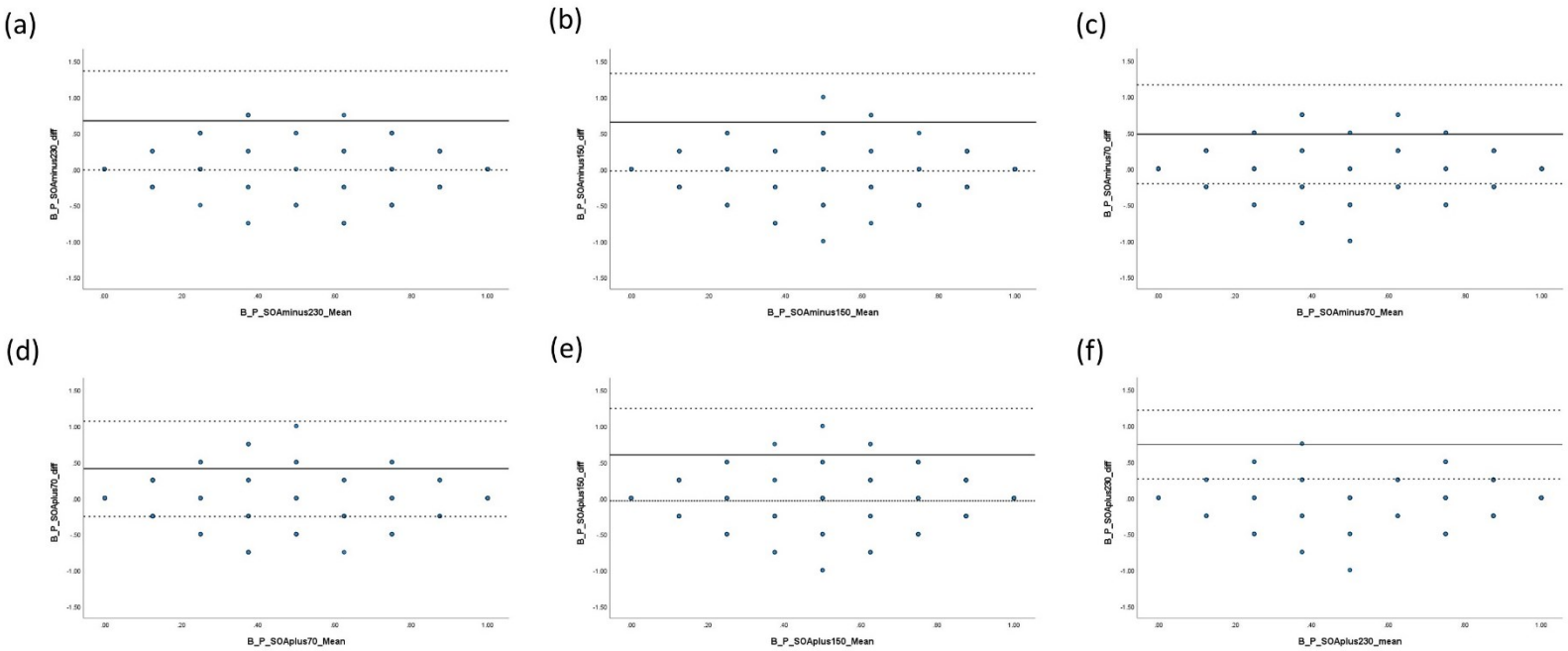

Figure 3.4. *Bland Altman Plots of each SOA condition for inter-session TEM reliability between Baseline and Pre-Moderate for the whole population.* **(a)** SOA-230, **(b)** SOA-150, **(c)** SOA-70, **(d)** SOA+70, **(e)** SOA+150, **(f)** SOA+230. The solid black line represents the mean, and the two dotted lines above and below represent the upper and lower limits of agreement.

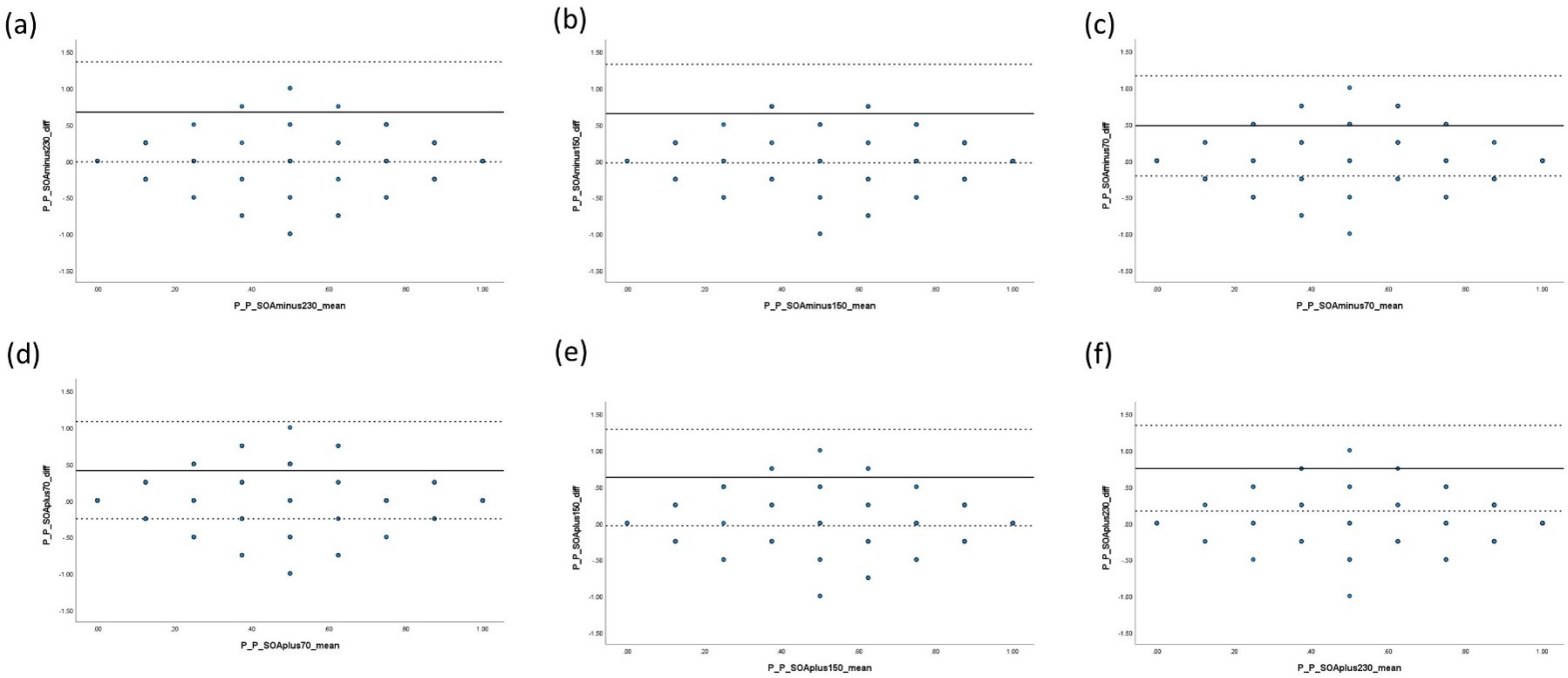

Figure 3.5. *Bland Altman Plots of each SOA condition for inter-session TEM reliability between Pre-Moderate and Pre-High for the whole population.* **(a)** SOA-230, **(b)** SOA-150, **(c)** SOA-70, **(d)** SOA+70, **(e)** SOA+150, **(f)** SOA+230. The solid black line represents the mean, and the two dotted lines above and below represent the upper and lower limits of agreement.

Table 3.5. *Within Session Reliability Analysis of SOA Conditions.*

	No. of Cases	Cronbach's Alpha	Sum of Squares	df	Mean Square	F	p-value
Baseline	191	**0.904**	13.877	5	2.775	52.373	**0.000*** **
Pre-Moderate	192	**0.916**	14.953	5	2.991	61.662	**0.000*** **
Pre-High	187	**0.916**	16.792	5	3.358	68.979	**0.000*** **

*Significance was set at p<0.05 (two-tailed).

Table 3.6. *Between Session Reliability Analysis of SOA Conditions.*

	Condition	No. of Cases	Cronbach's Alpha	Sum of Squares	df	Mean Square	F	p-value
Baseline – Pre Moderate – Pre High	*Overall*	186	**0.962**	44.236	17	2.602	45.851	**0.000*** **
	SOA-230	186	0.861	0.248	2	0.124	2.655	0.072
	SOA-150	186	**0.862**	0.405	2	0.202	4.287	**0.014***
	SOA-70	186	0.835	0.035	2	0.017	0.325	0.723
	SOA+70	186	0.812	0.163	2	0.081	1.409	0.246
	SOA+150	186	0.823	0.326	2	0.163	2.979	0.052
	SOA+230	186	**0.866**	0.252	2	0.126	3.627	**0.028***

*Significance was set at p<0.05 (two-tailed).

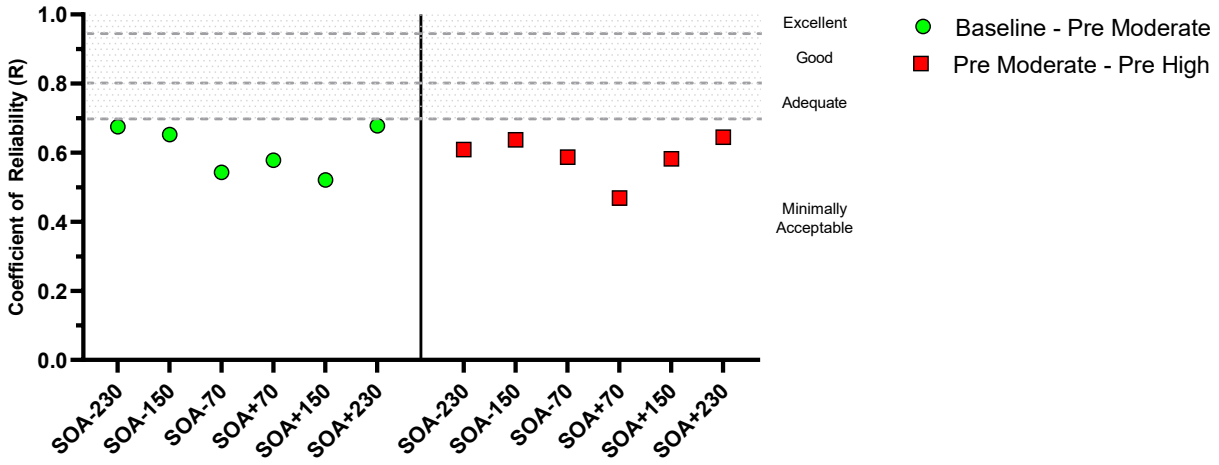

Figure 3.6. *Coefficients of Reliability across all SOA conditions between testing sessions.* Inter-session reliability across all 6 SOA conditions was found to be within the minimally acceptable range (i.e., <0.7).

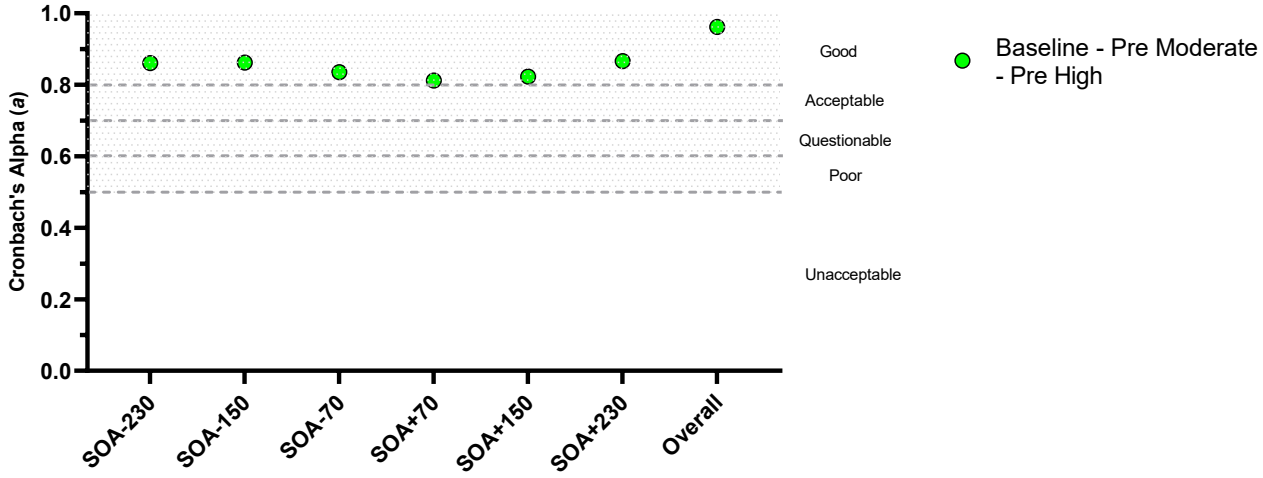

Figure 3.7. *Cronbach's Alpha across all SOA conditions and overall between testing sessions.* Inter-session reliability across all 6 SOA conditions was found to be within the acceptable-to-good range (i.e., >0.7).

Table 3.7. *Values of the expected agreement value.*

k	1	2	3	4	5	6	7	8	9	10	11	12	13
$\widehat{\gamma}_k$	1.00	1.00	1.00	1.00	0.96	0.99	1.00	0.97	1.00	1.00	0.26	0.00	0.00

All values of the expected agreement are listed for strata k = 1, . . . , 13 as shown in* ***Figure 3.10.*

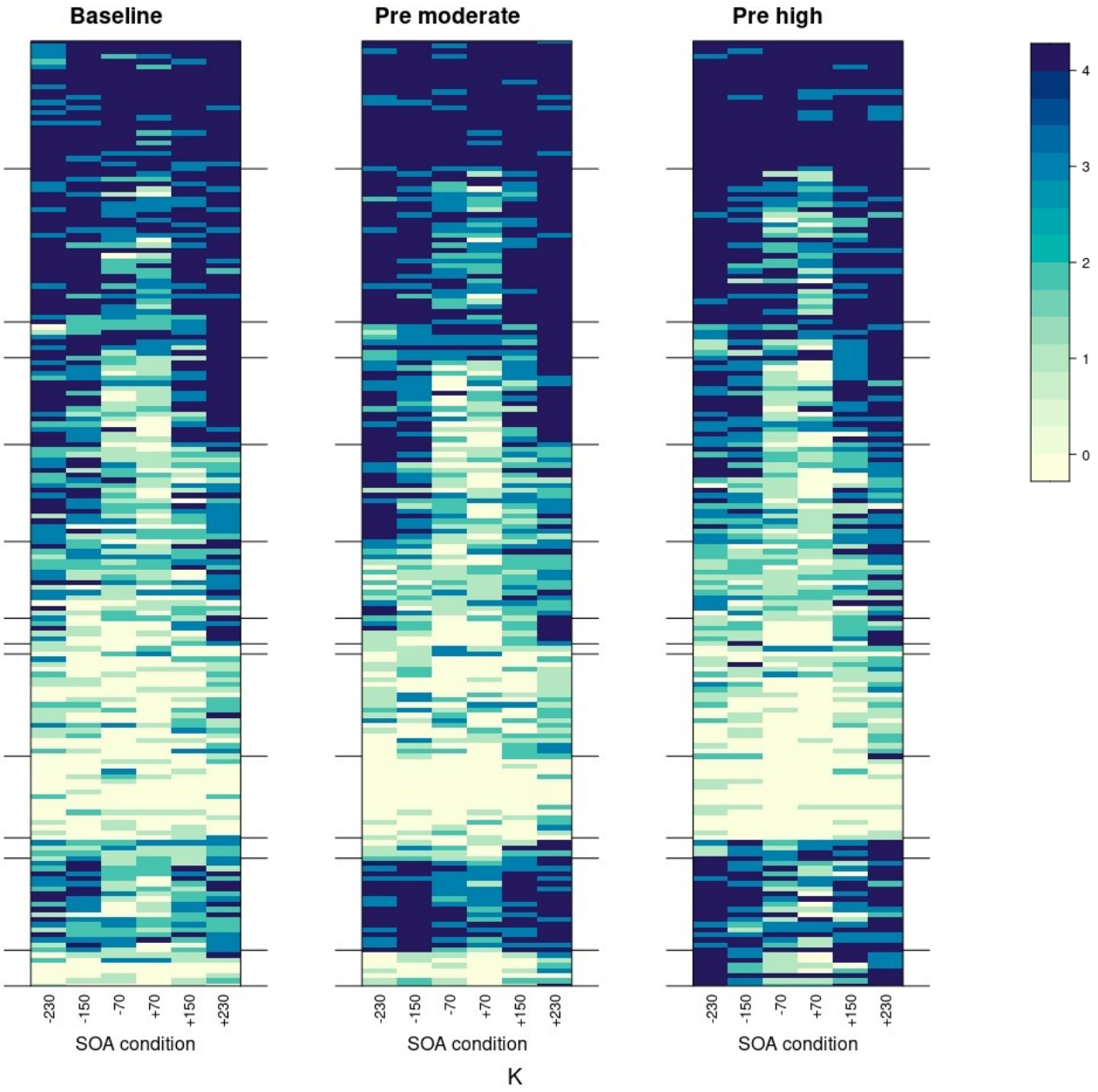

Figure 3.9. *Posterior probability of the number of strata connected over three time points demonstrating one aspect of the model uncertainty.*

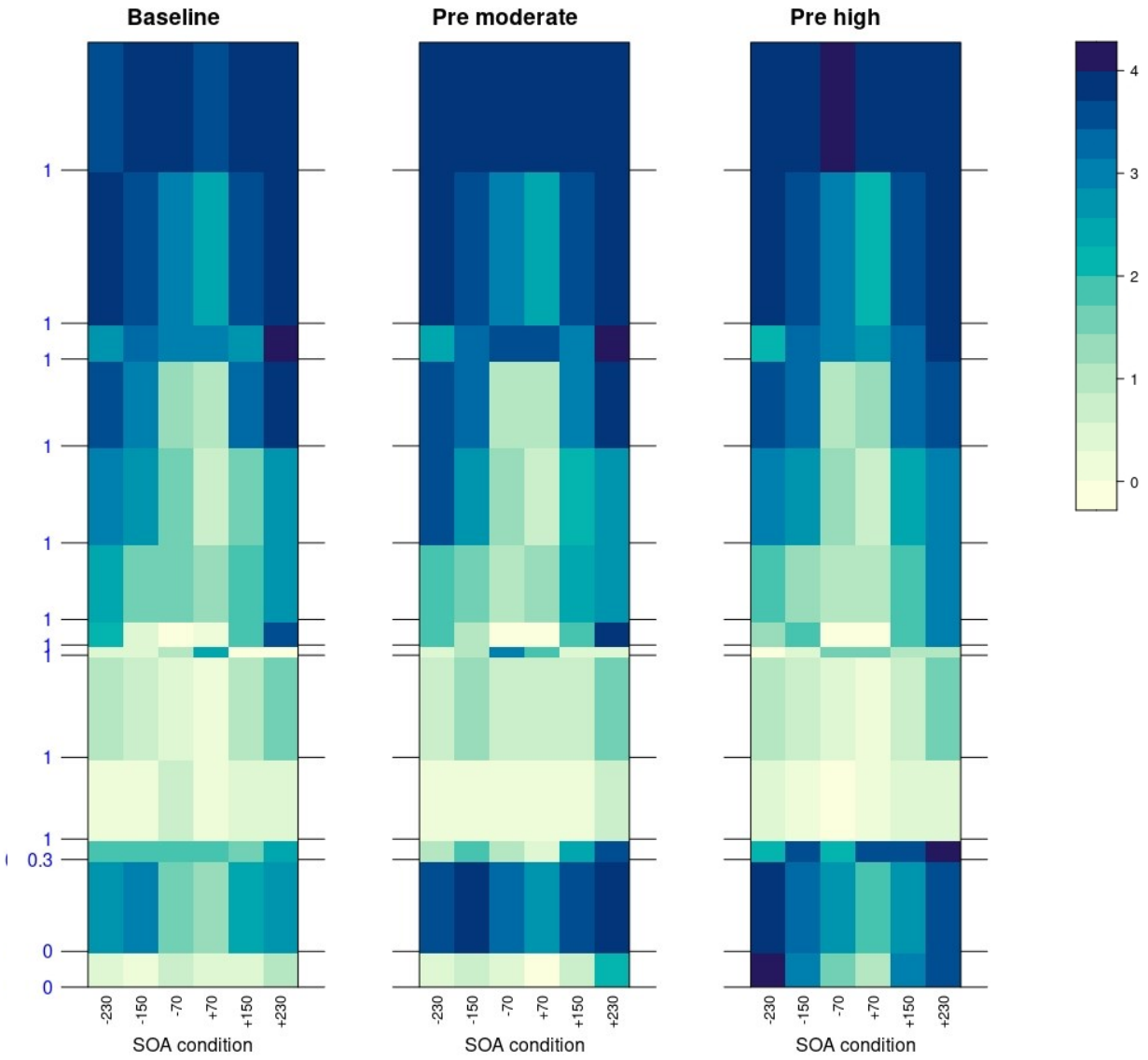

Figure 3.10. *Results of agreement model for most probable for $\hat{k}$ = 13 strata showing the mean of each variable within classes.* *Numbers on left are posterior agreement rounded to 1 place of decimals.

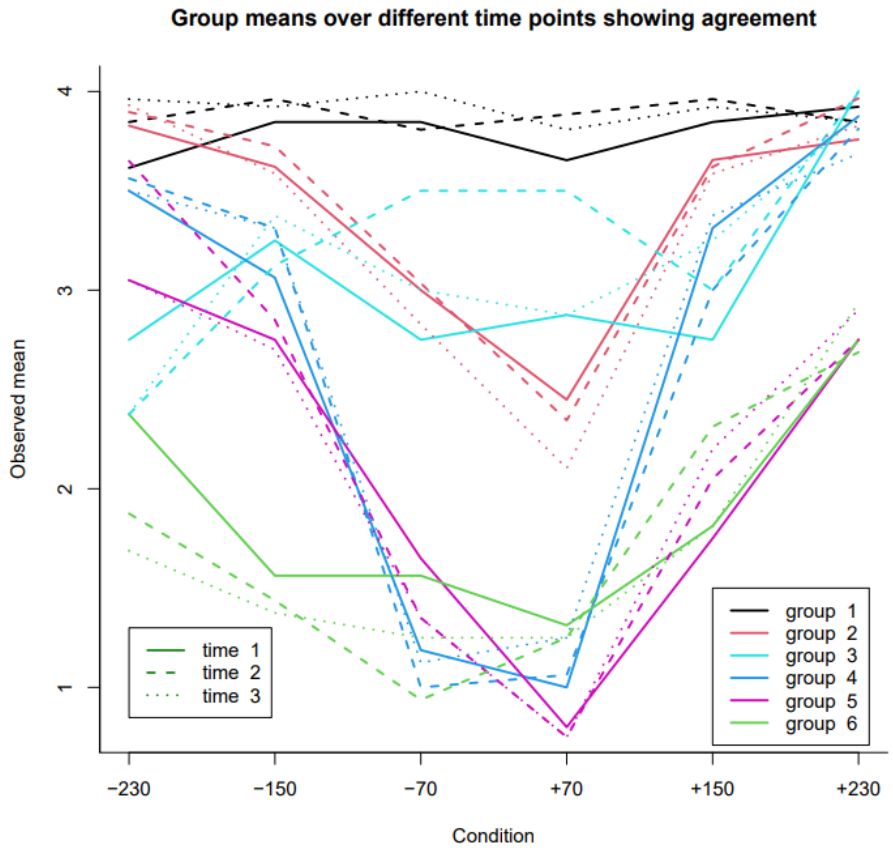

Figure 3.11. *Strata sample means for each SOA condition over the three time points (indicated by line type).*

3.5. Discussion

This is the first study to **(i)** determine the test-retest reliability of the SIFI test across separate testing sessions, **(ii)** assess the internal consistency of the SIFI test across each SOA condition using multiple measures of reliability, and **(iii)** determine the overall level of agreement between baseline testing sessions using a Bayesian Hierarchical Model of analysis. The results of our investigation found high levels of test-re-test reliability for the SIFI between and within sessions as evidenced by ICC and Cronbach's Alpha. However, there was a high level of measurement error across the SOA conditions. The highest level of susceptibility in those SOA conditions with the smallest level of temporal delay between presented stimuli produced the highest level of relative TEM. Likewise, those with the least perceptual susceptibility to the illusion were those SOA conditions with the largest temporal delay and concurrently showed the lowest level of relative TEM. Our Bayesian analysis concluded that,

although 13 strata were drawn, the reliability and level of agreement for SIFI was estimated at almost 80%.

Reliability and consistency of results during cognitive testing is essential, especially if the test is to be used as a possible diagnostic tool, e.g., for concussion. When no concussion is recorded there should be no identifiable change in cognitive performance across testing sessions. Baseline neuropsychological testing is now common practice among many amateur and professional sporting teams to create and archive a database across seasons for interindividual athlete comparison and comparison pre- to post-injury. By documenting an individual's level of cognitive function prior to a sporting season with the risk of concussive episodes, this accumulated data will also have the information to reliably model and determine both the presence and severity of cognitive decline following injury (596). Without a personal baseline of reference many sports clinicians are reliant on normative post-injury reference values which may not be suited for comparison thus increasing the risk of false positives. This may occur in those who are in fact healthy and void of injury, but their baseline cognitive scores fall below or sit at the lower end of what is deemed '*normal*'. Conversely, this may also incite false negatives in those with concussive injuries whose pre-injury cognitive function falls above the normative average and in fact have a decline in cognitive function. The assessment of the reliability of the SIFI test in the realm of sports is therefore essential for the creation of potential large-scale databases with personalised and normative reference values. Hence our investigation included presumed healthy individuals to ascertain the typical and atypical scoring trends of young adults in third level education for test interpretation in comparison to those who may be acutely suffering from a concussion.

The current diagnostic standard remains with subjective symptom reporting and clinical examination (2, 80). However, the consensus guidelines specifically recommend the assessment of cognitive function through computerised cognitive tools and conventional paper-and-pencil at baseline and following a concussive episode among athletes (2). Although the traditional paper-and-pencil neuropsychological tests are the gold standard for cognitive assessment, the rise of computer-based neurocognitive tools in recent years creates a platform for the adoption of new more objective measures of brain function and health such as the SIFI. Though widely used, computerised tests of cognition have been criticised for low reliability and poor sensitivity with coefficients as low as 0.22 for the ImPACT (597). The ImPACT test is a widely used computerised neuropsychological assessment tool for evaluating attention, memory, reaction time, and information-processing speed. Since its

inception in the 1990s, it has become the gold standard in assessing cognitive changes following a concussion, setting the benchmark for other evaluation tools. The psychometric characteristics of computerised neuropsychological testing platforms have been investigated, and the available evidence suggests a lack of reliability and validity across all existing computerised platforms. Aiming to best understand the test-retest reliability of the SIFI is crucial for clinical decision making and gaining invaluable insight as to what constitutes normal variation among a healthy population versus that attributed to concussion. Our results showed excellent levels of test-retest reliability and agreement, and high levels of internal consistency across testing sessions. Like most computerised tests used in the assessment of concussion and brain health, the SIFI is portable, easy to administer to large groups rather than just individuals and is low in cost. The SIFI test offers the additional advantage of assessing the additional cognitive functions of MSI beyond processing speed which are susceptible to concussion. However, any examination or procedure viewed as reliable would need to produce similar results regardless of time or environment.

When the items in a test relate to one another, the value of reliability and agreement, for example with Cronbach's alpha, rises. A high coefficient alpha, on the other hand, does not automatically imply a high degree of internal consistency. This is because the length of the test influences alpha. The value of alpha is lowered if the test length is too short. As a result, more similar items testing the same concept should be added to the test. In the context of the SIFI, there are six individual SOA conditions across a time period that is considered short in terms of practical utility, but in fact is lengthy enough to capture cognitive data from 64 trials across a time frame of around five minutes per participant. It is also worth noting that alpha is a characteristic of test scores from a given sample of participants or study population. As a result, researchers should not depend on published alpha estimates and instead assess alpha each time the test is given. Taking the SIFI three times across three separate testing sessions enabled us to gain valuable insight into the reliability and internal consistency of results across conditions. We found that the level of reliability and agreement was excellent overall and for each SIFI condition within an individual test and between several testing sessions. One point to consider is that the homogeneity among athletic sampled cohorts to date are likely to limit reliability estimates giving an inaccurate representation of results and possibly the utility of the test. Additionally, most tests of cognition assess specific domains which are susceptible to concussion but moreover susceptible to ageing. Over longer test-retest intervals various cognitive functions such as psychomotor processing speed, attention, memory develop through adolescence and young adulthood beginning to decline thereafter. Furthermore, evidence of low scores

performance on many traditional measures of cognition have shown up to 37% of healthy participants were atypical (598), which can lead to both an over- or underdiagnosis of concussion and associated cognitive decline. The SIFI test has shown in our analysis that it may be robust through its method of delivery over time with high levels of reliability to withstand the changes in cognition. Previous evidence shows that older adults produce similar scores to that of their younger counterparts highlighting a resilience and reliability of the SIFI with ageing (95, 114) making it a diverse tool in the assessment of cognition and brain health.

Recent systematic evidence however reports a wide range of test-retest coefficients ranging from 0.14 to 0.93 across computerized cognitive tools (599). It is most often assessed in the ImPACT with annual pre-season assessment and both short (six months, 0.35–0.86) and long term (four years, 0.29–0.69) follow ups. However, multiple attempts to increase the test–retest reliability of ImPACT have failed. What can be said from our current reliability results, although strong, and that of others to date is there is not sufficient evidence to claim that one computerised neurocognitive testing modality is superior to another in sports concussion, exercise, and future brain health. Reliability data on written neurocognitive tests over an 8-week interval across 48 athletes found test-retest correlations to range from .39 to .78, which did not reach the reliability estimates we found in our current investigation. Compared to only the second study to investigate the test-retest reliability of three commercially available computer neurocognitive tests used to assess SRC among healthy college age participants at clinically relevant time points, our results are similar but must be expanded upon across various sampled populations and larger cohorts (600). It was found that memory across neurocognitive tests had the lowest reliability coefficients, and reaction time and information processing had the highest values across all neurocognitive testing platforms. MSI can considered a measure of both reaction time and information processing when presenting an individual to various forms of sensory stimuli across pre-defined temporal windows of experience. Our results of reliability and agreement, although high and excellent, also pertain to many unidentified factors within the sampled cohort which must however be considered in future research and interpretation of results.

If a test's elements measure just one latent characteristic or concept, it is said to be unidimensional. Internal consistency is required but not sufficient for determining homogeneity or unidimensionality in a sample of test items (601). At its core, the idea of reliability requires that unidimensionality exists in a sample of test items, and if this assumption is broken, the dependability is greatly underestimated (602, 603). A multidimensional test does not necessarily have a lower

Cronbach's alpha than a unidimensional test for example, thus a more rigorous interpretation of alpha is that it cannot simply be taken as an indication of a test's internal consistency (604). As a result, rather than just measuring the unidimensionality of a group of items, alpha may be used to check whether or not a sample of objects is indeed unidimensional (601). Instead of calculating alpha for the entire test or scale, it should be therefore computed for each individual condition as was done for the six SOA conditions of the SIFI in this chapter. The results of this analysis showed high levels of internal consistency within all testing sessions highlighting the ever-increasing potential of the SIFI as a measure of cognition and brain health specifically among those in the sporting realm.

With close to 90% of all TBI's considered mild or concussive, symptoms will appear in the minutes, hours, or days after the event. Impairments of functional connectivity patterns within cognitive networks exist up to and beyond 6 months after the concussive injury, and the extent of reduced functional connectivity is associated with both symptomology and neuropsychological test performance (605). Due to the heterogeneity of the clinical aspects of concussion and sample populations, the literature necessitates an objective tool which considers underlying physiological function. Current and future assessments of cognition could employ and implement growing technology such as virtual reality and machine learning to improve the validity of results and subsequent clinical value. Making direct comparisons across neurocognitive tests is frankly problematic with each assessing their own domain of cognition, and although some may assess the same cognitive domain this is done using different tasks reducing our understanding of effectiveness in administration. Future research should aim to best understand the utility of the SIFI among other tests of cognition across an ageing, sports-based population ranging from adolescents and college students to middle aged, retired athletes and those in later life for the enhancement of test psychometrics such as test–retest reliability. For example, analysing the general population with non-sports related concussion and normal ageing would enhance testing utility and generalisability of results. Additionally, there is limited to no research assessing the outcome scores of neurocognitive tests when administered in languages other than English. This is an advantage of the SIFI provided it is a measure of stimuli response and perception, language ability is not a deciding factor in test administration making its utility in sport far more versatile.

3.6. Limitations

Several limitations have been noted in the current study. Although cognitive impairment would not be expected, atypical scores would be assumed to be false positives. However, a detailed medical history was not ascertained and the potential for underlying comorbid emotional and psychiatric factors or undiagnosed/unreported concussions may be present. If there was a significant decline in test performance between testing sessions it may be considered a false positive. For the purposes of this study a false positive would signify a healthy young adult who was identified as impaired based on the subsequent retest data. This may be the result of differences in physiological states between testing sessions, whether that be injury, lack of sleep, poor health, or ingestion of stimulants such as coffee. However, the testing protocol as outlined in **Chapter Two** for **Study One** ensured all participants refrained from heavy exercise, caffeinated drinks, and maintained a healthy sleep schedule prior to the day of testing giving credence to the reliability of the recorded SIFI results and subsequent high levels of reliability. Secondly, the current findings are limited to college students and thus may not generalise to additional cohorts. Future research should aim to further uncover the baseline reliability and incremental validity of the SIFI among other cohorts with neurological insults like acute concussion with Reliable Change Index-based comparisons for example. This form of conservative statistical analysis accounts for possible fluctuations in performance due to test-retest reliability and standard error of measurement (606). Thirdly, our time between baseline and retesting was on average 7-10 days which falls in line with the recovery of concussion but may significantly differ to the clinical timeline of follow-up assessments and ultimately return-to-play. The effect of these short retest intervals on estimates of reliability for the SIFI must be determined in future research. Reliability of testing output has been shown to depend highly on the length of time between testing sessions such that large variance in reliability coefficients were evident when the test-retest period was increased from 1-hour to 1-week (86, 607, 608). Fourthly, the level of attention and engagement engaged in by the participant undertaking the SIFI test is difficult to quantify and presents as a confounding factor. Those who may perform sub-optimally due to a lack of effort may alter the test-retest reliability as seen among previous clinical populations with TBI (609). This in turn would affect clinical decision-making with return-to-play and return-to-learn. Next, the use of the Pearson *'r'* calculation is limited by insensitivity to change in means over time due to practice or learning effects which can overestimate the relationship between measures when the sample size is constrained. However, we combatted this flaw by implementing ICC analysis as the univariate estimate of agreement on top of devising our Bayesian hierarchical model assessing the overall levels

of agreement. As ICCs values might vary depending on model, kind, and metrics utilised, this information should be supplied in the written text to avoid misinterpretation where ICC results may be influenced by causes other than measurement error. Finally, this is the first study to the authors knowledge assessing the reliability and level of agreement of the SIFI over several testing sessions. Further research declaring their statistical models, types and measurements is required to better understand the SIFI across a wider and more heterogenous population to comprehend the potential utility and applicability of the SIFI in the sporting realm.

3.7. Conclusion

Reliability is defined as producing the same or comparable findings in different clinical studies or statistical trials. Internal consistency, overall reliability, and level of agreement between testing sessions using the SIFI was excellent. The idea of reliability is essential because it may be utilised to eliminate mistakes during diagnostic examinations. Computerised neurocognitive tests do not, however, eliminate the necessity for clinical judgement and interpretation of results. Normative data should be increasingly used across clinical and research settings. The current evidence gathered for this chapter has created a new platform in the future assessment of measurement error and rates of impairment among larger cohorts involving the SIFI in a need for more translation to clinical relevance. A combination of assessments using tests that evaluate various cognitive functions would subsequently increase accuracy and overall efficacy of results to create a robust multimodal analysis of psychometric approaches. Clinicians and health-care professionals should therefore be careful about their choice of neurocognitive evaluation with emphasis placed on those tests with the highest level of reliability to suit their requirements.

3.8. Question(s) Raised

- How does sporting engagement, concussion history, and other covariates including age and sex affect perceptual performance during the SIFI neurocognitive assessment?

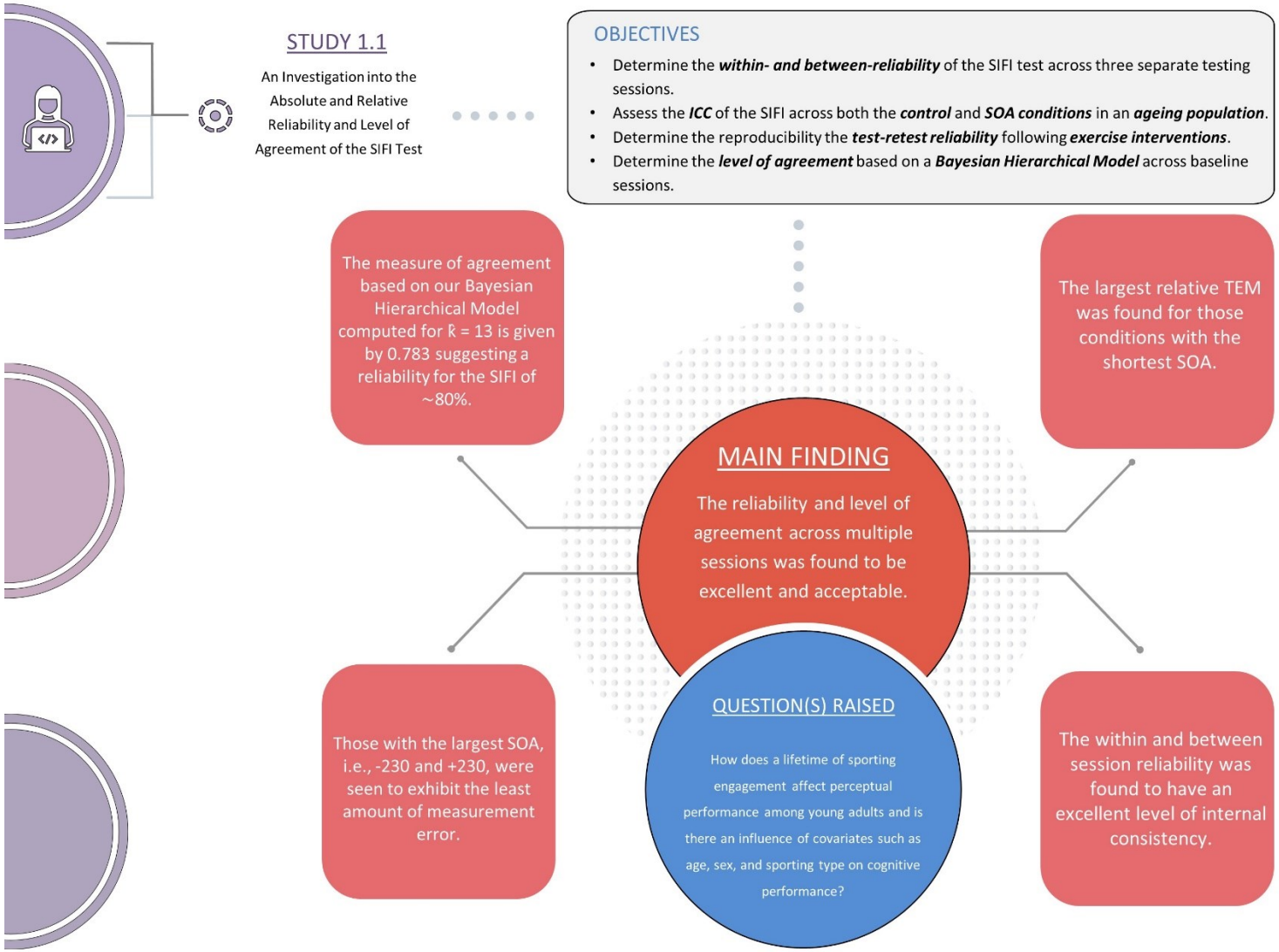

Figure 3.12. *Chapter Three – Study 1.1 Summary.*

4

Chapter Four:

Multisensory Performance and Brain Health among University Students with a History of Concussion and Various Sporting Backgrounds Assessed using the Sound induced Flash Illusion

4.1. Highlights

- Males had higher levels of perceptual performance capabilities in both the pilot and follow-up study.
- Contradictory to our hypothesis, those with and without a concussion history were found to perform with similar perceptual accuracy across all illusory SOA conditions.
- Those who participated in contact-based sports, who are expected to have higher exposure rates to possible concussive episodes, performed with higher levels of perceptual accuracy, although results were not statistically significant.
- Younger age and playing open skill sports appear to distinguish high scorers from low scorers in perceptual ability during the SIFI task.
- Sporting cohort, time since ceasing sporting engagement, and previous number of concussions were predictors of depression, and specifically a higher number of previous concussions was linked with increased metrics of somatisation, anxiety, and depression.
- Sex was a significant predictor variable of concussion knowledge, such that males were found to have a marginally better score of concussion knowledge and attitude compared to females.

4.2. Introduction

Multisensory Integration (MSI) is the process by which various sources of sensory input are combined to form a multisensory product that cannot be easily deconstructed into its constituent unisensory inputs (225). MSI is a unique ability that is possessed by most mammalian species which is not an innate attribute but is functionally created through sensory experience and neural plasticity. To navigate daily life, one must be capable of making accurate perceptual appraisals of their surrounding environment, especially among sporting populations.

An athlete's capability to make concise and informed behavioural decisions is vital to their performance during training and competition. For athletes this involves the constant accumulation of multiple sensory systems, identifying biologically significant stimuli, and translating them to produce desirable reactions that may otherwise not be achievable through a singular sensory input (201-204). Posterior-parietal regions of the brain have been cited to automatically fuse multisensory inputs, while the anterior parietal regions enable a more adaptable spatial representation of the event produced from statistical predictions of Bayesian causal inference (155, 156). Athletes are consistently

trained to heighten their current ability to process multiple forms of sensory input for sporting success. Indeed, consistent training heightens this ability to process multiple forms of sensory input for sporting success (205-207). A key sensory principle of MSI exploited by athletes is that of inverse effectiveness where decreasing the effectiveness of individual sensory stimuli increases the magnitude of multisensory enhancements (610). Sensory stimuli experienced at a high level of saliency are less likely to be integrated than multiple sensory stimuli presented simultaneously at low levels of saliency. This principle in a sporting context can be supported by attentional resource load theory which postulates that the recruitment of higher-order attentional networks may shield the observer from distractions when the perceptual load is high (247), with the intraparietal sulcus mediating additional attentional resource allocation for the integration of A-V stimuli (611, 612). Based on this theory, those participating in specific sports may experience a higher level of possible distractors and develop more fine-tuned perceptual and attentional abilities. Those who participate in fast-paced, dynamic sporting environments with arguably a higher level of multisensory stimuli may facilitate enhanced MSI capabilities. Countless sport-specific training programs can thus be developed incorporating multiple sensory modalities based on the key principles such as representative learning design (RLD) (205). Such training programs aim to create scenarios using performance-relevant sources of sensory information such as visual and/or verbal cues for the sporting athlete to perceive and subsequently act upon in hopes of transferring to competition (205). Moreover, humans are inherently neurobiological systems who use their knowledge and perception both of and about their environment; from the surfaces and surroundings to the people and noises; to provide the opportunity for action, and are ultimately relative to the capabilities of the sporting individual (202, 294). These moments of potentiality are known as affordances and are defined as *'*key ecological constraints on what humans perceive, what they learn and know, and how they decide and act' (204). Moments such as these in sporting environments can, however, be associated with a possible risk of injury such as concussion despite the development of sporting expertise.

Concussion and brain health are a major public health concern and considered a silent epidemic by many, with the annual incidence consistently rising across multiple settings (613). Although the brain's resilience to linear acceleration remains steadfast (614), the angular or rotational acceleration associated with concussion (1, 26) can have long-lasting effects including altering the ability to process multiple forms of stimuli (366, 615). Injury-induced executive hypofunction associated with a history of concussion could result in impaired performance in a fast-paced, dynamic game situation due to a reduction in relevant information processing (188, 363). The direct cortico-cortical pathways can be

negatively impacted by biomechanical forces manifesting in tensile shear and compressive tissue strain with the potential for diffuse axonal injury which can lead to the development of altered cerebral function and subsequent information processing (23, 24). Concussive events are known to affect perception, reducing MSI efficiency (616, 617), and negatively affecting neurocognitive functioning during immediate and long-term recovery (190, 618). TBI, including concussion, can result in long term negative effects on brain function, (619, 620), yet understanding of its pathophysiology remains limited and most concussions are undiagnosed or underreported (6-8).

Audio-visual (A-V) illusion paradigms enable assessment of MSI (95, 102). The SIFI occurs when a single visual flash that is shown in conjunction with two or more auditory tones is subsequently perceived as two or more flashes. Previous studies have used susceptibility to the SIFI as a measure of perceptual efficiency (97-99). Increased SIFI susceptibility has been reported in cohorts with neurological impairment, brain damage, and mild cognitive impairment supporting the potential application of the SIFI to delineate between healthy and injured populations (105, 174). For example, the work of Chan et al. compared 14 older adults with mild cognitive impairment (MCI), aged 65 to 78, to 16 age-matched healthy older adults (100). The results indicate that individuals with MCI perceived more illusions and over a broader range of auditory SOAs compared to the healthy controls. This suggests that not only are individuals with MCI more inclined to integrate A-V information, but they also integrate this information across a wider temporal window. The increased perception of illusory flashes in individuals with MCI is likely due to their inability to suppress irrelevant auditory stimuli such that the SIFI can serve as a valuable tool for assessing cortical connectivity between sensory regions. Moreover, a previous study from our laboratory found that history of concussion had no relationship with performance of the SIFI test in older athletes who had retired from sport (172). There is however an effect of the neurometabolite GABA and glutamate systems accounting for individual differences in multisensory processing (103). This may be linked with potential deficits among those previously affected by concussion. It is believed to have a role in axonal degeneration and long-lasting dysfunction post-injury (23, 24). Pathological processes such as these may have a significant impact on MSI during the SIFI test given that damage to axons and WM tracts from the initial injury may not fully recover, leading to impaired neurotransmission and reduced processing speed (182, 184, 185). The SIFI test, by evaluating specific brain regions handling the temporal synchrony and processing of A-V stimuli in the environment (621-623), could offer new insights into the biometric data associated with concussive impact pathology and the associative brain status (187, 624).

In sports, various situations require the integration of multiple senses to enhance rapid situational awareness and object detection. The SIFI test, a neurosensory challenge, could serve as a more effective method for assessing concussions compared to self-reports. It has the potential to distinguish between healthy and injured individuals and might help identify concussion risk provided impairments in MSI are common in those with a history of concussions. Consequently, the SIFI test may aid in diagnosing and predicting cognitive function impairments beyond identifiable structural damage. In this study, our objective was to investigate multisensory processing efficiency in young male and female collegiate athletes actively participating in either contact or non-contact sports. The aims of this study were to: **(i)** investigate MSI efficiency in collegiate sporting athletes in both sexes and, **(ii)** determine whether perceptual performance is influenced by self-reported history of concussion.

4.3. Methods

4.3.1. Study Design

The following study was cross-sectional in nature. It consisted of an initial pilot study and subsequent follow-up study which was conducted at two separate testing periods due to the COVID-19 pandemic; *see* ***Figure 2.1.*** in **Chapter Two** *for complete details*. Participants were recruited from the student population of Trinity College Dublin and collegiate sporting teams into three cohorts: contact athletes, non-contact athletes and non-athletic controls (*see* ***Table 4.1.***). The contact, non-contact and control cohorts were established based on the Rice categorisation tool of sporting cohorts (508). A power analysis for participant recruitment was not performed as the study was exploratory in nature. All participants were informed their participation in the study was voluntary and were supplied with a participant information leaflet. Written and informed consent was provided by all participants and ethical approval was granted by Trinity College Dublin Faculty of Health Sciences. The research was conducted in accordance with the Declaration of Helsinki and all subsequent amendments. Detailed information regarding participants, recruitment, eligibility criteria, ethical approval, and complete methodological descriptions for **Study One** is described in detail in **Chapter Two**.

4.3.2. Outcome Measures

4.3.2a Participant Demographics

Information regarding participant age, sex, height, weight, and BMI were obtained from all participants. Details of sporting history, time since sporting retirement, number of previous concussions, and knowledge & attitudes towards concussion were also gathered; see **Chapter Two Section 2.2. and 2.3.** for full details and assessment protocols.

3.3.2b Health-related Outcomes

The following health-related self-reported questionnaires, previously described in **Chapter Two**, were completed by all participants:

- The International Physical Activity Questionnaire (IPAQ) Short Form *(see **Section 2.4.2c**)*
- Brief Symptom Inventory (BSI) *(see **Section 2.4.2d**)*
- Generalised Anxiety Disorder Assessment (GAD-7) *(see **Section 2.4.2e**)*
- Patient Health Questionnaire (PHQ-9) *(see **Section 2.4.2f**)*
- Short Form 12 (SF-12) Health Survey *(see **Section 2.4.2g**)*

4.3.2c Cognitive Outcomes

The SIFI task involves participants reporting the number of visual flashes or auditory beeps presented to them. The experiment allows participants to set their own pace, with no emphasis on response speed. The fission illusion occurs when participants perceive two visual flashes after being presented with a single visual flash and two auditory beeps (1F2B) at specific time intervals. Control conditions include unimodal scenarios (1F0B, 2F0B, 0F1B, and 0F2B) and bimodal congruent conditions (1F1B and 2F2B) to establish a performance baseline for each participant. These baseline measures ensure that participants have similar abilities to perceive visual and auditory stimuli. In unisensory visual conditions, participants see either one or two flashes with no auditory beeps at a specific time interval (70ms) and report the perceived number of flashes. Unisensory visual conditions are mixed with multisensory conditions in the first block of stimuli. Auditory-only conditions are presented separately, indicated by a written command on the computer screen, and participants

report the number of perceived beeps. They hear either one (0F1B) or two (0F2B) auditory beeps at time intervals of 70ms, 150ms, and 230ms.

Multisensory trials consist of congruent or incongruent conditions. In congruent trials, the same number of visual flashes is presented with the same number of auditory beeps, either one flash with one beep (1F1B) or two flashes with two beeps (2F2B) at time intervals of 70ms, 150ms, and 230ms. In incongruent trials, one visual flash is always paired with two auditory beeps (1F2B) at time intervals of 70ms, 150ms, or 230ms. The first auditory beep is always synchronised with the visual flash, while the second auditory beep either precedes or follows the visual-auditory pairing, resulting in 6 time intervals (±70ms, ±150ms, and ±230ms). These time delays are referred to as negative and positive time intervals, indicating whether the beep comes before or after the visual-auditory pairing. Similar to unisensory visual trials, participants report the perceived number of visual flashes for all multisensory conditions and disregard the auditory beeps.

4.3.3. Statistical Analysis

Participant descriptive statistics were sub-categorised by sex and sporting type. A χ^2 test was used to establish proportions. Descriptive statistics of participant characteristics were reported as mean (SD) or median (IQR), as appropriate. For all analyses, $p < 0.05$ (two-tailed) was taken to be statistically significant.

The coding of responses for the SIFI task was done in accordance with previous reports (99, 268). Temporal parameters of SIFI susceptibility were examined across all 64 trials representative of 16 conditions including the 6 unisensory conditions and 10 multisensory conditions consisting of 4 congruent and 6 incongruent illusory trials with six distinct SOA's (±70,150, 230ms) (96, 99, 102). To eliminate possible effects caused by differences in the baseline performance of the control conditions, we determined whether groups differed in the unimodal (1F0B, 2F0B, 0F1B, and 0F2B) and bimodal congruent conditions (1F1B and 2F2B). The proportion of correct responses was calculated per condition (unisensory and multisensory) and SOA for each individual and used for all statistical analyses. Across each trial, the accuracy of response was scored as either 0 or 1 equating to a *'miss'* or *'hit'*, respectively, *i.e.,* when two flashes were presented during a unisensory condition (2F0B) or 2 flashes presented congruently with 2 auditory beeps (2F2B), a correct response was recorded for the participant when they indicated that two flashes (or beeps when flashes were not present) occurred.

During the multisensory incongruent conditions (1F2B), perception of one visual flash was noted as a correct response, and perception of two visual flashes was noted as an incorrect response, *i.e.*, when the fission illusion was experienced. A lower level of perceptual accuracy across the illusory incongruent conditions, *i.e.,* a high number of misses, indicated the participant had experienced a high number of fission illusions. With each trial repeated 4 times for each of the 16 conditions, the participants score of perceptual accuracy was either 0, 0.25, 0.5, 0.75, or 1 and acted as dependent variables of analysis. Between-group differences were determined using mixed model ANOVA's to uncover main effects and two-way ANOVA's were subsequently run to determine the effects of each multisensory SOA condition across between-subject factors. For all analyses, Greenhouse–Geisser correction was used when assumption of sphericity was violated (Mauchly's test) and the adjusted p-values alongside original degrees of freedom were reported. The violation from sphericity is denoted by epsilon (GGeps). For pairwise comparisons, Bonferroni correction was applied to maintain an α-level of 0.05, and adjusted p values are stated.

The mean proportion of correct responses across all conditions for each participant was established for cohort, sex and concussion history, range, and timing. Temporal parameters of susceptibility were examined across all six SOA's among the multisensory incongruent conditions known to generate the fission illusion (96, 99, 102). To eliminate possible effects that were caused by differences in the baseline performance of the control conditions, we determined whether groups differed in the unimodal (1F, 2F, 1B, and 2B) and bimodal congruent conditions (1F1B and 2F2B).

Logistic Regression was initially deemed appropriate to model the number of correct responses in each condition for each individual, allowing the probability of a correct response to vary according to group and condition based on the pilot study data. If there are *n* repetitions of the condition for each individual *i*, the number of correct responses *y* is binomial with success probability *pi*:

$$y_i \sim \text{Binomial}(n, p_i)$$

where p_i is modelled via

$$\log\left(\frac{p_i}{1-p_i}\right) = \alpha + \gamma_i + \beta_i$$

with γi the group effect for individual i (Contact, Non-Contact, Control) and βi is the effect for condition. A grouped logistic regression was used. To investigate whether concussion history was a predictor of performance on the SIFI a Logistic Regression Model was then devised to avoid issues associated with multicollinearity.

Due to the multivariate nature of the response for each individual (six SOA conditions), latent class analysis (LCA) or Bayesian classification was subsequently used to determine whether there were any consistent patterns across the SIFI conditions. LCA is a statistical approach for identifying qualitatively distinct subgroups within populations that frequently share certain external traits. The premise behind LCA is that patterns of scores across particular measurements can explain membership in unobserved groups (or classes). LCA is used to detect latent (or unobserved) heterogeneity in samples. LCA groups participants by their propensity to successfully respond to the various experimental conditions. Individuals' scores on a set of indicator variables are determined by their class membership, according to statistical theory. This is comparable to the concept of a latent construct driving scale item scores in factor analysis processes. Each group has its own set of six propensity (probabilities) of success. This form of analysis rooted in a cross-classification of two more observed categorical variables enabled us to classify a multidimensional discrete latent variable. Individuals are assigned to classes depending on their likelihood of being in those classes based on their pattern of scores on indicator variables. The actual number or percentage of sample members within each class cannot be known since class assignment is dependent on probability.

A six-group model was objectively selected by considering anything from one to eight groups and then determining that six groups gave the lowest value of the Bayesian Information Criterion (BIC). BIC is a criterion for comparing models which trades off model complexity and fit, through penalising against over-fitting. The number of actual correct responses and the LCA determined expected number of correct responses (shown by the bold line) were calculated and subsequently depicted.

Ideally, the patterns observed from the latent class analysis would be explained by sex, cohort and/or concussion history, range, or timing. An LCA which includes any combination of these covariates and anything from one to eight groups and all possible combinations of models were considered. To confirm the conclusions of the LCA analysis, a logistic regression model was fitted to the responses taking the group label as a covariate in the model.

All data were exported from Microsoft Excel, for statistical analysis using the Statistical Package for the Social Sciences (SPSS version 25; IBM, Ireland). Figures were generated using GraphPad Prism (GraphPad Software version 8, San Diego, CA).

4.4. Results

Pilot Study

4.4.1. Participant Demographics

130 participants (74 Males, age: 21.97 ± 2.38 years; 56 Females, age: 21.54 ± 1.69 years) were recruited for this pilot study from the student population of Trinity College Dublin and collegiate sporting teams.

Table 4.1. *Participant Demographics.*

	Male		Female		Total	
	Age (mean ± SD)	**n =**	**Age (mean ± SD)**	**n =**	**Age (mean ± SD)**	**n =**
Contact*	22.35 ± 2.19	37	23 ± 3.46	3	22.42 ± 2.29	40
Non-Contact*	21.75 ± 2.80	25	21.63 ± 1.85	40	21.68 ± 2.28	65
Control*	21.92 ± 1.33	11	21.39 ± 1.49	13	21.26 ± 1.45	24

**Note: A total of 9 participants excluded from demographic analysis due to missing data.*

4.4.2. Concussion History

A total of 105 self-reported concussions were recorded using the MTBIIM; see **Section 2.4.2a – Appendix 1.7**. The average age of injury was 17.37 ± 4.15 years. The most common mechanism of injury was sports-related (77.1% of all concussions), followed by a fall and blow to the head or neck. Just over half of all concussions were diagnosed by a clinician (50.5%). 43 of the 105 concussive events involved a loss of consciousness (LOC) with an average duration of 26.82 seconds. 54.3% of the concussions led to self-reported memory problems, lasting an average of 99.62 minutes. The average length of post-concussion symptoms experienced by the cohort was 9.78 ± 27.48 days.

Table 4.2. *Population Data of Concussion History, and Timing.*

	Female	Male	Total
	n =	n =	n =
Concussion History			
Yes	11	34	45
No	45	40	85
Concussion Timing			
No Concussion	45	40	85
Recent Concussion (≤6 months)	1	8	9
Past Concussion (>6 months)	10	26	36

Concussion history revealed that 85 participants had not suffered a previous concussion, 36 were recorded as having a previous concussion at the time of testing (>6 months prior to testing), and 9 were found to have a recent history of concussion, i.e., within 6 months of their testing session.

Sex was found to have a significant between-subjects effect (df = 1, F = 9.605, p=0.002) with a moderate effect size (η^2 = 0.07). Males (mean = 1.05, SD = 1.629) had significantly more concussions than females (mean = 0.32, SD = 0.789) with a mean difference of 0.733 concussions. Cohort had a significant between-subjects effect (df = 2, F = 6.944, p=0.001) with a moderate effect size (η^2 = 0.099). The Contact cohort had significantly more concussions than the non-contact cohort (Contact: mean = 1.37, SD = 1.609; Non-Contact: mean = 0.40, SD = 1.012; mean difference = 0.966, p=0.001), and the control cohort (Control: mean = 0.58, SD = 1.501; mean difference = 0.783, p = 0.068). Unexpectedly, the Control cohort (mean = 0.58, SD = 1.501) suffered more concussions than the Non-Contact cohort. The interaction of Sex x Cohort revealed a non-significant main effect (df = 2, F = 0.176, p = 0.839, η^2 = 0.003).

4.4.3. SIFI Results

4.4.3a Concussion History and Timing

Across the entire study population, those with no concussion history had a marginally higher performance accuracy (mean = 0.758, std = 0.011) than those with any history of concussion (mean = 0.755, std = 0.016) across the control conditions. However, a mixed model ANOVA test uncovered a non-significant main interaction of concussion history*control conditions (df = 4.547, F = 0.569, p=0.707, GGeps = 0.505, η^2 = 0.004), and non-significant between-subjects effect of concussion history

(df = 1, F = 0.028, p=0.868, η^2 = 0.000). Uncharacteristically, those with a past concussion history, *i.e.*, more than 6 months prior to testing, were found to perform better (mean = 0.765, std error = 0.018,) than those with no concussion history (mean = 0.758, std error = 0.011) and those with a recent concussion history (mean = 0.714, std error = 0.035); *see* ***Figure 4.1.*** *below*. Although, these results were shown through a mixed model ANOVA to have a non-significant main interaction of concussion timing*control conditions (df = 9.071, F = 0.758, p=0.656, GGeps = 0.504, η^2 = 0.012), and a non-significant between-subjects effect of concussion timing against the control conditions (df = 2, F = 0.869, p=0.422, η^2 = 0.013).

To determine the effect of concussion history on SIFI performance accuracy in respect to the fission illusion SOA conditions a mixed model ANOVA was undertaken. Those with and without a concussion history were found to perform with similar perceptual accuracy across all illusory conditions (mean difference = 0.043, SE = 0.05), although contradictory to our proposed hypothesis. A test of within-subjects effects revealed a significant main effect of SOA condition (df = 3.931, F = 46.458, p=0.000, GGeps = 0.786, η^2 = 0.266), and a non-significant interaction effect of concussion history (df = 3.931, F = 0.29, p=0.881, η^2 = 0.002). A test of between-subjects effects revealed a non-significant effect of concussion history on the SOA conditions (df = 1, F = 0.743, p=0.390, η^2 = 0.006). Similarly, concussion timing had a non-significant interaction effect of Concussion timing (df = 7.902, F = 0.042, p=0.808, GGeps = 0.79, η^2 = 0.009) and a non-significant between-subjects effect on SOA conditions (df = 2, F = 1.955, p=0.146, η^2 = 0.030); *see* ***Figure 4.2.*** *below*.

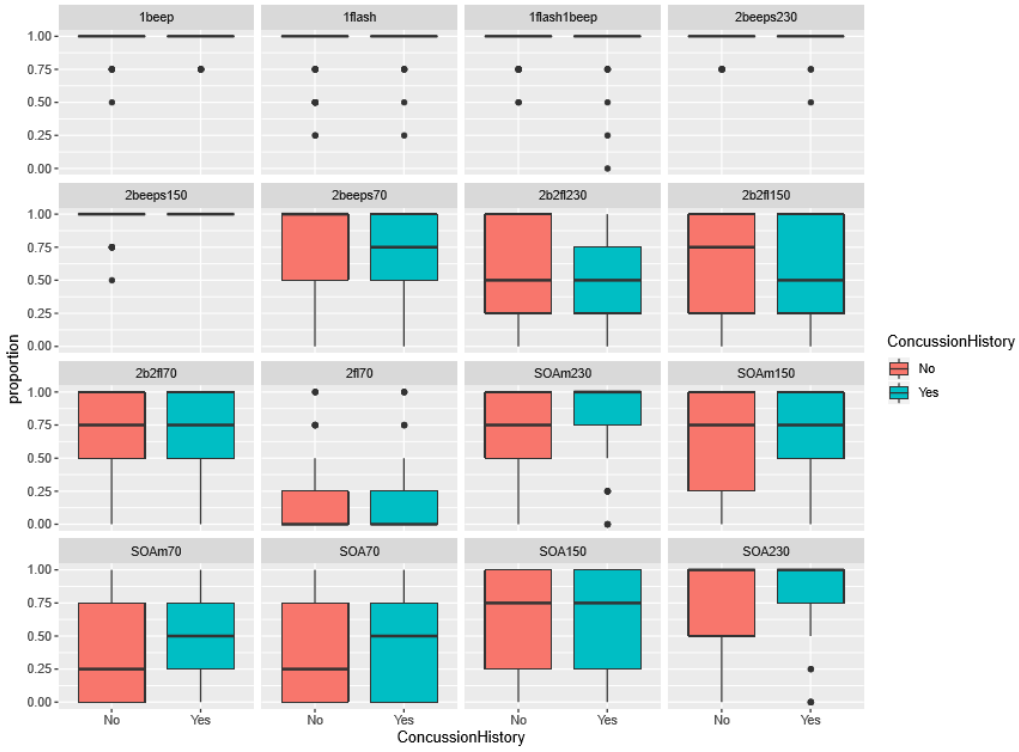

Figure 4.1. *Boxplots showing the Proportion of Correct Responses broken down by Concussion History.*

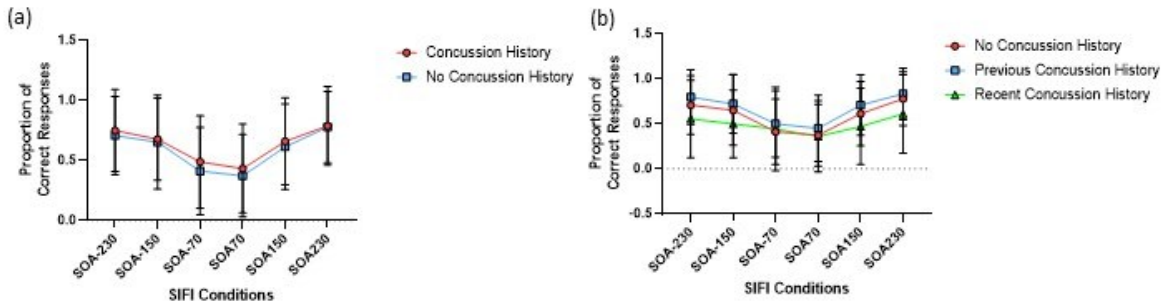

Figure 4.2. *Proportion of correct responses for entire cohort under the groupings of: Concussion History, Concussion Timing, and Concussion Range.* **(a)** Concussion History: No statistical differences were observed for any of the experimental SOA conditions between individuals reporting a history of concussion in their lifetime and those without any concussion history. **(b)** Concussion Timing: Although no significant differences were detected between groups, individuals with more recent concussions exhibited lower perceptual accuracy across all SOA conditions, particularly those with the largest SOA, i.e. SOA ±230.

4.4.3b Sporting Cohort

A mixed model ANOVA was run to investigate whether there were any differences between cohorts in the control conditions. There was a significant main effect of the control conditions (df = 4.54, F = 140.691, p=0.000, GGeps =0.504, η^2 = 0.526), a non-significant main interaction of cohort*control conditions (df = 9.081, F = 0.662, p=0.745, η^2 = 0.01), and a non-significant effect of cohort against the control conditions (df = 2, F = 1.198, p=0.305, η^2 = 0.019). Post-hoc analysis revealed that the only significant difference found was that the Non-Contact cohort performed significantly better than the Control cohort during the 0F2B SOA+70 control condition (Mean difference = 0.1619, SE = 0.0579, p=0.0157). Although not significant, the Non-Contact cohort (mean = 0.767, std error = 0.013) had a higher performance accuracy defined as group-averaged proportions than both the contact (mean = 0.759, std error = 0.016) and control (mean = 0.728, std error = 0.021) cohorts across control conditions.

To determine the effect of cohort on the fission illusion SOA conditions, a mixed model ANOVA revealed a significant main effect of SOA condition (df = 3.989, F = 47.789, p=0.000, GGeps = 0.780, η^2 = 0.273), a non-significant main interaction effect of cohort*SOA conditions (df = 7.796, F = 0.667, p=0.717, η^2 = 0.010), and a non-significant between-subjects effect of cohort (df = 2, F = 1.061, p=0.349, η^2 = 0.016). These results were unexpected given the proposed hypothesis that those who played contact sports may display reduced MSI due to greater history of concussion. This would have presented itself in significantly lower proportions of correct responses across the experimental SOA conditions compared to the non-contact and control cohorts given the heightened RHI and body contact exposure rates (499, 625). ***Figures 4.3. and 4.4.*** show that those in contact sports had a higher group-averaged proportion of correct responses across all fission illusion conditions. This may be in part explained by the types of training that are undertaken in comparison to non-contact sports, although not directly analysed in this pilot study.

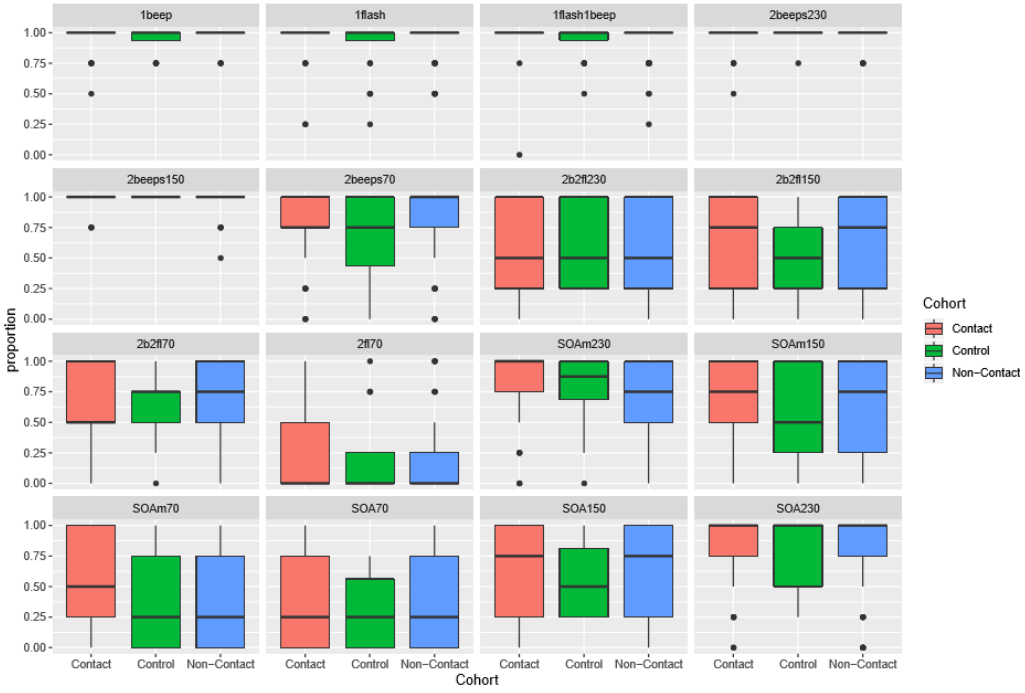

Figure 4.3. *Boxplots showing the Proportion of Correct Responses across all conditions broken down by Cohort.*

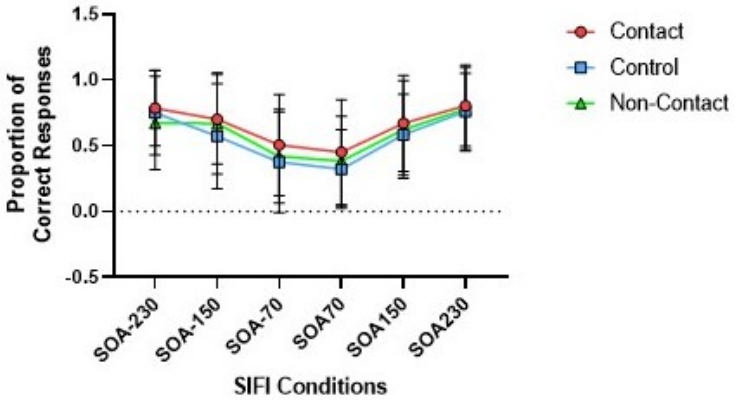

Figure 4.4. *Proportion of correct responses across SOA conditions among sporting cohorts.* No significant differences were found when the SOA conditions were analysed separately, with the control cohort performing worse across most SOA conditions, although not significant ($p>0.05$).

Given our hypothesis that concussion history may play a part in multisensory perceptual capacity, the group factor of cohort was analysed further against factors of concussion. A test of within subject effects revealed no significant main interaction effects between control

conditions*Cohort*Concussion History (p = 0.640), and Concussion Timing (p =0.275). No significant between subject interaction effects were found between control conditions*cohort*concussion history (p = 0.901), and Concussion Timing (p =0.671). Significant simple main effects were found for three of the control conditions (unimodal: [1F0B, 2F0B, 0F1B, and 0F2B] and bimodal congruent conditions [1F1B and 2F2B] with SOA's of 70, 150, or 230 ms). For the multisensory congruent 2F2B condition with an SOA of 70ms, those in the non-contact cohort with no history of concussion performed significantly better (mean = 0.67, std = 0.3007, p = 0.0088) than those with a recent history of concussion (mean = 0.25, std = 0.25).

No significant main interaction effects were uncovered between SOA*Cohort*Concussion history (p = 0.116), and Concussion Timing (p =0.284). Although, significant simple effects were revealed in Concussion Timing for SOA +230 in the Non-Contact cohort between those with no history and recent history of concussion (mean difference = 0.55, std error of difference = 0.2084, p = 0.0259) and recent vs past history of concussion (mean difference = 0.5625, std error of difference = 0.2263, p = 0.0401).

4.4.3c Sex

We first compared the performance between males and females to determine if there were any inherent sex differences for both the control and multisensory (congruent) conditions. A mixed model ANOVA of performance during the control conditions revealed significant main effects of control conditions (df = 4.583, F = 162.311, p=0.000, GGeps = 0.509, $\eta^2 = 0.559$) and a non-significant within-subject interaction effect of sex*control conditions (df = 1, , F = 0.057, p=0.475, $\eta^2 = 0.004$). A test of between-subjects effects revealed a non-significant effect of sex against the control conditions (df = 1, F = 0.057, p=0.475, $\eta^2 = 0.004$), although females (mean = 0.765, SE = 0.014) had a higher performance accuracy than their male counterparts (mean = 0.751, SE = 0.012). ***Figure 4.5.*** shows the group-averaged proportion of correct illusory responses for males and females as a function of SOA. Males can be seen to have an overall higher level of performance accuracy across all SOA conditions in comparison to females (mean difference = 0.125, std error = 0.047), whereby a mixed model ANOVA revealed a significant between-subjects effect of sex across SOA conditions (df = 1, F = 7.037, p=0.009, GGeps = 0.456, $\eta^2 = 0.052$). Females are more susceptible to the fission illusion across a wide range of SOA's, and most susceptible at the shorter SOA's, *i.e.*, SOA±70ms, however, they are as susceptible to the illusion when the secondary auditory beep follows the first at a larger SOA, *i.e.*, SOA+230. When

analysing the SOA conditions in isolation, a two-way ANOVA of the SOA conditions revealed a significant simple effect of sex for the SOA-230 condition (predicted mean difference = 0.188, p = 0.0131), with males performing significantly better (mean = 0.8041, std = 0.2845) than females (mean = 0.6161, std = 0.3598); *see* ***Figure 4.6.*** *below*.

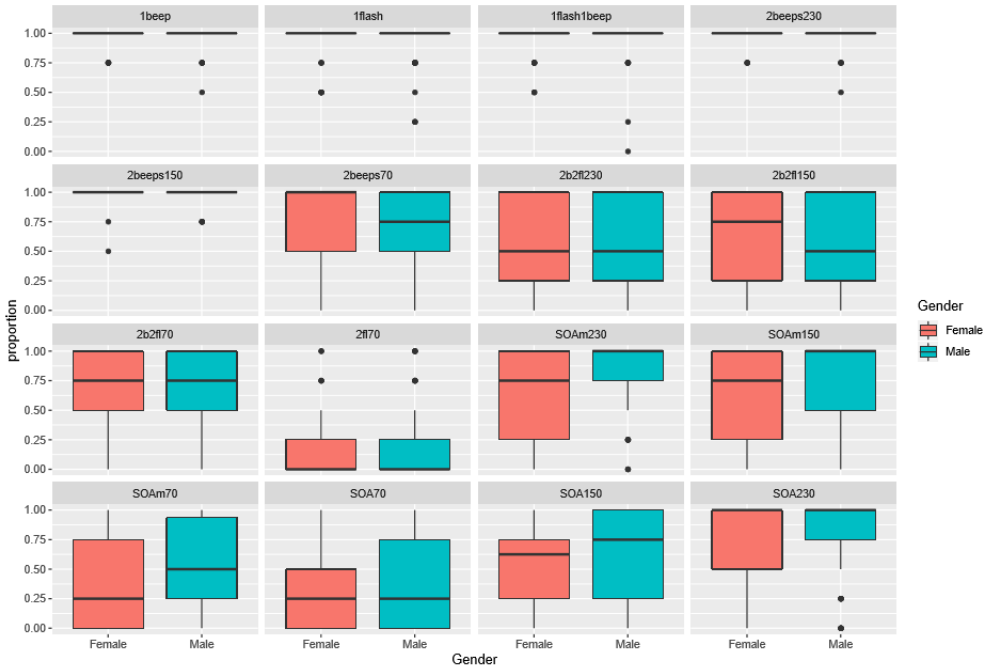

Figure 4.5. *Boxplots showing the Proportion of Correct Responses across all conditions broken down by Sex.*

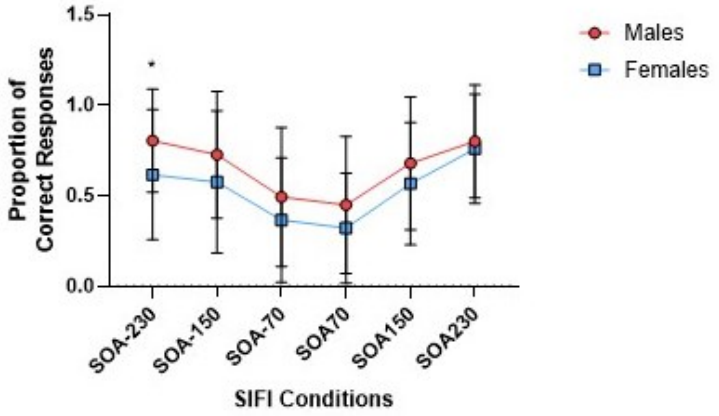

Figure 4.6. *Proportion of correct responses across SOA conditions grouped by sex.* A significant difference remained between males and females for the SOA-230 condition ($p<0.05$). Overall, males had a higher accuracy during the SIFI testing.

With a significant between-subjects effect of sex, a further analysis was conducted of sex compared to concussion factors of analysis. No significant within-subject interaction or between-subject effects of sex and concussion history or timing were uncovered for the control conditions. A mixed model ANOVA revealed no significant within-subject interaction (df = 3.935, F = 1.52, p=0.196, GGeps = 0.787, $\eta^2 = 0.012$) or between-subject effects of sex*concussion history (df = 1, F = 2.645, p=0.106, $\eta^2 = 0.021$). Across the range of SOA's producing a significant illusion effect, males with no concussion were less susceptible to the fission illusion than females at larger SOA's (SOA-230: mean difference = 0.2667, p = 0.0024, std error of difference = 0.0749; SOA-150: mean difference = 0.2542, p = 0.0044, std error of difference = 0.0749), highlighting a larger temporal binding window of young adult males when the second auditory beep precedes the A/V presentation.

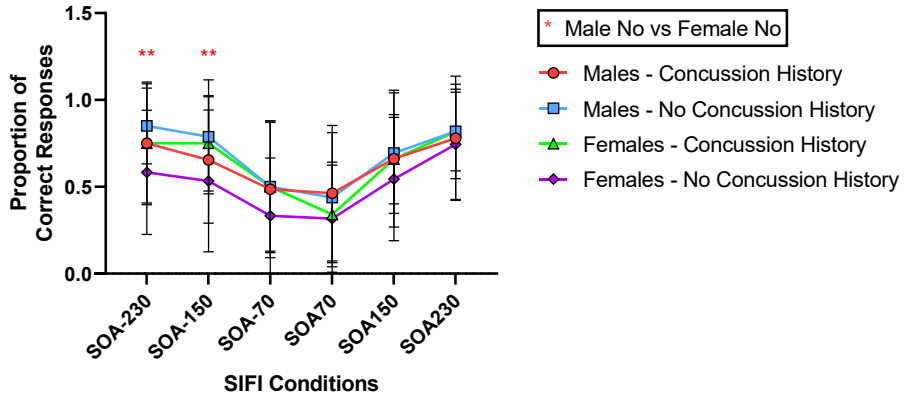

Figure 4.7. *Proportion of correct responses for Males and Females under the sub-groupings of Concussion History*. Significant differences were found between Males and Females both with no history of concussion for the SOA-230, and -150 conditions ($p<0.01$). Error bars represent standard deviation.

4.4.3d Latent Class Analysis (LCA)

Based on the comparative analysis above it was deemed more appropriate to model the data through a latent class analysis given the format and output of the SIFI data. **Table 4.3.** shows the Bayesian information criterion (BIC) for all models. The minimum BIC was obtained for the six-group model with no covariates (the model of **Figure 4.10.** below). Group 4 was found to be poor overall with the groups being fit using an expectation maximisation algorithm which constituted a likelihood function and a classification label of each participant. This would lead to questioning whether any of these covariates have a strong predictive quality with performance accuracy and susceptibility to the fission illusion of the SIFI despite the observed differences.

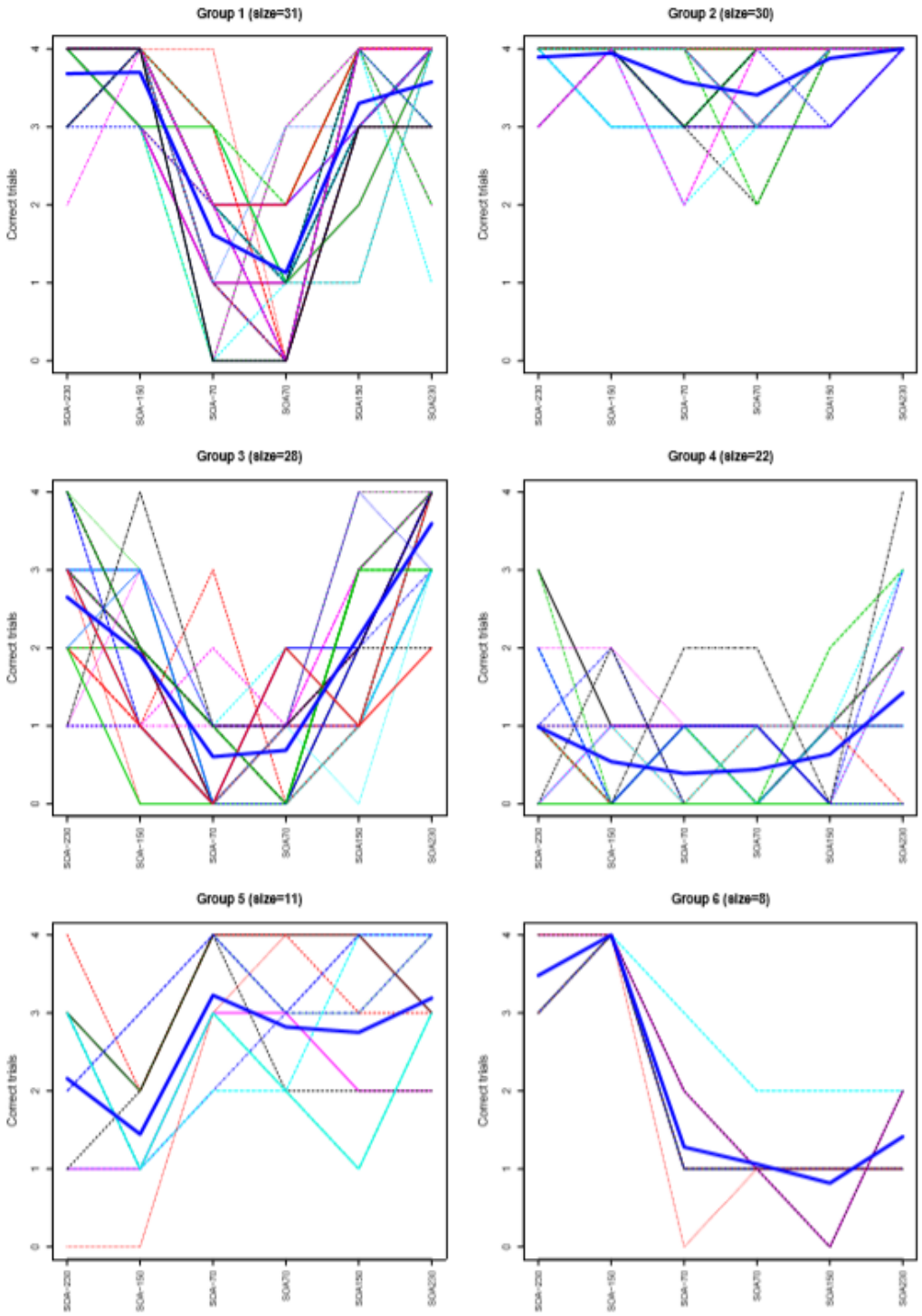

Figure 4.8. *Six Different Groupings from the LCA Model.* The thick blue line shows the expected number of correct trials from the estimated probability. The other lines indicate the individual participant's response.

Table 4.3. *Values of Bayesian Information Criterion (BIC) for all the entertained latent class analysis models*. Codes: I, intercept, G, gender, CT, cohort, CH, concussion history.

	I	I,G	I,CT	I,CH	I,G,CT	I,G,CH	I,CT,CH	I,G,CT ,CH
G=1	2838.90	2807.46	2873.50	2861.02	2856.39	2835.09	2900.50	2884.45
G=2	2263.10	2262.09	2324.57	2308.81	2307.87	2281.31	2377.82	2342.66
G=3	2131.99	2191.24	2243.81	2180.42	2250.21	2230.25	2301.81	2341.25
G=4	2094.64	2198.57	2259.08	2181.93	2308.40	2259.07	2351.24	2383.59
G=5	2078.53	2182.39	2285.47	2200.91	2354.35	2300.88	2396.86	2475.41
G=6	**2078.44**	2220.22	2344.95	2221.11	2418.78	2342.81	2466.58	2551.20
G=7	2097.01	2260.15	2392.52	2260.95	2506.35	2384.54	2553.32	2657.21
G=8	2112.81	2294.93	2460.24	2296.99	2615.53	2450.43	2624.42	2792.29

4.4.3e Logistic Regression Analysis

To confirm the conclusions of the LCA analysis above, a logistic regression model was fitted to the responses taking the group label from **Figure 4.10**. as a covariate in the model. **Table 4.3**. shows the results. Significant effects are attached only to the group label, the experimental condition label, and their interaction. This model had a deviance of 794.55 on 742 degrees of freedom (p =0.08) indicating a satisfactory fit to the data. It can be seen from **Table 4.4.** below that there are no significant effects attached to sex (p = 0.789) or concussion history (p = 0.2761). The odds-ratio table displays the fold change in the odds of having a correct response for each of the effects relative to SOA −230.

Table 4.4. *Effect estimates for logistic regression model.* Interactions are denoted using ×. (Effects are relative to SOA-230).

Variable	Estimate	Std. Error	z test	p-value
Intercept	2.5248	0.3507	7.20	**0.0000**
label 2	1.1147	0.6801	1.64	0.1012
label 3	-1.9146	0.3991	-4.80	**0.0000**
label 4	-3.8475	0.4327	-8.89	**0.0000**
label 5	-2.3929	0.4607	-5.19	**0.0000**
label 6	-0.5869	0.6375	-0.92	0.3572
Male	-0.0284	0.1062	-0.27	0.7890
Concussion History Yes	0.1211	0.1112	1.09	0.2761
SOA-150	0.1265	0.5035	0.25	0.8017
SOA-70	-2.9752	0.3919	-7.59	**0.0000**
SOA+70	-3.5643	0.4015	-8.88	**0.0000**
SOA+150	-1.0141	0.4185	-2.42	**0.0154**
SOA+230	-0.4032	0.4536	-0.89	0.3741
label 2 × SOA-150	0.2875	1.0507	0.27	0.7843
label 3 × SOA-150	-0.9329	0.5735	-1.63	0.1038
label 4 × SOA-150	-0.7829	0.6516	-1.20	0.2295
label 5 × SOA-150	-0.8690	0.6660	-1.30	0.1919
label 6 × SOA-150	16.2926	1042.6219	0.02	0.9875
label 2 × SOA-70	1.4192	0.7628	1.86	0.0628
label 3 × SOA-70	0.5552	0.5157	1.08	0.2816
label 4 × SOA-70	2.0923	0.5870	3.56	**0.0004**
label 5 × SOA-70	4.2979	0.6311	6.81	**0.0000**
label 6 × SOA-70	0.3813	0.7603	0.50	0.6159
label 2 × SOA+70	1.7019	0.7561	2.25	**0.0244**
label 3 × SOA+70	1.3480	0.5137	2.62	**0.0087**
label 4 × SOA+70	2.6815	0.5934	4.52	**0.0000**
label 5 × SOA+70	4.3634	0.6063	7.20	**0.0000**
label 6 × SOA+70	0.6787	0.7756	0.87	0.3816
label 2 × SOA+150	0.7178	0.8807	0.82	0.4150
label 3 × SOA+150	0.4942	0.5003	0.99	0.3233
label 4 × SOA+150	0.5511	0.5767	0.96	0.3393
label 5 × SOA+150	1.7014	0.6133	2.77	**0.0055**
label 6 × SOA+150	-2.2064	0.8024	-2.75	**0.0060**
label 2 × SOA+230	15.1043	538.3501	0.03	0.9776
label 3 × SOA+230	1.8972	0.5818	3.26	**0.0011**
label 4 × SOA+230	1.0340	0.5688	1.82	0.0691
label 5 × SOA+230	1.5799	0.6613	2.39	**0.0169**
label 6 × SOA+230	-2.1906	0.7938	-2.76	**0.0058**

Based on the results from this pilot study, neither sex nor concussion history could account for variance in multisensory performance as originally hypothesised. However, those in contact sports had a higher group-averaged proportion of correct responses across the illusory trials which may be in part explained by the types of training experienced, despite the higher rates of exposure to possible concussive injuries. It therefore begged the question of what additional factors of analysis could better explain the results. A follow up study was thus conducted asking the question as to whether lifetime of sporting exposure, time since ceasing sporting engagement, and sporting skill type (Open vs. Closed skill sports) could provide greater insight into MSI capabilities. To better understand the

population of university students, the association of sporting metrics and concussion history with mental and physical health was also assessed. This was undertaken alongside concussion knowledge and attitudes, and the results of this follow-up study are detailed below; see **Chapter Two** for full details on the methodology surrounding the additional mental and physical health surveys, and concussion knowledge and attitudes.

Follow -Up Study

4.4.5. Participant Characteristics and Demographics

192 participants (88 Males, age: 21.29 ± 1.47 years; 104 Females, age: 20.94 ± 1.59 years) were recruited from the student population of Trinity College Dublin and collegiate sporting teams. A power analysis for participant recruitment was not performed for the present prospective study as it is exploratory in nature. Participants who enrolled in the study attended the designated test centre on three separate occasions. All participants were informed their participation in the study was voluntary and were supplied with a participant information leaflet. This prospective, follow-up study was carried out on two sperate occasions due to the COVID-19 pandemic; *see* ***Chapter Two – Section 2.2.1 (Figure 2.1.)*** *for full details and corresponding timeline of data collection*. Written and informed consent was provided by all participants. Participant characteristics and demographics are detailed in ***Table 4.5.*** below.

Sex had a statistically significant between-subject effect on height (mean difference = 0.12 cm, df = 1, F = 69.87, SE = 0.01, $p<0.0001$) with a large effect size ($\eta^2 = 0.27$) and on weight (mean difference = 12.08 kg, df = 1, F = 33.88, SE = 2.08, $p<0.0001$) with a large effect size ($\eta^2 = 0.15$), whereby males were taller and heavier than females. There was no significant between-subject effect by sex on age (p=0.32) or BMI (p=0.15).

Cohort had a statistically significant between-subject effect on age (df = 2, F = 5.19, SE = 0.01, $p<0.05$) with a large effect size ($\eta^2 = 0.53$). Those in the control group were older than those in the non-contact group (mean difference = 0.86, SE = 0.27, p=0.005). No other significant comparisons for age were reported. Cohort did not have any significant between-subjects effect on height (p=0.29), weight (p=0.86), or BMI (p=0.85). No significant interaction effects of sex*cohort were reported by a multivariate general linear model.

Of the 192 participants, 98 were classified as *Closed skill*, 60 as *Open skill*, and the remaining 34 as *Control*. In accordance with the IPAQ scoring system, n = 117 were classified as having high levels of PA, n= 70 as moderate levels of PA, and n = 5 were classified as having low levels of PA. Sex was not found to have a significant between-subjects effect across all individual METs and total METs. However, cohort had a significant effect on vigorous PA (df = 2, F = 12.74, p=0.000, $\eta^2 = 0.12$) and total METs (df = 2, F = 4.33, p<0.05, $\eta^2 = 0.45$). The contact (mean difference = 1348.60, SE = 504.95, p=0.25) and non-contact (1713.73, SE = 341.56, p=0.000) groups reported significantly higher level of vigorous METs compared to the control group respectively. The control group reported significantly less total METs than the non-contact group (mean difference = 1466.24, SE = 500.66, p=0.11). When post-hoc comparisons were run with the Bonferroni correction method, further significant differences arose for both vigorous and total METs by cohort. The contact (mean difference = 1568.28, SE = 397.88, p=0.000) and non-contact (1619.14, SE = 326.12, p=0.000) groups remained significantly different to the control group with higher levels of vigorous METs. The control group reported significantly less total METs than the non-contact group (mean difference = 1538.66, SE = 478.03, p=0.005). No significant interaction effects of sex*cohort were reported by a multivariate general linear model for activity levels.

Lifetime sporting exposure and time since ceasing sporting engagement were two new critical factors of analysis that were explored from a sub-sample of 102 participants. Females were found to have spent longer playing sport than their male counterparts (mean difference = 2.484, SE = 1.207, 95% CI = 0.088 – 4.88, df = 1, F = 4.234, p<0.05, $\eta^2 = 0.042$), see **Figure 4.9**. Time since sporting retirement was not significantly different between females and males both ceasing regular sporting activities within ∼6 months of each other (mean difference = 0.449, SE = 0.68, 95% CI = -1.798 – 0.90, df = 1, F = 0.436, p>0.05, $\eta^2 = 0.005$). Sporting cohort was found to have a significant effect on lifetime sporting exposure (df = 2, F = 30.066, p<0.001, $\eta^2 = 0.385$).

The contact cohort had a longer mean duration of sports engagement (11.04 years, SD = 5.856) compared to both the non-contact cohort (mean difference = 8.554 years, SE = 1.316) and the control cohort (mean difference = 12.643 years, SE = 1.719), with both differences being statistically significant (p<0.001). Additionally, the non-contact cohort also spent more time in sports than the control cohort (mean difference = 4.089 years, SE = 1.368, p<0.05). There were no statistically significant differences among the sporting groups in terms of the time elapsed since they stopped participating in sports (df = 2, F = 0.537, p>0.05, $\eta^2 = 0.011$), see **Figure 4.9**. Similarly, no significant

interaction effect was observed between sex and sporting cohort for the assessed sporting metrics (both p>0.05). Individuals with a history of concussion had a longer mean duration of sports engagement (6.16 years, SD = 5.248) than those without a concussion history (mean (SD) = 4.75 years (6.00)), although this difference was not statistically significant (df = 1, F = 1.423, p>0.05, η^2 = 0.014). Furthermore, those with a history of concussion ceased their sporting participation earlier than those without, but again, this difference was not statistically significant (mean difference = 1.432 years, SE = 1.20, df = 1, F = 2.241, p>0.05, η^2 = 0.0122).

4.4.6. Mental Health & Well-Being

On the **PHQ-9** measure of depression, more than half (53.03%) of the total population reported some level of depression (*i.e., a PHQ-9 score of 5 or above*). There was no significant difference between females and males reporting similar scores (mean difference = 0.043, SE = 1.093, df = 1, F = 0.002, p>0.05, η^2 = 0.00), see **Figure 4.10**. The control cohort reported the highest score of depression (mean = 7.12, 95% CI = 4.95 – 9.44) compared to the contact (mean = 6.54, 95% CI = 4.38 – 8.70) and non-contact cohorts (mean = 5.06, 95% CI = 4.15 – 5.98), although these differences between sporting cohorts were not significant (df = 2, F = 2.029, p>0.05, η^2 = 0.031), see **Figure 4.11**. Similarly, there was no significant interaction effect between sex*sporting cohort (df = 2, F = 0.663, p>0.05, η^2 = 0.01). Those with and without a concussion history did not significantly differ in terms of depression on the PHQ-9 with similar scores reported (mean difference = 0.864, SE = 0.766, p=0.261), see **Figure 4.12**.

The **SF-12** was used to investigate mental and physical functioning and overall health-related-quality of life. Males and females reported similar scores of both physical and mental health and sex was found not to be a significant factor (PCS-12: df = 1, F = 0.70, p>0.05, η^2 = 0.006; MCS-12: df = 1, F = 0.77, p>0.05, η^2 = 0.006), see **Figure 4.10**. Across the sporting cohorts, similar scores of physical well-being were reported and no significant difference was found (df = 2, F = 0.51, p>0.05, η^2 = 0.008). However, sporting cohort was found to be significantly associated with poorer mental health (df = 2, F = 5.24, p=0.007, η^2 = 0.077). Lower scores represent poorer mental health. The control cohort reported significantly worse mental health scores than both the contact (mean difference = -5.90, SE = 1.93, p=0.008) and non-contact cohorts mean difference = -5.54, SE = 1.66, p=0.003) respectively, see **Figure 4.11**. Those with a concussion history reported significantly higher levels of physical discomfort than those without (mean difference = 1.388, SE = 0.70, p=0.049) but did not differ in

terms of mental health where those without concussion showed poorer results (mean difference = 1.256, SE = 1.316, p=0.342), see **Figure 4.12**.

The **BSI-18** as a self-report measure of depression and anxiety revealed no difference between females and males across all three subscales (**SOM**: df = 1, F = 0.01, p = 0.919, η^2 = 0.00; **DEP**: df = 1, F = 0.739, p = 0.392, η^2 = 0.006; **ANX**: df = 1, F = 0.699, p = 0.405, η^2 = 0.006) and overall GSI score (mean difference = 1.439, SE = 2.22, df = 1, F = 0.419, p = 0.518, η^2 = 0.003), see **Figure 4.10**. Levels of somatisation, anxiety, and overall depression/anxiety did not differ across the sporting cohorts (**SOM**: df = 2, F = 0.75, p = 0.475, η^2 = 0.012; **DEP**: df = 2, F = 0.403, p = 0.669, η^2 = 0.006; **ANX**: df = 2, F = 2.996, p = 0.054, η^2 = 0.045; **GSI**: df = 2, F = 1.423, p = 0.245, η^2 = 0.022), see **Figure 4.11**. No significant interaction effect between sex*sporting cohort was found (SOM: df = 2, F = 0.232, p = 0.794, η^2 = 0.004; DEP: df = 2, F = 0.185, p = 0.831, η^2 = 0.003; ANX: df = 2, F = 0.628, p = 0.535, η^2 = 0.01; GSI: df = 2, F = 0.098, p = 0.906, η^2 = 0.002). Those with concussion reported higher levels of depression and anxiety (**DEP**: mean difference = 1.008, SE = 0.507, p = 0.049; **ANX**: mean difference = 1.171, SE = 0.588, p = 0.049; **GSI**: mean difference = 3.605, SE = 1.527, p = 0.02), but did not differ in somatisation scores (**SOM**: mean difference = 1.426, SE = 0.729, p = 0.053), see **Figure 4.12**.

On the **GAD-7** as a measure of anxiety, males and females did not significantly differ from one another (mean difference = 1.06, SE = 0.84; df = 1, F = 0.805, p = 0.371, η^2 = 0.006), see **Figure 4.10**. No difference between the control group and both the contact and non-contact groups regarding levels of anxiety were seen (df = 2, F = 1.854, p = 0.161, η^2 = 0.029), see **Figure 4.11**. No significant interaction effect between sex*sporting cohort was found (df = 2, F = 0.135, p = 0.874, η^2 = 0.002). Those with a concussion history also did not differ regarding levels of anxiety on the GAD-7 to those without a history of concussion (mean difference = 1.213, SE = 0.832, p=0.147), see **Figure 4.12**.

4.4.7. Association between Sporting Metrics and Mental Health & Well-Being

Associations between common sporting metrics and mental health indicators were explored. Bivariate correlation analysis revealed significant correlations between various factors that are unique to an athletic population. Lifetime sporting exposure was significantly correlated with a higher number of previous self-reported concussions, and time since ceasing regular sporting activity was associated with worse physical health. Factors not unique to an athletic population such as the number of previous concussions was significantly correlated with worse outcomes across metrics of

somatization, anxiety, and depression (see **Table 4.10**.). Multiple linear regression modelling revealed sporting cohort, time since ceasing sporting activity, and previous number of concussions were significantly associated as significant predictors of depression across the entire cohort (see **Table 4.11.** and **Figure 4.14.**).

4.4.8. Concussion History

A total of 135 participants had no prior history of concussion, whereas 57 individuals reported having experienced a concussion in the past using the MTIIM; see **Appendix 1.7**. Some participants had sustained more than one previous concussion; hence a total of 135 self-reported concussions were recorded from a total of 57 participants. The number of concussions sustained by individual participants ranged from 1 to 13. The oldest and most recent recorded concussion was 15 years and 1-month from the day of testing, respectively. See **Table 4.6.** below for further details.

Sex did not have a significant between-subjects effect (df = 1, F = 1.48, p>0.05) with a small effect size (η^2 = 0.008). Cohort had a significant between-subjects effect (df = 2, F = 4.65, p<0.05) with a moderate effect size (η^2 = 0.05). The Contact cohort had significantly more concussions than the non-contact cohort (mean difference = 1.17, SE = 0.25, p=0.000), and the control cohort (mean difference = 1.34, SE = 0.27 p = 0.000). The interaction of Sex*Cohort revealed a non-significant main effect (df = 2, F = 0.86, p = 0.426, η^2 = 0.009).

4.4.9. Concussion Knowledge and Attitudes

Males were found to have a marginally better score of concussion knowledge and attitude towards concussion compared to females. All sporting groups performed similarly across both concussion knowledge and attitude to concussion (see **Table 4.7.**). Sex and study cohort were in fact found to have no significant between subject effect on both concussion knowledge (df = 1, F = 3.772, p>0.05, η^2 = 0.029; df = 2, F = 2.308, p>0.05, η^2 = 0.035) and concussion attitude (df = 1, F = 0.409, p>0.05, η^2 = 0.003; df = 2, F = 0.201, p>0.05, η^2 = 0.003) respectively, see **Appendix 1.9**. Those with a previous concussion history reported slightly higher knowledge and attitude scores than those without (mean difference = 0.190 & 0.571, respectively), although neither was statistically significant (p>0.05). Multiple linear regression analysis however showed that sex was in fact a significant predictor variable (p<0.05) of concussion knowledge (see **Table 4.16.**). The same cannot be said for

concussion attitude scores as there were no significant predictors based on the models (see **Table 4.17.**).

Table 4.5. *Participant Demographics of Follow-Up Study.*

	Contact			**Non-Contact**			**Control**			**Total**		
	Female (n=6)	*Male (n=35)*	*Total (n=41)*	*Female (n=63)*	*Male (n=34)*	*Total (n=97)*	*Female (n=35)*	*Male (n=19)*	*Total (n=54)*	*Female (n=104)*	*Male (n=88)*	*Total (n=192)*
Age (yrs; mean ± SD)	21.33 ± 1.21	20.91 ± 1.38	20.97 ± 1.35	21.14 ± 1.62	20.5 ± 1.81	20.92 ± 1.71	21.57 ± 1.17	21.79 ± 1.23	21.65 ± 1.18	21.29 ± 1.47	20.94 ± 1.59	21.14 ± 1.53
Height (m; mean ± SD)	1.69 ± 0.07	1.81 ± 0.07	1.79 ± 0.08	1.69 ± 0.07	1.80 ± 0.09	1.73 ± 0.09	1.67 ± 0.07	1.79 ± 0.08	1.71 ± 0.09	1.69 ± 0.07	1.80 ± 0.08	1.74 ± 0.09
Weight (Kg; mean ± SD)	65.82 ± 8.49	79.22 ± 14.79	77.26 ± 14.77	66.3 ± 9.23	77.05 ± 10.57	70.07 ± 10.95	64.94 ± 9.67	77.02 ± 11.67	69.19 ± 11.84	65.81 ± 9.23	77.91 ± 12.54	71.36 ± 12.43
BMI ($Kg.m^2$; mean ± SD)	22.91 ± 2.19	24.19 ± 3.99	24.01 ± 3.79	23.14 ± 2.63	23.59 ± 2.55	23.31 ± 2.59	23.27 ± 3.41	24.09 ± 3.12	23.56 ± 3.31	23.17 ± 2.87	23.94 ± 3.29	23.53 ± 3.09
***PA Levels (METs; mean* ± SD)**												
Vigorous	1888.00 ± 1361.18	2406.86 ± 1744.11	2329.76 ± 1689.02	2080.64 ± 2107.92	2936.47 ± 2670.89	2380.62 ± 2343.15	682.29 ± 997.55	907.37 ± 1264.77	761.48 ± 1092.44	1598.45 ± 1877.52	2287.73 ± 2193.62	1914.38 ± 2052.16
Moderate	1040.00 ± 979.79	746.29 ± 963.73	789.27 ± 959.43	838.92 ± 1177.16	779.41 ± 1026.52	817.94 ± 1121.59	338.29 ± 686.02	798.95 ± 783.79	500.37 ± 748.24	681.92 ± 1048.19	770.46 ± 943.51	722.50 ± 999.97
Walking	1540.00 ± 1458.53	1345.46 ± 908.79	1373.93 ± 986.30	1491.55 ± 1287.72	1483.93 ± 1127.07	1488.88 ± 1227.85	1676.21 ± 1318.61	2275.26 ± 1475.58	1886.99 ± 1392.21	1556.49 ± 1297.24	1599.71 ± 1177.28	1576.30 ± 1240.68
Total	4460.00 ± 2214.35	4498.66 ± 2461.81	4493.00 ± 2400.95	4410.98 ± 3083.16	5199.85 ± 3638.03	4687.49 ± 3291.19	2696.77 ± 1868.68	3981.58 ± 2344.71	3148.83 ± 2119.15	3836.91 ± 2789.02	4657.93 ± 2958.22	4213.21 ± 2889.47
n=	4	21	25	35	25	60	13	4	17	52	50	102
Lifetime Sporting Exposure (yrs)	15.00 ± 5.35	10.29 ± 5.75	11.04 ± 5.86	5.46 ± 5.19	2.72 ± 2.51	4.32 ± 4.47	-	-	-	4.83 ± 5.83	5.68 ± 5.73	5.25 ± 5.77
Time Since Ceasing Sporting Activity (yrs)	2.00 ± 0.82	4.43 ± 2.52	4.04 ± 2.49	3.51 ± 2.24	2.99 ± 2.45	3.30 ± 2.33	4.31 ± 2.84	3.75 ± 3.40	4.18 ± 2.88	3.60 ± 2.37	3.65 ± 2.59	3.63 ± 2.47

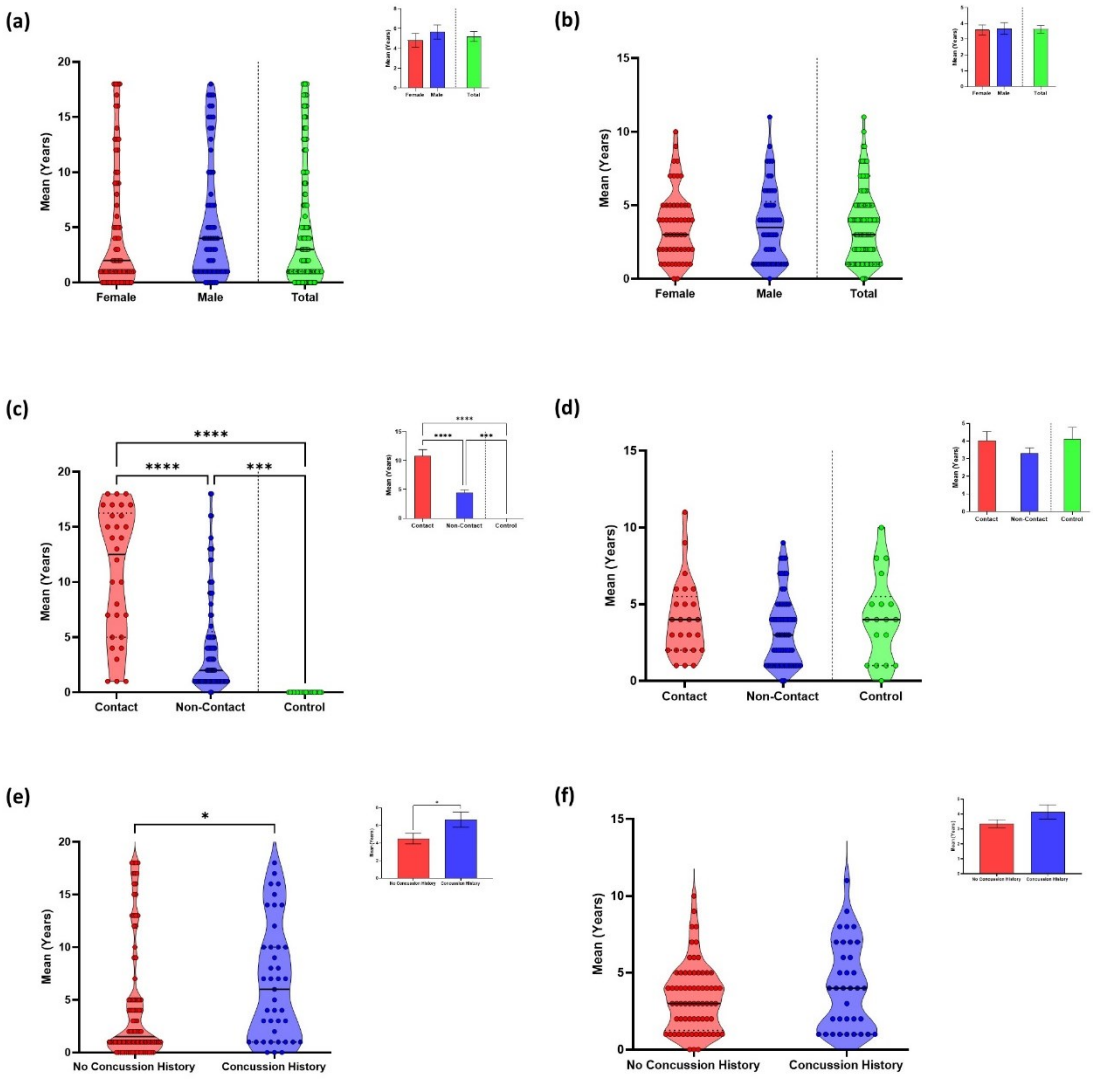

Figure 4.9. *Lifetime Sporting Exposure and Times Since Ceasing Sporting Activity*. These sporting metrics were broken down into: Sex (**a & b**), Sporting Cohort (**c & d**), and Concussion History (**e & f**), respectively. * - Signifcance at the p<0.05 level.

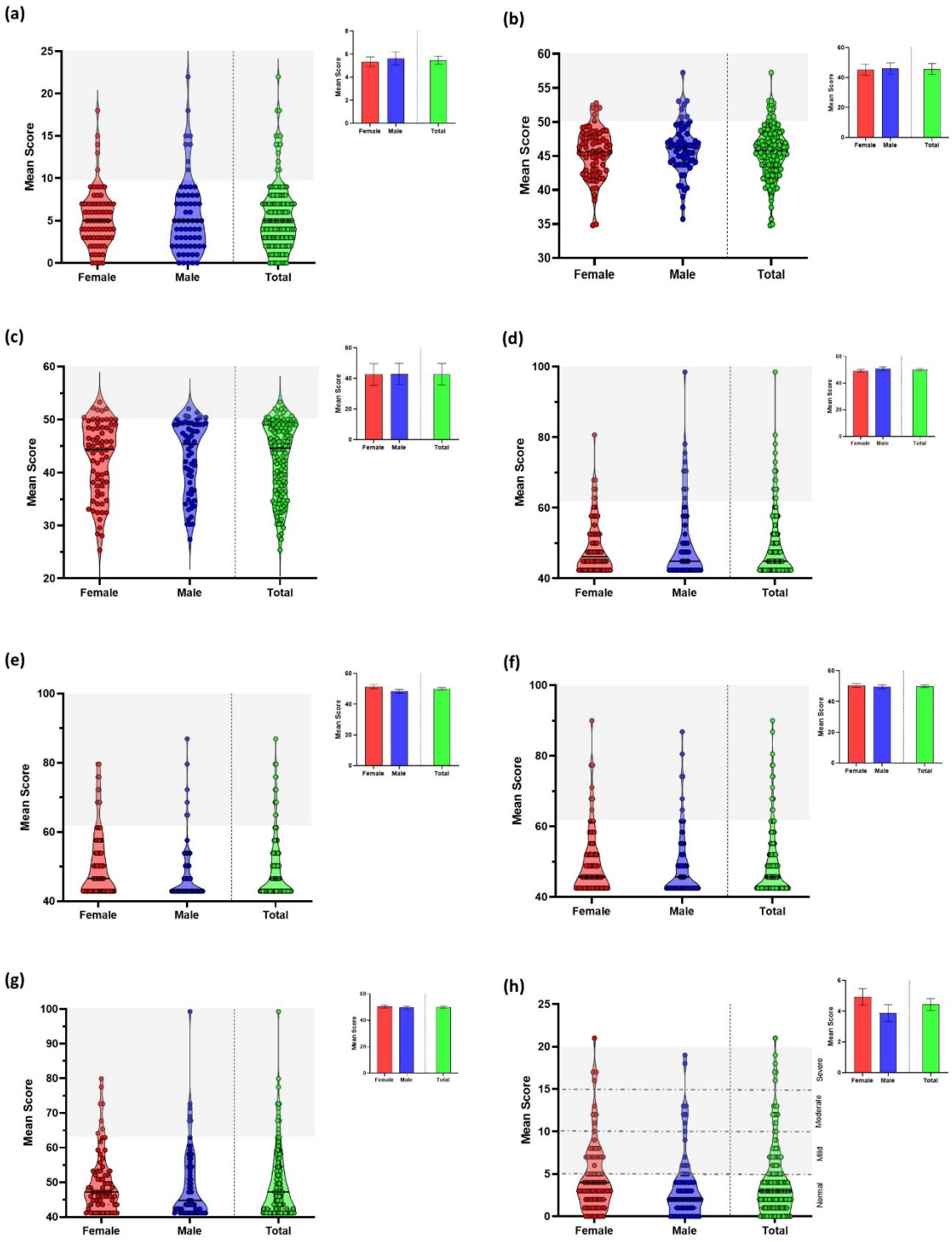

Figure 4.10. *Patient Reported Outcomes by Sex*. Abnormal scores are highlighted in grey. **(a)** PHQ-9, **(b)** PCS-12, **(c)** MCS-12, **(d)** SOM, **(e)** DEP, **(f)** ANX, **(g)** GSI, **(h)** GAD-7.

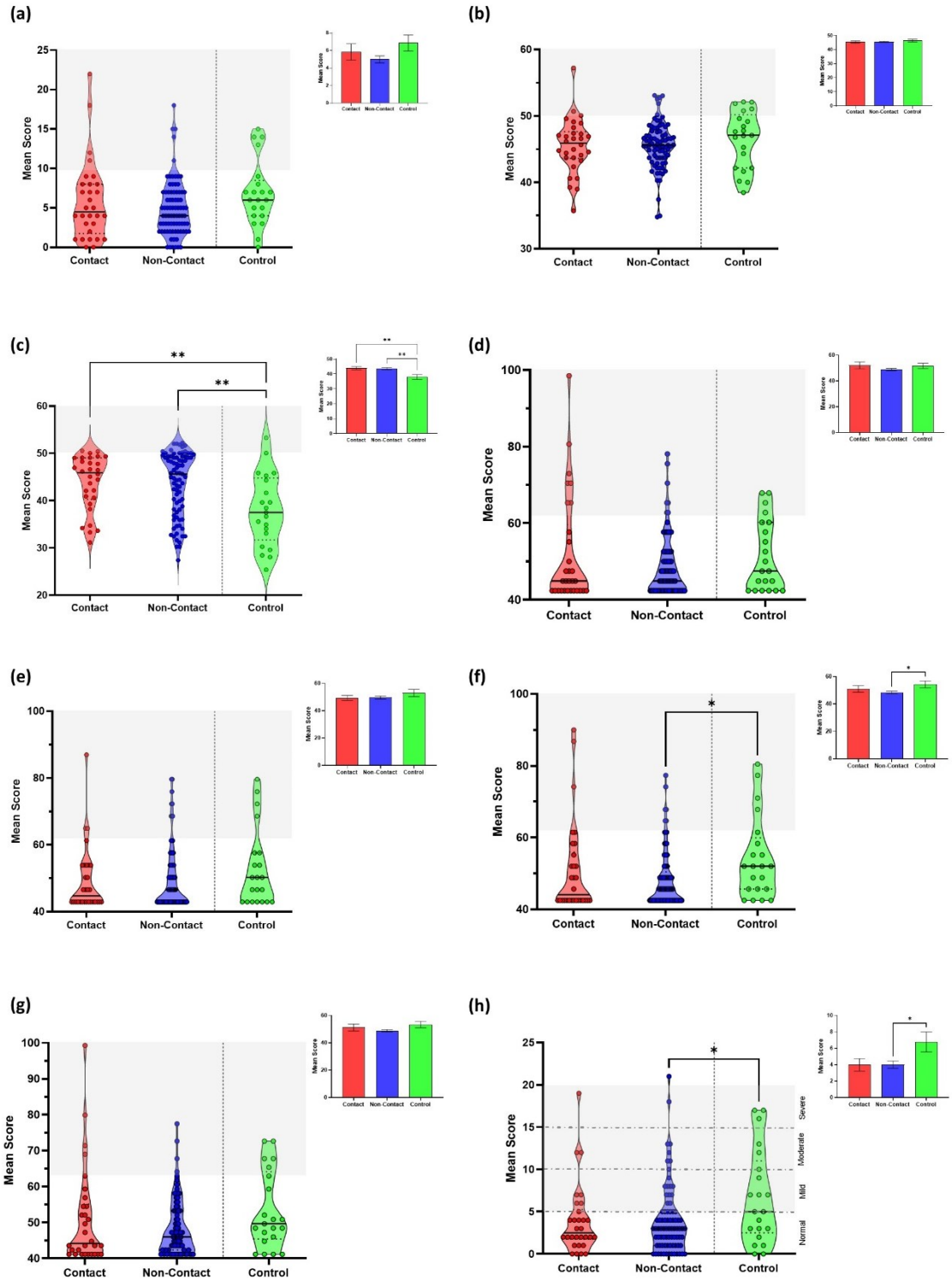

Figure 4.11. *Patient Reported Outcomes by Cohort*. Abnormal scores are highlighted in grey. **(a)** PHQ-9, **(b)** PCS-12, **(c)** MCS-12, **(d)** SOM, **(e)** DEP, **(f)** ANX, **(g)** GSI, **(h)** GAD-7. * - Significance at the p<0.05 level; ** - Significance at the p<0.01 level.

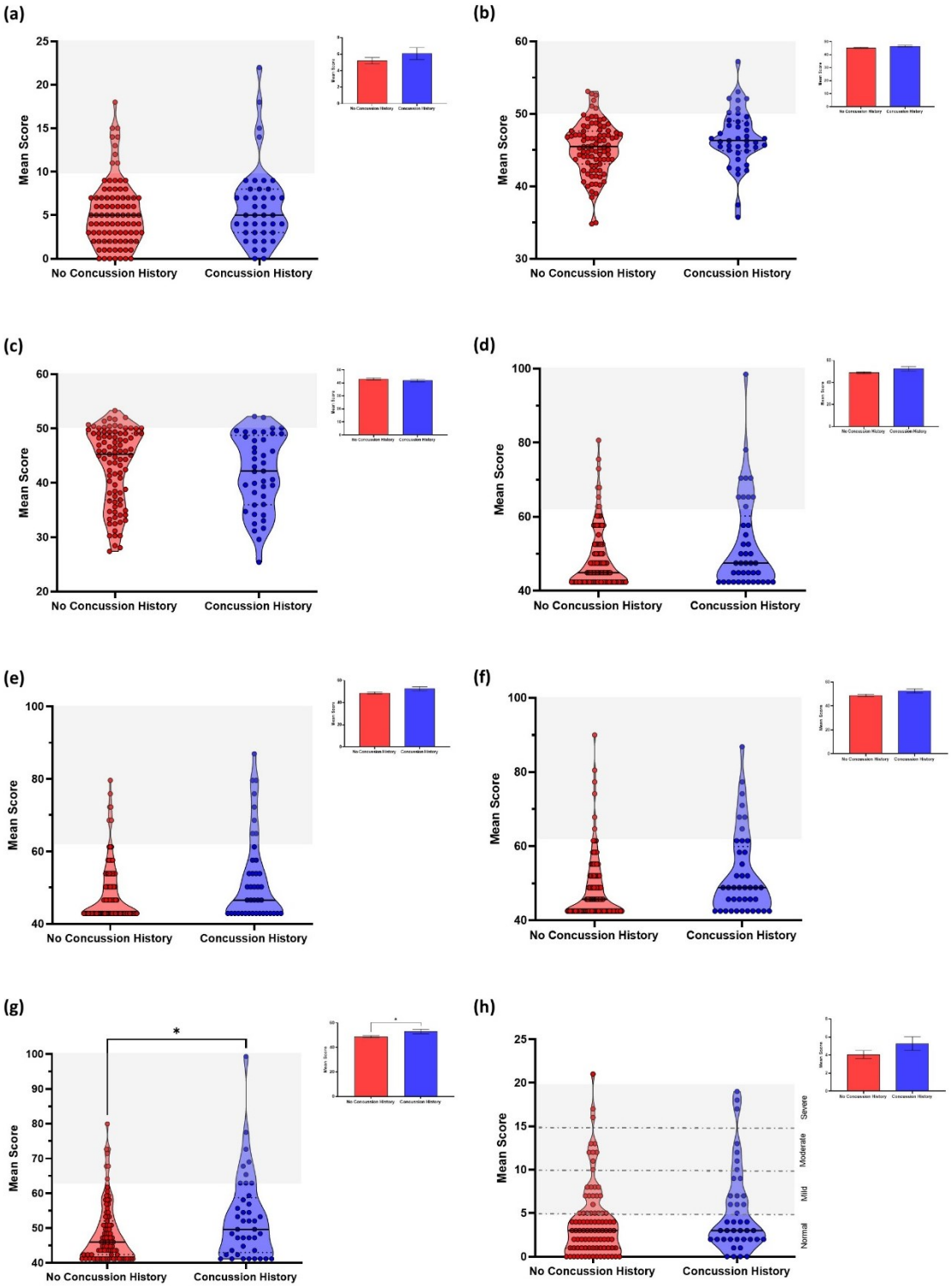

Figure 4.12. *Patient Reported Outcomes by Concussion History*. Abnormal scores are highlighted in grey. **(a)** PHQ-9, **(b)** PCS-12, **(c)** MCS-12, **(d)** SOM, **(e)** DEP, **(f)** ANX, **(g)** GSI, **(h)** GAD-7. * - Significance at the $p<0.05$ level.

Table 4.6. *Concussion History of Follow-Up Study.*

	Contact			Non-Contact			Control			Total		
	Female (n=6)	*Male (n=35)*	*Total (n=41)*	*Female (n=63)*	*Male (n=34)*	*Total (n=97)*	*Female (n=35)*	*Male (n=19)*	*Total (n=54)*	*Female (n=104)*	*Male (n=88)*	*Total (n=192)*
Concussion History												
Yes	2	21	23	12	13	25	6	2	9	21	36	57
No	4	14	18	51	21	72	29	17	45	83	52	135
No. of Previous Concussions	2.5 ± 2.12	2.81 ± 2.86	2.78 ± 2.76	1.92 ± 1.17	1.15 ± 0.38	1.52 ± 0.92	1.14 ± 0.69	2.00 ± 1.41	1.33 ± 0.87	1.71 ± 1.15	2.17 ± 2.32	2.00 ± 1.97
One Previous Concussion	1	9	10	6	11	17	4	1	5	11	21	32
Multiple Previous Concussions	1	12	13	6	2	8	2	1	3	9	15	24

Table 4.7. *Concussion Knowledge Score, Item Frequency of Correct Responses, and Concussion Attitude Score across a Subpopulation of the Follow-Up Study Cohort.*

	Contact			Non-Contact			Control			Total			Concussion History	
	Female (n=4)	Male (n=26)	Total (n=30)	Female (n=49)	Male (n=32)	Total (n=81)	Female (n=17)	Male (n=4)	Total (n=21)	Female (n=70)	Male (n=62)	Total (n=132)	No (n=91)	Yes (n=41)
Concussion Knowledge Score (mean ± SD)	12.25 ± 0.50	12.08 ± 1.62	12.10 ± 1.52	11.714 ± 1.225	12.22 ± 1.45	11.91 ± 1.33	11.94 ± 1.391	13.75 ± 0.50	12.29 ± 1.45	11.80 ± 1.235	12.26 ± 1.53	12.02 ± 1.39	11.96 ± 1.31	12.15 ± 1.57
Concussion Attitude Score (mean ± SD)	60.75 ± 10.44	65.73 ± 6.12	65.07 ± 6.82	65.02 ± 7.06	63.84 ± 7.55	64.56 ± 7.23	68.29 ± 4.58	61.00 ± 2.94	66.91 ± 5.17	65.57 ± 6.90	64.45 ± 6.81	65.05 ± 6.86	64.87 ± 6.59	65.44 ± 7.47
Recognition of Signs & Symptoms (n=)														
Headache (True)	3	26	29	47	32	79	16	4	20	66	62	128	87	41
Sensitivity to Light (True)	4	24	28	44	29	73	16	4	20	64	57	121	82	39
Difficulty Remembering (True)	3	25	28	44	31	75	16	4	20	63	60	123	84	39
In a "fog" (True)	4	25	29	42	28	70	16	4	20	62	57	119	81	38
Difficulty Concentrating (True)	3	24	27	46	31	77	16	4	20	65	59	124	87	37
Dizziness (True)	4	26	30	48	32	80	15	4	19	67	62	129	89	40
Hives (False)	4	26	30	49	32	81	17	4	21	70	62	132	91	41
Difficulty Speaking (False)	0	3	3	11	4	15	4	0	4	15	7	22	15	7
Arthritis (False)	3	26	29	47	32	79	17	4	21	67	62	129	88	41
Panic attacks (False)	3	21	24	38	24	62	13	3	16	54	48	102	72	30
Drowsiness (False)	0	5	5	9	6	15	4	1	5	13	12	25	18	7
Weight gain (False)	4	26	30	46	31	77	16	4	20	66	61	127	87	40
Feeling Slowed down (False)	0	3	3	9	3	12	4	0	4	13	6	19	12	7
Reduced breathing rate (False)	2	17	19	33	22	55	12	2	14	47	41	88	59	29
Excessive studying (False)	4	26	30	49	32	81	17	4	21	70	62	132	91	41
Hair loss (False)	4	26	30	48	32	80	17	4	21	69	62	131	90	41
General Concussion Knowledge (n=)														
There is a possible risk of death if a second concussion occurs before the first one has healed (True)	4	26	30	48	31	79	17	4	21	69	61	130	89	41
In order to be diagnosed with a concussion, you have to be knocked out (False)	4	26	30	48	32	80	17	4	21	69	62	131	90	41

A concussion can only occur if there is a direct hit to the head (False)	3	24	27	39	29	68	15	3	18	57	56	113	76	37
Symptoms of a concussion can last several weeks (True)	4	14	18	25	21	46	14	4	18	43	39	82	54	28
Concussions can sometimes lead to emotional disruptions (True)	4	25	29	47	30	77	17	4	21	68	59	127	88	39
There is rarely a risk to long-term health and well-being from multiple concussions (False)	4	24	28	41	31	72	17	4	21	62	59	121	84	37
Attitude Towards Concussion (n=)														
I would continue playing a sport while also having a headache that resulted from a minor concussion (Disagree)	2	15	17	44	22	66	17	3	21	6	16	22	73	30
I feel that concussions are less important than other injuries (Disagree)	4	25	29	46	29	75	17	3	21	67	57	124	86	38
I feel that a player has a responsibility to return to a game even if it means playing while still experiencing symptoms of a concussion (Disagree)	4	25	29	47	29	76	17	4	21	68	58	126	88	38
I feel that a player who is knocked unconscious should be taken to the emergency room (Agree)	3	23	26	40	28	68	16	4	20	60	55	115	80	35
I feel that Coach A made the right decision to keep Player R out of the game (Agree)	4	26	30	44	29	73	16	4	20	65	59	124	85	39
I feel that Player H should tell his coach about the symptoms (Agree)	4	26	30	48	31	79	16	4	20	69	61	130	90	40

***Note:** Some but not all questions from the questionnaire were chosen as representative figures for general concussion knowledge and attitudes towards concussion of the chosen sub-population; see **Appendix 1.9**. True/False or Agree/Disagree in brackets after each statement for both knowledge and attitude towards concussion is in reference to the correct response that varied for each question.*

***Note:** Figures for attitudes towards concussion (n=) represents the sum of those who strongly agree/agree and strongly disagree/disagree which was question dependent. Scenarios were provided within each section and a question posed to the participant about their attitude towards the decision of the key individual involved.*

Table 4.8. *Prevalence of Symptoms of Physical and Mental Health Disorders by Sex & Sporting Cohort of Sub-Population.*

	Contact			Non-Contact			Control			Total		
	Female (n=4)	*Male (n=26)*	*Total (n=30)*	*Female (n=49)*	*Male (n=32)*	*Total (n=81)*	*Female (n=17)*	*Male (n=4)*	*Total (n=21)*	*Female (n=70)*	*Male (n=62)*	*Total (n=132)*
PHQ-9	7.5 ± 7.42	5.58	5.83 ± 5.07	4.71 ± 2.62	5.41 ± 4.68	4.99 ± 3.57	6.65 ± 4.26	7.75 ± 4.27	6.86 ± 4.18	5.34 ± 3.51	5.63	5.48 ± 4.08
BSI-18												
GSI	10.25 ± 15.11	7.92 ± 11.01	8.23 ± 11.35	6.39 ± 6.10	6.00 ± 7.12	6.24 ± 6.48	10.35 ± 8.99	8.75 ± 9.36	10.05 ± 8.85	7.57 ± 7.61	6.98 ± 8.99	7.29 ± 8.26
SOM	4.00 ± 7.35	3.77 ± 5.47	3.80 ± 5.60	2.27 ± 2.33	2.94 ± 4.09	2.53 ± 3.14	3.77 ± 3.85	3.00 ± 2.94	3.62 ± 3.64	2.73 ± 3.18	3.29 ± 4.09	2.99 ± 3.92
DEP	1.75 ± 2.36	1.73 ± 2.77	1.73 ± 2.68	2.20 ± 2.63	1.22 ± 2.49	1.81 ± 2.61	2.88 ± 3.46	2.00 ± 1.83	2.71 ± 3.19	2.34 ± 2.82	1.48 ± 2.56	1.94 ± 2.72
ANX	4.50 ± 7.14	2.42 ± 3.54	2.70 ± 4.07	1.92 ± 2.45	1.84 ± 2.67	1.89 ± 2.52	3.71 ± 3.24	3.75 ± 5.56	3.71 ± 3.61	2.50 ± 3.11	2.21 ± 3.24	2.36 ± 3.16
SF-12												
PCS-12	44.03 ± 3.93	45.75 ± 4.22	45.52 ± 4.16	45.09 ± 3.44	46.27 ± 3.48	45.56 ± 3.48	46.52 ± 4.29	46.18 ± 4.98	46.46 ± 4.29	45.38 ± 3.69	46.04 ± 3.84	45.69 ± 3.76
MCS-12	45.27 ± 5.02	43.66 ± 6.01	43.87 ± 5.84	43.74 ± 6.33	43.17 ± 7.57	43.51 ± 6.81	38.47 ± 8.15	35.86 ± 4.70	37.97 ± 7.59	42.56 ± 7.06	42.90 ± 6.97	42.71 ± 6.99
GAD-7	5.50 ± 5.07	3.73 ± 4.09	3.97 ± 4.17	4.18 ± 3.75	3.72 ± 4.58	4.00 ± 4.08	6.94 ± 5.82	6.00 ± 4.83	6.76 ± 5.54	4.93 ± 4.49	3.87 ± 4.355	4.43 ± 4.44

Table 4.9. *Prevalence of Symptoms of Physical and Mental Health Disorders by Concussion History of Sub-Population.*

	No Concussion History	Concussion History	df, F	η^2	P-value
n=	91	41			
PHQ-9	5.21 ± 3.81	6.07 ± 4.61	1, 1.273	0.10	0.261
BSI-18					
GSI	6.18 ± 7.09	9.78 ± 10.06	1, 5.571	0.041	**0.020***
SOM	2.55 ± 3.31	3.98 ± 4.91	1, 3.826	0.029	0.053
DEP	1.63 ± 2.36	2.63 ± 3.33	1, 3.958	0.030	**0.049***
ANX	2.00 ± 2.92	3.17 ± 3.54	1, 3.966	0.030	**0.049***
SF-12					
PCS-12	45.26 ± 3.59	46.65 ± 3.99	1, 3.932	0.029	**0.049***
MCS-12	43.10 ± 6.99	41.85 ± 7.02	1, 0.911	0.007	0.342
GAD-7	4.06 ± 4.19	5.27 ± 4.92	1,		

***** *Significant at the p<0.05 level (2-tailed).*

Abbreviations: *PHQ-9: Patient Health Questionnaire; BSI-18: Brief Symptom Inventory; GSI: Global Symptom Index; SOM: Somatisation; DEP: Depression; ANX: Anxiety; SF-12: 12-item Short Form Survey; PCS-12: Physical Component Score; MCS-12: Mental Component Score; GAD-7: Generalised Anxiety Disorder Assessment.*

Table 4.10. *Bivariate Correlations (Pearson Correlation) between Mental Health Indicators and Unique and Non-Unique Factors to the Sporting and Non-Sporting Population of University Students.*

		Lifetime Sporting Exposure	Time Since Ceasing Sporting Activity	Number of Concussions	PHQ-9	PCS-12	MCS-12	SOM	DEP	ANX	GSI
Lifetime Sporting Exposure	*Pearson Correlation*										
	Sig. (2-tailed)										
	N										
Time Since Ceasing Sporting Activity	*Pearson Correlation*	0.058									
	Sig. (2-tailed)	0.563									
	N	102									
Number of Concussions	*Pearson Correlation*	**.184***	0.117								
	Sig. (2-tailed)	0.036	0.239								
	N	131	103								
PHQ-9	*Pearson Correlation*	-0.023	-0.190	**.274****							
	Sig. (2-tailed)	0.797	0.055	0.001							
	N	131	103	132							
PCS-12	*Pearson Correlation*	-0.149	**-.221***	-0.012	0.037						
	Sig. (2-tailed)	0.090	0.025	0.891	0.677						
	N	131	103	132	132						
MCS-12	*Pearson Correlation*	**.218***	0.063	-0.125	**-.612****	**-.346****					
	Sig. (2-tailed)	0.012	0.530	0.153	0.000	0.000					
	N	131	103	132	132	132					
SOM	*Pearson Correlation*	-0.008	0.001	**.380****	**.758****	0.115	**-.521****				
	Sig. (2-tailed)	0.926	0.991	0.000	0.000	0.189	0.000				
	N	131	103	132	132	132	132				
DEP	*Pearson Correlation*	0.003	0.038	**.400****	**.456****	-0.118	**-.296****	**.439****			
	Sig. (2-tailed)	0.970	0.704	0.000	0.000	0.177	0.001	0.000			
	N	131	103	132	132	132	132	132			
ANX	*Pearson Correlation*	0.066	-0.009	**.355****	**.622****	0.061	**-.415****	**.688****	**.526****		
	Sig. (2-tailed)	0.450	0.930	0.000	0.000	0.485	0.000	0.000	0.000		
	N	131	103	132	132	132	132	132	132		
GSI	*Pearson Correlation*	0.023	0.010	**.448****	**.748****	0.039	**-.503****	**.882****	**.739****	**.882****	
	Sig. (2-tailed)	0.797	0.923	0.000	0.000	0.657	0.000	0.000	0.000	0.000	
	N	131	103	132	132	132	132	132	132	132	
GAD-7	*Pearson Correlation*	0.006	-0.079	**.240****	**.686****	0.127	**-.562****	**.628****	**.496****	**.751****	**.749****
	Sig. (2-tailed)	0.944	0.430	0.006	0.000	0.148	0.000	0.000	0.000	0.000	0.000
	N	131	103	132	132	132	132	132	132	132	132

** Correlation is significant at the p<0.05 level (2-tailed).*

*** Correlation is significant at the p<0.01 level (2-tailed).*

Table 4.11. *Multiple Linear Regression Model illustrating Predictors of Depression (PHQ-9) among a Sub-Population of the Cohort.*

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for β	
		β	Std. Error	Beta			Lower Bound	Upper Bound
1	*(Constant)*	4.843	1.325		3.654	0.000	2.213	7.473
	Sex	0.618	0.843	0.073	0.733	0.465	-1.055	2.291
2	*(Constant)*	1.805	2.334		0.773	0.441	-2.827	6.437
	Sex	1.199	0.915	0.142	1.311	0.193	-0.616	3.013
	Sporting Cohort	1.131	0.718	0.170	1.576	0.118	-0.293	2.555
3	*(Constant)*	3.246	2.884		1.125	0.263	-2.477	8.969
	Sex	1.001	0.945	0.118	1.060	0.292	-0.873	2.876
	Sporting Cohort	1.229	0.728	0.185	1.689	0.094	-0.215	2.674
	Concussion History	-0.806	0.945	-0.091	-0.853	0.396	-2.681	1.069
4	*(Constant)*	2.976	3.556		0.837	0.405	-4.082	10.034
	Sex	1.032	0.979	0.122	1.055	0.294	-0.910	2.974
	Sporting Cohort	1.312	0.963	0.198	1.361	0.177	-0.601	3.224
	Concussion History	-0.807	0.950	-0.091	-0.849	0.398	-2.692	1.078
	Lifetime Sporting Exposure	0.013	0.097	0.017	0.131	0.896	-0.181	0.206
5	*(Constant)*	4.506	3.564		1.264	0.209	-2.569	11.581
	Sex	1.014	0.961	0.120	1.055	0.294	-0.893	2.921
	Sporting Cohort	1.448	0.948	0.218	1.527	0.130	-0.434	3.330
	Concussion History	-1.120	0.944	-0.126	-1.187	0.238	-2.994	0.754
	Lifetime Sporting Exposure	0.028	0.096	0.039	0.296	0.768	-0.162	0.219
	Time Since Ceasing Sporting Activity	-0.366	0.171	-0.213	-2.144	**0.035***	-0.705	-0.027
6	*(Constant)*	-2.368	3.730		-0.635	0.527	-9.774	5.038
	Sex	1.183	0.894	0.140	1.322	0.189	-0.593	2.958
	Sporting Cohort	2.419	0.914	0.364	2.646	**0.010****	0.604	4.234
	Concussion History	1.164	1.046	0.131	1.113	0.269	-0.913	3.240
	Lifetime Sporting Exposure	0.071	0.090	0.097	0.793	0.430	-0.107	0.249
	Time Since Ceasing Sporting Activity	-0.400	0.159	-0.233	-2.518	**0.013***	-0.716	-0.085
	Number of Concussions	1.170	0.292	0.476	4.011	**0.000*****	0.591	1.749

a. Dependent Variable: PHQ-9

** Predictor variable is significant at the p<0.05 level (2-tailed).*

*** Predictor variable is significant at the p<0.01 level (2-tailed).*

**** Predictor variable is significant at the p<0.001 level (2-tailed).*

Table 4.12. *Multiple Linear Regression Model illustrating Predictors of Physical Well-Being (PCS-12) among a Sub-Population of the Cohort.*

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for β	
		β	Std. Error	Beta			Lower Bound	Upper Bound
1	*(Constant)*	45.674	1.180		38.720	0.000	43.334	48.015
	Sex	0.054	0.750	0.007	0.072	0.943	-1.435	1.543
2	*(Constant)*	42.446	2.066		20.541	0.000	38.346	46.546
	Sex	0.670	0.810	0.089	0.828	0.410	-0.936	2.277
	Sporting Cohort	1.202	0.635	0.204	1.892	0.061	-0.059	2.463
3	*(Constant)*	45.548	2.506		18.177	0.000	40.576	50.521
	Sex	0.245	0.821	0.033	0.299	0.766	-1.384	1.874
	Sporting Cohort	1.414	0.633	0.240	2.235	**0.028***	0.159	2.669
	Concussion History	-1.736	0.821	-0.220	-2.114	**0.037***	-3.365	-0.107
4	*(Constant)*	47.263	3.076		15.367	0.000	41.159	53.367
	Sex	0.048	0.846	0.006	0.057	0.955	-1.632	1.727
	Sporting Cohort	0.892	0.833	0.151	1.071	0.287	-0.761	2.546
	Concussion History	-1.732	0.821	-0.219	-2.109	**0.038***	-3.362	-0.102
	Lifetime Sporting Exposure	-0.081	0.084	-0.124	-0.962	0.338	-0.248	0.086
5	*(Constant)*	48.900	3.043		16.069	0.000	42.860	54.941
	Sex	0.028	0.820	0.004	0.034	0.973	-1.600	1.657
	Sporting Cohort	1.038	0.810	0.176	1.282	0.203	-0.569	2.645
	Concussion History	-2.067	0.806	-0.262	-2.565	**0.012***	-3.667	-0.468
	Lifetime Sporting Exposure	-0.064	0.082	-0.098	-0.785	0.434	-0.227	0.098
	Time Since Ceasing Sporting Activity	-0.392	0.146	-0.257	-2.687	**0.008****	-0.681	-0.102
6	*(Constant)*	50.743	3.419		14.840	0.000	43.955	57.532
	Sex	-0.017	0.820	-0.002	-0.021	0.984	-1.644	1.610
	Sporting Cohort	0.778	0.838	0.132	0.928	0.356	-0.886	2.441
	Concussion History	-2.680	0.959	-0.339	-2.795	**0.006****	-4.583	-0.776
	Lifetime Sporting Exposure	-0.076	0.082	-0.116	-0.921	0.359	-0.239	0.088
	Time Since Ceasing Sporting Activity	-0.383	0.146	-0.251	-2.626	**0.010****	-0.672	-0.093
	Number of Concussions	-0.314	0.267	-0.144	-1.173	0.244	-0.844	0.217

a. Dependent Variable: PCS-12

***** *Predictor variable is significant at the p<0.05 level (2-tailed).*

****** *Predictor variable is significant at the p<0.01 level (2-tailed).*

Table 4.13. *Multiple Linear Regression Model illustrating Predictors of Mental Health (MCS-12) among a Sub-Population of the Cohort.*

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for β	
		β	Std. Error	Beta			Lower Bound	Upper Bound
1	*(Constant)*	40.977	2.322		17.651	0.000	36.371	45.583
	Sex	0.794	1.477	0.054	0.538	0.592	-2.136	3.725
2	*(Constant)*	51.210	3.948		12.972	0.000	43.377	59.044
	Sex	-1.160	1.547	-0.078	-0.750	0.455	-4.229	1.909
	Sporting Cohort	-3.810	1.214	-0.328	-3.138	**0.002****	-6.219	-1.401
3	*(Constant)*	47.273	4.848		9.751	0.000	37.652	56.894
	Sex	-0.620	1.588	-0.042	-0.391	0.697	-3.772	2.531
	Sporting Cohort	-4.079	1.224	-0.351	-3.333	**0.001***	-6.507	-1.650
	Concussion History	2.202	1.588	0.141	1.387	0.169	-0.950	5.355
4	*(Constant)*	42.676	5.924		7.203	0.000	30.918	54.434
	Sex	-0.092	1.630	-0.006	-0.056	0.955	-3.327	3.144
	Sporting Cohort	-2.680	1.605	-0.231	-1.670	0.098	-5.866	0.505
	Concussion History	2.192	1.582	0.141	1.385	0.169	-0.948	5.332
	Lifetime Sporting Exposure	0.217	0.162	0.169	1.339	0.184	-0.105	0.539
5	*(Constant)*	41.724	6.059		6.886	0.000	29.697	53.750
	Sex	-0.080	1.633	-0.005	-0.049	0.961	-3.323	3.162
	Sporting Cohort	-2.765	1.612	-0.238	-1.716	0.089	-5.965	0.434
	Concussion History	2.387	1.605	0.153	1.488	0.140	-0.798	5.572
	Lifetime Sporting Exposure	0.208	0.163	0.161	1.273	0.206	-0.116	0.531
	Time Since Ceasing Sporting Activity	0.228	0.290	0.076	0.785	0.434	-0.348	0.804
6	*(Constant)*	48.033	6.711		7.158	0.000	34.711	61.355
	Sex	-0.235	1.609	-0.016	-0.146	0.884	-3.429	2.959
	Sporting Cohort	-3.657	1.644	-0.315	-2.224	**0.029***	-6.921	-0.392
	Concussion History	0.291	1.882	0.019	0.154	0.878	-3.445	4.026
	Lifetime Sporting Exposure	0.168	0.162	0.131	1.041	0.300	-0.152	0.489
	Time Since Ceasing Sporting Activity	0.259	0.286	0.086	0.907	0.367	-0.308	0.827
	Number of Concussions	-1.074	0.525	-0.250	-2.047	**0.043***	-2.115	-0.032

a. Dependent Variable: MCS-12

***** *Predictor variable is significant at the p<0.05 level (2-tailed).*

****** *Predictor variable is significant at the p<0.01 level (2-tailed).*

******* *Predictor variable is significant at the p<0.001 level (2-tailed).*

Table 4.14. *Multiple Linear Regression Model illustrating Predictors of Depression & Anxiety (GSI) among a Sub-Population of the Cohort.*

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for β	
		β	Std. Error	Beta			Lower Bound	Upper Bound
1	*(Constant)*	51.501	3.170		16.245	0.000	45.211	57.791
	Sex	-1.184	2.017	-0.059	-0.587	0.559	-5.185	2.818
2	*(Constant)*	45.230	5.601		8.075	0.000	34.116	56.344
	Sex	0.014	2.194	0.001	0.006	0.995	-4.340	4.368
	Sporting Cohort	2.335	1.722	0.147	1.355	0.178	-1.083	5.752
3	*(Constant)*	55.703	6.707		8.306	0.000	42.394	69.012
	Sex	-1.422	2.197	-0.070	-0.647	0.519	-5.781	2.938
	Sporting Cohort	3.050	1.693	0.192	1.801	0.075	-0.310	6.410
	Concussion History	-5.859	2.197	-0.275	-2.666	**0.009****	-10.220	-1.498
4	*(Constant)*	53.604	8.263		6.487	0.000	37.204	70.004
	Sex	-1.180	2.274	-0.058	-0.519	0.605	-5.693	3.332
	Sporting Cohort	3.688	2.239	0.233	1.648	0.103	-0.755	8.131
	Concussion History	-5.864	2.207	-0.276	-2.657	**0.009****	-10.243	-1.484
	Lifetime Sporting Exposure	0.099	0.226	0.056	0.438	0.662	-0.350	0.548
5	*(Constant)*	54.191	8.472		6.396	0.000	37.374	71.008
	Sex	-1.187	2.284	-0.059	-0.520	0.604	-5.721	3.347
	Sporting Cohort	3.741	2.254	0.236	1.660	0.100	-0.733	8.214
	Concussion History	-5.984	2.244	-0.281	-2.667	**0.009****	-10.438	-1.530
	Lifetime Sporting Exposure	0.105	0.228	0.060	0.461	0.646	-0.347	0.558
	Time Since Ceasing Sporting Activity	-0.141	0.406	-0.034	-0.346	0.730	-0.946	0.665
6	*(Constant)*	30.337	7.972		3.805	0.000	14.511	46.163
	Sex	-0.602	1.911	-0.030	-0.315	0.753	-4.396	3.192
	Sporting Cohort	7.111	1.953	0.448	3.640	**0.000*****	3.232	10.989
	Concussion History	1.942	2.235	0.091	0.869	0.387	-2.496	6.380
	Lifetime Sporting Exposure	0.254	0.192	0.144	1.323	0.189	-0.127	0.635
	Time Since Ceasing Sporting Activity	-0.259	0.340	-0.063	-0.763	0.447	-0.934	0.415
	Number of Concussions	4.059	0.623	0.691	6.514	**0.000*****	2.822	5.297

a. Dependent Variable: GSI

** Predictor variable is significant at the p<0.05 level (2-tailed).*

*** Predictor variable is significant at the p<0.01 level (2-tailed).*

**** Predictor variable is significant at the p<0.001 level (2-tailed).*

Table 4.15. *Multiple Linear Regression Model illustrating Predictors of Anxiety (GAD-7) among a Sub-Population of the Cohort.*

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for β	
		β	Std. Error	Beta			Lower Bound	Upper Bound
1	*(Constant)*	6.466	1.466		4.409	0.000	3.557	9.376
	Sex	-1.293	0.933	-0.137	-1.386	0.169	-3.144	0.558
2	*(Constant)*	1.320	2.539		0.520	0.604	-3.717	6.358
	Sex	-0.310	0.995	-0.033	-0.312	0.756	-2.284	1.663
	Sporting Cohort	1.916	0.781	0.259	2.454	**0.016***	0.367	3.465
3	*(Constant)*	4.561	3.098		1.472	0.144	-1.587	10.709
	Sex	-0.755	1.015	-0.080	-0.744	0.459	-2.768	1.259
	Sporting Cohort	2.137	0.782	0.289	2.732	**0.007****	0.585	3.689
	Concussion History	-1.813	1.015	-0.183	-1.786	0.077	-3.828	0.201
4	*(Constant)*	2.455	3.803		0.646	0.520	-5.092	10.003
	Sex	-0.512	1.046	-0.054	-0.490	0.626	-2.589	1.564
	Sporting Cohort	2.778	1.030	0.376	2.696	**0.008****	0.733	4.822
	Concussion History	-1.818	1.016	-0.183	-1.790	0.077	-3.834	0.197
	Lifetime Sporting Exposure	0.100	0.104	0.121	0.956	0.342	-0.107	0.306
5	*(Constant)*	3.379	3.873		0.872	0.385	-4.310	11.067
	Sex	-0.523	1.044	-0.056	-0.501	0.617	-2.596	1.549
	Sporting Cohort	2.860	1.030	0.387	2.776	**0.007****	0.815	4.905
	Concussion History	-2.007	1.026	-0.202	-1.957	0.053	-4.043	0.029
	Lifetime Sporting Exposure	0.109	0.104	0.133	1.046	0.298	-0.098	0.316
	Time Since Sporting Retirement	-0.221	0.186	-0.115	-1.191	0.237	-0.589	0.147
6	*(Constant)*	-2.955	4.149		-0.712	0.478	-11.191	5.282
	Sex	-0.368	0.995	-0.039	-0.370	0.712	-2.342	1.607
	Sporting Cohort	3.755	1.017	0.508	3.693	**0.000*****	1.736	5.773
	Concussion History	0.097	1.163	0.010	0.083	0.934	-2.213	2.407
	Lifetime Sporting Exposure	0.148	0.100	0.181	1.486	0.141	-0.050	0.347
	Time Since Sporting Retirement	-0.252	0.177	-0.132	-1.428	0.157	-0.603	0.099
	Number of Concussions	1.078	0.324	0.393	3.323	**0.001****	0.434	1.722

a. Dependent Variable: GAD-7

***** Predictor variable is significant at the p<0.05 level (2-tailed).*

****** Predictor variable is significant at the p<0.01 level (2-tailed).*

******* Predictor variable is significant at the p<0.001 level (2-tailed).*

Table 4.16. *Multiple Linear Regression Model illustrating Predictors of Concussion Knowledge Score among a Sub-Population of the Cohort.*

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for β	
		β	Std. Error	Beta			Lower Bound	Upper Bound
1	(Constant)	12.716	0.387		32.858	0.000	11.951	13.482
	Sex	-0.458	0.240	-0.165	-1.905	0.059	-0.934	0.018
2	(Constant)	11.999	1.511		7.943	0.000	9.010	14.988
	Sex	-0.484	0.247	-0.174	-1.961	0.052	-0.973	0.004
	Age	0.036	0.073	0.044	0.491	0.624	-0.109	0.181
3	(Constant)	11.860	1.512		7.846	0.000	8.869	14.851
	Sex	-0.624	0.271	-0.224	-2.303	**0.023***	-1.160	-0.088
	Age	0.028	0.073	0.034	0.382	0.703	-0.117	0.173
	Study Cohort	0.269	0.217	0.120	1.242	0.217	-0.160	0.698
4	(Constant)	11.731	1.544		7.600	0.000	8.677	14.786
	Sex	-0.602	0.276	-0.216	-2.178	**0.031***	-1.148	-0.055
	Age	0.023	0.074	0.028	0.309	0.758	-0.124	0.170
	Study Cohort	0.289	0.222	0.129	1.302	0.195	-0.150	0.728
	Concussion History	0.124	0.277	0.041	0.448	0.655	-0.424	0.672

a. Dependent Variable: Concussion Knowledge Score

Table 4.17. *Multiple Linear Regression Model illustrating Predictors of Concussion Attitude Score among a Sub-Population of the Cohort.*

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for β	
		β	Std. Error	Beta			Lower Bound	Upper Bound
1	(Constant)	63.332	1.926		32.885	0.000	59.522	67.142
	Sex	1.120	1.196	0.082	0.936	0.351	-1.247	3.487
2	(Constant)	52.870	7.464		7.083	0.000	38.102	67.638
	Sex	0.740	1.220	0.054	0.606	0.545	-1.674	3.153
	Age	0.525	0.362	0.129	1.450	0.149	-0.191	1.241
3	(Constant)	52.705	7.511		7.017	0.000	37.842	67.567
	Sex	0.573	1.346	0.042	0.426	0.671	-2.090	3.236
	Age	0.515	0.365	0.127	1.414	0.160	-0.206	1.237
	Study Cohort	0.321	1.078	0.029	0.298	0.766	-1.812	2.453
4	(Constant)	51.865	7.665		6.766	0.000	36.697	67.033
	Sex	0.718	1.372	0.052	0.523	0.602	-1.997	3.433
	Age	0.483	0.370	0.119	1.306	0.194	-0.249	1.214
	Study Cohort	0.448	1.102	0.041	0.407	0.685	-1.733	2.629
	Concussion History	0.808	1.375	0.055	0.587	0.558	-1.914	3.529

a. Dependent Variable: Concussion Attitude Score

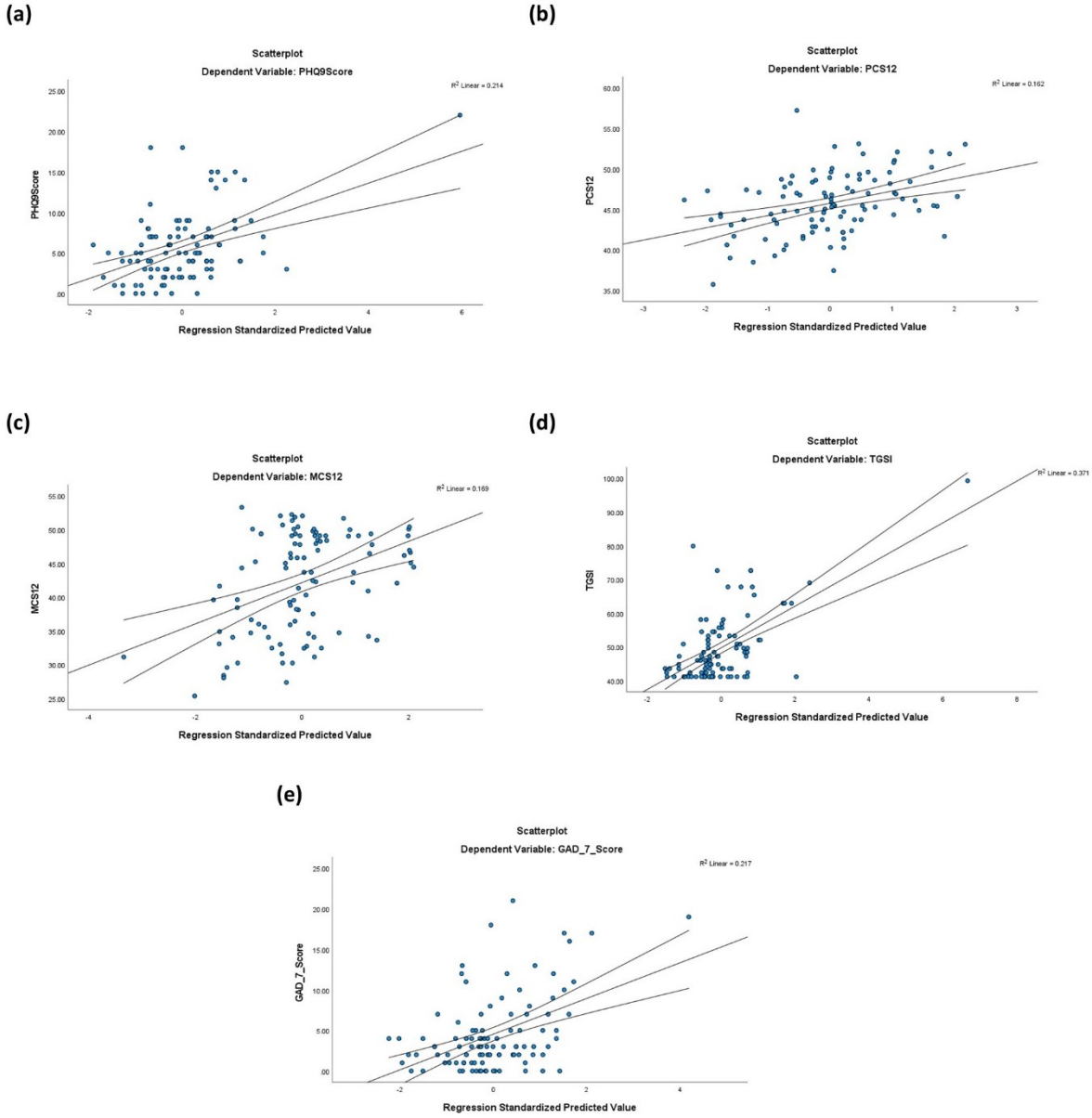

Figure 4.13. *Bivariate Regression Model Analysis Scatterplot of Depression and Anxiety Metrics.* **(a)** PHQ-9, **(b)** PCS-12, **(c)** MCS-12, **(d)** GSI, **(e)** GAD-7.

4.4.10. SIFI Results

SIFI scores are reported for 192 participants over the six multisensory incongruent SIFI illusory conditions. For each participant in each condition, the score is reported as a proportion (0, 0.25, 0.5, 0.75, 1) to represent the proportion of correct responses in four repetitions of the condition. A negative SOA signifies a sequence of A-V/A stimuli, while a positive SOA indicates V/A-A stimuli. A higher score indicates that the participant perceived one flash even when presented with two beeps, i.e., responded correctly,

whereas a lower score indicates that the participant experienced many illusions. The coding of responses was consistent with previous studies (268, 269, 626). In cases where two or more stimuli were presented, a correct response was one in which the participant reported that two or more flashes (or beeps when flashes were not present) occurred. All sub-populations showed a significant reduction in accuracy with decreasing SOA temporal pairings and is discussed in more detail below.

4.4.10a Concussion History

Similar to the pilot study, those with and without a concussion history were found to perform with similar perceptual accuracy across all illusory SOA conditions (mean difference = 0.064, SE = 0.048); see **Figure 4.14.** below. This is again contradictory to our proposed hypothesis but may be expected given no one in the present cohort sample was acutely suffering from a concussive injury to incite identifiable alterations in cognition and MSI. A test of within-subjects effects revealed a significant main effect of SOA condition (df = 3.964, F = 41.476, p=0.000, GGeps = 0.793, η^2 = 0.180), and a non-significant interaction effect of concussion history (df = 3.964, F = 0.489, p=0.742, η^2 = 0.003). A test of between-subjects effects revealed a non-significant effect of concussion history on the SOA conditions (df = 1, F = 1.185, p=0.179, η^2 = 0.01).

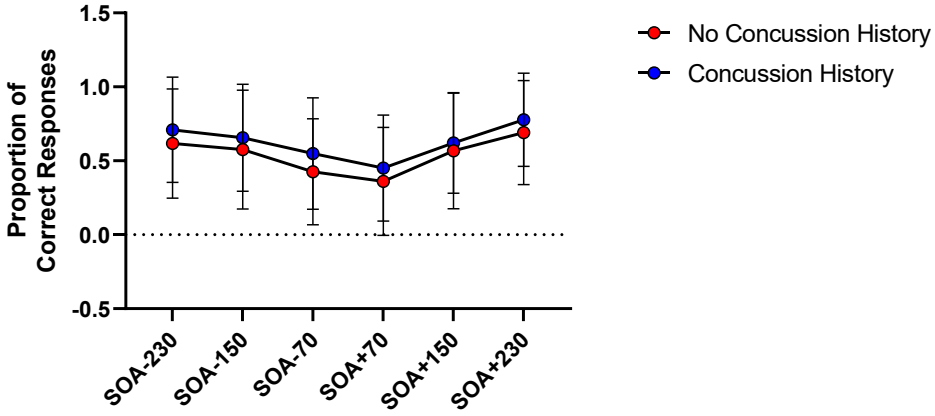

Figure 4.14. *Proportion of correct responses across SOA conditions by Concussion History.*

4.4.10b Sporting Cohort

To determine the effect of cohort on the fission illusion SOA conditions, a mixed model ANOVA revealed a significant main effect of SOA condition (df = 3.951, F = 45.246,

p=0.000, GGeps = 0.790, η^2 = 0.194), a non-significant main interaction effect of cohort*SOA conditions (df = 7.902, F = 0.422, p=0.907, η^2 = 0.004), and a non-significant between-subjects effect of cohort (df = 2, F = 2.651, p=0.073, η^2 = 0.027). Although, as seen in the pilot study, those who participated in contact-based sports in this cohort performed with higher levels of perceptual accuracy (mean = 0.654, SE – 0.047) versus the non-contact (mean = 0.526, SE = 0.031) and the control cohorts (mean = 0.568, SE = 0.041) across all SOA conditions, see **Figure 4.15.** below. A significant simple effect of cohort was however seen for the SOA+230 condition, where the Contact cohort showed higher levels of perceptual performance than the Non-Contact cohort (mean difference = 0.14, SE = 0.06, t = 2.53, DF = 95.58). All groups displayed a decline in perceptual with shorter temporal delays, and displayed a reduction across auditory stimulus onsets across pre and post flash presentations which is in line with previous studies demonstrating an increased susceptibility to the SIFI with decreasing SOAs (95, 269, 626).

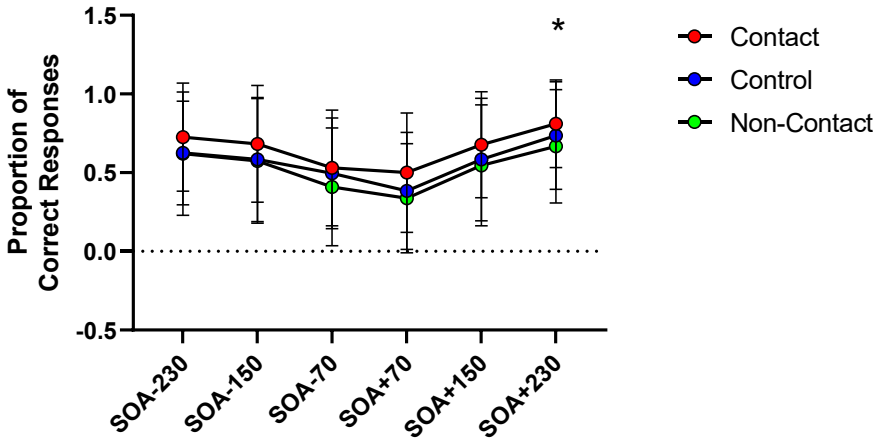

Figure 4.15. *Proportion of correct responses across SOA conditions by Sporting Cohort.* The Contact cohort displayed higher levels of MSI across all SOA conditions and were found perform significantly better than the Non-Contact cohort at the SOA+230 condition (p<0.05).

4.4.10c Sex

To determine the effect of sex on the fission illusion SOA conditions in the follow-up study, a mixed model ANOVA revealed a significant within-subject of SOA condition (df = 3.959, F = 52.363, p=0.000, GGeps = 0.792, η^2 = 0.217), a non-significant within-subject interaction effect of sex*SOA conditions (df = 3.959, F = 0.903, p=0.461, η^2 = 0.005). Similar to the pilot study, males can be seen to have an overall higher level of

performance accuracy across all SOA conditions in comparison to females (mean difference = 0.096, std error = 0.044), which revealed a significant between-subjects effect of sex (df = 1, F = 4.861, p=0.029, η^2 = 0.025) in line with what we previously found. **Figure 4.16.** below shows the group-averaged proportion of correct illusory responses for males and females as a function of SOA. Once again, females are more susceptible to the fission illusion across a wide range of temporal asynchronies, but in this cohort irrespective of whether the second auditory beep precedes or follows the A-V pairing.

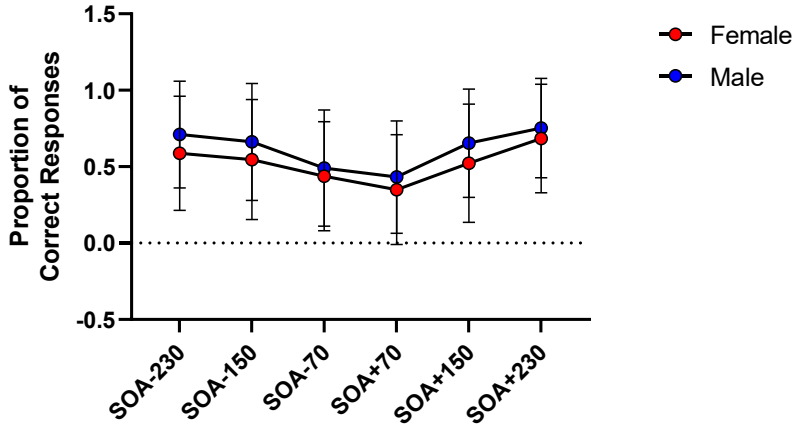

Figure 4.16. *Proportion of correct responses across SOA conditions by Sex.* No significant differences between males and females were found for individual SOA conditions. Although males tended to exhibit higher multisensory performance than females across all illusory SOA conditions.

4.4.10d Open Skill vs Closed Skill

To further determine the effect of sporting skill type on MSI, we compared those who participated in open skill versus closed skill-based sports. We hypothesised that those involved in sports that are predominantly open skill would pertain to higher levels of perceptual ability due to the fast-moving and high-paced sporting environment than those in more stationary, closed skill sports.

To determine the effect of sporting type on the fission illusion SOA conditions, a mixed model ANOVA revealed a significant within-subjects effect of SOA condition (df = 3.955, F = 45.227, p=0.000, GGeps = 0.791, η^2 = 0.194), and a non-significant within-subject interaction effect of sporting type*SOA conditions (df = 7.910, F = 0.359, p=0.940, η^2 = 0.004). But in line with our hypothesis based on the MSI capabilities from the pilot study population, a significant between-subjects effect of sporting type was

found (df = 2, F = 3.123, p=0.046, η^2 = 0.032), see **Figure 4.17.** below. An isolated analysis of the SOA conditions revealed a significant simple effects of sporting type where, in line with our hypothesis, those classified as open skill sports performed significantly better among those conditions with a larger temporal asynchrony at the SOA+230 condition compared to closed skill sports (mean difference = 0.15, t = 2.75, DF = 139.3, p<0.05).

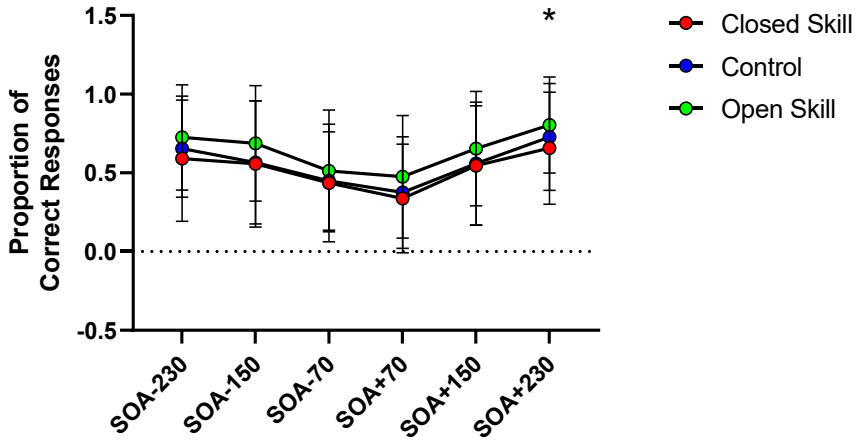

Figure 4.17. *Proportion of correct responses across SOA conditions by Skill Type.* A significant difference between Open and Closed Skill was found for the SOA+230 condition, where open skill sporting type led to enhanced MSI capabilities (p<0.05).

4.4.10e Predicting SIFI Performance and Patterns from Baseline Data

In order to assess whether baseline variables measured in participants can accurately predict overall patterns in response to the SIFI, the data was categorised into defined strata; see **Chapter Three** for further details. The following four categories were derived from the strata: "*High*" performers constitute the three strata at the top, "*Good*" performers are strata 4 and 5 (from the top), "*Low*" performers are strata 6 to 10. The final category is termed "*Learners*" who appear to get better over time, and they are strata 11 to 13. A category label z_i for each participant is formed:

$$z_i = \begin{cases} 0 & \text{“High”} \\ 1 & \text{“Good”} \\ 2 & \text{“Low”} \\ 3 & \text{“Learner”} \end{cases}$$

We then performed a multinomial logistic regression using the baseline variables recorded:

Variable	Explanation
Age	Participant age
Sex	Participant sex Female (reference) / Male
Sport type	Control (reference) / Open Skill / Closed Skill
Cohort	Control(reference) / Non-Contact / Contact
Concussion history	No (reference) / Yes
Activity level	Low (reference) / Moderate / High

For multinomial regression we need to choose one category to be the reference category, and all interpretations are in comparison to that category - here we take $z_i = 2$ ("*Low*") to be the reference category, so that we can distinguish if any of the baseline variables can distinguish "*High*" from "*Low*" scorers. The model is fit through Markov Chain Monte Carlo using a Bayesian Pólya-Gamma sampler as discussed in Polson et al. (627), adopting non-informative independent $N(0, 10^2)$ priors on all coefficients.

Figure 4.18. shows credible intervals for categories "*High*", "*Good*", "*Learners*" in reference to the "*Low*" category. The only notable result is that being younger and playing an open skill sport makes a participant more likely to be a high performer than a low scorer (*as the intervals do not cross zero, indicated with a vertical line*). There are no other noteworthy interpretations from this plot, but further understanding of the data can be gathered from **Figure 4.19.** depicting the spread of High, Good, Low performers, and Learners from the baseline SIFI data of the follow-up study.

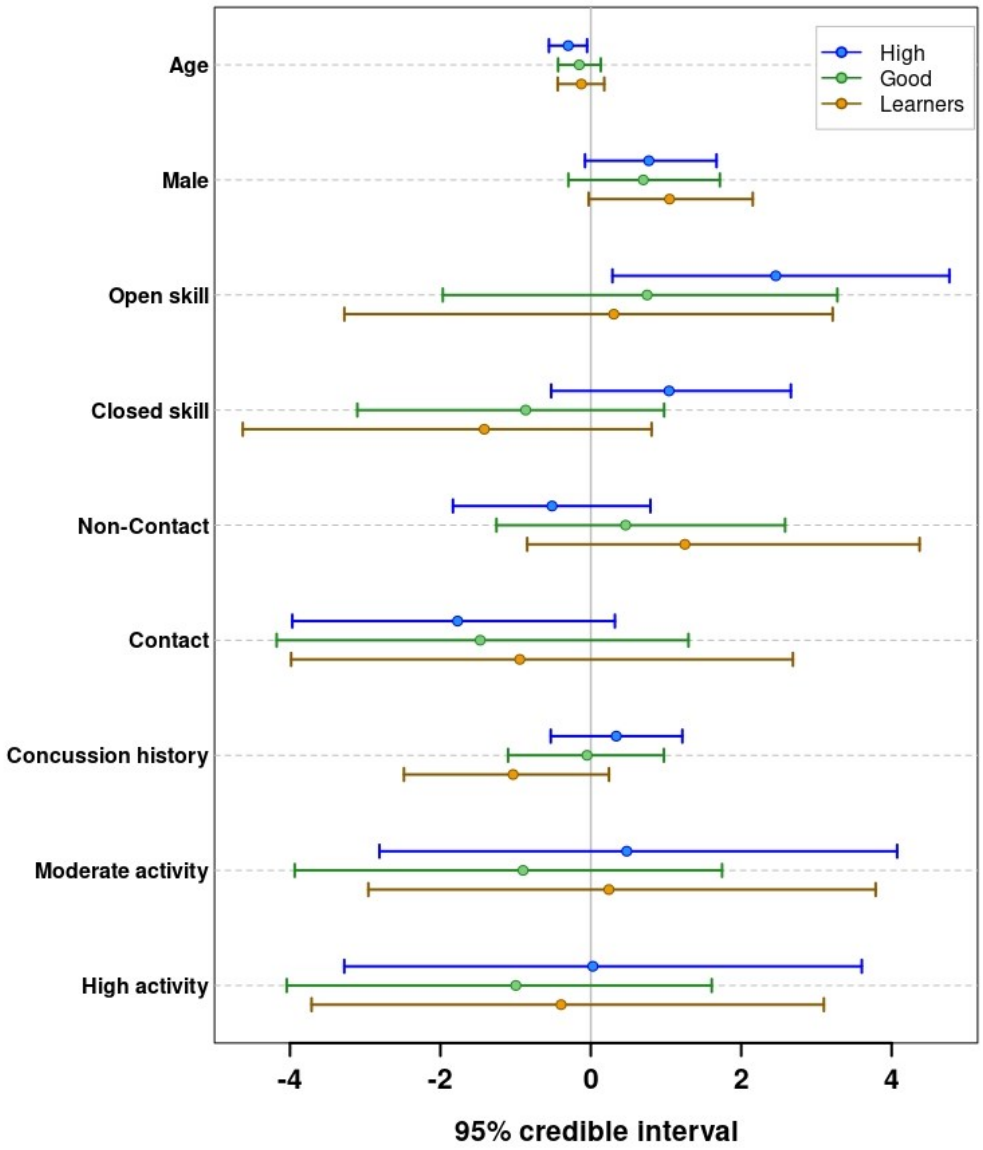

Figure 4.18. *Credible intervals (95%) from Bayesian multinomial regression on the categories.*

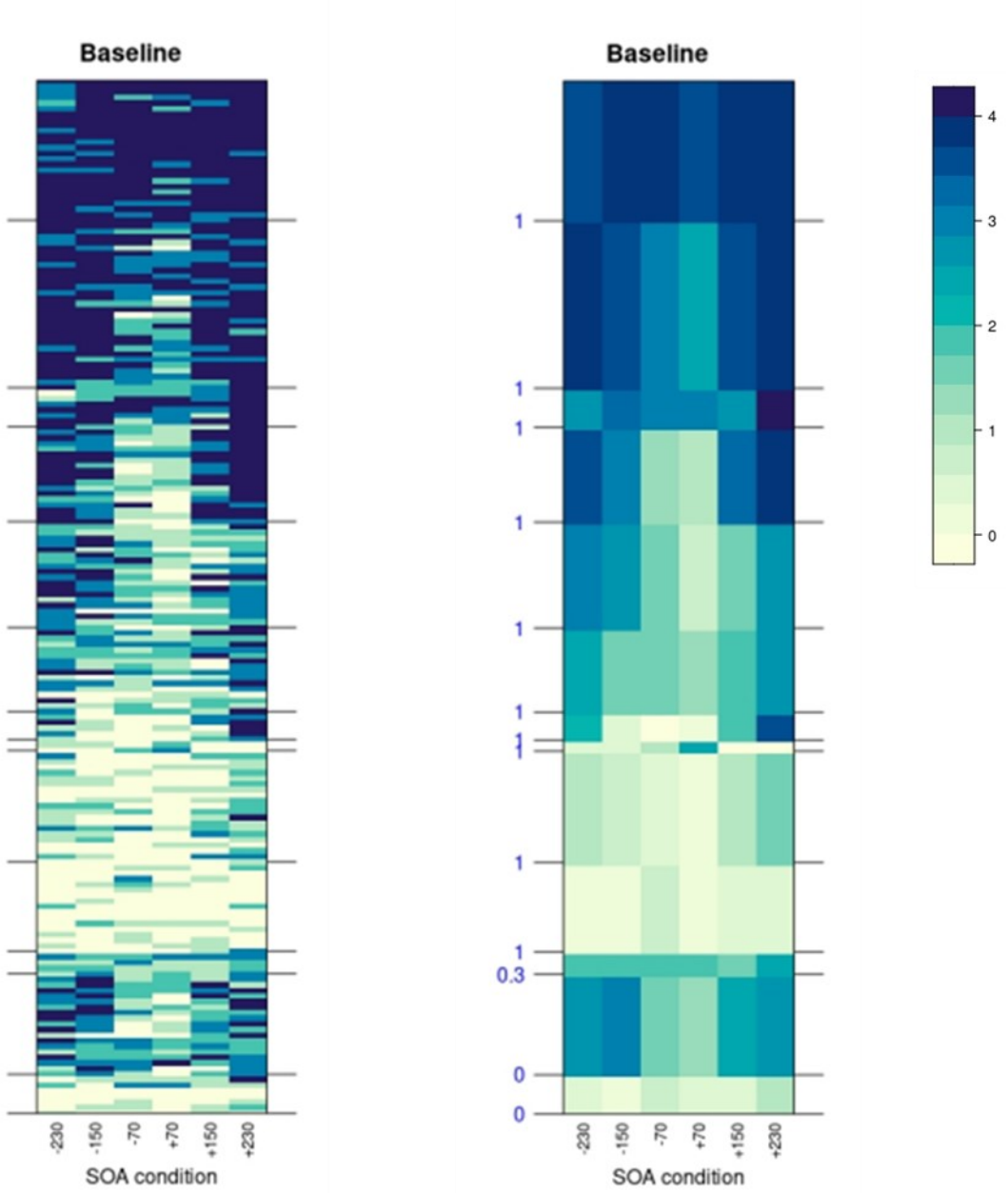

Figure 4.19. Results of the agreement model for $\hat{k}$ = 13 strata showing the mean of each variable within classes as a heatmap for baseline results of the follow-up study.

4.5. Discussion

The results of the present study revealed that young adults who participate regularly in contact sport had the highest level of reported concussions compared to those who play non-contact sport or do not participate in sport. However, no significant differences were found in susceptibility to the SIFI task between these groups. Males had a significantly higher number of reported concussions than females, although surprisingly males also had a significantly higher level of perceptual performance during the SOA illusory conditions. It was originally hypothesised that those participating in contact sports would perform with reduced levels of MSI during the SIFI task due to greater exposure to sustained RHI's. However, the present findings show that, independent of regular participation in contact sport, a greater number of previous concussions did not result in impaired performance in the SIFI task across the illusory conditions. Finally, there are identifiable and significant differences in performance of the SIFI task between those in open skill versus closed skill sports alongside age based on our regression modelling which may better account for the higher rates of performance accuracy among those in the contact cohort. This study has provided an insight into the perceptual performance of young adults with and without a history of concussion across a range of sporting types determined using the MTBIIM (518, 628) and the SIFI as an assessment of MSI (96-98).

In line with recent data (95, 172), we report a significant between-subject effect of sex against the illusory SOA conditions, although this significance did not persist during our multinomial logistic regression using baseline variables. Males were found to perform better (*i.e.*, less susceptible to SIFI) than females, and specifically during the negative SOA pairing at 230ms. The present findings, although mixed, are in line with recent research documenting sex differences in the SIFI, with females being more susceptible to the fission illusion than males (95). Although it the cause of the sex difference in perceptual performance could not be determined, the authors suggest that "a smaller temporal binding window in males than in females might lead to different levels of susceptibility to the illusion" and the increased level of susceptibility was not the result of auditory processing imbalances (95). Early investigations have postulated the idea that females rely more heavily on visual information during spatial tasks of perceived orientation (629). However, further investigations will be required to determine the sex differences during SIFI performance and if concussion history could be a potential factor of determination.

Athletes who play contact-based sports are more prone to concussions given the nature of reoccurring physical contact. The incidence of concussion in rugby union is as high as 8.9/1000 hours of play over two seasons (630), where rugby players are cited to endure up to 100 direct head impacts over a competitive season (499). In the present study, those in contact sports had higher levels of MSI across the illusory SOA conditions despite numbers of self-reported concussions being highest in this group, although the results were not significant. This may be in part accounted for in the types of training that are specific to those in contact sports and moreover those in open skill sports with high levels of player movement and dynamic play to compete with. Our results however did highlight those who participate in open skill sports showed higher MSI capabilities in support of our hypothesis following on from our pilot study. Contact sporting athletes and those in open skill sports can be seen to develop the highest level of decision-making, attentional, and memory-related skills due to the high-speed, competitive nature of the sporting environment (166) which may support our present findings. Perceptual-cognitive skills that are honed and developed include pattern recognition and situational probabilities based on a priori expectations (167, 301). Participating in sports throughout one's life is associated with enhanced cognitive abilities and executive processing, particularly in field-based athletes who rely on advanced visuospatial skills and precise motor coordination. Repeat engagement in multisensory training can subsequently lead to adaptive and proficient neural modifications in the prefrontal cortex increasing rate of perceptual and information processing (631). The work by Dux et al. involved subjects performing either one sensory-motor task (single-task) or two distinct tasks (dual-task) that didn't overlap in sensory or output types. Three models were considered regarding how training affects sensory-motor processing. The first model suggested a shift from deliberate to efficient processing in specific pathways. The second model proposed the segregation of neural ensembles for independent task processing. The third model suggested improved multitasking efficiency through prefrontal cortex processing. The results from four analyses across two data sets align predominantly with the idea that training improves efficiency in prefrontal cortex information processing, supporting the notion of improved multitasking efficiency (631). This may in part explain our results of enhanced MSI in open skill, contact sports with long term exposure to dynamic sporting environments that leads to cognitive changes at a functional and structural level. Sports-specific perceptual training is achieved by methods of distant-dependent

modulations by increasing the spatial distance between the A-V stimulus pairing to reduce the signal saliency, thus increasing the temporal disparity between them, and decreasing the size of the object/event as perceived by the retina. These temporo-spatial modulations of objects and environmental events will alter the size of the TBW and cause a shift in the PSS (118, 119, 128). It may be that those in contact sports which are predominantly open skill are better equipped to alter their PSS providing them with heightened MSI abilities despite higher rates of concussion. Neurobehavioural enhancements such as these may derive from re-organisation through neuroplastic mechanisms to alter sensory cortical maps and perceptual abilities reflective of our present findings (161).

Of significant note in the present study, participating in certain sports with high levels of information processing classified as open skill sports may enhance SIFI task performance, i.e., render a participant less susceptible to the illusion, provided skill type was a significant variable in our regression analysis. Task specific learning and training in daily activities and sports causes focal changes in task-relevant brain regions. With an initial recruitment of a few regions in the learning and development stages as a novice, approaching expert level performance enables isolation of region-specific neural networks creating a functional neural topography that is most efficient for a given task (632). This differentiation, recruitment, and specialisation tend to dissipate with age, recruiting more brain regions for the given task (443). This 'unlearning' of MSI could be brought upon earlier in those who are female, present with a history of concussion, and did not participate in fast paced dynamic sporting environments. The spatiotemporal dynamics of MSI thus present differently between young and old where increases in self-reported visual temporal discrimination with age predicted susceptibility to the SIFI (114). This was also exemplified among females where older age and lower cognitive capabilities were indicative of higher SIFI susceptibility (95). Furthermore, an assessment of A-V integration through MEG recordings revealed that older adults had faster reaction times to A-V stimuli relative to unisensory stimuli in comparison to young adults by 158 ms with the associative increase sourced in the activity of the right medial pre-frontal cortex (245). The enhanced reaction times of the older adults were correlated with reductions in GM volume, specifically in frontal, parietal, temporal and occipital lobes, and translates to a functional reorganisation due to age-related cerebral structural decline (245). It may be that the SIFI would allow for the detection of subtle and persistent deficits in MSI from a history of concussion and

exposure to RHI. Compared to a single concussive incident, experiencing one or more concussions over a number of years has been cited to decrease cognitive capabilities (88). However, due to the heterogeneity of the clinical aspects of concussion, literature necessitates an objective tool which considers underlying physiological and neurological function in the preservation of brain health. The inclusion of the SIFI task as part of a multidimensional approach to concussion diagnosis and rehabilitation may provide the means of a reliable and sensitive perceptual assessment tool of current and future brain health.

The most comparable results to our study to date are those of Cunningham and colleagues (172). Former rugby players of middle-age were found to have a significantly larger amount of self-reported concussions than their rowing counterparts, and concurrently were more susceptible to the fission illusion. Cunningham et al., however, did report that their study was limited by 'self-reported concussion history' which may account for the inherent outlier discrepancy. These results pose the question as to whether long-term engagement in sport may in fact establish longer lasting and coherent MSI, protecting against neurocognitive decline with age. Or could RHI exposure in contact sport accelerate associative ageing cognitive decline and counteract these benefits? There are many different age-related auditory and visual deficits, including, but not limited to, reduction in auditory sensitivity to temporal fine structures (256, 257), and impaired auditory and visual temporal order judgements (258, 259). Temporal and spatial perception of A-V sensory input diminish with normal ageing with the potential to impact daily life and, when coupled with sporting pathophysiological decrements and being female, could impact optimal sporting performance. With cognitive ageing reported to begin as early as 20 years of age and progressively declining until an acceleration around the age of 60 (59), concussion and RHI may only exacerbate the natural cognitive decline with a reorganisation of functional brain networks and additional recruitment of neural resources to counteract any injury-induced neurodegeneration (60-62).

Previous systematic evidence has stated that the length of time since the most recent concussion and retirement (i.e., time elapsed since potential head impact exposure) is also important when attempting to distinguish between the potential long-term effects of a sporting career and the more short-term effects of a recent concussion (482). We found that sporting cohort, time since ceasing regular sporting activity, and previous number of concussions were predictors of depression, and

specifically a higher number of previous concussions was linked with increased metrics of somatisation, anxiety, and depression. Cunningham and colleagues report that greater concussion exposure is linked to higher total PCS scores, including the memory post-concussion symptom (PCS) scale. This link, however, was seen only in retired and recreational athletes and not competitive players. The authors highlight that given the possibility of recollection bias in self-report, it is challenging to evaluate the relationships between self-reported concussion history and subjective PCSC scores (482). Our results suggest that mental health scores among athletes in early adulthood are comparable to those in mid and later life. While it is widely acknowledged that physical activity and sports engagement yield notable physical and mental health advantages, there is a growing body of compelling evidence indicating that involvement in certain athletic activities, particularly contact sports, carries potential drawbacks, leading to adverse effects on brain function and overall brain health. However, we found was there was a significant toll on player physical well-being even at an early age. A significant association between time since ceasing sporting activity and worse physical health was also found in our results. This is not unexpected considering the nature of sport and particularly contact sport which involves high-impact body contact owing to repeated physical collisions with opponents and the terrain at fast speeds, as well as regular twisting and manoeuvring. For example, those athletes who are forced to retire involuntarily due to injury report reduced physical component scores, and similar mental component scores, in comparison to those who were not forced to retire (633). Musculoskeletal injuries such as joint, ligament, muscle, and tendinous injuries, as well as fractures and dislocations, are common among contact sports like rugby union (634). Previous injury records were not recorded in the present study which could provide more substantial insight in future research. The link between a history of concussion and repetitive head impacts with symptoms of mental and physical health issues later in life and the long-term consequences of a higher injury incidence in contact sport on longer-term health-related quality of life is lacking. However, emerging evidence suggests former male rugby athletes are just as likely as the general population, if not more at risk, of experiencing anxiety or depression (3). This may be due to other psychological factors such as decreased satisfaction with life, decreased resilience, and greater athletic identity leading to a loss of purpose. The latter in particular may be associated with increased distress on retirement and necessitates further empirical extension to younger adults retired from recreational

sport given our current findings. Awareness of the hidden symptoms surrounding physical and mental health among current and retired players should be a priority to put the appropriate support structures in place.

In the context of the concussion literature what must however be acknowledged is that greater knowledge of concussion is not necessarily associated with increased reporting behaviour (635). The problem of underreporting potential serious concussion-related events is worsened by the widespread use of imprecise and seemingly harmless terms such as "dings" or "bell ringers". Based on the concern surrounding self-report of concussion history, good knowledge of concussion alone is insufficient. In the present results we found sex to be a significant predictor variable of concussion knowledge but not attitudes. Males reported marginally higher scores than females. Although there was no significant difference in concussion history by sex, females had longer sport participation rates than males potentially exposing them to a higher risk of sustaining a concussive injury which could be viewed as contradictory. Previous evidence found that male student-athletes had lower probabilities of having greater levels of self-reporting intents than female student-athletes which may substantiate our present findings and is reflective of potential differences between male and female sporting environments by type and level of competitive play (636). In contrast, males have been cited to express higher negative results from concussion reporting and lower intention to disclose concussions than females, although no significant sex differences in concussion-reporting behaviours were observed (637). Athletes require adequate knowledge to have the confidence to report concussion symptoms when they arise. Nevertheless, having greater knowledge alone does not guarantee improved concussion-reporting behaviours. Indeed, attitudes towards concussion reporting have been demonstrated to be a crucial factor linked to self-reporting behaviours (638). Interventions have been designed in the hopes of influencing actual behaviour by enhancing user understanding of indications and symptoms, attitudes, and reporting intentions. The extent to which different educational models and theories are incorporated and reaching appropriate audiences to account for the variance in our results is unclear. In a previous cohort of athletes with a history of concussion, more than 70% continued to play following their most recent concussion, and over half stated that they would not report suspected concussions if they occurred during an important game (522). Nonetheless, the authors found that having a higher concussion knowledge score did not translate into differences in reporting or continuing to play after a concussion. In contrast, those who received

previous concussion education had higher knowledge scores and were less likely to continue playing after a concussion. Recent systematic evidence suggests that future concussion educational programs should be created with expert input, theoretical foundations, and a knowledge transfer framework such as the knowledge-to-action cycle in mind to ensure improvement (639). It is unfortunately challenging to develop and implement targeted concussion-prevention strategies given the multidimensional nature of such a complex injury.

4.6. Limitations

Participants in the present study were undertaking third level education and engaging in collegiate sports, and thus are not fully representative of the general population. Participants were not assessed for normal or correct-to-normal vision and hearing or handedness upon recruitment. Concussion history was determined retrospectively by self-report and may not be an accurate representation of concussion history. Further research into the potential utility of the SIFI in sport should be undertaken to meet the current requirement for more objective diagnostic tests of concussion to reduce, if not remove, the subjectivity surrounding some of the current assessment protocols and that of the patient (640). Additionally, the convenience sampling technique and the geographic constraint of our sample limit the external validity of our findings.

4.7. Conclusion

Our results indicate that young adults participating regularly in contact sports had the highest reported concussion rates, while no differences were observed in susceptibility to the SIFI task among different sport groups. Despite males reporting more concussions than females, males also showed better perceptual performance in the SOA illusory conditions. Contrary to our initial hypothesis, a greater history of concussion did not translate to poorer performance in the SIFI task, regardless of contact sport involvement. Moreover, significant performance variations in the SIFI task were linked to open vs. closed skill sports and age as revealed by regression analysis. With an ageing population, increased research into the female athlete, and the cumulative effects of concussion across the lifespan with the ability to predict brain health and cognitive status is paramount. A divergent movement from the past

symptom-base model of concussion diagnosis is needed, and towards a more extensive multi-dimensional and temporal assessment (641). The results of the current study highlight the need for more broadened research on the use of measures of perceptual function in sport and concurrent assessment of an athletes' current and future cognitive well-being.

4.8. Question(s) Raised

- If the SIFI is to be introduced as a pitch side assessment tool of SRC, how do results possibly differ in the context of varying exercise intensities experienced during match play?

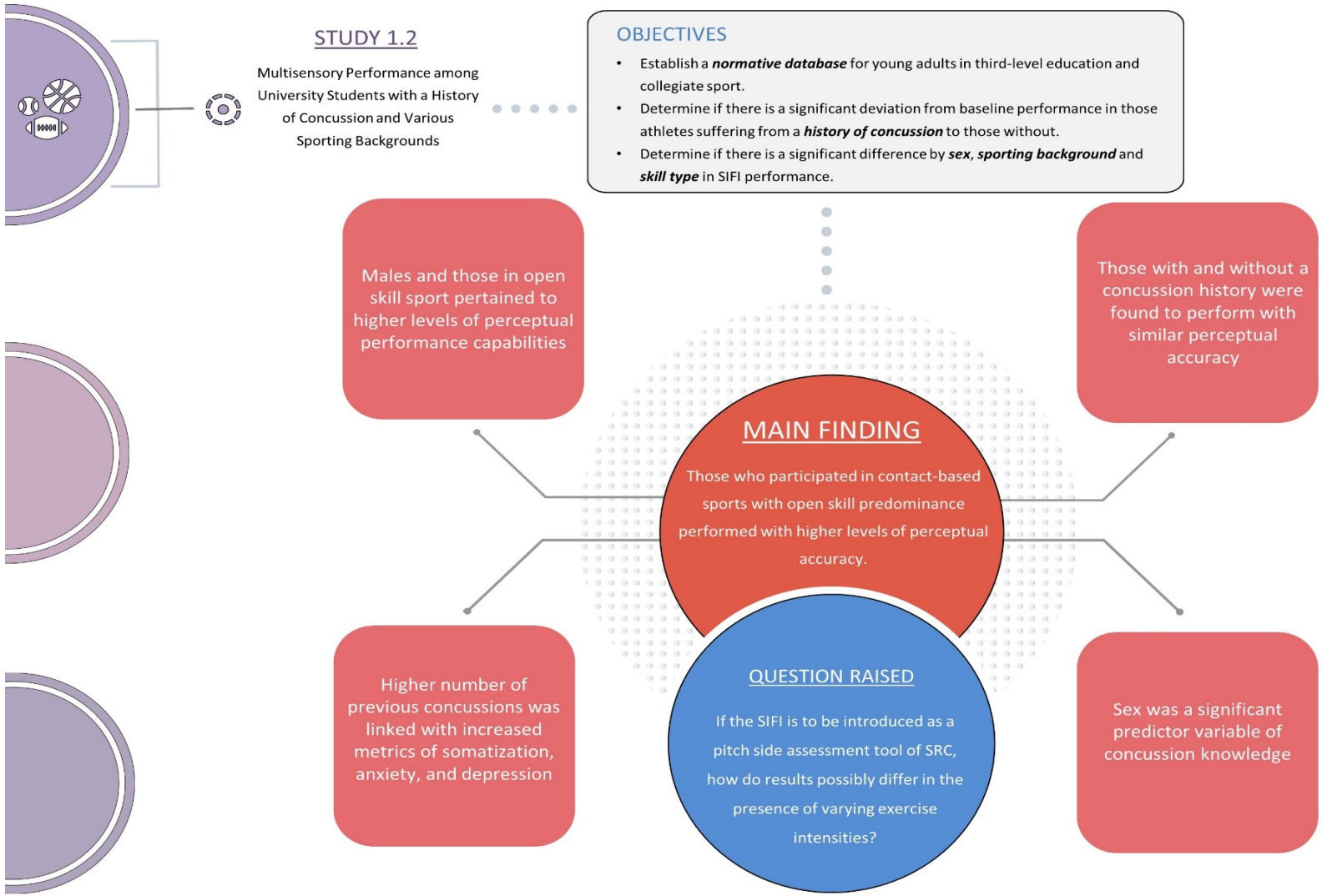

Figure 4.20. *Chapter Three – Study 1.2 Summary.*

5

Chapter Five:

Does Moderate or High Intensity Exercise Influence Multisensory Integration Capabilities among a Cohort of University Students: An Exploratory Study

5.1. Highlights

- Central and peripheral fatigue were induced during the high intensity exercise protocol as evidenced by significant differences in lactate, heart rate, and RPE post exercise compared to moderate intensity exercise, supporting our study design and methodology.
- Exercise of moderate and high intensity replicating the physiological demands of a sporting environment did not impact the susceptibility to the SIFI illusion.
- The agreement measure for pre and post moderate exercise, which is 0.8428, is comparable to the agreement results in **Chapter Three**. However, the agreement measure for pre and post high intensity (0.9928) surpasses the moderate intensity. This might suggest a learning effect, at least in a small proportion of participants who demonstrate increased consistency in their performance, as they become more familiar with the task following multiple testing sessions irrespective of possible confounding factors such as exercise.
- Similar to our findings in **Chapter Four**, sex was found to be a significant variable in perceptual performance of the SIFI task, where males were less susceptible to the illusory conditions.
- An isolated analysis of the individual illusory conditions revealed that those in open skill sports had a higher capacity for MSI, irrespective of exercise intensity.

5.2. Introduction

Exercise and physical activity (PA) are an integral part of healthy ageing, linked with the safeguarding of both cognitive and cardiovascular (CV) function in young and old (642, 643). The seminal work of Spirduso in the mid-1970's paved the way for exercise and cognition research after finding that active, older adults possessed similar cognitive capabilities to that of young adults (644). Higher levels of aerobic fitness contribute to enhance cognitive performance across the lifespan from childhood into adulthood and later life (645, 646). The chronic effects of PA on enhanced cognitive function are also exhibited in sporting athletes (647). Exercise is known to modulate cognitive and perceptual abilities in both the acute and long term associated with both positive and negative outcomes in task performance. Even a single bout of acute exercise can induce immediate positive effects on cognitive performance irrespective of age (648). Arousal of the CNS through exercise is

proposed to provide positive enhancements in the sensitivity and flexibility of the human sensory systems in response to environmental stimuli such as the case in sporting events (157). But what remains to be determined is how influential exercise is on MSI capabilities.

Previous evidence investigated the effects of open and closed skill exercise on cognitive function in older adults, using the SIFI test and the Forward Digit Span task (158). The open skill and control groups performed better than the closed skill group, and the open skill group improved after exercise engagement. The study found that habitual PA was not the main factor in perceptual improvements and the differences seen were "*due to a quantitative difference in the intensity of the exercises or a qualitative difference between the two exercise modes"* (158). Exercise modality and intensity can significantly impact cognition, but there is a great disparity in the literature regarding the exercise intensity used, making it difficult to draw meaningful conclusions about cognitive performance outcomes (514). Methodological differences such as sample size, types of exercise performed, and timing of cognitive testing post-exercise may also account for contradictory and varying outcomes in the literature. For example, running requires more attention and energetic constraint to maintain balance and whole-body co-ordination than stationary cycling (649), where temporal dynamics linked to motor co-ordination and more inherent multisensory environments than others may come into play. The interplay between these modifiable yet confounding factors are of utmost importance in the decline or enhancement of specific cognitive domains following exercise.

In young adults, acute exercise to volitional exhaustion significantly improves hippocampal and prefrontal cortex functional performance, which lasts up to 2 hours after exercise cessation, demonstrating experience-dependent plasticity to exercise (650, 651). The assessment protocol involved a graded exercise test with an initial workload set at 75 W and then increased by 50 W increments every 3 minutes, up to 9 minutes. Beyond the 9-minute mark, the workload was increased by 25 W increments every minute to determine the maximal rate of oxygen consumption. Submaximal exercise lasting up to 60 minutes can improve cognition such that exercise protocols longer than 11 minutes can result in cognitive enhancements with the largest benefits in cognition seen with protocols longer than 20 minutes (514, 652, 653). However, some studies show contradictory findings that cognitive enhancements from acute exercise occur only within the first 15 minutes post-exercise (654). Cognitive-specific enhancements in working memory can last up to 30 minutes following acute aerobic exercise (655), making an appropriate experimental testing methodology crucial by including all relevant factors such as fatigue and its potential impact on short term cognitive outcomes. Critical

threshold of peripheral fatigue and sensory tolerance limit models regulate performance through neural feedback, tied to psychological and physiological factors with peripheral and central fatigue (656, 657). As peripheral locomotor muscular fatigue increases during exercise, central (motor) commands increase in compensation, highlighting the link between the two fatigue states via neural feedback mechanisms (126, 127). Research shows that moderate-intensity exercise does not alter muscle activation to the same extent as exhaustive high-intensity exercise which more recruits muscle fibres to counter force reduction from peripheral neuromuscular fatigue during dynamic exercise (658-660) , with energy depletion and metabolic by-products affecting power and muscle contractility. Inhibitory neural feedback from active muscles hinders performance, causing unfavourable sensations of pain and fatigue, and reduced exercise engagement due to an increasing workload (660). Exercise-induced psychological modulators are identifiably influenced by metabolic correlates of fatigue and physical performance, including protons, lactate, and ATP activating both the ASIC and purinergic P2X receptors, which in turn may lead to changes in cognitive performance.

The most common investigations of exercise in human participants are short in duration and look to investigate the impact on pre-frontal cortex-dependent functions in tasks related to executive function, attention, and reaction time (514). Most studies investigating the effects of acute exercise and cognition have focused predominantly on exercise of low-to-moderate intensity, 50-76% of heart rate max, to bring about small but significant improvements in cognitive performance. Two meta-analyses have shown acute exercise leads to a significant, small, positive effect on cognitive performance with the effect sizes ranging from 0.10 to 0.20 (514, 654). Even the timing of neurocognitive assessment in exercise-cognition studies is crucial given that those tests performed within 10 minutes directly following exercise performance pertained to negative effects, but with a delay of 11-20 minutes a large positive effect was observed (514). Any transient increases in cognition are likely brought about by coherence among different neuronal populations inciting enhancements in functional connectivity across connected regional networks mediated by distinct temporo-spatial properties. A single bout of moderate intensity exercise for 20-minutes at 70% of their age-predicted maximum heart rate has brought about intermediary changes in the functional connectivity patterns in the auditory resting state network, and the sensorimotor network (513). Exercise intensity-dependent modulatory effects exist for the right fronto-parietal and sensorimotor networks where high intensity exercise set at 20% above lactate threshold for 30 minutes brought about significant decreases in the post-central gyrus of the sensorimotor network, and the low intensity exercise performed at 35% of individual lactate threshold for 30 minutes induced significant increases in the

right fronto-parietal network (661). Findings such as these suggest potential benefits for sporting athletes in the performance of perceptual tests of cognition such as the SIFI. Whether exercise acts a confounding factor when taking the SIFI test if it is to be implemented as a concussion and brain health pitch side assessment tool remains unanswered.

Therefore, the aim of this study was to determine the acute effects of both moderate and high intensity exercise bouts on perceptual cognitive performance among a cohort of healthy university students. This would evaluate the feasibility of the SIFI to employed as an tool in the immediate sideline assessment and subsequent cognitive recovery for concussion.

5.3. Methods

No a-priori sample size calculations were conducted as this was an exploratory study which will capture the available sample. Written and informed consent was provided by all participants and ethical approval granted by Trinity College Dublin Faculty of Health Sciences (**Ref no.** 191204 & 221101; see **Appendix 1.1.** and **1.2.** for further details. Due to the impact of COVID-19 two separate ethical approvals were sought for this study protocol, although nothing differed other than the new experimenters listed. The experimental procedure was conducted in accordance with principles outlined in the Declaration of Helsinki (1964).

5.3.1. Participants

A total of 192 participants (Males = 88; Females = 104) included in the present chapter took part in the *follow up study* of **Study One – Protocol 1.2.** Due to the COVID-19 pandemic, there were 5 participants with missing SIFI data from the follow-up study and as such were removed from all relevant analysis for this chapter. A total of four SIFI assessments, pre and post moderate and high intensity exercise respectively, were included for analysis in this chapter. The initial baseline SIFI assessment was used for analysis in **Chapter Four** of **Study One**. For full details on participant characteristics, demographics, and recruitment see **Chapters Two and Four**.

5.3.2. Assessment Protocol

All participants engaged in both moderate and high intensity exercise on two separate occasions after a minimum of seven days following the initial laboratory familiarisation session. The two exercise sessions were separated by at least 48 hours and took place in the same testing environment. Participants undertook a high-intensity, interval anaerobic exercise protocol adapted from the work of Pearcey and colleagues (511) or a moderate-intensity, steady-state exercise protocol adapted from the work of MacInnis and colleagues (512). The high-intensity protocol of Pearcey and colleagues consisted of participants completed a series of 10 maximal 10-second sprints, with each sprint followed by 180 seconds of recovery. The session started with a 5-minute warm-up at 60–70 rpm and 50 watts workload. To accommodate the longer sprint duration, a 3-minute rest period was employed. Prior to each sprint, a 20-second warm-up phase at 50 watts and 100 rpm was performed, followed by verbal encouragement to exert maximum effort during the 10-second sprint. Participants were instructed to accelerate only after the initiation of the electromechanical brake to avoid overestimating power due to increased acceleration before brake engagement. The moderate-intensity protocol of MacInnis and colleagues involved a 5 min warm-up at 25 W where subjects were then instructed to cycle at the same cadence (∼80 rpm) throughout each session. Upon arrival at the lab for their second and third sessions, and once participants were satisfied, they completed the PAR-Q and Wellness questionnaire. This was followed by pre-exercise assessments consisting of a SIFI cognitive test, resting lactate measurement, and resting heart rate (HR). Immediately following each exercise bout all participants retook the SIFI test, had their lactate and HR measured, and provided their level of subjective fatigue by RPE. The anaerobic high intensity protocol consisted of a 3-minute warm-up on stationary wind-braked ergometric bike at 60-70 rpm and 10 x 10 second sprints at maximal volition interspersed with 180 seconds (3 minutes) of active rest/recovery. The moderate intensity protocol consisted of a 5-minute warm-up at 60-70 rpm with no resistance, 20-minutes of steady state continuous cycling at 60 rpm maintained at a HR of 70% of each participant's predicted maximal HR, followed by a 5-minute cool down with no resistance was performed at a cadence of 60 rpm. Blood lactate and HR were taken, and the SIFI test was performed after completion of both exercise bouts. For further information on the exercise protocols see **Chapter Two Section 2.3.5.** for full details on **Protocol 1.2.**

5.3.3. Statistical Analysis

Participant descriptive statistics were sub-categorised by sex and sporting type. A χ^2 test was used to establish proportions. Descriptive statistics of participant characteristics were reported as mean (SD) or median (IQR), as appropriate. One-way ANOVA or T-test where appropriate were used to assess for multiple comparison across groups. Two-way analyses of variance (ANOVA) and post-hoc multiple comparison tests were performed to enable adjustment for any possible Type-1 errors and investigate inter-group variations across multiple measures of HR, lactate, and RPE across both exercise sessions. For all analyses, $p < .05$ (two-tailed) was taken to be statistically significant. For all ANOVAs, Bonferroni's post hoc test was used when a significant main effect was measured to isolate differences in means. Effect sizes for pairwise comparisons were assessed using Cohen's d, calculated in the standard manner, and interpreted according to Cohen's scale (small effect: $0.2 < d < 0.5$, medium effect: $0.5 < d < 0.8$, and large effect: $d > 0.8$).

The coding of responses for the SIFI task was done in accordance with previous reports (99, 268). Temporal parameters of SIFI susceptibility were examined across all 64 trials representative of 16 conditions including the 6 unisensory conditions and 10 multisensory conditions consisting of 4 congruent and 6 incongruent illusory trials with six distinct SOA's (±70,150, 230ms) (96, 99, 102). The proportion of correct responses was calculated per condition (unisensory and multisensory) and SOA for each individual and used for all statistical analyses. With each trial repeated 4 times for each of the 16 conditions, the participants score of perceptual accuracy was wither 0, 0.25, 0.5, 0.75, or 1 and acted as dependent variables of analysis. The strata established for the SIFI performance in **Chapter Three and Four** wherein we analyse the agreement levels over the baseline, pre moderate and pre high intensity exercise, we now fix these strata assignments and examine whether there is agreement in SIFI results pre and post exercise. To reiterate, we now fix the assignment to strata shown in **Figure 3.8.** so that K and the c_i can be assumed fixed in the model. We look at the posterior of γ_k now measuring model agreement between $t = 1$ (pre-exercise) and $t = 2$ (post-exercise).

$$[y_{tij} \mid c_i = k, \boldsymbol{\theta}, \gamma_k = 0] \quad \sim \quad \text{Binomial}\,(r, \theta_{tkj})$$

$$[y_{tij} \mid c_i = k, \boldsymbol{\theta}, \gamma_k = 1] \quad \sim \quad \text{Binomial}\,(r, \theta_{kj})$$

Between-group differences were determined using mixed model ANOVA's to uncover main effects and two-way ANOVA's were subsequently run to determine the effects of each multisensory SOA condition across between-subject factors pre- and post-exercise. All data were exported from Microsoft Excel, for statistical analysis using the Statistical Package for the Social Sciences (SPSS version 25; IBM, Ireland). Figures were generated using GraphPad Prism (GraphPad Software version 8, San Diego, CA).

5.4. Results

5.4.1. Exercise intensity Metrics

5.4.1a Lactate

A paired samples T-test revealed no significant differences between pre-moderate and pre-high intensity exercise as expected (**Moderate**, **High** [mean (SD)]: 1.82 (0.66), 1.94 (0.75) mmol/L; t = -1.89, df = 185, p = 0.06, effect size = -1.39). Our findings revealed a significant distinction between the post-moderate and post-high-intensity exercise sessions. This outcome validates our chosen methodology and study design, confirming that the high-intensity exercise protocol indeed delivered the intended high-intensity challenge. (**Moderate**, **High** [mean (SD)]: 3.00 (1.58), 11.19 (4.00) mmol/L; t = -25.55, df = 182, p = 0.00, effect size = -1.89). From the post-high intensity data, sex was found to be a significant variable of interest where males had higher recorded lactate values (mean (SD): 11.92 (3.65) mmol/L) than females (mean (SD): 10.59 (4.19) mmol/L; df = 1, F = 5.06, p = 0.026, partial eta-squared = 0.027). No significant difference by sex was found post-moderate intensity exercise (p = 0.141); see **Figure 5.1.** and **Figure 5.4.** below.

5.4.1b Heart Rate

Similar to lactate, no significant differences were revealed from a paired samples T-test between pre-moderate and pre-high intensity exercise (**Moderate**, **High** [mean (SD)]: 78.86 (15.64), 80.93 (12.52) bpm; t = -1.96, df = 185, p = 0.051, effect size = -0.14). However, a significant difference was observed between post-moderate and post-high intensity exercise sessions in support of each protocol, both moderate and high, corresponding to their intended level of intensity (**Moderate**, **High** [mean (SD)]: 137.88 (14.30), 165.86 (13.42) bpm; t = -22.12, df = 183, p = 0.000, effect size = -1.63). Similar HR values which were not significantly different were recorded for males and females at post-

moderate (**Males**, **Females** [mean (SD)]: 135.78 (14.04), 139.75 (14.36), p = 0.072) and post-high (**Males**, **Females** [mean (SD)]: 164.72 (13.81), 166.79 (13.09), p = 0.299) intensity exercise sessions; see **Figure 5.1.** and **Figure 5.4.** below.

5.4.1c RPE

A significant mean difference of subjective fatigue was found post moderate and high intensity exercise in line with the lactate and HR results above indicating that fatigue scores were higher post high intensity versus moderate intensity exercise (**Moderate**, **High** [mean (SD)]: 4.65 (1.39), 7.19 (1.25); t = -19.292, df = 183, p = 0.000, effect size = -1.42). Sex was found to be a significant fixed factor of analysis post-moderate intensity exercise (p = 0.005) such that females reported higher RPE ratings (mean (SD): 4.90 (1.35)) than males (mean (SD): 4.33 (1.37)). No significant difference between males and females was found post-high intensity exercise (p = 0.576), see **Figure 5.2.** and **Figure 5.4.** below.

5.4.1d Fatigue Index, Performance Decrement Score, & Fatigue Metrics

As seen in **Figure 5.3.** all measures of physical performance including power output, time to peak power, anaerobic power and capacity, and maximal velocity were seen to decrease as the number of sprints increased during the high intensity exercise trial. No significant difference was observed between males and females by Fatigue index (**Males**, **Females** [mean (SD)]: 68.34 (18.89), 69.68 (18.57), p = 0.684) and Decrement Score (Mean (SD): 10.89 (5.82)) than males (Mean (SD): 9.29 (3.99), p = 0.08). Similarly, no differences by sex were found for fatigue and performance metrics of Anaerobic capacity (p = 0.479), Anaerobic power (p = 0.851), Fatigue slope (p = 0.761), and Rate of Fatigue (p = 0.545).

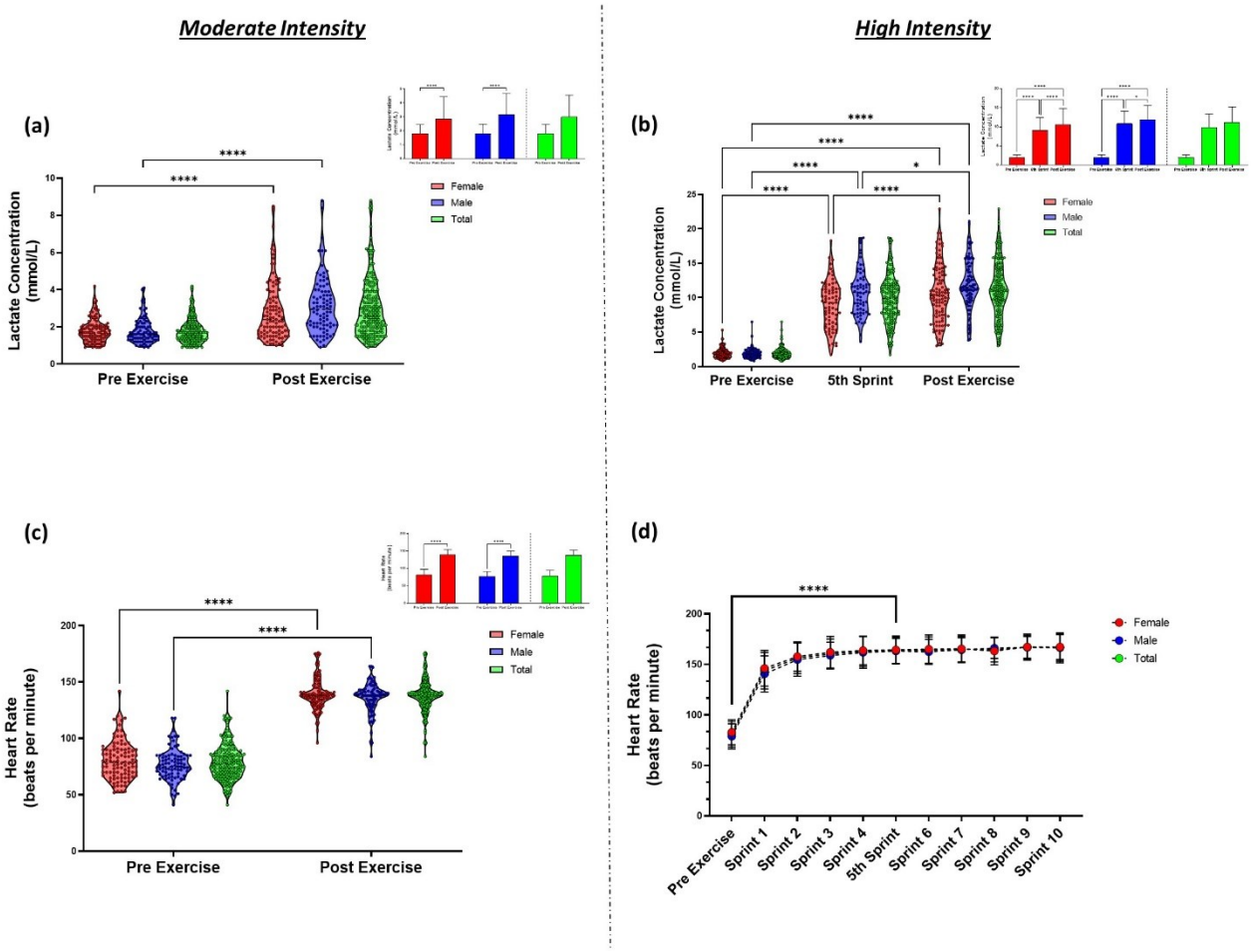

Figure 5.1. *Physiological Metrics of Fatigue and Exhaustion across Moderate and High Intensity Exercise Protocols.* **(a)** Lactate concentrations for moderate intensity exercise: a significant difference pre-to-post exercise was exhibited for both males and females ($p<0.0001$); **(b)** Lactate concentrations for high intensity exercise: significant differences for males and females were seen from pre-exercise to the 5th sprint and post exercise respectively. **(c)** HR values for moderate intensity exercise: a significant rise in HR was seen from pre-to-post exercise by sex ($p<0.0001$). **(d)** HR values for high intensity exercise: a significant increase in HR values were exhibited from pre-exercise to the 5th sprint by sex after which it plateaued around the age predicted max HR.

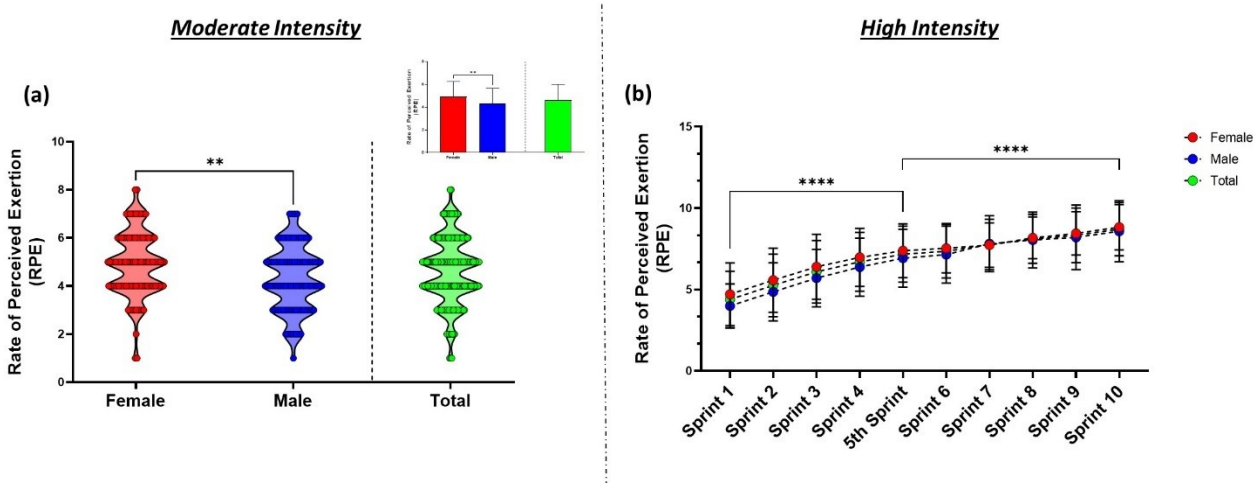

Figure 5.2. *Self-Perceived, Subjective Metric of Fatigue and Exhaustion across Moderate and High Intensity Exercise Protocols*. **(a)** Females reported significantly higher rates of RPE compared to males during the moderate intensity exercise ($p<0.01$). **(b)** Both males and females were found to exhibit a significant increase in RPE as it rose incrementally for the duration of the high intensity exercise protocol ($p<0.0001$).

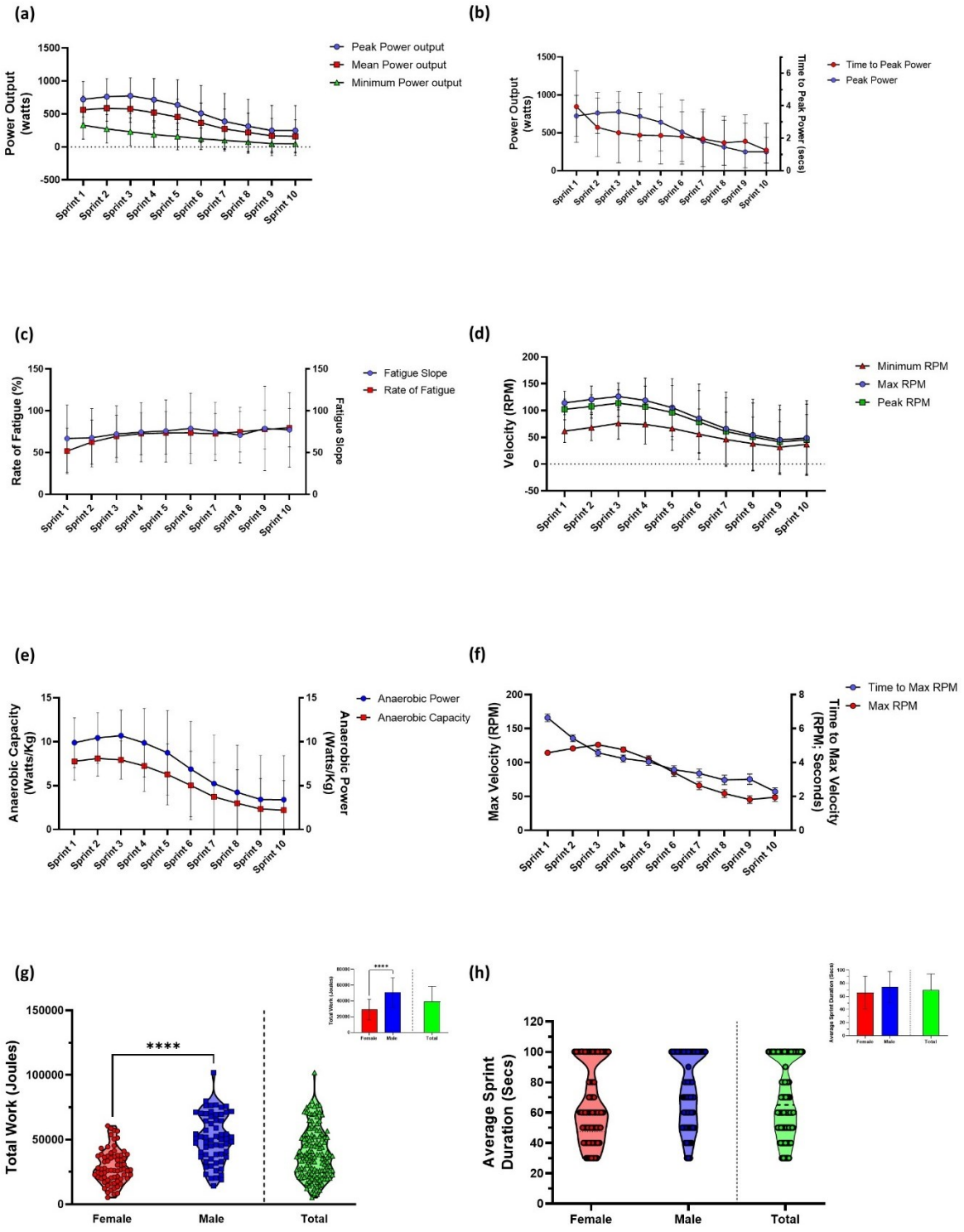

Figure 5.3. *Exercise Performance Metrics across the <u>High Intensity</u> Exercise Protocol*. **(a)** The peak, mean and minimum power output of the cohort was seen to decrease over the course of the high intensity exercise protocol indicative of central and peripheral fatigue. **(b)** Time to peak power and corresponding peak power output were seen to steadily decline over the protocol representative of fatigue over time. **(c)** The rate of fatigue was seen to moderately increase over the duration of the

trial. **(d)** Minimum, max, and peak RPM evidently decreased over the duration of the trial similar to that of power output highlighting the onset of fatigue. **(e)** Both anaerobic power and capacity were seen to drop extensively from the beginning of the high intensity protocol to the final sprint. **(f)** A coinciding decrease in time to max RPM and max RPM was evident over the course of the exercise bout. (g) Males performed a significantly larger volume of work compared to females ($p<0.0001$). **(h)** No significant differences were seen in the duration of the high intensity exercise bouts between males and females.

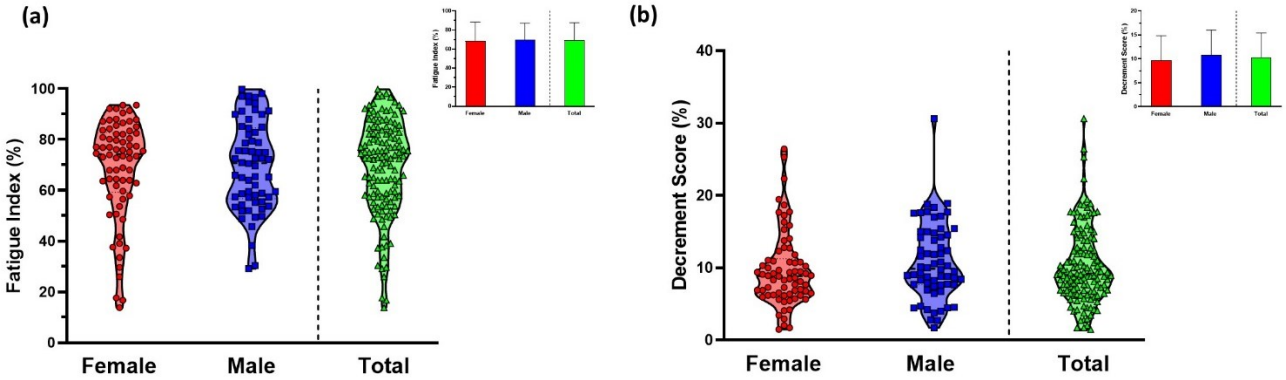

Figure 5.4. *Summary Score Metrics of Physiological Fatigue for <u>High Intensity</u> Exercise Protocol*. **(a, b)** Summary scores of fatigue and performance during the high intensity exercise protocol – No differences by sex were found, but the results represent a high rate of exercise induced fatigue representative of sporting demands.

5.4.2. SIFI Results

5.4.2.1. Moderate Intensity Effect of Exercise on MSI Performance (Pre vs Post)

The SIFI was administered to participants pre and post moderate exercise. **Figures 5.5. and 5.6.** show participants organised into strata over the two runs of the experiment. The agreement measure for pre and post moderate exercise is 0.8428 indicating a similar level of agreement to that seen in **Chapter Three**. Note that there is some rearrangement of strata here which is purely a plotting incidence. The results show that moderate intensity exercise did not impact perceptual performance accuracy during the SIFI.

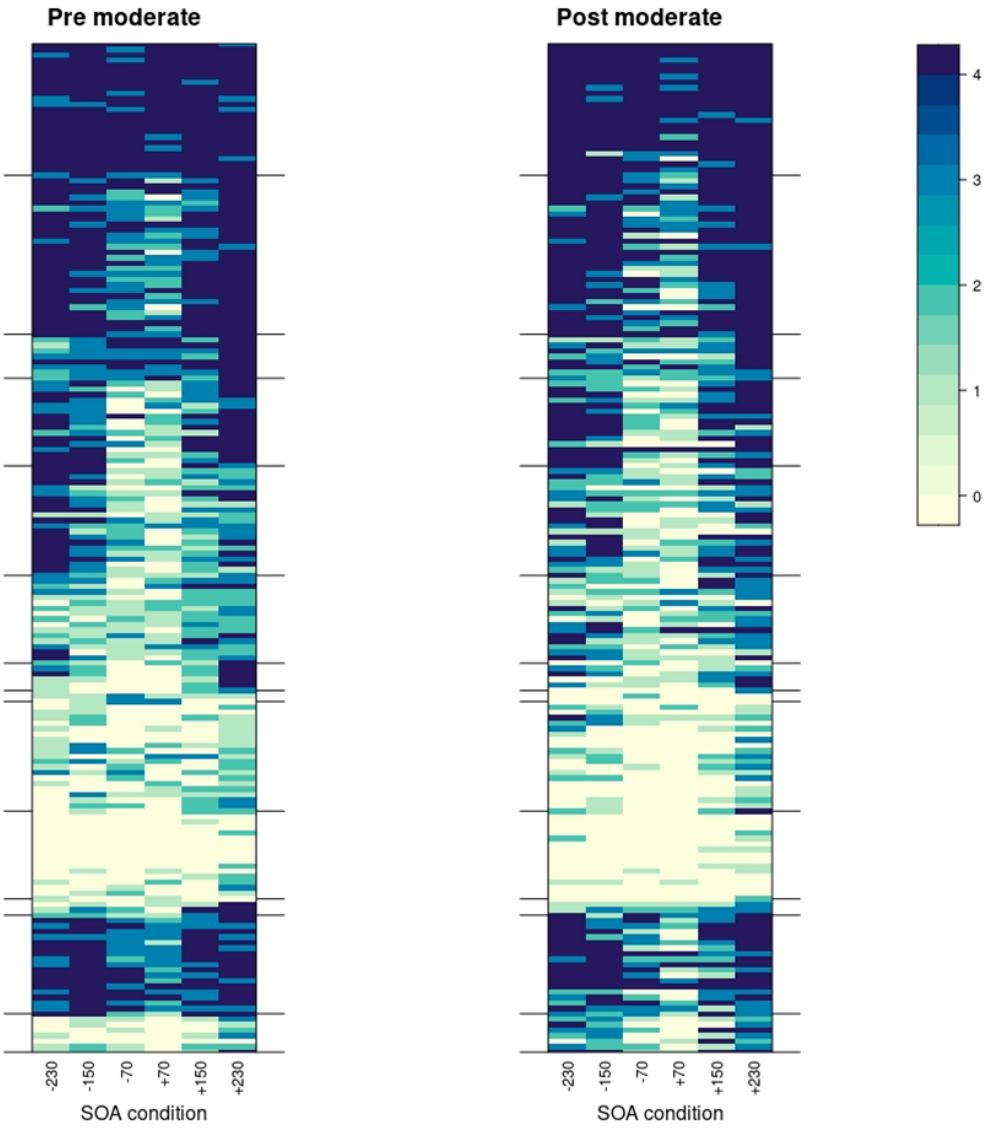

Figure 5.5. *Comparison of pre to post moderate exercise in same strata arrangement to Figure 3.8.*

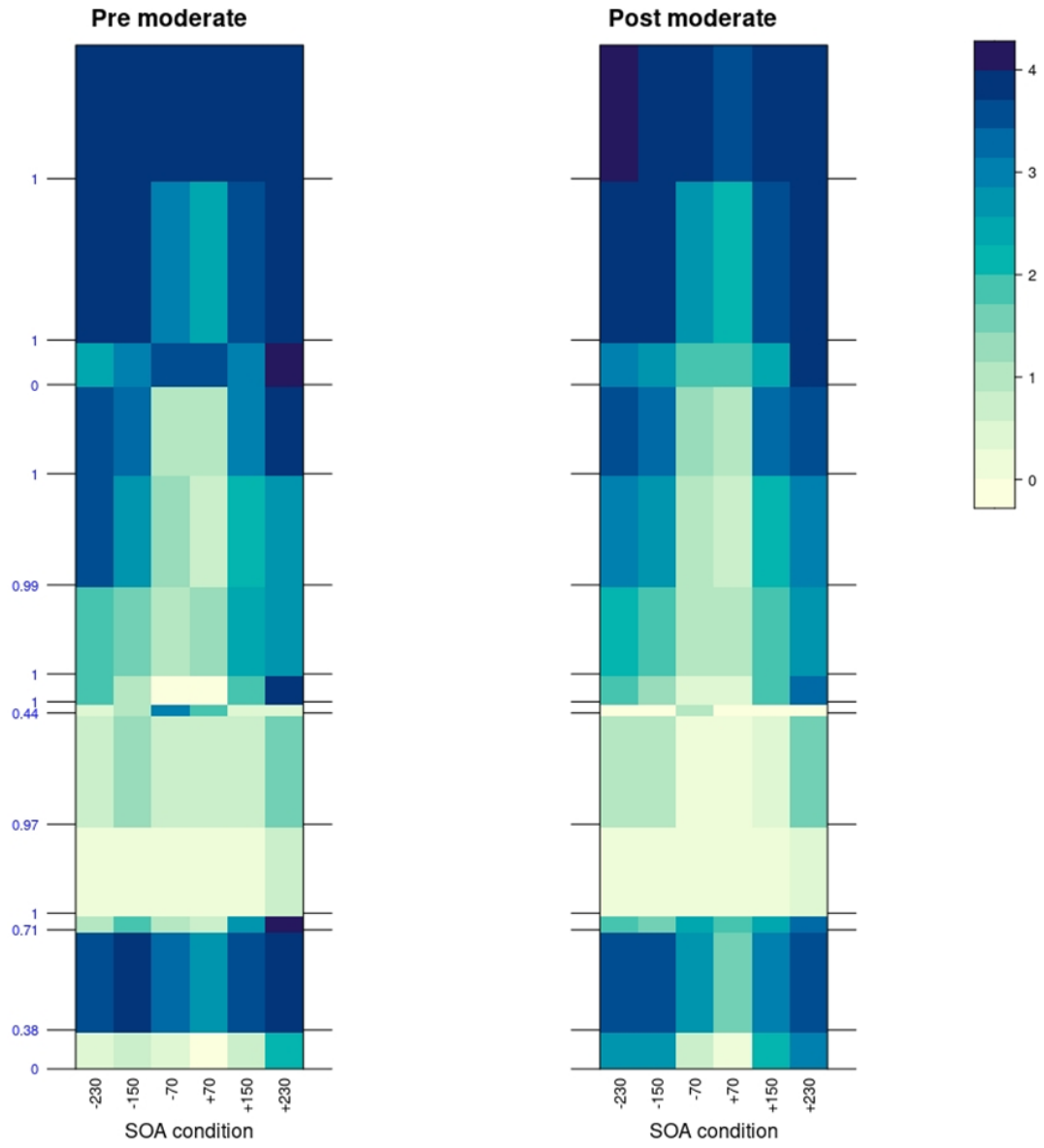

Figure 5.6. *Pre to post moderate summary plot with agreement scores indicated in blue text on left for each stratum.*

5.4.2.2. High Intensity Effect of Exercise on MSI Performance (Pre vs Post)

Figures 5.7. and 5.8. show participants organised into strata over the two runs of the experiment. The agreement measure for pre and post high intensity exercise is higher than the moderate intensity at 0.9928, still indicating that exercise at a higher intensity did not affect perceptual performance. The

higher measure of agreement could be indicative of a learning effect at the post high intensity session, wherein the "learners" (n=30) begin to perform more consistently following repeated exposure to the SIFI undertaking 5 tests over three separate testing sessions irrespective of exercise interventions.

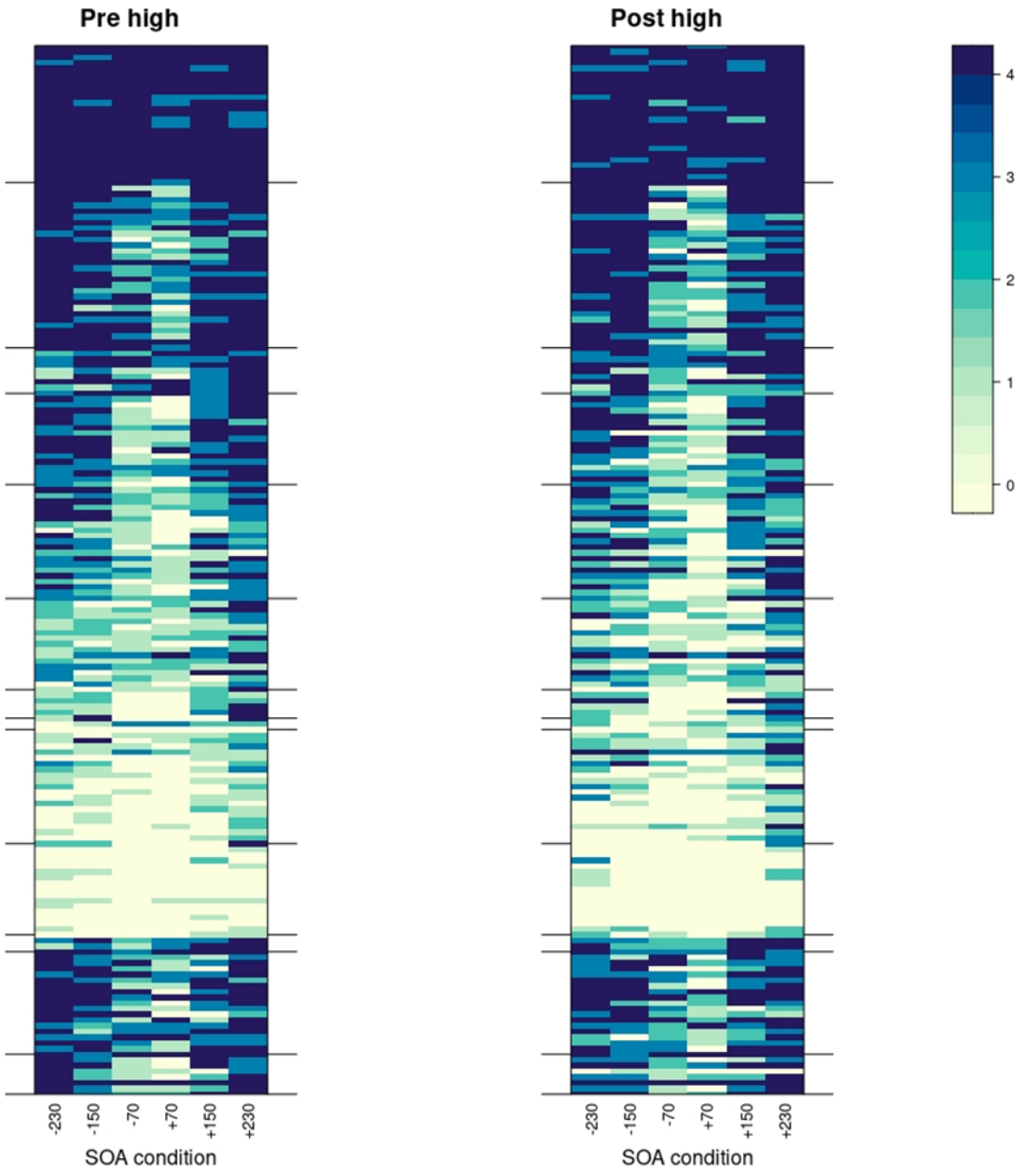

Figure 5.7. *Comparison of pre to post high intensity in same strata arrangement to Figure 3.8.*

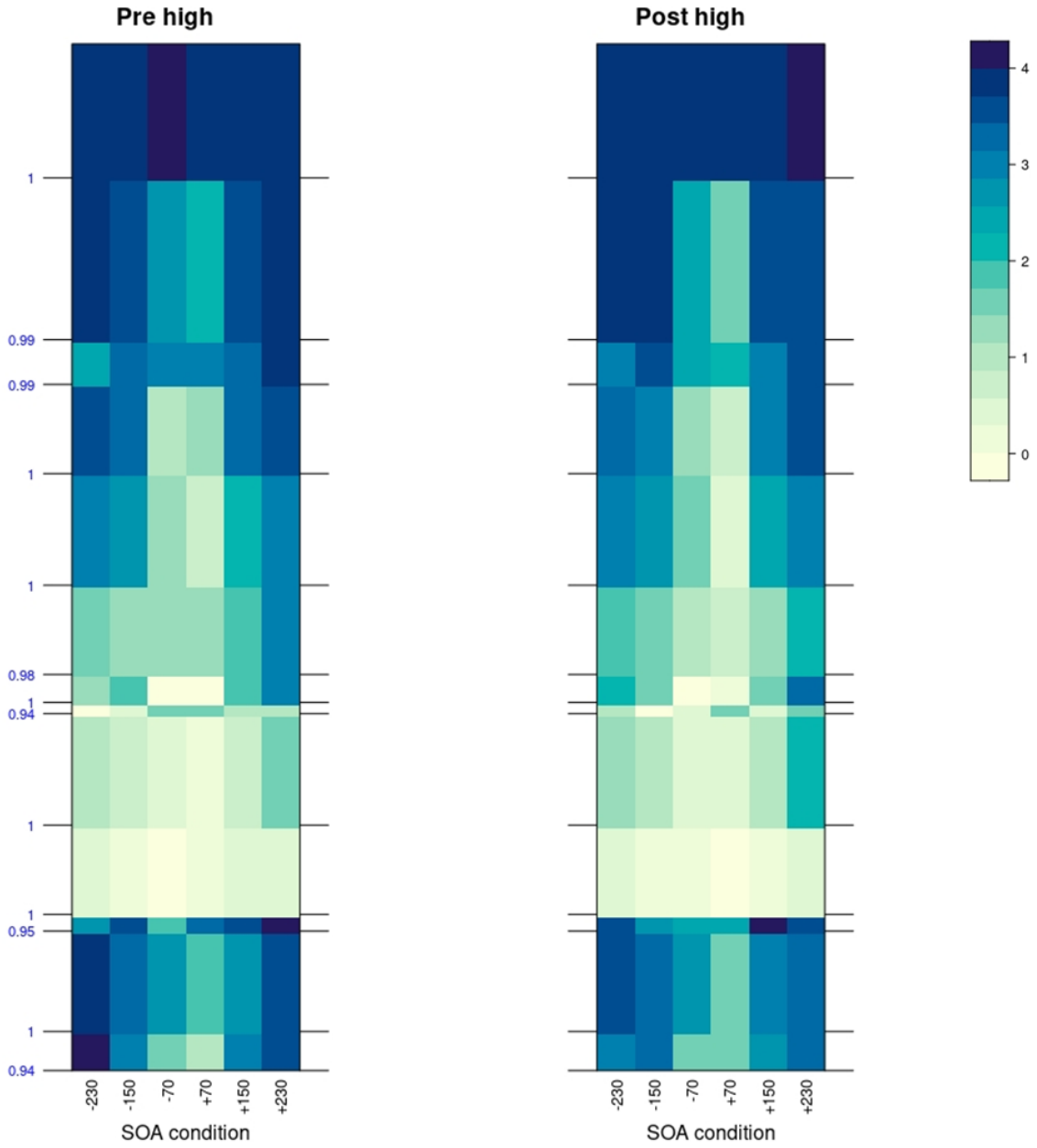

Figure 5.8. *Pre to post high intensity summary plot with agreement scores indicated in blue text on left for each stratum.*

5.4.2.3. Isolated Analysis of Pre and Post Exercise SIFI Trials

The rationale for the selected statistical methods above was rooted in the careful consideration of data stratification and label consistency. **Study One** involved the application of these statistical techniques across three distinct time points: baseline, pre-moderate, and pre-high-intensity exercise.

The decision to condition on the same labels for both the pre and post exercise trials was made to account for the complex interplay of various factors within the study providing stability by maintaining consistent labels across these time points, thereby simplifying the analysis process.

Given our current understanding from the results of **Chapter Four**, we acknowledge the interpretation of the strata's significance is not yet fully defined. As a result, a conscious choice was made to maintain uniform labels and baseline variables (such as age, sex, and sporting type) to avoid introducing additional complexity. This ensures that the categories used in the multinomial analysis remain unchanged, as they are directly mapped from the established labels. The decision to keep the labels and baseline variables constant between pre and post phases reflects a pragmatic approach that streamlines the analysis process, making it more manageable and reducing potential confounding variables. While a more nuanced exploration of each stratum's meaning could potentially lead to separate stratification for pre and post phases in the present chapter, this avenue is considered a separate and more time-intensive endeavour. Therefore, the present research aimed to provide meaningful insights using the current approach, accounting for the intricacies of the data while acknowledging the potential for deeper analysis in the future. Thus, an isolated analysis of the pre and post exercise trials at moderate and high intensity was also undertaken centred on the covariates of *concussion history, cohort, sex,* and *sporting type* in line with the previous analysis of the *pilot study* in **Chapter Four**.

Moderate Intensity Exercise

5.4.2.3a Concussion History

No identifiable differences were observed between those with and without a history of concussion across all fission illusory conditions at both pre and post moderate exercise (**Pre**: mean difference = 0.14, SE = 0.049; **Post**: mean difference = 0.008, SE = 0.05). Both pre and post exercise revealed a non-significant between subject effect across all six SOA conditions (**Pre**: df = 1, F = 0.086, p=0.770, $\eta^2 = 0.000$; **Post**: df = 1, F = 0.028, p=0.867, $\eta^2 = 0.000$); see **Figure 5.9.** below.

5.4.2.3b Cohort

Those in the contact cohort were seen to display higher levels of MSI across all temporal SOA's in line with the results of **Chapter Four**, and was a non-significant between subject effect for both pre

(df = 2, F = 0.759, p=0.470, η^2 = 0.008) and post (df = 2, F = 1.545, p=0.216, η^2 = 0.016) moderate exercise indicating no effect of exercise based on cohort; see **Figure 5.9.** below. However, looking at the SOA conditions in isolation, the contact cohort performed significantly better at the SOA+70 condition post-moderate intensity exercise in comparison to both the non-contact and control cohorts (both p<0.05).

5.4.2.3c Sex

Males were seen to have a higher level of performance accuracy across those SOA conditions with a larger temporal asynchrony, *i.e.,* SOA-/+230, -/+150, across both pre and post exercise (**Pre**: mean difference = 0.077, SE = 0.045; **Post**: mean difference = 0.118, SE = 0.045); see **Figure 5.9.** below. Although, a non-significant between-subjects effect of sex (df = 1, F = 2.978, p=0.086, η^2 = 0.015) was seen for the pre-moderate trial, a significant between-subjects effect of sex was found post exercise (df = 1, F = 6.827, p<0.01, η^2 = 0.035). More specifically, females had worse MSI capabilities at the longer temporal asynchrony of the SOA+230 condition than males post-moderate intensity exercise (p<0.05).

5.4.2.3d Sporting Type

Across both pre and post exercise, those in the open skill cohort were found to display higher levels of perceptual performance accuracy (**Pre**: mean = 0.675, SE = 0.04; **Post**: mean = 0.657, SE = 0.04) across a wide range of temporal asynchronies than both the closed skill (**Pre**: mean = 0.561, SE = 0.031; **Post**: mean = 0.542, SE = 0.032), and control groups (**Pre**: mean = 0.581, SE = 0.053; **Post**: mean = 0.548, SE = 0.054); see **Figure 5.9.** below. Although our analysis revealed a non-significant between subject effect (**Pre**: df = 2, F = 2.628, p=0.075, η^2 = 0.027; **Post**: df = 2, F = 2.726, p=0.068, η^2 = 0.028), those classified as participating in open skill sports performed better during the SOA+70 condition compared to the closed skill and control cohorts post-moderate intensity exercise (both p<0.05).

High Intensity Exercise

5.4.2.4a Concussion History

No differences between those with and without a history of concussion across all SOA's for pre and post high intensity exercise were seen (**Pre**: mean difference = 0.043, SE = 0.05; **Post**: mean difference = 0.044, SE = 0.049), with a non-significant between subject effect (**Pre**: df = 1, F = 0.746, p=0.389, η^2 = 0.004; **Post**: df = 1, F = 0.777, p=0.379, η^2 = 0.004); *see* ***Figure 5.10.*** *below.*

5.4.2.4b Cohort

Those in the contact cohort were seen to exhibit higher levels of MSI across all SOA conditions pre-exercise (**Pre**: mean = 0.671, SE = 0.049; **Post**: mean = 0.68, SE = 0.048) and appeared to be enhanced post exercise compared to the non-contact (**Pre**: mean = 0.592, SE = 0.031; **Post**: mean = 0.555, SE = 0.031) and control (**Pre**: mean = 0.563, SE = 0.044; **Post**: mean = 0.554, SE = 0.044) cohorts; see **Figure 5.10.** below. Although a mixed model ANOVA revealed a non-significant between-subjects effect of cohort (Pre: df = 2, F = 1.429, p=0.242, η^2 = 0.015; Post: df = 2, F = 2.634, p=0.074, η^2 = 0.028), those in the contact cohort performed with a higher level of MSI at the SOA-230 condition ($p<0.05$).

5.4.2.4c Sex

Males were seen to perform at a higher level of perception across a wide range of SOA's at both pre (mean difference = 0.104, SE = 0.045) and post (mean difference = 0.104, SE = 0.045) high intensity exercise. A mixed model ANOVA most notably revealed a significant between-subjects effect of sex (**Pre**: df = 1, F = 5.343, p=0.022, η^2 = 0.028; **Post**: df = 1, F = 5.372, p=0.022, η^2 = 0.028). The group-averaged proportion of correct illusory responses for males and females as a function of SOA highlight the increased susceptibility of females across all conditions, where males performed significantly better at the SOA+230 condition during the pre-high intensity exercise ($p<0.05$).

5.4.2.4d Sporting Type

Those who participate in open skill sports were found to exhibit higher levels of MSI capabilities across a wide range of temporal asynchronies, irrespective of whether the second auditory beep precedes or follows the A-V pairing pre (mean = 0.678, SE = 0.04) and post (mean = 0.658, SE = 0.04) high intensity exercise compared to the closed skill (**Pre**: mean = 0.568, SE = 0.032; **Post**: mean = 0.555, SE = 0.031) and control (**Pre**: mean = 0.563, SE = 0.053; **Post**: mean = 0.525, SE = 0.053) cohorts.

Contrary to our hypothesis and results in **Chapter Four**, a non-significant between-subjects effect of sporting type was found (**Pre**: df = 2, F = 2.630, p=0.075, η^2 = 0.028; **Post**: df = 2, F = 2.733, p=0.068, η^2 = 0.029). However, when looking at the SOA conditions in isolation, open skill participants appeared to perform better than the control and closed skill cohorts at the SOA-70 and SOA+150 conditions respectively during the pre-high intensity trial. Similarly, during the post-high intensity trial, open skill participants performed significantly better than the closed skill and control cohorts at the SOA-230 and SOA+150 conditions respectively.

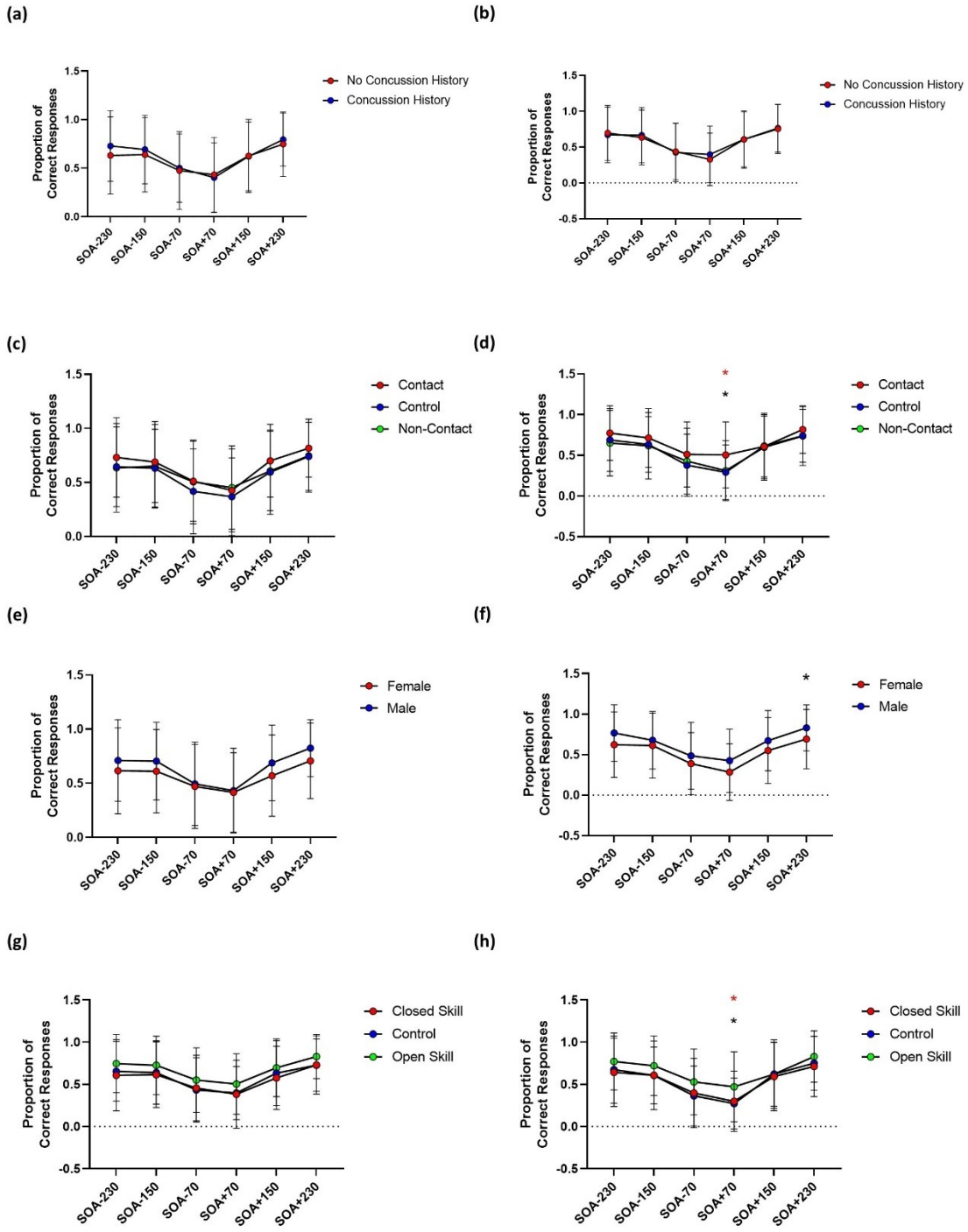

Figure 5.9. *Moderate Intensity Exercise SIFI Performance.* Pre- and Post exercise MSI accuracy for **(a & b)** concussion history, **(c & d)** sporting cohort, **(e & f)** sex, and **(g & h)** sporting type, respectively. The contact cohort had higher levels of perceptual performance that were statistically significant at the SOA+70 condition post-moderate intensity exercise (both $p<0.05$). Females had worse MSI capabilities at longer temporal asynchrony (SOA+230) condition than males post-moderate intensity exercise ($p<0.05$). Those in open skill sports performed significantly better at the SOA+70 condition compared to the closed skill and control cohorts post-moderate intensity exercise (both $p<0.05$).

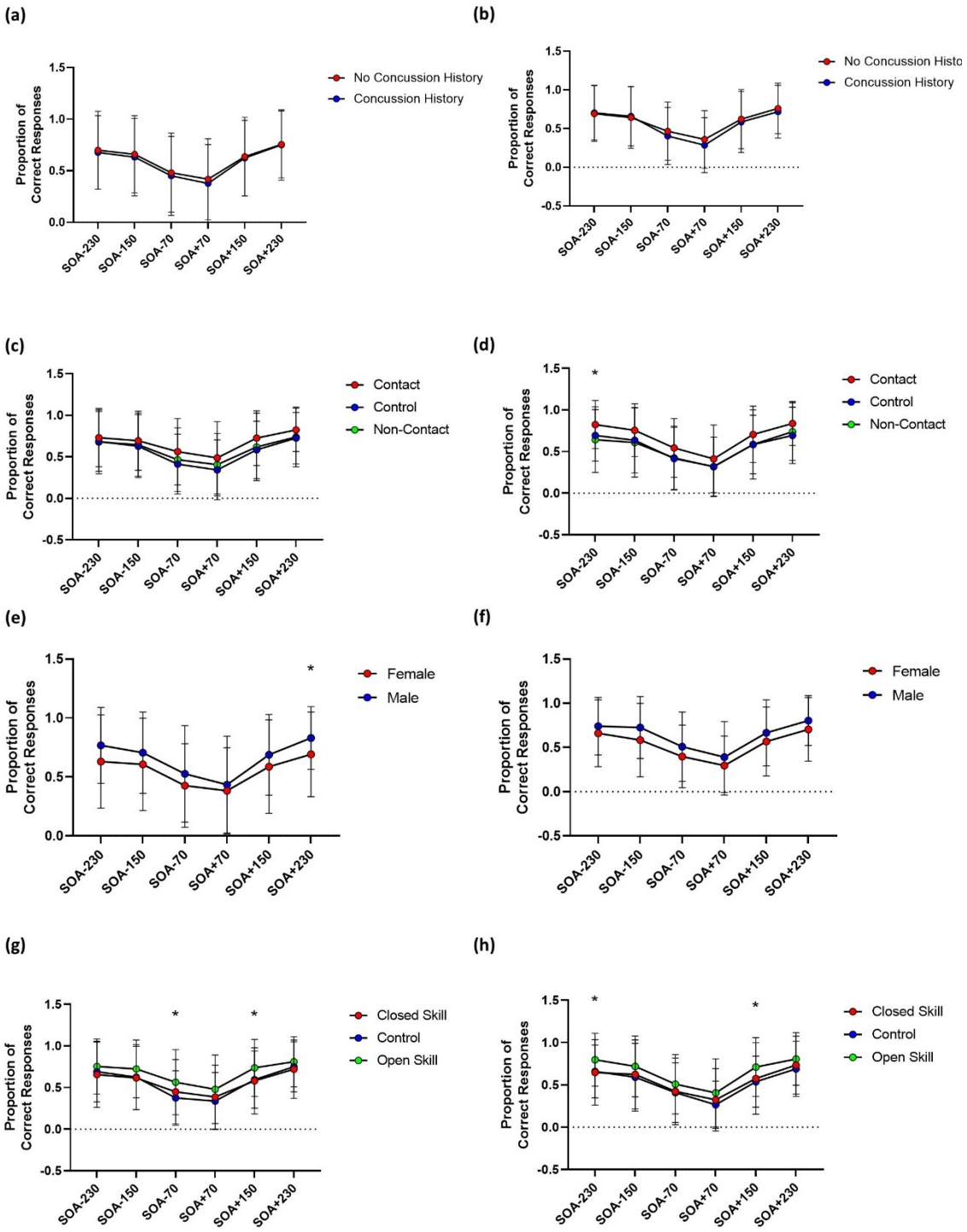

Figure 5.10. *High Intensity Exercise SIFI Performance.* Pre- and Post exercise MSI accuracy for **(a & b)** concussion history, **(c & d)** sporting cohort**, (e & f)** sex, and **(g & h)** sporting type, respectively. The contact cohort were seen to perform with a higher level of perceptual accuracy at the SOA-230 condition post-high intensity exercise ($p<0.05$). Males were found to perform significantly better at the SOA+230 condition compared to females pre-high intensity exercise ($p<0.05$). Pre-high intensity exercise, open skill participants performed significantly better than the control and closed skill cohorts

at the SOA-70 and SOA+150 conditions respectively (both $p<0.05$). Similarly, post-high intensity exercise open skill participants performed significantly better than the closed skill and control cohorts at the SOA-230 and SOA+150 conditions respectively (both $p<0.05$).

5.5. Discussion

This study aimed to investigate the effect of moderate and high intensity exercise on MSI assessed using the SIFI neurocognitive test. Our results show that the selected protocols did in fact replicate the appropriate physiological parameters of exercise intensity experienced during a sporting event. Significant increases in lactate, HR, RPE and fatigue index were exhibited to a greater extent during the high intensity protocol alongside parallel declines in power output, anaerobic capacity, and decrement scores distinguishing it from the lower levels of physiological stress and subjective fatigue experienced during the moderate intensity exercise. There was no difference in perceptual performance pre-to-post exercise for both moderate and high intensity, suggesting the SIFI is tolerant of exercise's impact on perceptual performance. The agreement measure for pre and post high intensity surpassed moderate intensity, suggesting a learning effect, with participants showing increased performance consistency as they became more familiar with the task since their first baseline assessment. In line with our findings in **Chapter Four**, females and those in closed skill sports were more susceptible to the illusion during both pre and post exercise conditions. Overall, our results indicate that the SIFI test is resistant to a performance decrement or enhancement in cognitive ability following exercise.

Those in open skill sports performed significantly better across conditions of shorter SOA's, which was not affected by exercise intensity. Task-related factors such as whether participants are instructed to detect or discriminate task-relevant stimuli, as well as whether they are visually or acoustically dominant, can create sensory bias. Professional drummers exhibit enhanced spatial understanding and more complex memory traces of temporal characteristics in auditory stimuli, which may be due to their musical proficiency (161, 162). Similarly, athletes with developed expertise may exhibit enhanced cognitive and sensory motor performance (166). Differences in sensory task performance could be mediated by the type of sport, with attention and anticipation playing a role in the dynamic interplay between perceptual cognitive processes and sensory dominance which was not seen in our results (167, 168). However, potential variance in sensory perceptual accuracy may be explained by the '*Dual-mode model*'. This theoretical framework proposes that during exercise below individual ventilatory threshold (VT), individuals are better able to sustain functional activation of the

pre-frontal cortex, which can override negative affective responses to incoming sensory information. Though, exercise above the VT can lead to a struggle of activation between executive function of the pre-frontal cortex and the subcortical regions associated with sensory perception (169). In line with this model, one would expect a difference in SIFI performance with VT more likely to be surpassed during the high intensity protocol. Despite this, neither moderate nor high intensity exercise herein was found to alter perceptual performance indicating alternative processes for the preservation of cognitive ability were at play.

Given the extensive variation regarding the index of exercise intensity and protocol timing in cognition-based research, it is difficult to make meaningful conclusions concerning cognitive performance following exercise. Exercise intensity frequently serves as a pivotal factor in acute exercise studies due to its significant relevance in elucidating underlying mechanisms. Notably, very light exercise (<50% HRmax) exhibited a substantial negative impact on cognitive performance, whereas all other intensity levels yielded positive effects significantly distinct from zero (514). Consideration of physiological factors such as heart rate, catecholamines, and BDNF underscores the significance of exercise intensity in determining alterations in these mechanisms, which, in turn, may be crucial in predicting behavioural outcomes, notably cognitive function. Additionally, the specific timing of cognitive test administration plays a vital role in understanding these effects, given the distinct impact of exercise on mechanisms, suggesting a potential dependence on the cognitive task's nature. Early studies utilised simple cognitive paradigms such as choice reaction time, whereas current studies focus on high-order executive functions (662, 663). Hockey's Compensatory Control Model explains the allocation of cognitive resources during perceptual tasks like the SIFI, regulating behavioural performance and cognitive executive functions through upper-level executive loops processing conscious information and lower-level loops processing non-conscious information (664). The lower-level loop monitors and regulates variations in learned motor skills and allocates greater cognitive resources to maintain task performance during a task in a stressful environment such as sport (664). Rerouting of energy resources and cerebral blood flow (CBF) to the activated brain regions may have conferred the so-called cognitive resistance we report for the SIFI. Explanatory theoretical constructs like the inverted-U hypothesis posit the correlation between performance and arousal, known as the Yerkes-Dodson Law, related to the effect of exercise on cognition (665). Information processing speed improvements are linearly correlated with exercise intensity, while multisensory task performance exhibits an inverted-U relationship with moderate intensity designated as 70% of the participant's 10 rep-maximum showing the largest improvement (666). Our results however

display no difference in MSI performance capabilities between moderate and high intensity exercise, but more importantly show a learning effect due to the cumulative exposure to the SIFI task which was evident by a small number of participants during their final SIFI trial, i.e., post high intensity. Moderate intensity (64–76% HRmax) exercise lasting between 10-45 mins has been found to positively impact cognitive function, but it is unclear which specific functions are improved (514, 667). One identified aspect of cognition affected is set shifting, which involves attention control and inhibition (668, 669). Acute bouts of moderate intensity exercise have been found to disproportionately improve executive processes (642, 670). Nonetheless, exercise has also been found to improve basic information processing (649, 667). These improvements are related to altered neural function and improved behavioural outcomes, such as shorter response times across flanker conditions deemed as being task-irrelevant, indicating improved inhibition and attention processes as a measure of immediate response conflict in response to presented stimuli (671). In contrast, our results indicate neither intensity nor task duration were affective variables in cognitive ability, and it may in fact be due to a compounding rate of task exposure over time creating a learned experience to bolster perceptual performance. A more detailed analysis of the functional neurocognitive mechanisms at play following exercise at varying intensities is required.

Our results show that exercise intensity did not impact perceptual performance. However, those participating in open skill sports performed significantly better at shorter A-V stimulus pairings, i.e., SOA+70 condition, post-moderate intensity exercise in line with the results of **Chapter Three**. Arousal plays a critical role in increasing cognitive uptake of available resources, as reflected by the P3 ERP complex amplitude (672, 673), diminishing the behavioural response to task-irrelevant stimuli (672). After moderate intensity aerobic exercise, a large P3 amplitude correlates with enhanced attentional processing (655, 674, 675). Optimal arousal for cognitive benefits is achieved through moderate intensity exercise for 20 minutes (649, 676). This level of exercise may lead to higher contingent negative variation compared to high intensity exercise, positively correlated with arousal and attention (677). The inverted-U hypothesis suggests that the decline in cognitive performance seen in closed skill and control cohorts, irrespective of exercise intensity, may result from competition for neural resources between muscle movement activation and the prefrontal cortex. (664, 678, 679). In a recent electrophysiological assessment, exercise intensity did not affect reaction time and target detection of incoming sensory information with no alterations in the P3a and P3b amplitude (680). Exercise-induced changes in sensory processing may therefore be mediated by mechanisms of sensory gain control, which can shift excitation towards the visual cortex and remaining sensory

cortices (680, 681). These changes in sensory processing and cognition are linked to glutamate and GABA cerebral release in the form of the *'excitation-inhibition balance'* (682, 683). The GABA and glutamate systems have been identified as factors contributing to individual differences in MSI (103), potentially linked to those with a history of concussion. However, our study aligns with previous evidence that concussion history does not affect SIFI performance (172). Despite the hypothesised metabolic cascade of concussion, physiological disturbances may persist beyond the clinical recovery period (27, 28). These findings have implications for perceptual performance after exercise with co-activation in the visual and anterior cingulate cortices (684, 685). Exercise-induced metabolic changes, particularly glucose and lactate uptake, may thus contribute to maintaining perceptual performance in the wake of exercise (686).

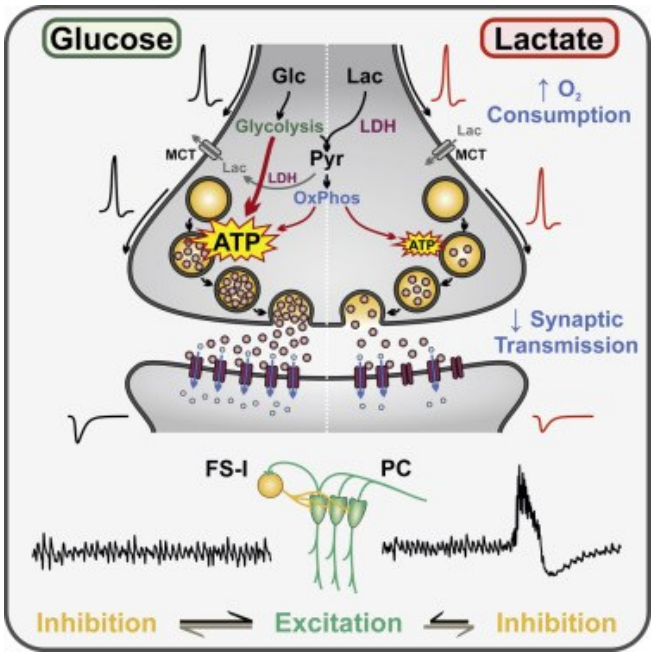

Figure 5.11. *Proposed mechanism for lactate-evoked attenuation of synaptic transmission.* **Left:** During glycolysis, glucose is converted to pyruvate, which rapidly supplies ATP for filling neurotransmitter vesicles. This ATP production is also supported by oxidative phosphorylation in the mitochondria. Some of the pyruvate is converted to lactate-by-lactate dehydrogenase, and the lactate exits the presynaptic terminal through monocarboxylate transporters along the concentration gradient. **Right**: Lactate is converted to pyruvate through lactate dehydrogenase (LDH), and pyruvate is subsequently metabolised by oxidative phosphorylation (OxPhos) in mitochondria to provide a slower supply of ATP. This slow ATP supply leads to reduced levels of neurotransmitters in presynaptic vesicles, resulting in decreased postsynaptic responses at both excitatory and inhibitory synapses when quick glycolytic ATP supply is absent. **Adapted from the work of* Hollnagel and colleagues (687).

Lactate, a key biomarker for neuronal metabolism and excitability, is released during exercise due to reduced oxygen availability (688). The accumulation of lactate depends on exercise intensity, duration, and individual fitness levels where lactate can become the preferred energy source for maintaining task performance. Trained individuals tend to have lower lactate levels due to differences in lactate production and removal (689-691). Lactate has been shown to acutely enhance cognitive function by supporting long-term potentiation, memory formation, and synaptic activity through central astrocytic support and exercise-induced peripheral supplies which may extend to later life if such exercise is chronically conducted as part of an active lifestyle (688, 692, 693). High-intensity exercise offers the best advantages in cognitive function with sufficient rest following exercise bouts (514), in line with the design of our study methodology. High-intensity interval exercise increases neuronal activity in the pre-frontal cortex, requiring high levels of energy consumption that can be met by lactate, leading to enhanced cognitive function including increases in inhibitory control (694-696). However, lactate levels exceeding 12 mmol/L can negatively affect attentional processes and cognitive abilities, such as working memory and visuomotor abilities; notably independent of biological sex (697, 698). The lactate levels recorded during moderate-intensity exercise in this study did not exceed 3.37 mmol/L. However, during high-intensity exercise, lactate levels approached the upper end of this threshold (mean = 11.1 mmol/L), potentially negating any cognitive performance enhancements. Astrocytic glycogen-derived lactate is a sufficient energy source for scaling of computational and high cognitive load, while less demanding cognitive paradigms rely on glucose (699). Factors such as sporting background, fitness levels, and genetics could contribute to these metabolic conditions affecting cognition, but the results suggest a potential association with biological sex.

Males and females have distinct physiological and metabolic characteristics affecting exercise performance (700, 701). The underlying mechanisms of exercise-induced fatigue responses in both sexes are however not fully understood (702, 703). Sex-based differences in fatigue characteristics are evident, with women often displaying distinct patterns compared to men. Notably, when performing submaximal fatiguing contractions at the same relative intensity, women tend to exhibit prolonged endurance before reaching task failure, in contrast to men. Moreover, women tend to experience less reduction in maximal force during sustained or intermittent maximal contractions which may be due to women maintaining a greater perfusion of blood flow to the working muscle during the contraction than men at the same relative intensity (701). Recent interest in this area has emphasised that the extent of sex differences in fatigue depends on the specific task at hand and that

these differences can evolve over one's lifespan. Males may experience higher fatigability over sprint trials attributed to factors such as body weight, BMI (700, 704, 705) and leading to a greater reliance on anaerobic metabolism and higher rates of fatigue. Males' higher muscle mass demands more oxygen consumption, but force production can constrain the supply of oxygen and nutrients through intramuscular vasculature (706). This mismatch creates an anaerobic dependency leading to higher rates of fatigue and ultimately increased lactate production (707, 708). Females, however, rely more on oxidative metabolism for ATP production, resulting in less metabolic lactate production (709). Our results showed higher lactate levels in males during high-intensity exercise, potentially impacting cognitive performance. Some studies report better cognitive enhancement in females following exercise than males, while others report worse cognitive performance in females after aerobic exercise (710-712). Our results support the latter whereby males were less susceptible to the illusory conditions at both moderate and high intensity. Subjective measures of fatigue following high-intensity exercise, particularly among individuals unacquainted with the specific exercise modality, regardless of fitness level, can lead to a faster onset of physiological fatigue. This rapid fatigue may result in reduced attention and arousal, which could explain the variations observed in perceptual performance, thus confirming the validity of our exercise protocol and performance outcomes.

One drawback of our present findings but of notable interest is that 103 participants were unable to complete the high intensity protocol with an average completion of 6.96 sprints leading to their voluntary cessation. Repeated high intensity exercise is a crucial determinant of performance in select sports. Fatigue during such exercise is caused by neuromuscular fatigue, which is characterised by exercise-induced reduction in a muscle's ability to generate force or power due to central and peripheral factors (713, 714). Central fatigue occurs toward the end of the repeat sprint protocol, leading to decreased voluntary activation (715, 716). While peripheral fatigue occurs early and lasts the duration of the repeated sprints, it leads to metabolic and ionic perturbations and decreased evoked twitch force of the knee extensors during cycling by 10-50% (511, 717). Fatigue induced by exercise involves both central and peripheral fatigue processes that interact through humoral and non-humoral mechanisms (658). The induction of central fatigue may result from increased afferent input from groups III and IV, which are sensitive to muscle contraction and intramuscular metabolic changes, respectively (658-660). Changes in power output metrics suggest the onset of central fatigue as seen in the present study is in line with previous research on repeat sprints (511, 657, 715). The activation of muscle groups that are not typically used during cycling (compared to field-based sports like running) may cause higher central motor drive limitations, which induced the necessary fatigue

replicating a sport-setting required for assessment of the SIFI (718). Our findings of physiological fatigue, both centrally and peripherally by objective and subjective assessment, provide credence in validating the chosen protocols for determining the effect of exercise on perceptual abilities and its utility in sport.

5.6. Limitations

Several limitations are evident in this study. Firstly, the cohort consisted of healthy young adults in higher education, which restricts the generalisability of the findings. Secondly, due to the exploratory nature, no sample size calculation was conducted, and a convenience sample was used, potentially introducing recruitment bias. Thirdly, the study did not assess energy pathways or neuromuscular function alterations, which could have provided a more comprehensive understanding of the cycle ergometer's task specificity for repeat sprint ability. Fourthly, the recovery time between sprints may have counteracted the onset of neuromuscular fatigue, as longer recovery periods have been shown to improve muscle buffering capacity and skeletal muscle function recovery (719, 720). For example, peak power was previously found to significantly decrease by 12.5 ± 2% when sprints were interspersed with 30 sec compared to 180 sec of recovery and decreased by 14 ± 2% and 20 ± 2% from sprint 1 to sprints 5 and 10, respectively (721). Despite this, subjective RPE ratings increased, and average peak power decreased over the completed sprints, indicating significant fatigue in all participants. Fifth, each participant undertook the moderate intensity exercise trial followed 48 hours later by the high intensity trial after completing their initial baseline assessment and familiarisation session, which was a total of 7-10 days between baseline and their final assessment. We found that cumulative exposure to the SIFI over the three testing sessions led to a learning effect among a small proportion of the study population. We acknowledge that randomisation for undertaking each exercise session at either moderate or high intensity would be preferable, but due to operational logistics and access to equipment it was necessary to complete the moderate intensity session prior to the high intensity. We acknowledge this as a limitation of the study **Protocol 1.2.** and future research is required to address this. Sixth, perturbations in the neuroendocrine system and neuromuscular changes were not assessed using sEMG, potentially affecting participants' RPE during high-intensity workload giving more insight into fatigue components. Moreover, the contribution of varying BDNF genotypes was not directly assessed, providing valuable insights into possible sex differences in the exercise-cognition relationship. Finally, the reduced data collection timeframe due

to the impact of COVID-19 highlights the need to assess the effect of sport-specific exercise on SIFI test performance. Finally, for a more accurate quantification of moderate and high intensity exercise it would have been most suitable to perform a standard (double-legged) ramp test to exhaustion on an electronically-braked cycle ergometer to determine whole-body peak oxygen uptake VO_{2peak}, peak power output, and lactate threhsolds. Expired gases would be analysed to determine from the greatest 30 s average of VO_{2peak}. A future study is therefore required with the following objectives:

- Determine if the use of a sports-specific exercise protocol will affect perceptual performance at designated time points in the acute stages following the intervention.
- Assess the validity of the sport-specific protocol to simulate the physiological and cognitive demands placed on the athlete during training and competitive match play.
- Determine the translational capacity of the SIFI from a lab setting to pitch-side clinical assessment of multisensory performance.
- Determine cerebral hemodynamics using fNIRS, which may shed light on the hypofrontality hypothesis and the protocol's impact on cognition.

5.7. Conclusion

Exercise is a known moderator of cognitive performance across multiple domains and is dependent upon methodological considerations, metabolic factors, and biological sex differences. We found that exercise intensity did not significantly affect perceptual performance assessed using the SIFI. However, females were more susceptible to the illusion both before and after exercise, and those in open skill sports exhibited higher levels of MSI post-exercise. From the present findings we can suggest that perceptual performance as measured by the SIFI test is not impacted following exercise making it potentially a suitable measure as an assessment tool for sports related concussion and brain health among athletes. Given the complexity of the exercise-cognition relationship, future research is warranted investigating the theoretical frameworks and neurobiological underpinnings on exercise-induced changes in sensory processing and cognitive performance.

5.8. Question(s) Raised

- How does a lifetime of sporting engagement affect perceptual performance and is there an influence of CV health on cognition with advancing age?

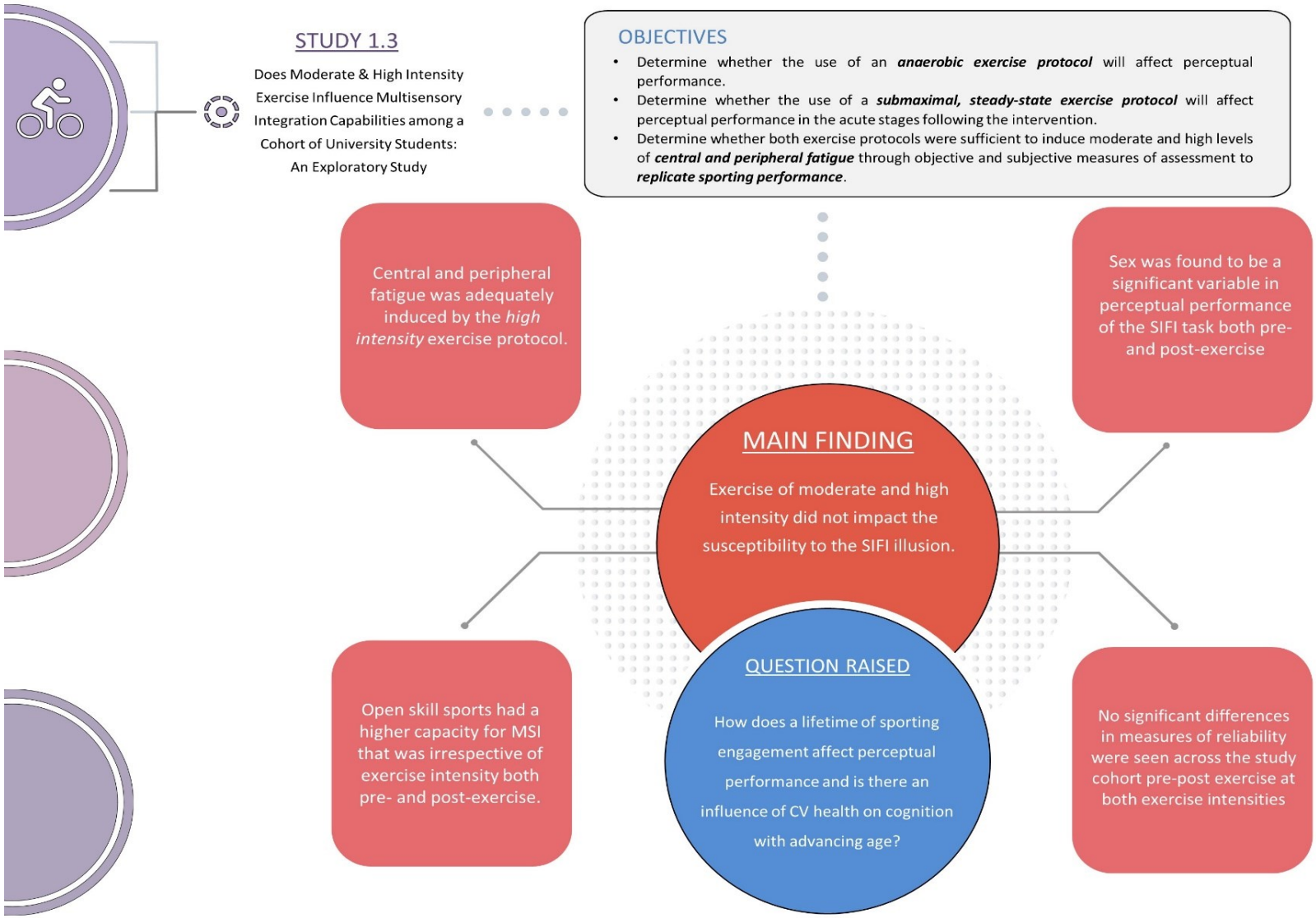

Figure 5.12. *Chapter Five – Study 1.3 Summary.*

6

Chapter Six:

Midlife Hypertension Negatively affects Select Domains of Cognitive Function at both Mid and Later Life: A Systematic Review and Meta-Analysis

6.1. Highlights

- 150 studies (67 individual study cohorts and 83 studies from 12 larger national cohorts) across 26 countries between 1992 and 2022 were included.
- 129,845 middle aged adults were included with a weighted mean age of 54.5 ± 3.9 yrs.
- Hypertension was most commonly defined using the ESC definition.
- Midlife hypertension negatively affected 3 select domains of cognition (memory, executive function, and global cognition) in later life.
- The same 3 cognitive domains were adversely impacted at midlife by hypertension.

6.2. Introduction

The worldwide prevalence of age-related cognitive decline is a major public health concern, especially in the context of an ageing population. Globally, the number of people living with dementia and cognitive impairment is expected to rise from 24.3 million in 2001 to 81.1 million in 2040, almost doubling every 20-years (722, 723). Current evidence from the Lancet Commission on dementia prevention, intervention, and care suggests that up to 40% of all dementia cases can be linked to modifiable risk factors (414). Identification of such risk factors and strategies to modify their negative influence on cognitive function therefore has the potential to protect and improve quality of life for a significant proportion of the global population, now and in the future.

Hypertension, which affects at least 1 billion people globally (724), has emerged as an important risk factor for cognitive deterioration and vascular dementia (430), and age of onset may impact on overall risk to brain health and function later in life (725). Specifically, there is evidence that hypertension during midlife could accelerate brain ageing (428, 430), potentially inducing premature cognitive decline via vascular and structural change (726). Interestingly, blood pressure (BP) exceeding optimal values even in the absence of a diagnosis of hypertension during young adulthood and midlife has been found to increase the risk of cognitive impairment in later life (379). Therefore, midlife may be the optimal time point for appropriate treatment and management of BP to mitigate the associated trajectory of cognitive decline with age. Cognitive function can be measured clinically and experimentally across several domains including but not limited to memory, attention, executive function, and global cognition. Different studies have assessed the effects of hypertension on one or more of these functions, yet there is no consensus on the impact of midlife hypertension on any of

these domains at mid- or later life; the systematic analysis and meta-analysis presented here aims to address this issue.

As the world's population over the age of 60-years is expected to double by 2050 (727), there is a growing need to investigate the association between midlife hypertension and cognitive decline, including any parallels in the time course of progression of each domain, to help inform public health policy. Though midlife hypertension has the potential to increase risk of later life cognitive decline, it is unclear at what point in the lifespan this decline begins and whether it is apparent during midlife. The purpose of this systematic review was to perform an analysis of the published evidence to explore the relationship between midlife hypertension status and cognitive function at both later life and at midlife, and to assess whether any negative impact was evident across different cognitive domains.

6.3. Methods

This review was conducted in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses guidelines (PRISMA; www.prisma-statement.org) and was recorded in PROSPERO, a registry of systematic reviews. Registration of this review can be found at https://www.crd.york.ac.uk/prospero/ (registration number: CRD42021238293). The present review is a subset analysis of the registered review.

6.3.1. Search strategy

Online electronic databases were searched, and relevant articles retrieved from the following: EMBASE, MEDLINE, PubMed, Web of Science and CINAHL, from their inception to May 2022. All search strategies were conducted by a medical librarian with methodological experience and the full search strategy can be found in the **Supplementary material**. The search strategy comprised of key words, MeSH terms, common medical terms, and a combination of these including, but not limited to, middle age, midlife, cardiovascular disease, cardiovascular risk, hypertension, high BP, cognition, and cognitive defect. The search strategy focused on the inclusion of longitudinal, prospective and follow-up studies to ensure later life cognition was captured. No search restrictions for language or publication date were implemented. The search of electronic databases was supplemented by a manual literature search of the reference lists of included studies and appropriate databases to

ensure all relevant studies were captured. The stepwise process of the search methodology can be seen in **Figure 6.1**.

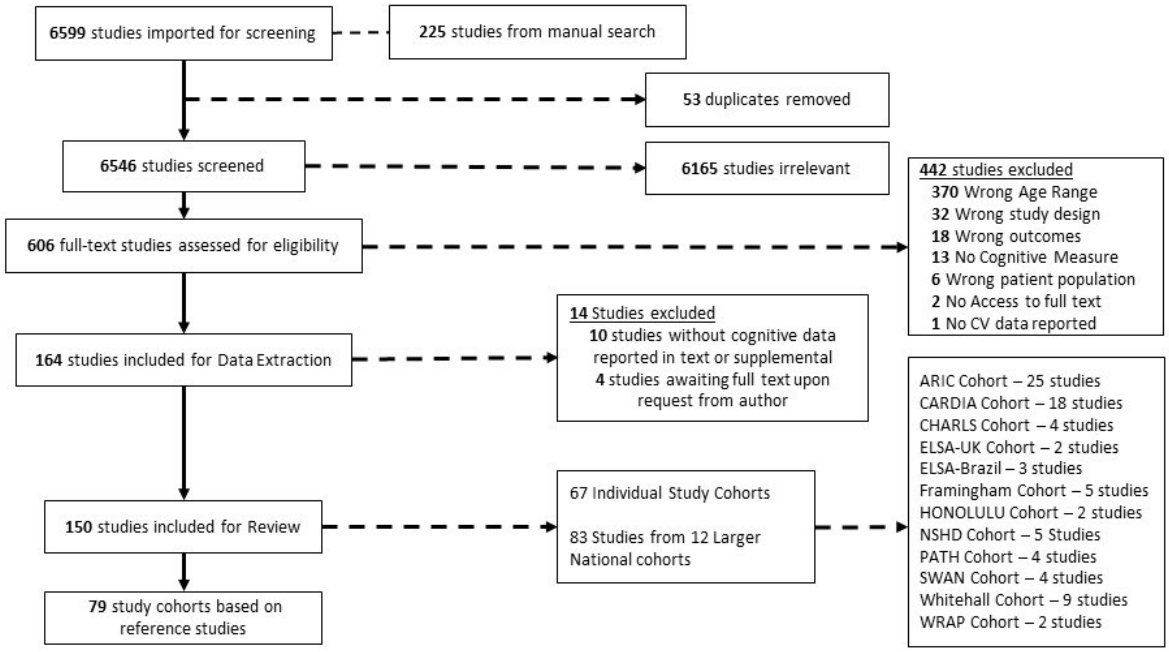

Figure 6.1. *Flow chart of the study selection process.*

6.3.2. Eligibility criteria

Studies were deemed eligible based on the following inclusion criteria: human participants, adults between ages of 40-65 years were classified as middle-aged [World Health Organisation (WHO) definition of middle age], hypertension and/or BP reported as an outcome measure at later life, midlife, or both for determination of the longitudinal association with midlife hypertension and cognition across domains including memory, attention, executive function, intelligence, and global cognitive functioning (see **Supplementary file**). Hypertension was considered an outcome of elevated BP where diagnosis by clinician, self-report, and/or by recorded BP metric in line with accepted definitions were considered eligible for inclusion and data analysis. For full details of the eligibility criteria please see **Chapter Two Section 2.5.2**.

6.3.3. Data extraction

Data extraction was carried out in accordance with the STROBE guidelines (546). To ensure accurate reporting the data extraction pro-forma was piloted against a selection of papers. All BP values reported are classified according to the European Society of Cardiology (ESC) classification in order to determine hypertension status (554). For full details of the data extraction process please see **Chapter Two Section 2.5.3**.

6.3.4. Risk of bias and methodological assessment

The methodological quality of included studies was evaluated using the Appraisal Tool for Cross sectional Studies (AXIS) (547). This tool employs 20 questions to determine quality of study design and risk of bias with questions being answered as 'Yes', 'No' or 'Unsure'. Study quality was then classified as either low, moderate, or high; see **Chapter Two Section 2.5.4.** for further details.

6.3.5. Statistical analysis

The weighted mean for demographics, cognitive measures (cognitive-specific domains and associated neuropsychological tests), systolic and diastolic BP values were calculated across studies to better understand the relationship with hypertension diagnosis. Weighted means were calculated using the following formula: $\sum^{n}_{i=1}$ (xi*wi)/$\sum^{n}_{i=1}$ wi; where ∑ denotes the sum, *w* denotes the weights, and *x* is the corresponding value (728).

$$\bar{x} = \frac{\sum_{i=1}^{n} (x_i * w_i)}{\sum_{i=1}^{n} w_i}$$

Cognitive outcome measures were grouped according to cognitive domain. Qualitative analysis assessed the relationship between midlife hypertension status and cognition at later life and midlife; positive, negative, or neutral, across studies.

A random effects meta-analysis was conducted to compare the difference across each cognitive domain between two independent groups, hypertension vs normotension. This meta-analysis was

deemed appropriate to calculate the pooled summary effect of midlife hypertension on cognition at midlife across the domains of memory, attention, executive function, and global cognition. For full details on the meta-analysis procedures please see **Chapter Two Section 2.5.5**.

6.4. Results

6.4.1. Literature Search

Figure 6.1 displays details of the study selection. The initial search and manual search yielded 6,824 records. Following the removal of duplicates and title and abstract screening, 606 full texts were screened, and 442 studies were excluded. The authors of four studies were contacted for access to full texts and were later recorded as 'studies awaiting classification' due to lack of response. All studies were imported in Endnote version 20 and an appropriate database was created from all extracted data in Microsoft Excel. Overall, 150 studies published between 1992 and 2022 were included.

6.4.2. Methodological and Risk of Bias Assessment

Of the 150 included studies, 35 were deemed low quality, 59 moderate quality and 56 high quality. Overall, studies were deemed of moderate-to-high quality with negative commonalities arising across several domains (*see* ***Supplementary Material***). The most common domains that were absent or unclear from studies included sample size justification (n= 128), categorisation of non-responders (n= 131), information about non-responders (n= 138), clear determination of statistical significance (n= 56) discussion of limitations (n= 22) and disclosure of ethical approval or consent (n= 21).

6.4.3. Characteristics of Included Studies

Of all included studies, 131 assessed males and females, 11 assessed males only and 8 assessed females only. Eighty-three studies assessed subsets of data from 12 prospective longitudinal cohorts (*see* ***Supplementary Material***). The remaining 67 studies assessed data from individual study cohorts. Studies were conducted across 26 countries with the top 5 including USA (n= 68), UK (n= 23), China (n= 7), Australia (n= 4) and Brazil (n= 4).

6.4.4. Participant Characteristics

129,845 participants were included and pooled for analysis. The weighted mean age of participants was 54.5 ± 3.9 yrs, weighted mean BMI was 27.2 ± 4.6 kg/m^2 and weighted mean height and weight were 171.4 ± 7.1 cm and 78.7 ± 14.6 kg, respectively. In studies that provided data according to sex (n= 56), 39,325 males and 40,678 females were included. Weighted mean BMI for males and females was 25.7 ± 3.4 and 24.5 ± 4.3 kg/m^2, respectively. Weighted mean age for males was 58.9 ± 1.8 and 56.7 ± 1.9 yrs for females. Mean height and weight were not available.

6.4.5. Blood pressure and Hypertension

The pooled weighted mean systolic and diastolic BP for all participants were 130.35 ± 12.3 and 80.8 ± 7.6 mmHg, respectively. Males had a higher systolic (128.2 ± 6.2 vs. 121.8 ± 8.2 mmHg) and diastolic BP (82.7 ± 0 vs. 77.4 ± 1.5 mmHg) compared with females. Hypertension was most commonly defined using the ESC definition (n= 30). Alternative definitions included American Heart Association (AHA) (n= 8), use of anti-hypertensive medication (n=12) and self-reported hypertension (n= 10). One study applied their own definition (systolic BP >150 mmHg or diastolic BP >95 mmHg), and seven studies did not provide a working definition (see **Table 6.2**). A total of 46,706 participants were classified as hypertensive, with 1,553 classified as pre-hypertensive; 3,968 were taking anti-hypertensive medication. A greater number of females were classified as hypertensive (8,423 vs. 7,516) and pre-hypertensive (108 vs. 0) compared with males. A higher number of males than females reported taking anti-hypertensive medication (951 vs. 800). A total of 18,931 participants were normotensive, with a higher proportion of females than males reporting normal BP (2,849 vs. 2,682).

6.4.6. Associations between Hypertension Status at Midlife and Measures of Cognition at Later Life

Of the 12 longitudinal study cohorts, 10 evaluated midlife hypertension and cognitive function at later life. A negative relationship was reported among domains including, memory (n= 8) (410, 729-735), executive function (n= 4) (410, 729, 734, 735), attention (n= 3) (729, 730, 736), global cognition (n= 5) (421, 730, 732, 734, 736), visuospatial organization (n= 1) (729), and psychomotor speed (n= 1) (730).

From the 67 independent study cohorts, 10 evaluated the relationship between midlife hypertension and later life cognition. Three studies reported negative relationships for memory and visuospatial organisation (737-739) and a further three studies also found a negative relationship for executive function, global cognition, and psychomotor speed (416, 740, 741). No relationship was found between hypertension and any measure of cognition in four studies (742-745).

Findings on the relationship between midlife hypertension and later life cognition did not differ by study quality. Longitudinal studies of moderate-to-high quality reported a negative relationship between midlife hypertension and later life cognition mainly in memory, executive function, and global cognition (see **Table 6.1**).

In summary, midlife hypertension was found to negatively impact on cognitive function across multiple domains at later life, irrespective of study design or quality.

6.4.7. Associations between Hypertension and Measures of Cognition at Midlife

Table 6.2 details mean pooled weighted outcomes for all measures of cognition and associated BP and hypertension values.

Conflicting findings were reported on the relationships between midlife hypertension and cognitive function at midlife (*see* ***Supplementary Material***). A similar number of studies reported no relationship or a negative relationship for cognitive domains, including attention, memory, inductive reasoning, and visuospatial organisation. Reports of no relationship were more common in the case of intelligence (n= 5, 83%), global cognition (n= 17, 74%) and executive function (n= 25, 75%). A negative relationship was more commonly reported for psychomotor speed (n= 5, 71%).

There were no discernible differences in reported relationships between midlife hypertension and midlife cognition based on study design (individual cohorts vs. large cohorts) or by study quality (low vs. moderate vs. high) (*see* ***Supplementary Material***).

6.4.8. Meta Analyses

All meta-analysis performed reflects the association between midlife hypertension diagnosis and midlife cognition. There was insufficient data available for meta-analyses including later life cognition. Fifteen studies across four cognitive domains (memory, executive function, attention, and global

cognition) were suitable for meta-analysis. A total of 12,919 participants were classified as hypertensive and 21,342 as normotensive. High levels of heterogeneity (I^2 ≥75%) were identified for all four cognitive domains. Hypertension diagnosis had a negative effect on memory compared to normotension [MD = -0.06; 95% CI = -0.20 to 0.08; I^2 = 0%] (see **Figure 6.2**). Hypertension diagnosis had no effect on attention compared to normotensives [MD = 0.41; 95% CI = 0.26 to 0.56; I^2 = 18%] (see **Figure 6.3**). Hypertension diagnosis had a negative effect on executive function, [MD = -0.02; 95% CI = -0.08 to 0.03; I^2 = 36%] (see **Figure 6.4**). Hypertension diagnosis negatively impacted global cognition compared to normotensive status, [MD = -0.24; 95% CI = -0.28 to -0.21; I^2 = 12%] (see **Figure 6.5**). Study quality or study design had no influence on meta-analyses findings for all four measures.

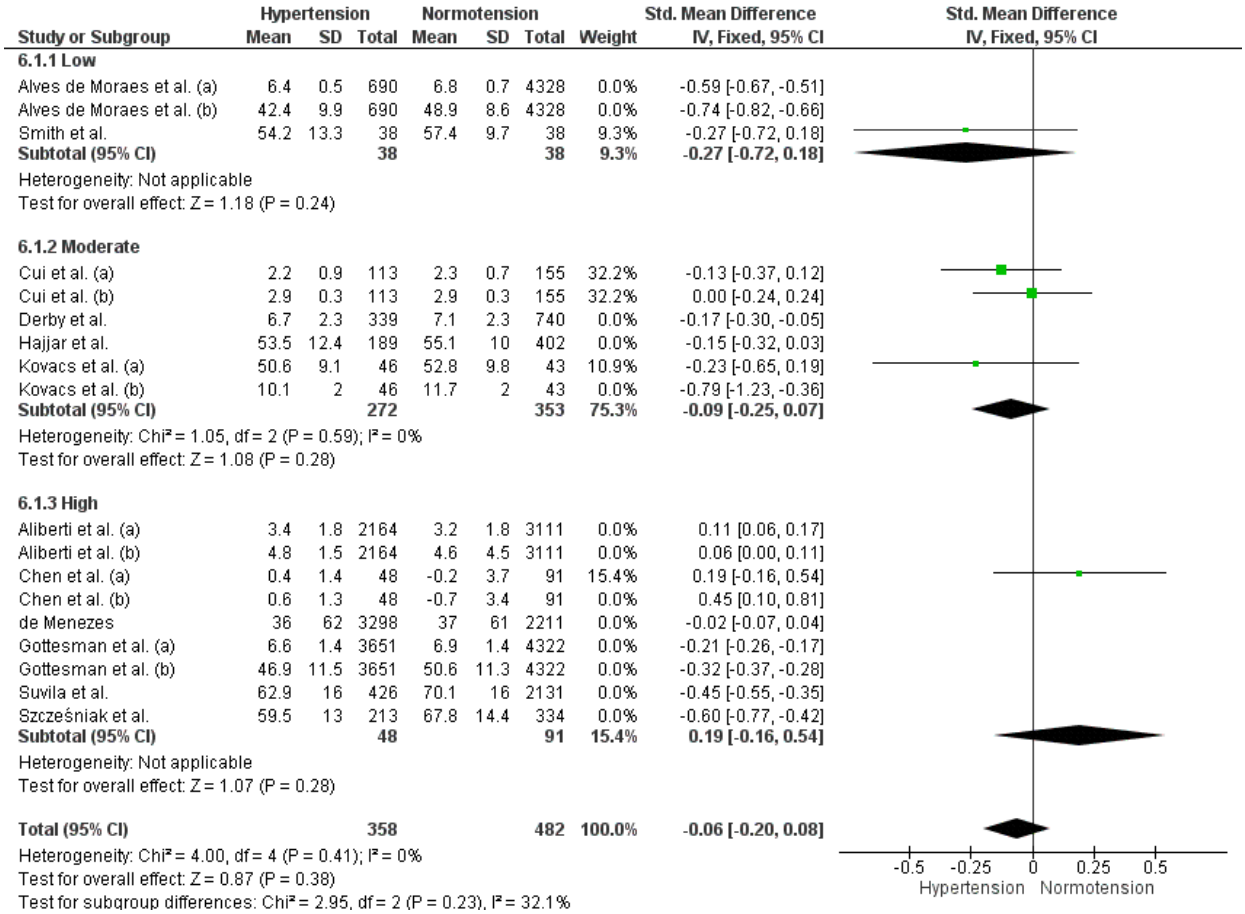

Figure 6.2. *Forest plot examining the overall effect of hypertension status versus normotension status on memory function.*

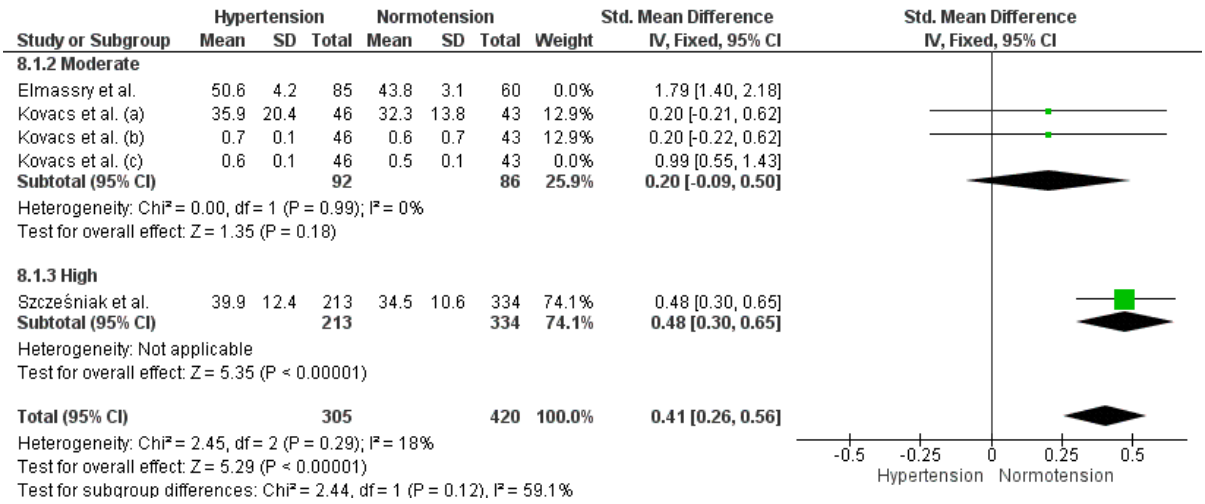

Figure 6.3. *Forest plot examining the overall effect of hypertension status versus normotension status on attention.*

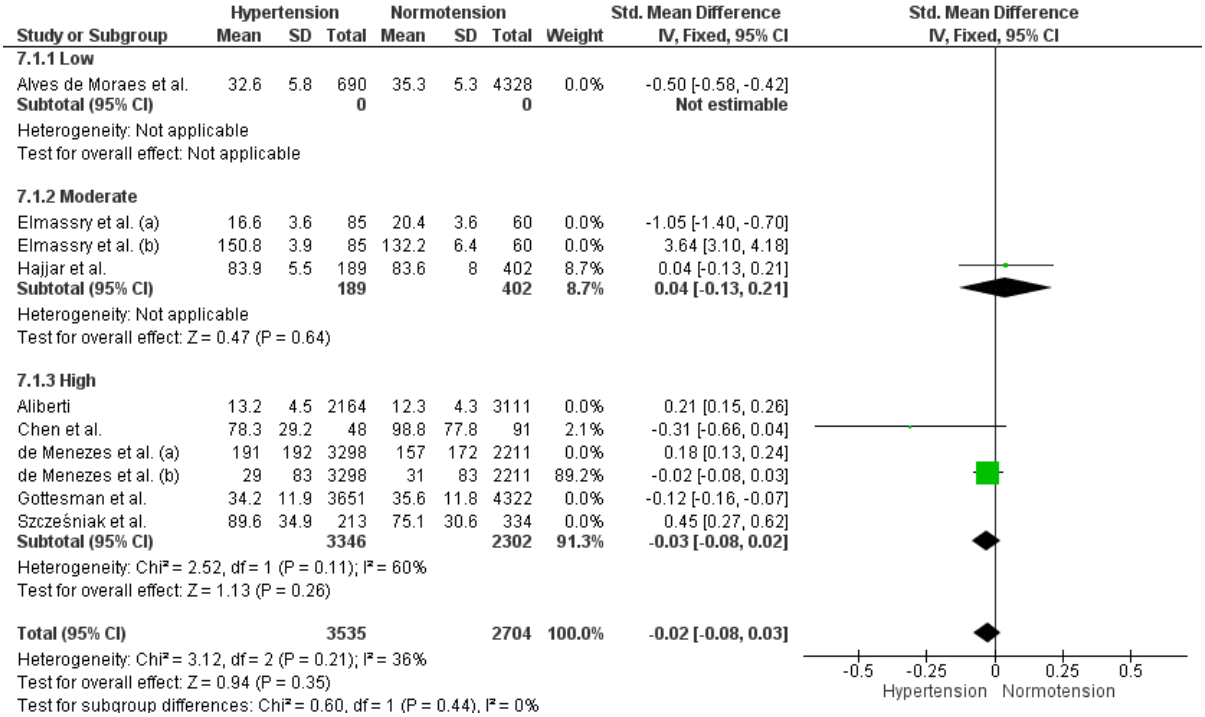

Figure 6.4. *Forest plot examining the overall effect of hypertension status versus normotension status on executive function*.

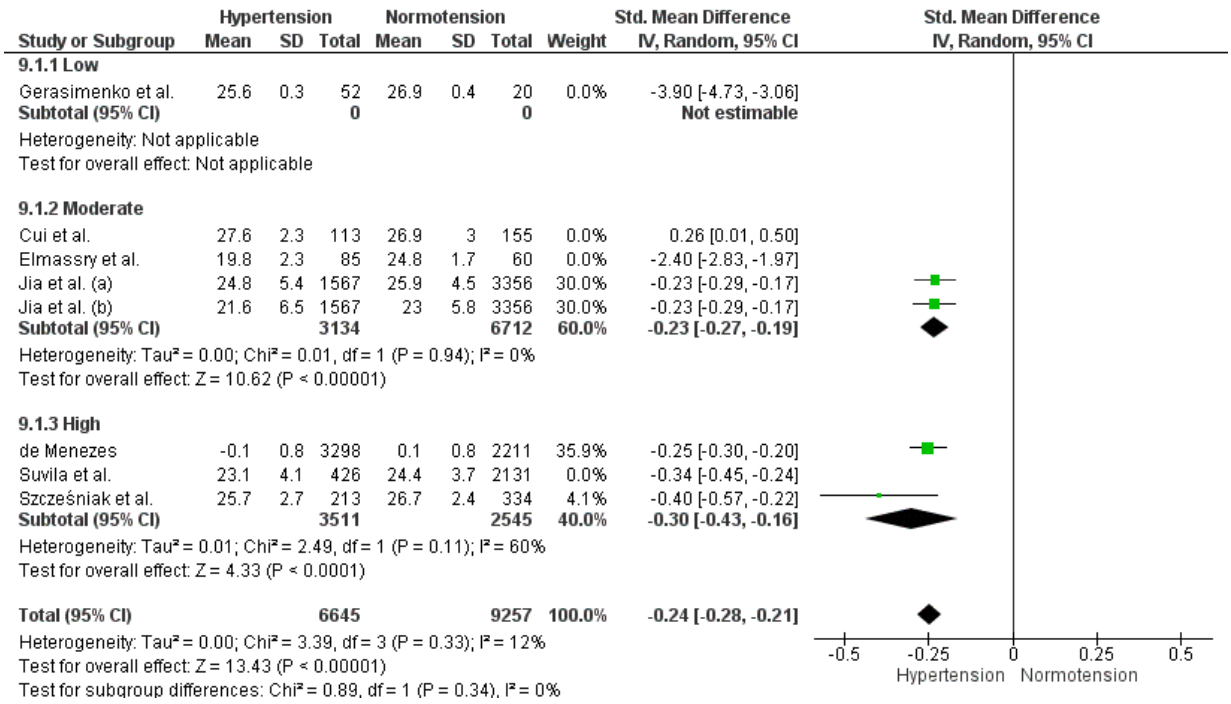

Figure 6.5. *Forest plot examining the overall effect of hypertension status versus normotension status on global cognition.*

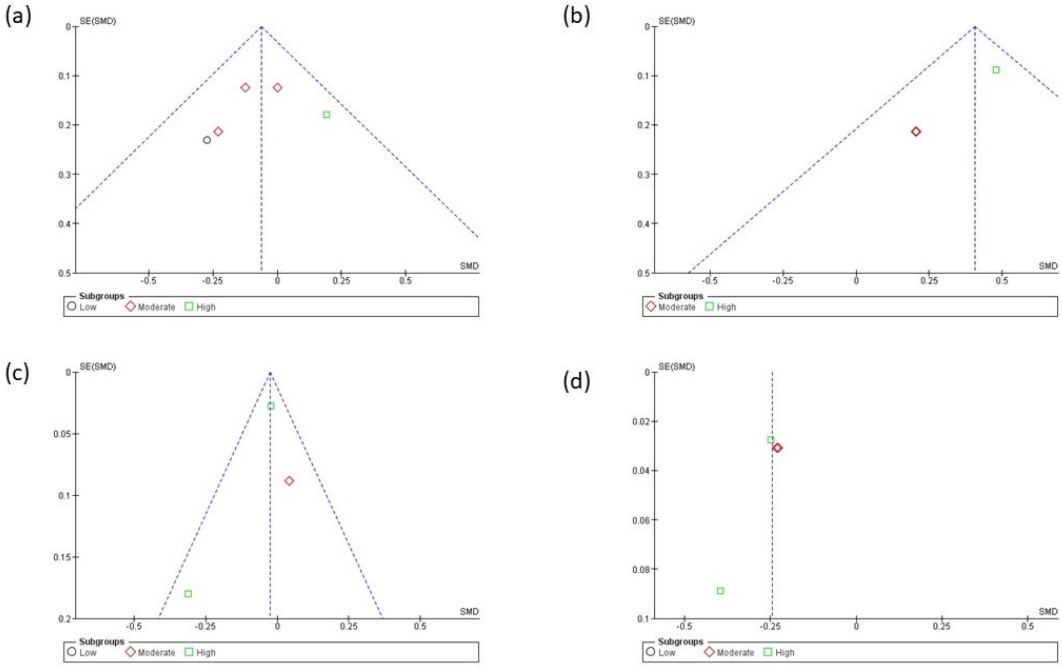

Figure 6.6. *Funnel plots representing hypertensives versus normotensives and their effect on cognition in midlife.* **(a)** memory; **(b)** attention; **(c)** executive function; **(d)** global cognition.

SMD = standardised mean difference; *SE* = standard error.

Table 6.1. Summary of longitudinal studies with negative or null relationship between hypertension and cognitive measures at later life.

Author	Year	Setting	Study Quality	Cognitive Variables	Relationship
Anstey et al.	2014	PATH through Life; Australia	High	Memory, attention, executive function, global cognition, and psychomotor speed	**- (Memory, attention, global cognition, psychomotor speed)**
Bangen et al.	2013	Framingham Study; USA	High	Memory, executive function, global cognition, and visuospatial organisation	**- (Executive function, attention, visuospatial organisation)**
Bayes-Marin et al.	2020	Edad con Salud; Spain	High	Memory	**- (Memory)**
Brunner et al.	2017	Whitehall II Study; UK	Low	Global Cognition	**- (Global Cognition)**
de Menezes et al.	2021	ELSA Study; Brazil	High	Memory, executive function, and global cognition	**- (Memory, executive function, global cognition)**
Derby et al.	2021	SWAN, USA	Moderate	Memory and executive function	**- (Women only: Memory and executive function)**
Dixon et al.	2021	SWAN, USA	Moderate	Memory and executive function	**- (Memory and executive function)**
Hajjar et al.	2016	USA	Moderate	Memory, executive function, attention, global cognition, and visuospatial organisation	**0**
Hoffmann et al.	2021	Recall Study; Germany	High	Memory, executive function, and visuospatial organization	**- (Memory)**
Kazlauskaite et al.	2020	SWAN; USA	Moderate	Memory and psychomotor speed	**- (Memory, executive function)**
Kesse-Guyot et al.	2015	SU.VI.MAX study; France	High	Memory, attention, executive function, and global cognition	**0**
Kivipelto et al.	2001	North Karelia Project and FINMONICA study; Finland	High	Memory, attention, executive function, and global cognition	**- (Global Cognition)**
Leong et al.	2020	TILDA; Ireland	Moderate	Attention, Global cognition	**- (Global cognition)**
Lin et al.	2020	KALS; Taiwan	High	Global cognition, memory, executive function, visuospatial orientation and attention	**0**
Lutski et al.	2019	BIP Neurocognitive Study; Israel	High	Memory, executive function, attention, global cognition and visuospatial organisation	**0**
Olaya et al.	2019	ELSA; UK	High	Memory	**- (Memory)**
Power et al.,	2017	ARIC Study; USA	High	Memory and executive function	**- (Memory, global cognition)**
Rouch et al.	2019	VISAT Cohort Study; France	Moderate	Memory, attention, executive function, global cognition, and psychomotor speed	**- (Global cognition)**
Swan et al.	1998	NHLBI Twin Study; USA	Moderate	Memory, executive function, global cognition, and psychomotor speed	**- (Global cognition, psychomotor speed)**

Swan et al.	1998	Western Collaborative Group Study, USA	Moderate	Memory, executive function, and psychomotor speed	**- (Global cognition)**
Szoeke et al.	2016	WHAP; Australia	Moderate	Memory	**- (Memory)**
Zhang et al.	2019	CHARLS; China	Low	Memory, executive function, and global cognition	**- (Memory, executive function)**

0, no association; -, negative association; +, positive association

Abbreviations: *ACE*, Akershus Cardiac Examination; *APAC,* Asymptomatic Polyvascular Abnormalities Community; *ARIC, Atherosclerosis Risk in Communities*; *ASCEND*, A Study of Cardiovascular Events in Diabetes; *Barcelona-AsIA*, Asymptomatic Intracranial Atherosclerosis; *BHS*, Bogalusa Heart Study; *BIP,* Bezafibrate Infarction Prevention; *BP,* blood pressure; *CARDIA,* Coronary Artery Risk Development in Young Adults; *CHARLS*, China Health and Retirement Longitudinal Study; *DBP*, diastolic blood pressure; *ELSA*, English Longitudinal Study of Ageing; ELSA, Brazilian Longitudinal Study of Adult Health, *FINMONICA*, Finnish Multinational Monitoring of Trends and Determinants in Cardiovascular Disease; *HAALSI*, Health and Aging in Africa; *HANDLS, healthy Aging in Neighborhoods of Diversity Across the Life Span*; *HHP*, Honolulu Heart Program; *IHDB*, Institute of Human Development in Berkeley; *KALS,* Kaohsiung Atherosclerosis Longitudinal Study ; *KEEPSCog*, Kronos Early Estrogen Prevention cognitive; *KIHD,* Kuopio Ischaemic Heart Disease Risk Factor Study; *MACS,* Multicentre AIDS Cohort Study; *MADT*, Middle-Aged Danish Twins; *MDCS,* Malmö Diet and Cancer Study; *MORGEN*, Monitoring Project on Cardiovascular Disease Risk Factors; *MRC,* Medical Research Council; *NHLBI*, National Heart, Lung, and Blood Institute; *NSHD,* National Survey of Health and Development; *PATH,* Population Assessment of Tobacco and Health*; PURE,* prospective Urban and Rural Epidemiological; *RECALL*, Risk Factors, Evaluation of Coronary Calcium and Lifestyle; *SBP*, systolic blood pressure; *Swan,* Study of Women's Health Across the Nation; TILDA, The Irish Longitudinal Study on Ageing; *VETSA,* Vietnam Era Twin Study of Aging; VISAT, Vieillissement Santé Travail (Aging, Health and Work); *WHAP,* Women's Health Aging Project

Table 6.2. Summary Table of pooled weighted average for all cognitive measures and associated BP metrics at baseline (i.e., midlife).

Cognitive Variable	No. of Studies	Weighted Average (Mean ±SD)	Age (Mean ±SD; Years)	Systolic BP (Mean ±SD; mm Hg)	Systolic BP Category Status (ESC)	Diastolic BP (Mean ±SD; mm Hg)	Diastolic BP Category Status (ESC)
Memory *Verbal Memory*	*Total*: n= 21 ***Immediate***: n= 3, ***Delayed***: n= 11, ***STW***: n= 1, ***EBM***: n= 2, ***RAVLT (Immediate & Delayed recall, Learning & Summary Score):*** n= 2, ***SRT*:** n= 1, ***ROCF (Immediate & Delayed):*** n= 1, ***CERAD (Immediate & Delayed):*** n= 1, ***CVLT (Immediate & Delayed):*** n= 1 ***WLL***: n= 1	***Immediate:*** Total = 9.9, males = 5.7 ± 1.1, females = 4.6 ± 1.5 ***Delayed:*** Total = 6.2 ± 1.5 males = 9.9 ± 2.9, females = 12.7 ± 3.7 ***STW:*** Total = 50.5 ***EBM***: Total = 10, Female = 10.2 ***RAVLT (Immediate & Delayed recall):*** Total = 7.1 ± 2.7 ***RAVLT (Learning Score):*** Total = 36.8 ± 8.3 ***RAVLT (Summary Score):*** Total = 8.8 ± 3.2 ***SRT:*** Total = 34.3 ***ROCF (Immediate):*** Total = 16.1 ± 7.6 ***ROCF (Delayed):*** Total = 14.9 ± 7.8 ***CERAD (Immediate):*** Total = 7.2 ± 1.1 ***CERAD (Delayed):*** Total = 7.7 ± 1.5 ***CVLT (Immediate):*** Total = 8.8 ± 2.1 ***CVLT (Delayed):*** Total = 8.8 ± 3.2 ***WLL***: Total = 20.5 ± 0.5	Total = 52.7 ± 4.8 males = 53.2 ± 4.9, females = 52.4 ± 4.5	Total = 124.2 ± 16.8, males = 123.7 ± 15.6, females = 120.9 ± 16.8 ***Delayed:*** Total = 121.6 ± 17.1, males = 123.7 ± 15.6, females: 120.9 ± 16.8 ***Immediate:*** - ***STW:*** - ***EBM:*** - ***RAVLT (Immediate & Delayed recall, Learning Score):*** Total = 131.2 ± 16.1 ***SRT:*** Total = 134.1 ± 16.9 ***ROCF (Immediate & Delayed):*** Total = 131.2 ± 16.1 ***CERAD (Immediate & Delayed):*** Total = 127.9 ± 20.7 ***CVLT (Immediate & Delayed):*** Total = 127.9 ± 20.7 ***WLL***: Total = 123.7 ± 17.6	Total = Normal, males = Normal, females = Normal ***Delayed:*** Total = Normal, males = Normal, females = Normal ***Immediate:*** - ***STW***: - ***EBM:*** - ***RAVLT (Immediate & Delayed recall, Learning Score):*** Total = High Normal ***SRT:*** Total = High Normal ***ROCF (Immediate & Delayed):*** Total = High Normal ***CERAD (Immediate & Delayed):*** Total = Normal ***CVLT (Immediate & Delayed):*** Total = Normal ***WLL***: Normal	Total = 82.7 ± 10.3 ***Delayed:*** Total = 74.8 ± 10.5 ***Immediate:*** - ***STW:*** - ***EBM:*** - ***RAVLT (Immediate & Delayed recall, Learning Score):*** Total = 83.4 ± 10.7 ***SRT:*** Total = 88.8 ± 10.2 ***ROCF (Immediate & Delayed):*** Total: 83.4 ± 10.7 ***CERAD (Immediate & Delayed):*** Total = 88.8 ± 10.6 ***CVLT (Immediate & Delayed):*** Total = 88.8 ± 10.6 ***WLL***: 77.2 ± 8.1	Total = Normal ***Delayed:*** Total = Optimal ***Immediate:*** - ***STW:*** - ***EBM:*** - ***RAVLT (Immediate & Delayed recall, Learning Score):*** Total = Normal ***SRT:*** Total = High Normal ***ROCF (Immediate & Delayed):*** Total = Normal ***CERAD (Immediate & Delayed):*** Total = High Normal ***CVLT (Immediate & Delayed):*** Total = High Normal ***WLL*:** Optimal
Episodic Memory	*Total:* n= 4	*Total* = 5.9 ± 2.3	Total = 51.7 ± 6.1, males = 50.3 ± 8, females = 51 ± 8.1	-	-	-	Total = High Normal
Semantic Memory	*Total*: n= 1	*Total* = 15.6 ± 2.9 males = 15.2 ± 3.0, females = 16 ± 2.8	Total = 50.7 ± 8, males = 50.3 ± 8, females = 51 ± 8.1	-	-	-	-
Working Memory	*Total:* n= 22 ***DSST:*** n= 8 ***Composite Score***: n= 1 ***CMS Score:*** Total: n= 1 ***DSB Test***: n= 7 ***McNS:*** n= 1 ***WDS:*** n= 1	***DSST:*** Total = 47.8 ± 7.9 ***Composite Score:*** male = 6.9 ± 2.3, females = 6.9 ± 2.72 ***CMS Score:*** Total = 76.6 ± 12.9 ***DSB Test:*** Total = 6.2 ±	Total = 52.3 ± 4.2, males = 54.2 ± 4.9, females = 56 ± 4.9	Total = 123.4 ± 15.9, males = 123.5, females = 121.7 ***DSST:*** Total = 119.3 ± 15.6 ***Composite Score:*** male = 123.5, female = 121.7 ***CMS Score:*** -	Total = Normal, males = Normal, females = Normal ***DSST:*** Total = Optimal ***Composite Score:*** male = Normal, female = Normal ***CMS Score:*** - ***DSB Test:*** Total = High	Total = 77.2 ± 9.9, males = 78.3, females = 75.1 ***DSST:*** Total = 73.2 ± 9.8 ***Composite Score:*** male = 78.3, female = 75.1 ***CMS Score:*** - ***DSB Test:*** Total = 80.5 ±	Total = Optimal, males = Optimal, females = Optimal ***DSST:*** Total = Optimal ***Composite Score:*** male = Optimal, female = Optimal

	MIS (MoCA): n= 1 ***VRT***: n= 1	1.9, males = 5, females = 5.3 ***McNS:*** Total = 27.7 ± 1.9 ***WDS:*** male = 38.6 ± 3.9, females = 38.9 ± 4.7 ***MIS (MoCA):*** Total = 12.72 ± 2.4 ***VRT:*** Total = 11.3		***DSB Test:*** Total = 132.7 ± 14.8 ***McNS:*** - ***WDS:*** - ***MIS (MoCA):*** - ***VRT:*** Total = 134.1 ± 16.9	Normal ***McNS:*** - ***WDS:*** - ***MIS (MoCA):*** Not available ***VRT:*** Total = High Normal	9.1 ***McNS:*** - ***WDS:*** - ***MIS (MoCA):*** - ***VRT:*** Total = 88.8 ± 10.5	***CMS Score:*** - ***DSB Test:*** Total = High Normal ***McNS:*** - ***WDS:*** - ***MIS (MoCA):*** - ***VRT:*** Total =High Normal
Attention	*Total:* n= 11 ***TMT-A:*** n= 10 ***CRT***: n= 4 ***SiRT:*** n= 3 ***DSF Test:*** n= 5 ***5-CMT***: n= 1	***TMT-A:*** Total = 24.8 ± 8.4 ***CRT:*** Total = 733.4 ± 153.8 ***SiRT:*** Total = 296.5 ± 64.6 ***DSF Test:*** Total = 7.5 ± 1.9 ***5-CMT:*** Total = 370.5	Total = 51.9 ± 4.3, males = 56.1 ± 3.7, females = 56.5 ± 3.6	Total = 130.5 ± 16.13 ***TMT-A:*** Total = 129.4 ± 15.9 ***CRT:*** Total = 134.1 ± 15.8 ***SiRT:*** Total = 134.8 ± 15.7 ***DSF Test:*** Total = 132.8 ± 17.1 ***5-CMT:*** Total = 129.4 ± 5.03	Total = High Normal ***TMT-A:*** Total: =Normal ***CRT:*** Total = High Normal ***SiRT:*** Total = High Normal ***DSF Test:*** Total = High Normal ***5-CMT:*** Total = High Normal	Total = 81.04 ± 9.4 ***TMT-A:*** Total = 82.4 ± 9.3 ***CRT:*** Total = 79.03 ± 8.8 ***SiRT:*** Total = 78.2 ± 8.4 ***DSF Test:*** Total = 80.4 ± 10.8 ***5-CMT:*** Total = 83.4 ± 3.2	Total = Normal ***TMT-A:*** Total = Normal ***CRT:*** Total = Optimal ***SiRT:*** Total = Optimal ***DSF Test:*** Total = High Normal ***5-CMT:*** Total = Normal
Intelligence	*Total:* n= 6 ***WAIS:*** n= 1 ***IQ***: n= 2 ***MR:*** n= 1	***WAIS:*** Total = 17 ± 3 ***IQ:*** Total = 104.08 ± 18.5 ***MR:*** Total = 18.13	Total = 54.7 ± 4.7	Total = 125.7 ± 17.5 ***WAIS:*** Total = 124 ± 18 ***IQ:*** Total = 149.9 ± 13.4 ***MR:*** Total = 126.7 ± 13.7	Total = Normal ***WAIS:*** Total = Normal ***IQ:*** Total = Grade 1 Hypertension ***MR:*** Total = Normal	Total = 84.4 ± 7.3 ***WAIS:*** - ***IQ:*** Total = 90.3 ± 6.9 ***MR:*** Total = 77.3 ± 6.9	Total = High Normal ***WAIS:*** - ***IQ:*** Total = Grade 1 Hypertension ***MR:*** Total = Optimal
Executive Function *Letter Cancellation*	*Total:* n= 2 ***LSST***: n= 1 ***LCCS***: n= 1	***LSST:*** Total = 282 ***LCCS:*** Total = 50 ± 7.3	Total = 52.66 ± 2.59	*Total* = 129.7 ± 17.1 ***LSST:*** Total = 134.8 ± 17.9 ***LCCS:*** Total: 118.58 ± 15.25	Total = High Normal ***LSST:*** Total = High Normal ***LCCS:*** Total = Optimal	Total = 77.2 ± 9.7 ***LSST:*** Total = 77.2 ± 9.7 ***LCCS:*** -	Total = Optimal ***LSST:*** Total = Optimal ***LCCS:*** -
Verbal Fluency	*Total:* n= 15 ***WFT***: n= 10 ***BNT***: n= 2 ***MVT***: n= 2 ***PFT***: n= 2 ***SFT***: n= 2 ***VIS (MoCA):*** n= 1 ***BeDT:*** n= 1 ***BuDT:*** n= 1	***WFT:*** Total = 31.3 ± 8.2, male = 25.7 ± 6.4, female = 24.8 ± 6.2 ***BNT:*** Total = 27.1 ± 1.9 ***MVT:*** Male = 25.8 ± 3.7, female = 23.3 ± 5.4 ***PFT:*** male = 17.1 ± 4.3, female = 16.8 ± 4.8 ***SFT:*** male = 16.7 ± 3.9, female = 16.02 ± 4.6 ***VIS (MoCA):*** Total = 6.48 ± 0.92 ***BeDT:*** male = 12, female: 12 ***BuDT:*** male = 7, female: 6	Total = 52.9 ± 5.3, male = 51.2 ± 4.9, female = 52.9 ± 4.9	Total = 123.7 ± 16.5, male = 128.7 ± 16.2, female = 122.6 ± 17 ***WFT:*** Total = 123.7 ± 16.4, male = 128.7 ± 16.2, female = 122.6 ± 17 ***BNT***: Total = 123.7 ± 17.6 ***MVT:*** - ***PFT:*** - ***SFT:*** - ***VIS (MoCA):*** - ***BeDT:*** Total = 126.7 ± 12.9 ***BuDT:*** Total = 126.7 ± 12.9	Total: Normal, male: Normal, female: Normal ***WFT:*** Total = Normal, male = Normal, female = Normal ***BNT:*** Total = Normal ***MVT:*** - ***PFT:*** - ***SFT:*** - ***VIS (MoCA):*** - ***BeDT:*** Total = Normal ***BuDT:*** Total = Normal	Total = 77.8 ± 12.1 ***WFT:*** Total = 77.8 ± 12.1 ***BNT:*** 77.2 ± 8.1 ***MVT:*** - ***PFT:*** - ***SFT:*** - ***VIS (MoCA):*** - ***BeDT:*** Total = 77.2 ± 7.8 ***BuDT:*** Total = 77.2 ± 7.8	Total = Optimal ***WFT:*** Total = Optimal ***BNT:*** Optimal ***MVT:*** - ***PFT:*** - ***SFT:*** - ***VIS (MoCA):*** - ***BeDT:*** Total = Optimal ***BuDT:*** Total = Optimal
Processing Speed	*Total*: n= 19 ***TMT-B:*** n= 9 ***TrB-A:*** n= 1 ***STIT:*** n= 2 ***WMT:*** n= 1 ***CES***: n= 3	***TMT-B:*** Total = 93.9 ± 2.7 ***TrB-A:*** Total = 1.14 ***STIT:*** Total = 42.9 ± 1.5 ***WMT:*** Total = 2.25 ± 1.09 ***CES:*** Total = 57.1 ± 0.1 ***RVP (CANTAB):*** Total =	Total = 52.5 ± 5.1, male = 56.9 ± 3.8, female = 54.1 ± 3.5	Total = 130.4 ± 14.6, male = 128.7 ± 16.2, female = 122.7 ± 16.9 ***TMT-B:*** Total = 131.6 ± 16.9, Female = 123.3 ± 16.3	Total = High Normal, male = Normal, female = Normal ***TMT-B:*** Total = High Normal, female = Normal ***TrB-A:*** Total = High	Total = 85.7 ± 8.7, female = 77.4 ± 9.34 ***TMT-B:*** Total = 87.5 ± 10.4, Female = 77.4 ± 9.34 ***TrB-A:*** Total = 86.1 ± 6.5 ***STIT:*** Total = 83.3 ± 10.7	Total = High Normal, female = Optimal ***TMT-B:*** Total = High Normal, female = Optimal ***TrB-A:*** Total = High

	RVP (CANTAB & Isolated): n= 1 ***SCWT:*** n= 1 ***EIS (MoCA):*** n= 1 ***VSS:*** n= 1 ***LT***: n= 1	0.92, 333.61 ± 88.01 ***SCWT:*** Total = 19.1 ***EIS (MoCA):*** Total = 11.64 ± 1.42 ***VSS:*** male = 302.02 ± 74.5, female = 323.5 ± 74.5 ***LT:*** male = 39.77 ± 17.8, female = 45.51 ± 26.6		***TrB-A:*** Total = 138.3 ± 8.4 ***STIT:*** Total = 131.2 ± 16.1, male = 128.7 ± 16.2,female = 122.6 ± 17 ***WMT:*** Total = 133.3 ± 16.9 ***CES:*** Total = 121 ***RVP (CANTAB):*** - ***SCWT:*** male = 128.7 ± 16.2, female: 122.6 ± 17 ***EIS (MoCA):*** - ***VSS:*** Total = 138.3 ± 8.4 ***LT:*** -	Normal ***STIT:*** Total = High Normal, male = Normal, female = Normal ***WMT:*** Total = High Normal ***CES:*** Total = Normal ***RVP (CANTAB):*** - ***SCWT:*** male = Normal, female = Normal ***EIS (MoCA):*** - ***VSS:*** Total = High Normal ***LT:*** -	***WMT:*** Total = 84.1 ± 12.3 ***CES:*** Total = 76.5 ***RVP (CANTAB):*** - ***SCWT:*** - ***EIS (MoCA):*** - ***VSS:*** Total = 86.1 ± 6.5 ***LT:*** -	Normal ***STIT:*** Total = Normal ***WMT:*** Total = Normal ***CES:*** Total = Optimal ***RVP (CANTAB):*** - ***SCWT:*** - ***EIS (MoCA):*** - ***VSS:*** Total = High Normal ***LT:*** -
Global Cognition	*Total*: n= 23 ***MMSE:*** n= 13 ***MoCA:*** n= 7 ***IQCODE:*** n= 1 ***CAMCOG:*** n= 1 ***NART:*** n= 1 ***MINT:*** n= 1 ***IST:*** n= 1 ***BPP:*** n= 1 ***ACE:*** n= 1 ***HRS-CS:*** n= 1 ***CERAD:*** n= 1	***MMSE***: Total = 27.8 ± 0.6 ***MoCA***: Total = 24.9 ± 3.1 ***IQCODE***: Total = 43.38 ± 3.01 ***CAMCOG:*** Total = 90 ***NART:*** Total = 28, male = 35.13 ± 9.5, female = 35.5 ± 9.1 ***MINT:*** Total = 30.25 ***IST:*** Total = 32.4 ***BPP:*** Total = 46.9 ***ACE:*** Total = 94.9 ***HRS-CS:*** Total = 14.31 ± 4.06, male = 14.2 ± 4.15, female = 14.44 ± 3.96 ***CERAD:*** Total = 81.6 ± 0.9	Total = 54.5 ± 5.3, males = 58.6 ± 2.8, females = 58.2 ± 2.8	Total = 131.5 ± 16.6 ***MMSE:*** Total = 133.2 ± 16.7 ***MoCA:*** Total = 118.3 ± 15.03 ***IQCODE:*** Total = 124.05 ± 15.6 ***CAMCOG***: Total = 140.8 ± 19.3 ***NART:*** Total = 140.8 ± 19.3 ***MINT***: Total = 126.7 ± 12.9 ***IST***: - ***BPP:*** - ***ACE:*** - ***HRS-CS:*** - ***CERAD:*** Total = 119.9 ± 11.8	Total = High Normal ***MMSE:*** Total = High Normal ***MoCA:*** Total = Optimal ***IQCODE:*** Total = Normal ***CAMCOG:*** Total = Grade 1 Hypertension ***NART:*** Total = Grade 1 Hypertension ***MINT:*** Total = Normal ***IST:*** - ***BPP:*** - ***ACE:*** - ***HRS-CS***: - ***CERAD:*** Total = Optimal	Total = 81.14 ± 10.08 ***MMSE:*** Total = 82.6 ± 10.02 ***MoCA:*** Total = 72.4 ± 10.4 ***IQCODE:*** Total = 73.9 ± 9.06 ***CAMCOG:*** Total = 88.7 ± 12.6 ***NART:*** Total = 88.7 ± 12.6 ***MINT:*** Total = 77.2 ± 7.8 ***IST:*** - ***BPP:*** - ***ACE:*** - ***HRS-CS:*** - ***CERAD:*** Total = 77.2 ± 8.1	Total = Normal ***MMSE:*** Total = High Normal ***MoCA:*** Total = Optimal ***IQCODE:*** Total = Optimal ***CAMCOG:*** Total = High Normal ***NART:*** Total = High Normal ***MINT:*** Total = Optimal ***IST:*** - ***BPP:*** - ***ACE:*** - ***HRS-CS:*** - *CERAD:* **Total = Optimal**
Inductive Reasoning	*Total:* n= 4	***AH-4:*** Total = 52.02 ± 8.5. male = 49.2 ± 9.5, female = 42.9 ± 11.6	Total = 52.56 ± 2.95, males = 49.5 ± 5.9, females = 49.86 ± 5.9	Total = 126.6 ± 15.2, male = 122.4 ± 15.5, female = 119.6 ± 16.7	Total = Normal, male = Normal, female = Optimal	Total = 82.4 ± 10.3	Total = Normal
Psychomotor Speed	*Total:* n= 5	***SDMT:*** Total = 56.1 ± 11.2, male = 48.2 ± 13.7, female = 50.5	Total = 52.3 ± 5.2, females = 50.01 ± 2.6	Total = 133.8 [SE: 0.3], Female = 123.3 ± 16.3	Total = High Normal, female = Normal	Total = 82.9 [SE: 0.2], female = 77.4 ± 9.34	Total = Normal, female = Optimal
Visuospatial Organisation	*Total:* n= 5 ***BDT:*** n= 2 ***VIS MoCA:*** n= 1 ***CDT:*** n= 1	***BDT:*** Total = 16.9 ± 0.1 ***VIS MoCA:*** Total = 6.48 ± 0.92 ***CDT:*** male = 28 ± 5, female = 55 ± 10	Total = 52.2 ± 5.7	Total = 128.5 ± 16.2 ***BDT:*** Total = 131.1 ± 15.2 ***VIS MoCA:*** - ***CDT:*** -	Total = Normal ***BDT:*** Total = High Normal ***VIS MoCA:*** - ***CDT:*** -	Total = 82.9 ± 9.4 ***BDT:*** Total = 82.9 ± 9.4 ***VIS MoCA:*** - ***CDT:*** -	Total = Normal ***BDT:*** Total = Normal ***VIS MoCA:*** - ***CDT:*** -

Abbreviations: *ACE*, Addenbrooke's cognitive examination, *AH-4*, Alice Heim 4-I, *BDT:* Block Design Test; *BeDT:* Benson Delay Test; *BNT:* Boston Naming Test; *BP*, Blood Pressure, *BPP*, Børge Priens Prøve, *BuDT: Buschke Delay Test; CAMCOG*, Cambridge Cognition Examination, *CANTAB*, Cambridge Neuropsychological Test Automated

Battery, *CDT:* Clock Drawing Test; *CERAD*, Consortium to Establish a Registry for Alzheimer's Disease, *CMS*, Chinese Clinical Memory Scale, *CRT:* Choice Reaction Time; *CVLT*, California Verbal Learning Test *DSB*, Digit Span Backwards, *CES:* Composite Executive Score; *DSF*, Digit Span Forward, *DSST*, Digit Symbol Substitution Test, *EBM:* East Boston Memory Test; *EIS*, Executive Index Score, *HRS-CS*, U.S. Health and Retirement Study Composite Score, *IST*, Intelligenz-Struktur-Test, *IQCODE*, Informant Questionnaire on Cognitive Decline in the Elderly, *IQ*, Intelligence Quotient, *LCCS:* Letter Cancellation Composite Score; *LSST*, Letter Search Speed Test, *LT:* Labyrinth Test*; McNS:* McNair Survey; *MINT*, Multilingual Naming Test, *MIS:* Memory Index Score; *MoCA*, Montreal Cognitive Assessment, *MMSE*, Mini-Mental State Exam, *MR:* Mental Rotation Test; *MVT:* Mill Hill Vocabulary Test; *NART*, National Adult Reading Test, *PFT: Phonemic Fluency Test; RAVLT*, Rey Auditory Verbal Learning Test, *ROCF*, Rey–Osterreith complex figure, *RVP:* Rapid Visual Processing*; SCWT:* Stroop Colour Word Test; *SDMT*, Symbol Digits Modalities Test, *SFT:* Semantic Fluency Test; *SRT:* Selective Reminding Test; *SiRT*: Simple Reaction Time; *STIT:* Stroop Test (Interference Time)*; STW:* Spot the Word Test; *TMT-A*, Trail making Test Part A, *TMT-B*, Trail making Test Part B, *TrB-A*, Trail making Test Difference between Part B and A, *VIS*, Visuospatial Index Score, *VRT:* Visual Reproduction Test; *VSS:* Visual Search Speed; *WFT:* Word Fluency Test; *WDS:* WAIS-IV Digit Sequencing*; WAIS*, Wechsler Adult Intelligence Scale; *WLL:* Word List Learning; *WMT:* Word Matching Test*; 5-CMT:* -Choice Movement Test.

Table 6.3. Summary of negative relationships between hypertension and cognition at midlife by study design and quality.

Study Design	Memory	Attention	Executive Function	Global Cognition	Psychomotor Speed	Intelligence	Visuospatial Organisation
Individual Study Cohorts	(426, 737, 738, 742, 746-752)	(749, 750, 752-754)	(426, 742, 746-750, 752, 755, 756)	(741, 746, 748, 755-759)	(426, 750)	(757)	**(750)**
Longitudinal Study Cohorts	(410, 731, 760)	-	(410, 760)	(736, 760)	(760)	-	-
Study Quality (n=)	Low: 9 Moderate: 12 High: 11	Low: - Moderate: 4 High: 3	Low: 8 Moderate: 13 High: 8	Low: 3 Moderate: 6 High: 10	Low: - Moderate: 1 High: 3	Low: - Moderate: 1 High: -	Low: - Moderate: 1 High: 1

6.5. Discussion

This review aimed to investigate the relationship between midlife hypertension status and cognitive function at later life and midlife. Using qualitative analysis our results indicate mixed and inconsistent findings across all cognitive domains, but predominantly favour a negative relationship between midlife hypertension and later life cognition in some but not all domains. This relationship was most notably found for memory, executive function, and global cognition although no relationship was observed for attention, inductive reasoning, visuospatial organization, or temporal orientation. There was conflicting evidence on the relationship between hypertension and cognitive function at midlife, irrespective of study quality and study design. Although qualitative analysis suggested a larger number of studies reported no relationship between hypertension and memory or global cognition at midlife, findings from our meta-analyses indicate a negative relationship in the case of memory, executive function, and global cognition and no relationship with attention.

The finding in this review that midlife hypertension affects later life cognition is consistent with previous research (422, 761, 762), indicating accelerated cognitive decline in the presence of midlife hypertension, specifically memory, executive function, and global cognition. Growing evidence highlights that the hypertension-cognition relationship is age-dependent (407, 763). Prolonged exposure to hypertension over a period of 25-30 years, beginning in middle-age, results in markedly higher risk of cognitive impairment in later-life (762). Evidence suggests that elevated BP even during young adulthood can have deleterious effects on cognition among middle-aged adults (764). Ageing plays a key role in functional adaptation to elevated BP over time by the cerebral blood vessels which precedes hypertension-induced microvascular damage and subsequent vascular cognitive impairment. Hypertension and ageing create a state of vulnerability that is suggested to alter the hippocampal expression of genes associated with cognitive decline and AD (765). The findings presented here confirm previous reports that midlife hypertension negatively affects cognition in later life. However, further to this our analysis reveals that select domains, including higher order processes like memory, are more notably affected than others. The hippocampus and entorhinal cortex are structures associated with learning and memory that are vulnerable to pathoanatomical and pathophysiological change in the presence of CV risk factors such as hypertension (766, 767). Impairments in working memory and the encoding of new long-term memories are reported in those experiencing age-related cognitive decline (768). Working memory declines with age are in line with the Baddeley model where the processing or central executive component are negatively impacted

(769). Therefore, hypertension may be contributing to the limitation of attentional capacity, where older adults are less able to inhibit irrelevant information and cognitive correlates of efficiency and arousal become impaired with ageing, a process that may begin as early as midlife (59, 770-773).

Neurocognitive tests can enable the subtle detection of cognitive change before the presentation of observable signs and symptoms, acting as robust indicators of pathological ageing. Our results provide evidence of the significant consequences of midlife hypertension for the time course and progression of cognitive impairment, and possible neurological comorbidities including dementia and AD (774, 775). Similar to our later life findings, meta-analysis indicated that memory, executive function, and global cognition at midlife were negatively affected by hypertension. This finding acts as a potential indicator of divergence from normal healthy ageing and a decline of brain health. The select cognitive domains of memory and executive function rather than overall cognitive function may be affected first and more extensively in the presence of hypertension with advancing age. In line with this finding, hypertension status among 207 late middle-aged adults was reported to be associated with age-related decline in verbal learning and memory, although hypertension was reported in only 19% of the study population (776). Various forms of memory are thus subject to age-related and pathological decline with the rates of change highly varied (777). Moreover, executive function, encompassing high-order cognitive processes, also declines with age and is accelerated in late midlife i.e. after 65 years of age (778). Decline in executive function is believed to precede reductions in memory by up to 18 years before diagnosis of AD and cognitive impairment in later life (779), with longitudinal evidence from women in midlife showing a mean decline of 2% per year in memory (780). Although the exact cause of accelerated regional decline in midlife is unclear, several hypothesised mechanisms are reported in support of our results, including higher aortic stiffness (781, 782), adaptive vascular changes in CBF and arterial pressure, and hypertension-induced neurovascular uncoupling (429-431). However, the timing and onset of these pathological features and parallel cognitive decline from midlife onwards is yet to be adequately determined, suggesting that a focus on midlife as a target area of intervention could yield benefit to cognitive function.

Similar to previous research, we found conflicting evidence on the impact of hypertension and cognitive function at midlife (422, 761, 783). Notably, our meta-analyses indicated no relationship between hypertension and attention at midlife, contradicting those of Ou and Colleagues (761). These conflicting findings are possibly explained by the lower number of studies included in the previous review (range: 2-4 studies) compared with 15 studies in the present review. It is probable rather than

possible that in the studies where no relationship was reported, the negative implications consequent to hypertension are not identifiable through cognitive function testing at midlife. Greater duration of time since the onset of hypertension is therefore associated with increased cognitive impairment, independent of age (762, 784). Theories of cognitive ageing and the dynamics of neural networks, however, postulate that the most basic of cognitive functions, such as attention, are affected by age. Attention at midlife was unaffected by the presence of midlife hypertension in our review. The majority of cognitive tests tend to incorporate more than one domain of cognitive function in any given task (785). Deficits in the early processing stages may influence additional co-domains in the later processing cognitive streams ultimately affecting global cognition from midlife into later life as seen in the present review. It is well-known that attention is involved in most cognitive processes, therefore any impact on attention potentially causes downstream consequences affecting the ability to complete normal daily tasks efficiently. Early evidence reports those with hypertension exhibit deficits in memory and executive function but no apparent decline in continuity of attention, similar to our present findings (786). Similarly, a decline in attention in response to a synergy between age and hypertension has been found to increase with age but did not significantly differ between those with hypertension and those without (787). Deficits in higher order processes, like attention, in the pre-frontal cortex can impact memory function in later life with significant impairment in divided attention or switching attentional focus (788-790). This may be accounted for by the so-called '*central executive control*' which has a role in virtually all cognitive functions from the allocation of attentional resources to the inhibition of irrelevant stimuli in working memory (791, 792). However, the stage in the lifespan from midlife onwards when cognitive changes begin to be exacerbated by the presence of hypertension, and how the declining trajectory across select domains can be targeted with intervention strategies, remains to be identified at a population or an individual level.

Significant cognitive impairment should not be considered a normal part of the ageing process. As a modifiable risk factor, hypertension represents a key target for the prevention, delayed progression, and reduction of cognitive impairment in aging populations (430). Attention at midlife was not negatively impacted by midlife hypertension as evident by meta-analysis but was affected in later life. Studies of ageing and neurocognition have reported age-related declines in attention (793-796). Our results highlight an inconsistent relationship between hypertension at midlife and a decline in attention in later life, similar to previous reports (739, 740, 797). Of notable concern, recent evidence from the National Health and Nutrition Examination Survey reports 70% of older adults are living with hypertension in comparison to just 32% of adults aged 40-59 years (798). Management of

previously untreated hypertension later in life cannot correct for the negative impact of decades of uncontrolled hypertension on cognitive function (799, 800). Hypertension may therefore contribute to, and even exacerbate, brain ageing via deterioration of neuroanatomical substrates and modulators among certain cognitive domains from midlife onwards (801-804). Our results support the hypothesis of significant variation of age-related cognitive trajectories across several domains which may be exacerbated with long-term exposure to hypertension across the lifespan.

6.6. Limitations

There are several limitations within this current study. A range of tools were used for assessing cognitive function and were broadly categorized across all studies for one or several cognitive domains. We are aware that some tests will incorporate the use of multiple cognitive domains but for the purpose of this review each test was organized according to the core cognitive function tested. Additionally, inadequate reporting of data limited meta-analysis abilities, therefore a cautionary interpretation of the results presented here is required. Subsequently, defined associations may not indicate a causal relationship and should be interpreted with caution, given the number of eligible studies included. We have examined the qualitative relationship between hypertension status and cognitive function at two distinct points across the adult lifespan, but we did not investigate or analyse the biological underpinnings or physiological causality leading to cognitive impairments. Studies were of varied design and quality, limiting the ability to establish definitive cause and effect of midlife hypertension on cognitive function in later life. Studies applied different BP values for the classification of hypertension, which impedes our ability to generalise findings related to specific BP values. In studies where the reported associations are used to interpret findings the true causal effect may not be represented due to the presence of selection bias, reverse causation, or misclassification of observational studies. Finally, although our evidence indicates that hypertension at midlife may increase the risk of cognitive impairment from midlife into later life, the currently available data do not enable us as to yet define an age or an age range at midlife where hypertension diagnosis or duration of hypertension can provide a clinically meaningful intervention point for the introduction of management strategies.

6.7. Conclusions

The risks of midlife hypertension to cognition across the adult lifespan are of considerable concern in the context of an aging population. The variability across cognitive domains is apparent, such that some domains are impaired more than others in those with a diagnosis of hypertension. Midlife hypertension adversely affected memory, executive function, and global cognition in later life, and negatively affected the same select domains of memory, executive function, and global cognition, but not attention, at midlife. With a lack of clarity as to when certain domains begin to decline with age from midlife onwards, more longitudinal, and prospective studies are needed to establish a window of hypertension duration at which cognitive impairment and cognitive domains that are impacted earliest become apparent. There is ultimately a need to develop public health policies and prevention strategies for hypertension in relation to cognitive function across all domains.

6.8. Question(s) Raised

- With a lack of clarity as to when certain domains begin to decline with age from midlife onwards, what are other contributing CV risk factors potentially affecting cognitive function in mid- and later life?

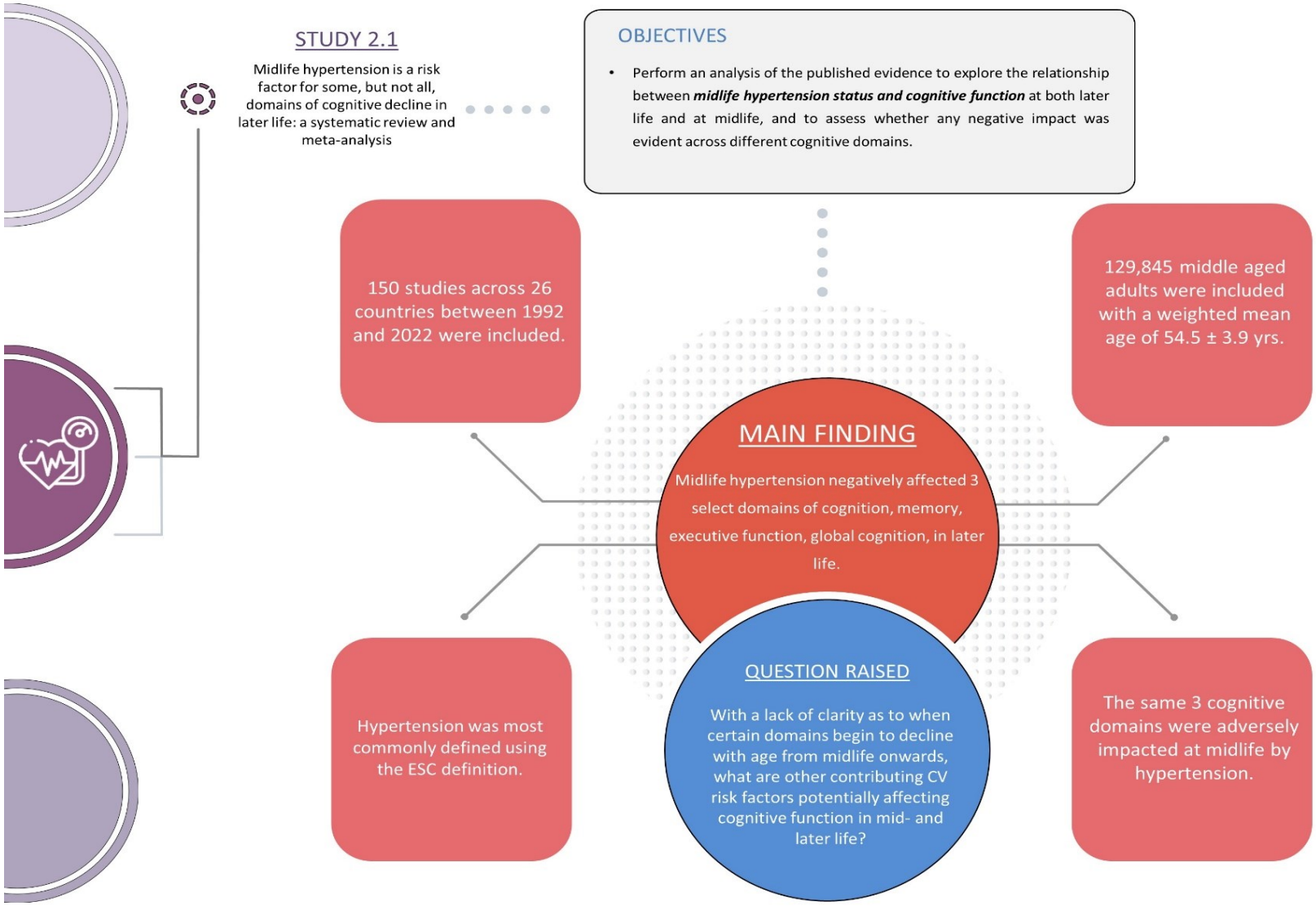

Figure 6.7. *Chapter Six – Study 2.1 Summary.*

7

Chapter Seven:

The influence of Type 2 Diabetes Mellitus and its Metabolic Correlates in Middle-Aged Adults on Cognitive Function in Mid and Later Life: A Systematic Review and Meta-Analysis

7.1. Highlights

- 143 studies were included for review published between 1995 and 2022.
- Seventy-three studies examined data from twelve prospective longitudinal cohorts and the remaining 70 studies examined data from independent study cohorts.
- A total of 110,906 participants with a weighted mean age of 53.9 ± 3.8 years were included.
- The majority of longitudinal studies reported negative relationships between midlife T2DM and later cognition, specifically executive function and global cognition. Conflicting findings were however reported for memory.
- There were no discernible differences in findings reported based on study design with most studies reporting no relationship between T2DM and its metabolic correlates with cognition at midlife.

7.2. Introduction

Type 2 diabetes mellitus (T2DM), a chronic metabolic illness also known as adult-onset diabetes, is linked with increased rates of morbidity and mortality. Once confined to older populations, there is a growing prevalence among younger adults (449). Worldwide there were 463 million people with a diagnosis of T2DM in 2019 and numbers are expected to exceed more than 700 million by 2045 (805). High levels of blood glucose with advancing age can impact the circulatory, renal and peripheral nervous systems with almost 20% of older adults aged 65-69 years diagnosed with T2DM (806). Exposure to T2DM during midlife accelerates neurodegeneration and structural neuropathology with advancing age (807). Those with diabetes are 1.5-2.0 times more likely to develop some form of cognitive deterioration across one or several domains with impairments ranging from subtle to severe and even leading to dementia in some instances (808). As the global prevalence of T2DM is expected to exceed more than 10% by 2030 (809), most notably among those aged 75-79 years (810, 811) the management of T2DM during midlife has the potential to slow the progression of cognitive ageing and development of dementia in this population (465).

Altered glucose metabolism has been linked to minor reductions in cognition with more severe impairment found in those with prolonged T2DM (454, 456). For example, mild reductions in cognitive function linked to T2DM are observed across various age brackets, even in individuals below the age

of 60 to 65, and these changes exhibit a gradual progression over the course of time. The rate of cognitive deterioration among individuals diagnosed with T2DM closely parallels or slightly surpasses the pace of cognitive decline typically associated with the natural aging process. Although, the physiological mechanisms underlying neurodegeneration and associated cognitive decline in the presence of T2DM are not fully understood but are considered multifactorial (475). It is proposed that sustained hyperglycaemia can lead to macro- and micro- vasculature injury, specifically endothelial dysfunction, in the periphery possibly resulting in cerebral vasculature injury (451, 452) and impairment of neuronal function (812, 813). It is well-established that changes in resting-state brain activity and cerebral blood flow (CBF) occur in T2DM. Recent findings show that combining regional homogeneity and CBF reveals persistent alterations in regional neurovascular coupling in T2DM patients. This study suggests that T2DM may accelerate NVC decline in specific brain regions, like the left insula, contributing to memory loss (814).Further research suggests that this regional hypoperfusion, with reductions in CBF to the fronto-temporal and limbic regions of the brain may be responsible for this impairment (815, 816). Neuroimaging studies report that cognitive impairment in diabetic patients mirrors the pathology of vascular dementia and Alzheimer's disease (AD); e.g. lower CBF (817, 818), reduced total brain volume (819), and structural alterations to the hippocampus and basal ganglia (467). The contribution of this cerebral regional atrophy is linked with reduced executive function and memory (468, 472). Additionally, T2DM increases oxidative stress and endothelial dysfunction associated with reduced psychomotor speed, mental flexibility, and attention (820). It is likely that in the presence of T2DM structural deterioration of brain tissue begins at midlife and precipitates diminished cognitive reserve and/or resilience (819).

The relationship between T2DM and cognitive function in middle age has been previously investigated in middle aged adults (821) but no review has yet evaluated the impact of T2DM at midlife on cognitive function across the lifespan to include midlife and later life. Greater understanding of the relationship between T2DM and cognition during ageing could inform appropriate age-dependent and time sensitive management of T2DM (822, 823). The aim of this systematic review was to assess the impact of midlife T2DM on cognitive function, as measured across different domains, at midlife and later life. The secondary aim was to determine the relationship between metabolic correlates of T2DM (fasting blood glucose [FBG], glycosylated haemoglobin [HbA_1c] and metabolic syndrome [MetS]) and cognitive function.

7.3. Methods

This systematic review was undertaken in accordance with the PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) statement (PRISMA; www.prisma-statement.org) and registered with PROSPERO (CRD42021238293; https://www.crd.york.ac.uk/prospero/). The presented review is a sub-analysis of a larger registered review of midlife CV health and later life cognition.

7.3.1. Search Strategy

Electronic databases, including MEDLINE, PubMed, Web of Science, and CINAHL were searched from their inception to January 2022; searches were not restricted for language or publication date. Predefined keywords, MeSH terms and a combination of these were used such as midlife, cardiovascular risk, cognition, and a focus on all those with "*diabetes*". A manual search of reference lists of included studies was undertaken to identify any additional articles for inclusion. The full search strategy can be found as supplementary material (see **Supplementary file**). The step-by-step search process can be seen in **Figure 7.1**.

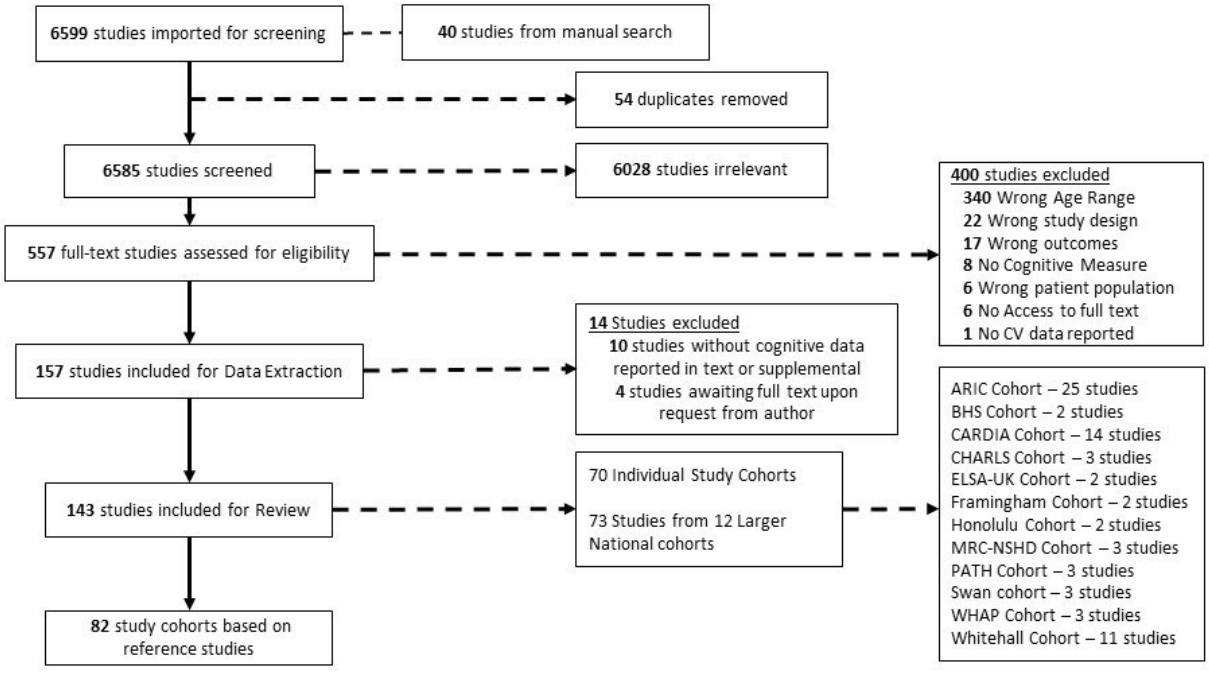

Figure 7.1. *Flow chart of the study selection process*.

7.3.2. Eligibility Criteria

Studies were deemed eligible for inclusion based on the following criteria: human participants, middle aged adults between ages of 40-65 years in line with the definition set by the World Health Organisation, at least one outcome measure of diabetes according to the European Society of Cardiology (ESC) and American Heart Association (AHA), including diabetes status (Yes or No) and metabolic metrics; FBG, HbA1c levels, and the presence or absence of MetS reported at later life, midlife, or both for determination of the longitudinal association with midlife diabetes and cognition across domains including memory, attention, executive function, intelligence, and global cognitive functioning (*see* ***Supplementary file***). Studies were excluded if cognitive testing was undertaken by a proxy or designated respondent, such as a friend or family member, if the participant cohorts included those with midlife dementia or any form of pre-existing cognitive impairment.

7.3.3. Data Extraction

Relevant data was extracted in accordance with STROBE guidelines (546). Where multiple studies from prospective longitudinal cohorts were included, the most recent publication was chosen as the reference study for the determination of baseline data. In the presence of uncertainties during data extraction corresponding authors were contacted for clarification. For full details on the data extraction process please see **Chapter Two Section 2.5.3**.

7.3.4. Risk of Bias and Methodological Assessment

Methodological quality was determined using the AXIS tool (Critical appraisal tool to assess the quality of cross-sectional studies) (547) independently by two authors. This appraisal tool utilised a series of 20 questions to determine study quality and risk of bias by means of answering 'Yes', 'No' or 'Unsure' as seen in previous reviews (484). For full details please see **Chapter Two Section 2.5.4**.

7.3.5. Statistical analysis

The weighted mean of all cognitive measures, FBG, and HbA_1c values were calculated. Qualitative analysis was used to determine associations between diabetes (T2DM, FBG and HbA_1c) and cognition (positive, negative, or neutral) at both midlife and later life. Cognitive outcome measures were

grouped according to cognitive domain and further sub-divided where appropriate based on included cognitive tests. Qualitative analysis assessed the relationship between midlife diabetes diagnosis and cognition at later life and midlife; positive, negative, or neutral, across studies.

A random effects meta-analysis was undertaken to compare the difference across each cognitive domain between two independent groups, T2DM vs no diabetes. The pooled summary effect of midlife T2DM diagnosis on cognition at midlife was deemed appropriate. For full details please see **Chapter Two Section 2.5.5**.

7.4. Results

7.4.1. Literature Search

Figure 7.1 provides full detail of study selection. The initial search returned 6,585 records. Upon duplicate removal and screening of titles and abstracts, 557 studies remained and 400 were subsequently excluded (*see **Figure 7.1.***). A further 14 studies were excluded during data extraction as authors who were contacted for full text or access to raw cognitive not reported did not respond. All included studies were then imported into Endnote version 20 to create a database of studies for data extraction in Microsoft Excel. In total, 143 studies were included for review published between 1995 and 2022.

7.4.2. Methodological and Risk of Bias Assessment

Overall, studies included were deemed to be of moderate- to- high quality; 49 high, 59 moderate and 33 low quality studies. Studies shared common weaknesses, including failing to justify sample size (n= 131), to address non-responders (n= 126), to provide information about non-responders (n= 133), to provide a clear assessment of statistical significance (n= 49), and/or failing to state whether ethical approval was granted (n= 17).

7.4.3. Study Characteristics

123 studies included both males and females, with 12 and 8 studies assessing males or females only, respectively. Seventy-three studies examined data from twelve prospective longitudinal cohorts, with the largest numbers derived from the Atherosclerosis Risk in Communities (ARIC) cohort (n= 25),

Coronary Artery Risk Development in Young Adults (CARDIA) cohort (n= 14), and Whitehall cohort (n= 11) (see Supplementary file). The remaining 70 studies examined data from independent study cohorts. Studies were undertaken across 28 nations, including United States (n= 563), the United Kingdom (n= 20), China (n= 8), Australia (n= 6), and India (n= 6).

7.4.4. Participant Characteristics

There was a total of 110,906 participants. The weighted mean age of the participants was 53.9 ± 3.8 years. The weighted mean BMI was 27.3 ± 4.4 kg/m2. The average height and weight were 167.7 ± 2 cm and 79.2 ± 15.0 kg, respectively. Of studies that provided data by sex (n= 55), 42,460 males and 46,831 females were included. Weighted mean BMI was 25.3 ± 3.2 kg/m2 and 26.5 ± 4.9 kg/m2 for males and females, respectively. Males and females were of similar weighted mean age (53.5 ± 3.6 vs 53.5 ± 3.1 years) and females were shorter in mean height than males (175 ± 7.9 vs 162 ± 6 kg). The average body mass was unavailable.

7.4.5. Measures of T2DM and its Metabolic Correlates

Tables 7.2 and 7.3 details all measures of T2DM and MetS across studies. Mean FBG was 93.2 ± 19.2 mg/dl and 4.7 ± 0.5 for HbA1c assessed across 45 studies and 19 studies, respectively. Of studies that assessed MetS (n= 6), 1,195 participants were found to present with MeTS and 8,128 had no MeTS. Of studies that assessed diabetes status (n= 57) 14,841 were diabetic and 104,205 were classified as non- diabetes. Males and females had similar levels of mean FBG (95.4 ± 10.08 vs 93.6 ± 9.72 mg/dl) and HbA1c levels (5.6 ± 0.5 vs 5.6 ± 0.4 mg/dl). 635 females presented with MeTS compared to 1,514 did not. No data was available for males. A higher number of males were classified as diabetic than females (1,000 vs 790).

7.4.6. Associations between Midlife T2DM and Cognition in Later Life

Studies assessing midlife T2DM and later life cognition were of moderate-to-high quality. Of the 7 longitudinal cohort studies, a negative relationship between midlife T2DM and later life cognition was reported across several domains, including memory (n= 2, 29%), executive function (n= 3, 43%), and global cognition (n= 3, 43%). In two of the studies, no relationship was found between midlife

T2DM and memory, attention, executive function, processing speed, or global cognition (730, 824) (*see* ***Table 7.1***).

Of the individual longitudinal studies (n= 3), two studies (66%) reported no relationship (825, 826) and one study (33%) reported a negative relationship with memory (737) (*see* ***Table 7.1***).

Overall, the majority of longitudinal studies reported negative relationships between midlife T2DM and later cognition, specifically executive function and global cognition. Conflicting findings were however reported for memory (**Table 7.1**).

Table 7.1. *Summary of longitudinal studies with negative or null relationship between diabetes and cognitive measures at later life.*

Author	Year	Setting	Study Quality	Cognitive Variables	Relationship
Anstey et al.	2014	PATH, Australia	Moderate	Memory, attention, executive function, processing speed, global cognition	0 Diabetes
Bancks et al.	2017	ARIC, USA	High	Memory, executive function	- Diabetes (executive function, global cognition)
Bangen et al.	2013	Framingham, USA	High	Memory, executive function, global cognition, and visuospatial organisation	- Diabetes (executive function)
Bayes-Marin et al.	2020	Edad con Salud, Spain	High	Memory	- Diabetes (Memory)
Blodgett et al.	2020	MRC NSHD, UK	Moderate	Verbal Memory	0 Diabetes
Cherbuin et al.	2009	PATH, Australia	High	Global Cognition	- Diabetes (global cognition)
Dixon et al.	2021	SWAN, USA	Moderate	Executive function, working and episodic memory	0 Diabetes (working memory) - Diabetes (executive function, episodic memory)
Kaffashian et al.	2013	Whitehall II, UK	High	Memory, executive function, attention, global cognition, inductive reasoning	- Diabetes (global cognition)
Nunley et al.	2017	Pittsburgh Epidemiology of Diabetes Complications Study; USA	High	Memory, attention, executive function, global cognition, intelligence, and psychomotor speed	0 Diabetes
Tuligenga et al.	2014	Whitehall II study; UK	Moderate	Memory, executive function, inductive reasoning, and global cognition	- Diabetes (Memory, inductive reasoning, global cognition)

7.4.7. Associations between Measures of T2DM and its Metabolic Correlates and Cognition at Midlife

Tables 7.3, 7.6 and Supplemental Table 7.4 and 7.5 provide the details of the relationship between T2DM, MetS, FBG, and HbA1c levels and measures of cognitive function.

Most studies reported no relationship between T2DM and memory (n= 29, 66%), attention (n= 16, 76%), executive function (n= 26, 67%), global cognition (n=24, 73%), psychomotor speed (n= 4, 80%) and visuospatial organisation (n=7, 86%). All studies reported no relationship between T2DM and intelligence (n= 2) or temporal orientation (n= 1)

All studies reported no relationship between FBG and inductive reasoning (n=2), intelligence (n= 5), and temporal orientation (n= 1), visuospatial organisation (n= 7). The majority of studies reported no relationship between FBG and memory (n= 25, 78%), attention (n= 15, 83%), executive function (n= 23, 79%), global cognition (n=24, 87%), and psychomotor speed (n= 4, 80%).

In the case of HbA_1c, studies predominately reported no relationship with memory (n= 9, 69%), attention (n= 6, 75%), executive function (n= 9, 64%) and global cognition (n=5, 56%). All studies reported no relationship with intelligence (n= 1), and psychomotor speed (n= 1), visuospatial organization (n= 1).

For MeTs and cognitive function, findings were conflicting and limited due to a small number of studies. A negative relationship was reported with memory (n= 2/3,75%), attention (n= 1/2, 50%), executive function (n= 1/2, 50%), global cognition (n= 1/3, 25%), inductive reasoning (n= 1/1, 100%), and psychomotor speed (n= 1/2, 50%). Whereas no relationship was reported for intelligence (n= 1/1, 100%).

There were no discernible differences in findings reported based on study design with most studies reporting no relationship (individual cohorts: n=23/32, 72%; large cohorts: n= 6/10, 60%). Studies of moderate and high quality consistently reported no relationship (n = 82, 80%) whereas findings from low quality studies were inconsistent.

7.4.8. Meta-analysis

Meta-analysis was conducted to assess the relationship between midlife T2DM and midlife cognition; similar analysis was not possible in the case of later life cognition as limited or no raw cognitive data were available.

Ten studies across four cognitive domains (memory, executive function, attention, and global cognition) were suitable for meta-analysis. A total of 3,586 participants were diagnosed with T2DM while 30,167 participants did not have a diagnosis of T2DM.

A meta-analysis using random effects and sensitivity analysis was deemed appropriate and sub-divided by study quality. T2DM had a negative effect on memory function compared to no diagnosis across 4 studies of low and moderate quality [MD = -0.19; 95% CI = -0.26 to -0.11; I^2 = 25%] (see **Figure 7.2**). T2DM had no effect among 4 studies on attention across all levels of study quality with high heterogeneity [MD = 0.03; 95% CI = -0.15 to 0.20; I^2 = 87%] (see **Figure 7.3**). T2DM had a negative impact on executive function across 2 studies of moderate quality [MD = -0.14; 95% CI = -0.25 to -0.04; I^2 = 0%] (see **Figure 7.4**), and global cognition across two studies of moderate quality [MD = -0.26; 95% CI = -0.34 to -0.17; I^2 = 0%] (see **Figure 7.5**).

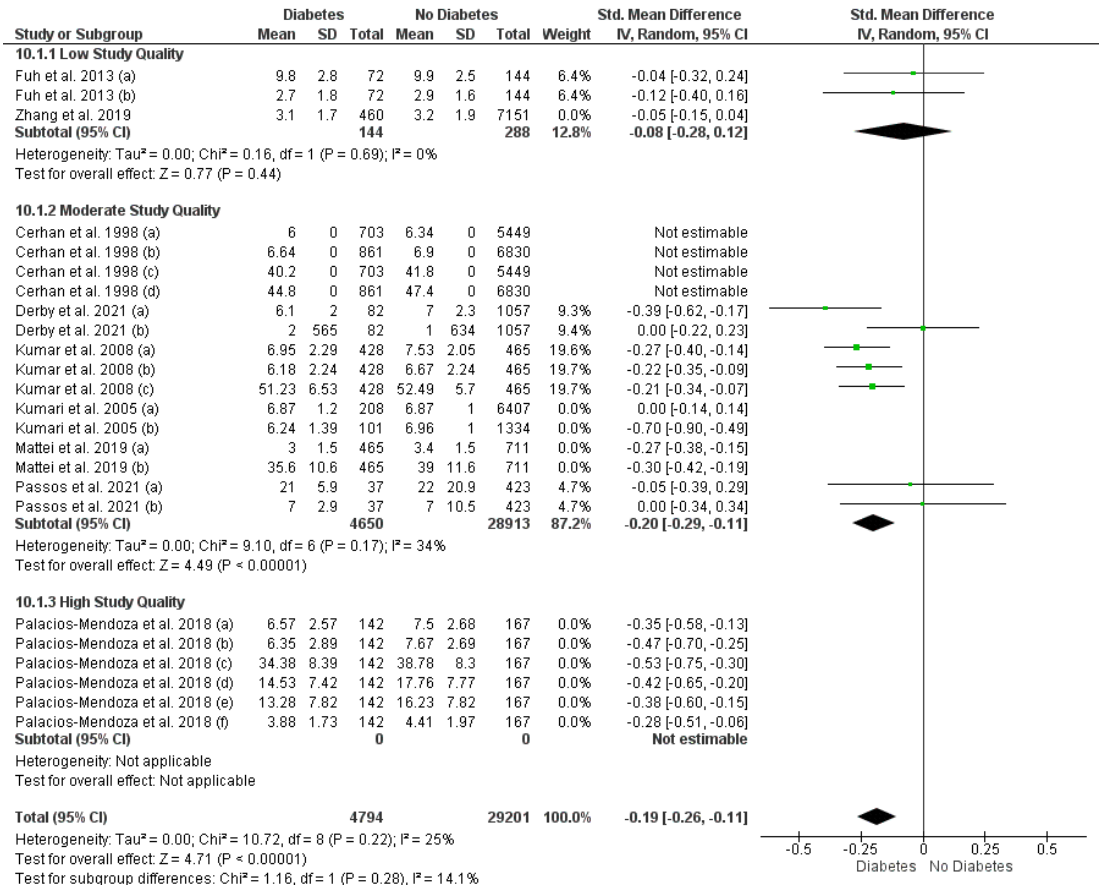

Figure 7.2. *Forest plot examining the overall effect of diagnosed diabetes versus no diabetes on memory function at midlife.*

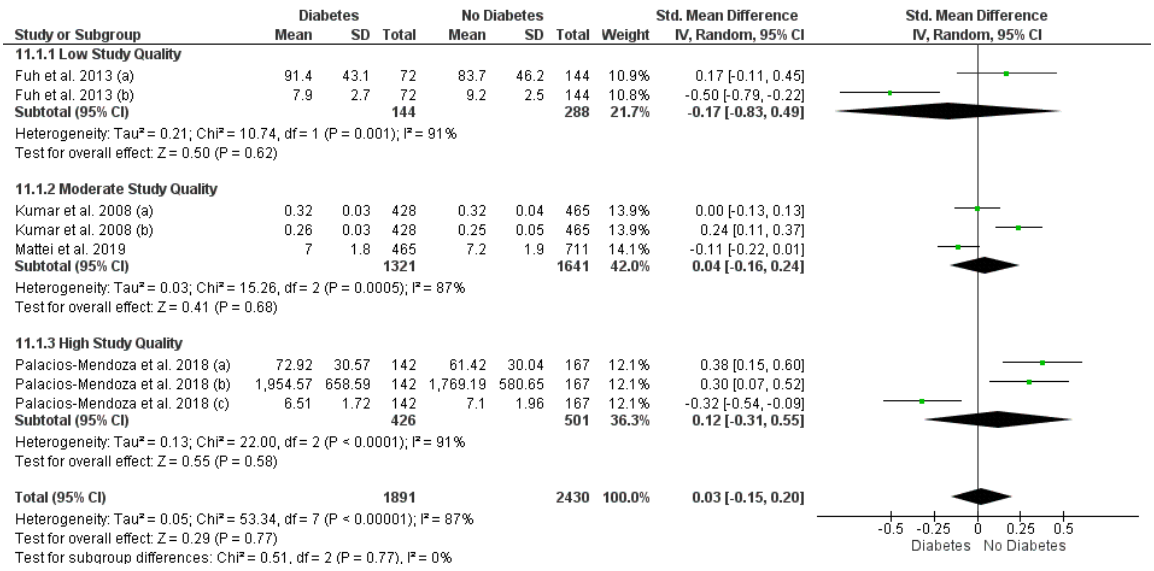

Figure 7.3. *Forest plot examining the overall effect of diagnosed diabetes versus no diabetes on attention.*

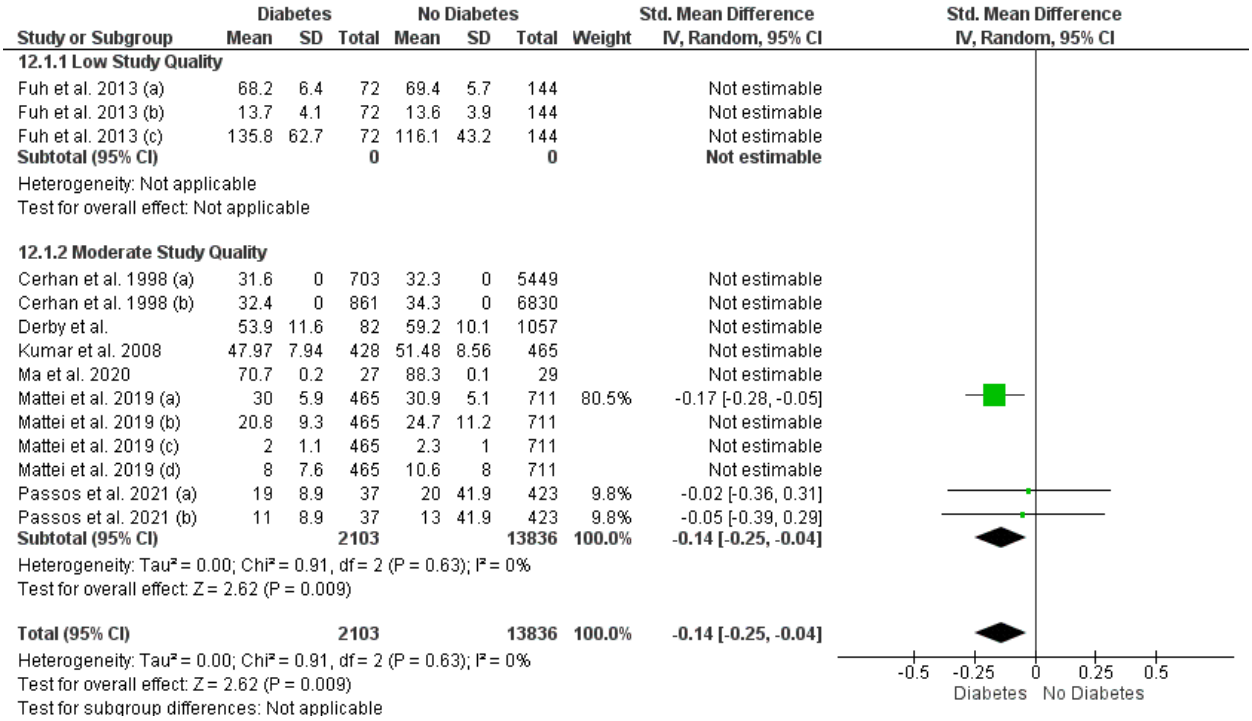

Figure 7.4. *Forest plot examining the overall effect of diagnosed diabetes versus no diabetes on executive function.*

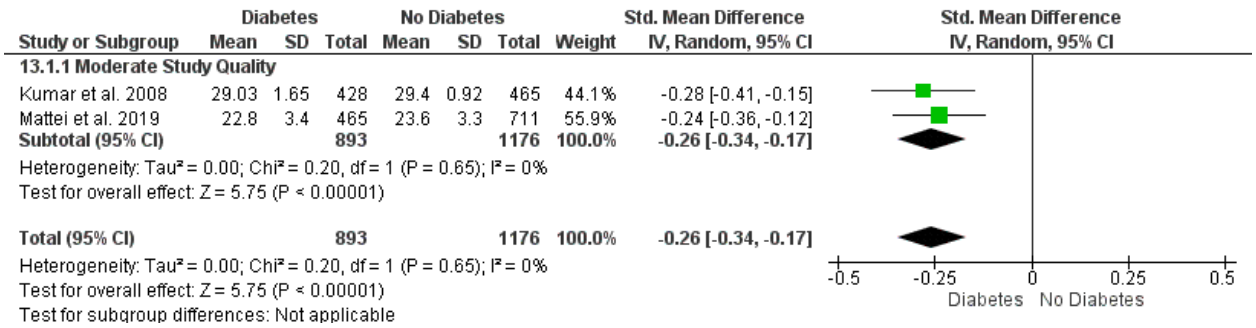

Figure 7.5. *Forest plot examining the overall effect of diagnosed diabetes versus no diabetes on global cognition.*

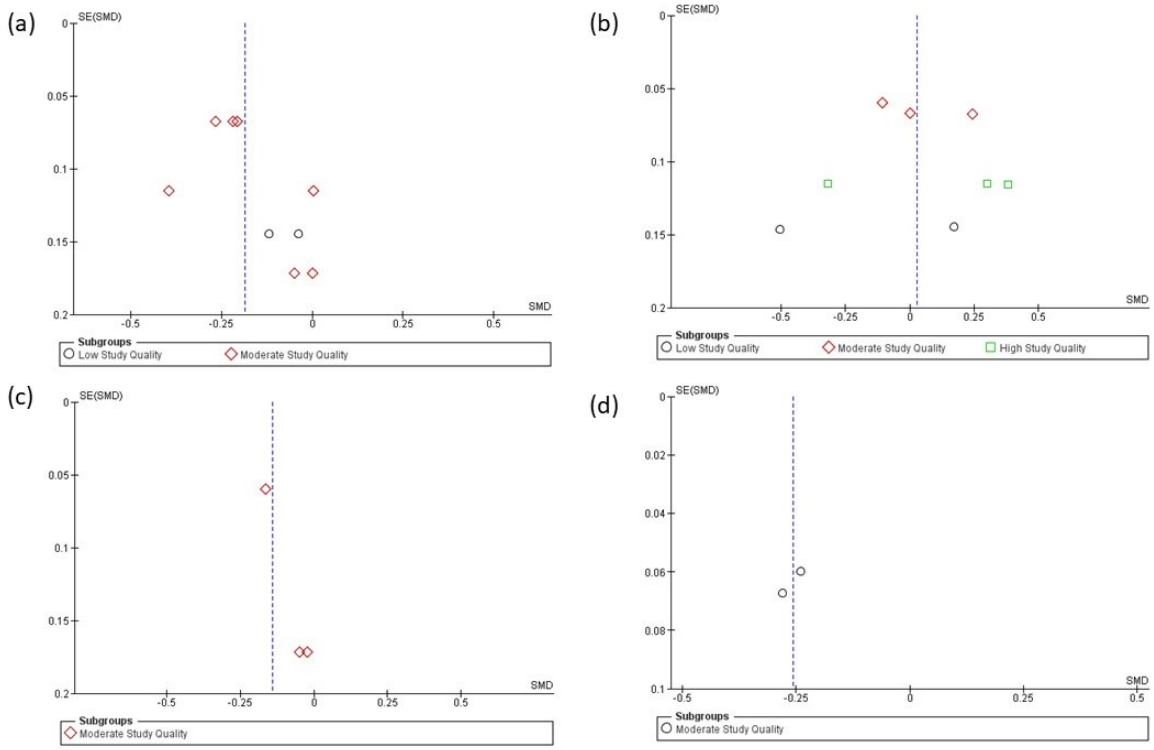

Figure 7.6. *Funnel plots of meta-analysis assessing the differences between those with diabetes versus those without diabetes.* **(a)** Memory, **(b)** Attention, **(c)** Executive Function, **(d)** Global Cognition.

Table 7.2. *Summary Table of weighted average for all cognitive measures and associated diabetes metrics at baseline (i.e., midlife).*

Cognitive Variable	No. of Studies (n =)	Weighted Average (Mean ±SD)	Age (Mean ±SD; Years)	FBG (mg/dl)	HbA1c (mg/dl)
Memory	*Total*: n= 17	***Immediate:*** Total = 9.9, Males = 15.8 ± 4, Females = 17.6 ± 4.1	Total = 50.9 ± 4.7, Males = 56.6 ± 7.1, Females = 56.2 ± 7.1	Total = 90.9 ± 19.3	**Total =** 5.7 ± 0.4
Verbal Memory	***Immediate***: n= 5, ***Delayed***: n= 5, ***STW***: n= 1, ***EBM***: n= 2, ***RAVLT (Immediate & Delayed recall):*** n= 2, ***RAVLT (Learning Score):*** n= 1, ***RAVLT (Summary Metric):*** n= 3 ***SRT*:** n= 1, ***ROCF (Immediate & Delayed recall):*** n= 1, ***CERAD (Immediate & Delayed):*** n= 1, ***CVLT (Immediate & Delayed):*** n= 1 ***RCF (Delayed recall):*** n = 1	***Delayed:*** Total = 8.6 ± 1.4, Males = 4.1 ± 1.6, Females = 4.2 ± 1.7 ***STW***: Total = 50.5 ***EBM***: Total = 10.2 ± 1.1 ***RAVLT (Immediate & Delayed recall):*** Total = 7.1 ± 2.7, 9.1 ± 3.2 ***RAVLT (Learning Score):*** Total = 36.8 ± 8.3 ***RAVLT (Summary metric):*** Total = 8.9 ± 3.2 ***SRT:*** Total = 34.3 ***ROCF (Immediate & Delayed recall):*** 16.1 ± 7.6, 14.9 ± 7.8 ***CERAD (Immediate & Delayed):*** Total = 7.2 ± 1.1, 7.7 ± 1.5 ***CVLT (Immediate & Delayed):*** Total = 8.8 ± 2.1, 8.8 ± 3.2 ***RCF (Delayed recall):*** Total = 21.1 ± 7.6		***Delayed:*** Not available ***Immediate:*** Not available ***EBM:*** Not available ***RAVLT (Summary metric):*** Total = 100.9 ± 17.2 ***RAVLT (Immediate & Delayed recall, Learning Score):*** Total = 101.5 ± 12.5, 131.9 ± 48.8 ***SRT:*** Total = 86.2 ± 20.9 ***ROCF (Immediate & Delayed recall):*** Total = 131.9 ± 48.8 ***CERAD (Immediate & Delayed):*** Total = 95.22 ± 21.96 ***CVLT (Immediate & Delayed):*** Total = 95.22 ± 21.96 ***RCF (Delayed recall):*** Total = 91	***Delayed:*** Not available ***Immediate:*** Not available ***EBM:*** Not available ***RAVLT (Summary metric):*** Not available ***RAVLT (Immediate & Delayed recall, Learning Score):*** **Total =** 7.0 ± 1.4, 5.7 ± 0.4 ***SRT:*** Not available ***ROCF (Immediate & Delayed recall):*** **Total =** 7.0 ± 1.4 ***CERAD (Immediate & Delayed):*** Not available ***CVLT (Immediate & Delayed):*** Not available ***RCF (Delayed recall):*** **Total =** 5.2
Episodic Memory	*Total:* n= 4	*Total* = 6.7 ± 2.1	Total = 56.9 ± 5.9, Males = 50.3 ± 8, Females = 51 ± 8.1	Total = 106.5 ± 19.8, Males = 95.4 ± 10.08, Females = 93.6 ± 9.72	**Total =** 5.3 ± 0.6
Semantic Memory	*Total*: n= 2	*Total* = 19.4 ± 0.3 Males = 15.2 ± 3.0, Females = 15.9 ± 2.8	Total = 50.7 ± 8, Males = 50.3 ± 8, Females = 51 ± 8.1	Total = 95.4 ± 9.9, Males = 95.4 ± 10.08, Females = 93.6 ± 9.72	Not available
Working Memory	*Total:* n= 21 ***DSST:*** n= 7 ***CMS Score:*** Total: n= 1 ***DSB Test***: n= 8 ***MIS (MoCA):*** n= 1 ***VRT***: n= 1 ***LMT:*** n = 1 ***Composite:*** n = 1 WAIS: n = 1	***DSST:*** Total = 45.2 ± 11.03 ***CMS Score:*** Total = 76.6 ± 12.9 ***DSB Test:*** Total = 6.2 ± 1.9 ***MIS (MoCA):*** Total = 12.7 ± 2.4 ***VRT:*** Total = 11.3 ***LMT:*** Total = 17.2 ± 6.9 ***Composite:*** 12.88 ± 0.55 WAIS: Males = 38.6 ± 3.9, Females = 38.9 ± 4.7	Total = 52.2 ± 4.6	Total = 98.7 ± 24.9 ***DSST:*** Total = 108 ± 27 ***CMS Score:*** 89.54 ± 9.5 ***DSB Test:*** Total = 104.9 ± 19.3 ***MIS (MoCA):*** Not available ***VRT:*** Total = 86.2 ± 21.4 ***LMT:*** Total = 95.1 ± 11.7 ***Composite:*** **176.8 ± 5.58** WAIS: Not available	**Total =** 7.0 ± 1.4 ***DSST:*** Not available ***CMS Score:*** Not available ***DSB Test:*** **Total =** 7.0 ± 1.4 ***MIS (MoCA):*** Not available ***VRT:*** Not available ***LMT:*** Not available ***Composite:*** Not available **WAIS:** Not available
Attention	*Total:* n= 14 ***TMT-A:*** n= 9 ***CRT***: n= 4 ***SiRT:*** n= 3 ***DSF Test:*** n= 5 ***AI (MoCA):*** n = 1 5-CMT: n= 1	***TMT-A:*** Total = 28.6 ± 10.03 ***CRT:*** Total = 783.1 ± 167.5 ***SiRT:*** Total = 250.9 ± 56.7 ***DSF Test:*** Total = 7.2 ± 1.9 ***AI (MoCA):*** 16.3 ± 1.8 5-CMT: 370.5 ACE-III: 13.58 ± 0.29	Total = 51.4 ± 6.1, Males = 55.1 ± 6.8, Females = 56.9 ± 6.3	Total = 115.5 ± 25.2 ***TMT-A:*** Total = 101.2 ± 12.9 ***CRT:*** Total = 114.3 ± 25.8 ***SiRT:*** Total = 94.7 ± 5.3 ***DSF Test:*** Total = 126.0 ± 38.04 ***AI (MoCA):*** Not available 5-CMT: Not available	**Total =** 5.7 ± 0.2 ***TMT-A:*** **Total =** 5.7 ± 0.4 ***CRT:*** **Total =** 6.1 ± 0.3 ***SiRT:*** **Total =** 5.8 ***DSF Test:*** **Total =** 7.0 ± 1.4 ***AI (MoCA):*** Not available **5-CMT:** Not available

	ACE-III: n = 1			ACE-III:176.8 ± 8.6	**ACE-III:** Not available
Intelligence	*Total:* n= 5 ***IQ***: n= 3 ***MR:*** n= 1 ***AFQT:*** n = 1	***IQ:*** Total = 102.3 ± 10.6 ***MR:*** Total = 18.1 ***AFQT:*** Total = 61.8 ± 0.9	Total = 54.8 ± 3.9	Total = 91.4 ± 11.2 ***IQ:*** Total = 100.6 ± 19.1 ***MR:*** Not available ***AFQT:*** Not available	**Total =** Not available ***IQ:*** Not available ***MR:*** Not available ***AFQT:*** Not available
Executive Function *Letter Cancellation*	*Total:* n= 3 ***LSST***: n= 1 ***LCCS***: n= 1 ACE-Language: n = 1	***LSST:*** Total = 282 ***LCCS:*** Total = 50 ± 7.3 ACE-Language: Total = 21.63 ± 0.37	Total = 49.27 ± 2.15	*Total* = 97.5 ± 9.7 ***LSST:*** Not available ***LCCS:*** *Total* = 89.17 ± 9.77 ***ACE-Langugae:*** *Total* = 176.8± 9.7	**Total =** 5.8 ***LSST:*** **Total =** 5.8 ***LCCS:*** Not available **ACE-Language:** Not available
Verbal Fluency	*Total:* n= 9 ***WFT***: n= 4 ***VIS (MoCA):*** n= 1 ***BeDT:*** n= 1 ***BuDT:*** n= 1 BNT: n = 1 WRT: n = 1 ACE-VF: n = 1	***WFT:*** Total = 29.7 ± 4.7, Male = 25.67 ± 6.4, Female = 24.81 ± 6.2 ***VIS (MoCA):*** Total = 6.48 ± 0.92 ***BeDT:*** Male = 12, Female: 12 ***BuDT:*** Male = 7, Female: 6 BNT: Total = 28 ± 2 WRT: Total = 30.4 ± 5.6 ACE-VF: Total = 4.38 ± 0.28	Total = 54.9 ± 5.8, Male = 56.6 ± 7.1, Female = 56.2 ± 7.1	Total = 97.6 ± 25.5 ***WFT:*** Total = 86.2 ± 20.9 ***VIS (MoCA):*** Not available ***BeDT:*** Not available ***BuDT:*** Not available BNT: Not available WRT: Total = 129.5 ± 39.9 ACE-VF: Total = 176.8 ± 5.58	**Total =** Not available ***WFT:*** Not available ***VIS (MoCA):*** Not available ***BeDT:*** Not available ***BuDT:*** Not available **BNT:** Not available **WRT:** Not available **ACE-VF:** Not available
Processing Speed	*Total*: n= 20 ***TMT-B:*** n= 8 ***TrB-A: n = 2*** ***STIT:*** n= 4 ***WMT:*** n= 1 ***CES***: n= 3 ***RVP (CANTAB):*** n= 1 ***SCWT:*** n= 1 ***EIS (MoCA):*** n= 1 ***LT***: n= 1 VSS: n = 2 SWME: n = 1	***TMT-B:*** Total = 94.6 ± 3.9 ***TrB-A: Total = 0.81 ± 0.69*** ***STIT:*** Total = 34.2 ± 5.14 ***WMT:*** Total = 2.3 ± 1.1 ***CES:*** Total = 48.9 ± 0.1, Male = 52.73 ± 14.37, Female = 54.66 ± 0.05 ***RVP (CANTAB):*** Total = 0.9 ***SCWT:*** Total = 19.1 ***EIS (MoCA):*** Total = 11.6 ± 1.4 ***LT:*** Male = 39.8 ± 17.8, Female = 45.5 ± 26.6 VSS: Male = 329.13 ± 78.4, Female = 348.58 ± 78.27 SWME: Total = 2.66 ± 1.75	Total = 53.26 ± 5.29, Male = 56.6 ± 7.1, Female = 56.2 ± 7.1	Total = 98.66 ± 26.53 ***TMT-B:*** Total = 89.74 ± 22.76 TrB-A: Not Available ***STIT:*** Total = 89.74 ± 22.76 ***WMT:*** Total = 94.4 ± 9.35 ***CES:*** Not Available ***RVP (CANTAB):*** Not Available ***SCWT:*** Not Available ***EIS (MoCA):*** Not available ***LT:*** Not available VSS: Not Available SWME: Not Available	**Total =** 7.0 ± 1.37**, Male =** 5.58 ± 0.19, **Female = 5**.52 ± 0.12 ***TMT-B:*** **Total = 7.0 ± 1.37,** ***STIT:*** **Total = 7.0 ± 1.37,** ***WMT:*** **Not available** ***CES:*** **Male = 5.58 ± 0.19, Female = 5.52 ± 0.12** ***RVP (CANTAB):*** **Not available** ***SCWT:*** **Not available** ***EIS (MoCA):*** **Not available** ***LT:*** **Not available**
Global Cognition	*Total*: n= 23 ***MMSE:*** n= 14 ***MoCA:*** n= 7 ***IQCODE:*** n= 1 ***CAMCOG:*** n= 1 ***NART:*** n= 1 ***IST:*** n= 1 ***BPP:*** n= 1 ***ACE:*** n= 2 ***HRS-CS:*** n= 1 ***MINT:*** n = 1	***MMSE***: Total = 26.9 ± 1.3 ***MoCA***: Total = 24.7 ± 3.7, Male = 25 ± 2.9, Females = 25.5 ± 2.9 ***IQCODE***: Total = 43.4 ± 3.01 ***CAMCOG:*** Total = 90 ***NART:*** Total = 28 ***IST:*** Total = 32.4 ***BPP:*** Total = 46.9 ***ACE:*** Total = 87.5 ± 0.4 ***HRS-CS:*** Total = 14.31 ± 4.06, Male = 14.2 ± 4.15, Female = 14.44 ± 3.96 ***MINT:*** Total = 31	Total = 54.4 ± 5.8, Males = 63.9 ± 0.7, Females = 63.9 ± 0.6	Total = 97.5 ± 20.3 ***MMSE:*** Total = 97.1 ± 20.4 ***MoCA:*** Total = 102.9 ± 13.7 ***IQCODE:*** Total = 96.4 ± 13.8 ***CAMCOG***: Total = 86.4 ***NART:*** Total = 86.4 ***IST***: Not available ***BPP:*** Not available ***ACE:*** Total = 176.8 ± 8.6 ***HRS-CS:*** Not available ***MINT:*** Total = 176.8 ± 8.6	**Total = 5.02 ± 0.8** ***MMSE:*** **Total = 5.02 ± 0.8** ***MoCA:*** **Not Available** ***IQCODE:*** **Not Available** ***CAMCOG:*** **Not Available** ***NART:*** **Not Available** ***IST:*** **Not available** ***BPP:*** **Not available** ***ACE:*** **Not available** ***HRS-CS:*** **Not available** ***MINT:*** **Not Available**
Inductive Reasoning	*Total:* n= 2	***AH-4:*** Total = 73.1, Male = 49.2 ± 9.5, Female = 42.9 ± 11.6	Total = 52.8, Males = 55.1 ± 5.9, Females = 55.3 ± 5.9	Total = 86.4	**Not available**

Psychomotor Speed	*Total:* n= 6	***SDMT:*** Total = 54.3 ± 10.02, Female = 50.5	Total = 46.7 ± 2.6, Females = 50.1 ± 2.6	Total = 92.9 ± 1.7	**Not available**
Visuospatial Organisation	*Total:* n= 7 ***BDT:*** n= 1 ***VIS MoCA:*** n= 1 ***Vr-D:*** n = 1 ***VSA:*** n = 1 HVOT: n = 1 CDT: n = 1 FC: n = 1	***BDT:*** Total = 25.6 ± 0.6 ***VIS MoCA:*** Total = 6.48 ± 0.92 ***Vr-D:*** Total = 8.62 ± 3.24 ***VSA: Total = 11.65 ± 0.34*** ***HVOT: 25 ± 3*** ***CDT:*** Total = 2.1 ± 1.1, Male = 28 ± 5, Female = 55 ± 10 FC: Total = 9.1 ± 7.8	Total = 51.2 ± 7.4	Total = 127.9 ± 35.7 ***BDT:*** Total = 92.9 ± 1.6 ***VIS MoCA:*** Not available VSA: Total = 176.8 ± 5.58 HVOT: Not available ***Vr-D:*** Not available CDT: Total = 129.5 ± 39.9 FC: Total = 129.5 ± 39.9	**Total = Not available** ***BDT:*** **Not available** ***VIS MoCA:*** **Not available** **VSA: Not available** **HVOT: Not available** ***Vr-D:*** **Not available** **CDT: Not available** **FC: Not available**
Temporal Orientation	*Total: n = 2*	*Total = 6.6* ± 0.9	Total = 54.5 ± 7.1	Not available	**Total = 6.95 ± 1.5**

Abbreviations: *ACE*, Addenbrooke's cognitive examination, *AH-4*, Alice Heim 4-I, *BDT:* Block Design Test; *BeDT:* Benson Delay Test; *BNT:* Boston Naming Test; *BP*, Blood Pressure, *BPP*, Børge Priens Prøve, *BuDT: Buschke Delay Test; CAMCOG*, Cambridge Cognition Examination, *CANTAB*, Cambridge Neuropsychological Test Automated Battery, *CDT:* Clock Drawing Test; *CERAD*, Consortium to Establish a Registry for Alzheimer's Disease, *CMS*, Chinese Clinical Memory Scale, *CRT:* Choice Reaction Time; *CVLT*, California Verbal Learning Test *DSB*, Digit Span Backwards, *CES:* Composite Executive Score; *DSF*, Digit Span Forward, *DSST*, Digit Symbol Substitution Test, *EBM:* East Boston Memory Test; *EIS*, Executive Index Score, *GCA,* General Cognitive Ability; *HRS-CS*, U.S. Health and Retirement Study Composite Score, *IST*, Intelligenz-Struktur-Test, *IQCODE*, Informant Questionnaire on Cognitive Decline in the Elderly, *IQ*, Intelligence Quotient, *LCCS:* Letter Cancellation Composite Score; *LSST*, Letter Search Speed Test, *LT:* Labyrinth Test*; McNS:* McNair Survey; *MINT*, Multilingual Naming Test, *MIS:* Memory Index Score; *MoCA*, Montreal Cognitive Assessment, *MMSE*, Mini-Mental State Exam, *MR:* Mental Rotation Test; *MVT:* Mill Hill Vocabulary Test; *NART*, National Adult Reading Test, *PFT: Phonemic Fluency Test; RAVLT*, Rey Auditory Verbal Learning Test, *ROCF*, Rey–Osterreith complex figure, *RVP:* Rapid Visual Processing*; SCWT:* Stroop Colour Word Test; *SDMT*, Symbol Digits Modalities Test, *SCS:* Spatial Composite Score; *SFT:* Semantic Fluency Test; *SRT:* Selective Reminding Test; *SiRT*: Simple Reaction Time; *STIT:* Stroop Test (Interference Time)*; STW:* Spot the Word Test; *TMT-A*, Trail making Test Part A, *TMT-B*, Trail making Test Part B, *TrB-A*, Trail making Test Difference between Part B and A, *VIS*, Visuospatial Index Score, *VRT:* Visual Reproduction Test; *VSS:* Visual Search Speed; *WFT:* Word Fluency Test; *WDS:* WAIS-IV Digit Sequencing*; WAIS*, Wechsler Adult Intelligence Scale; *WMT:* Word Matching Test*; 5-CMT:* - Choice Movement Test.

Table 7.3. *Summary of weighted FBG and HbA1c levels, and prevalence of diabetes and MetS across included studies (reference studies applied).*

	FBG (mg/dl)	No Studies (n=)	No. Participants (n=)	HbA1c (mg/dl)	No Studies (n=)	No. Participants (n=)	Diabetes (n=)	No Studies (n=)	MeTS (n=)	No Studies (n=)
Total	93.2 ± 19.2	28	34,814	4.67 ± 0.54	10	21,219	With: 14,841 Without: 104,205	50	With: 1,195 Without: 8,128	**6**
Male	95.4 ± 10.1	1	127	5.59 ± 0.45	2	1,763	With: 1,000 Without: 7,084	8	-	**-**
Female	93.6 ± 9.7	1	164	5.56 ± 0.41	2	1,735	With: 790 Without: 7,952	7	With: 635 Without:1,514	**1**

Table 7.4. *Summary of negative relationships between diabetes metrics and cognition at midlife by study design and quality.*

Study Design	Memory	Attention	Executive Function	Global Cognition	Psychomotor Speed	Inductive Reasoning	Intelligence	Visuospatial Organisation	Temporal Orientation
Diabetes									
Individual Study Cohorts	(428, 735, 737, 751, 827-833)	(751, 828, 832)	(428, 735, 751, 827-832, 834)	(736, 749, 759, 835-838)	(832)	(839)	-	-	-
Longitudinal Study Cohorts	(410, 729, 730, 840)	(729, 730)	(410, 730, 840)	(730)	-	-	-	(729)	-
Study Quality (n=)	Low: 5 Moderate: 6 High: 3	Low: 1 Moderate: 2 High: 2	Low: 4 Moderate: 7 High: 2	Low: 1 Moderate: 6 High: 2	Low: - Moderate: 1 High: -	Low: - Moderate: 1 High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: 1	Low: - Moderate: - High: -
FBG									
Individual Study Cohorts	(735, 828, 841-843)	(828, 832, 842)	(735, 828, 832, 842, 843)	(836, 843, 844)	(832)	-	-	-	-
Longitudinal Study Cohorts	(410, 733)	-	(410)	-	-	-	-	-	-
Study Quality (n=)	Low: 3 Moderate: 3 High: 1	Low: 1 Moderate: 1 High: 1	Low: 3 Moderate: 2 High: 1	Low: 1 Moderate: 2 High: -	Low: Moderate: High:	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -
HbA1c									
Individual Study Cohorts	(751, 832, 843)	(751, 832)	(751, 832, 843, 845)	(836-838, 843)	-	-	-	-	-
Longitudinal Study Cohorts	(410)	-	(410)	-	-	-	-	-	-
Study Quality (n=)	Low: 2 Moderate: 1 High: 1	Low: 0 Moderate: 1 High: 1	Low: 2 Moderate: 1 High: 2	Low: 2 Moderate: 2 High: 0	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -
MeTS									
Individual Study Cohorts	(843, 846)	(847)	(843)	(839, 843)	-	(839)	-	-	-
Longitudinal Study Cohorts	-	-	-	-	(848)	-	-	-	-
Study Quality (n=)	Low: 2 Moderate: - High: -	Low: - Moderate: 1 High: -	Low: 1 Moderate: - High: -	Low: 1 Moderate: 1 High: -	Low: - Moderate: 1 High: -	Low: - Moderate: 1 High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -

7.5. Discussion

The present review aimed to systematically determine the impact of midlife T2DM on cognitive function at mid and later life. Qualitative analysis revealed 66.6% of longitudinal cohort studies found a negative relationship between midlife T2DM and some measures of cognition, including memory, executive function, and global cognition in later life. In contrast, findings from the 3 independent longitudinal studies were inconsistent with both negative and null findings reported. The majority of studies reported no identifiable relationship between midlife T2DM and cognition at midlife across all cognitive domains. Most studies also found no relationship between the midlife metabolic correlates of T2DM, FBG and HbA_1c levels, and cognition at midlife across all cognitive domains. Conflicting findings were found between cognition and MetS at midlife, although limited to very few studies. Findings from our meta-analysis revealed that midlife T2DM had a negative effect on memory, executive function, and global cognition at midlife. These meta-analytic findings include studies ranging from low to high quality evidence across each domain.

With functional changes first reported in memory, processing speed and arithmetic ability in the early 20^{th} century, growing evidence suggests that sub-domains of cognition are impacted at different rates among those with diabetes (849). Our results support the idea that some but not all cognitive domains are affected at different rates from midlife into later life with long-term exposure to T2DM. The majority of the studies in our review found a negative relationship between midlife T2DM and later life cognition across select domains of memory, executive function, and global cognition. The negative relationships in later life may be related to the structural changes associated with advancing age. Although this review did not include assessment of imaging data, previous research has shown that cognitive decline is associated with higher grey matter volume loss among various cortical regions relating to executive and memory function among middle-aged diabetic adults (850). Subtle cortical atrophy identified among T2DM is unequally distributed and presents with spatially-specific and regional abnormalities among the hippocampus and middle temporal gyrus with notable symmetry (851). Structural-related alterations between cognitive decline and T2DM are evident with neurological abnormalities such as lacunar infarcts and cerebral atrophy (473, 852), and regional atrophy of the hippocampus and occipital lobes (853). These localized and widespread brain abnormalities in T2DM collectively may be attributable to several mechanisms including chronic elevations in blood glucose causing oxidative stress, neuronal dysfunction and subsequent apoptosis, endothelial dysfunction, chronic inflammation damaging the blood brain barrier (BBB), and divergent

changes in CBF (808, 854) . Domain specific rates of decline rather than global cognitive decline from midlife T2DM are apparent from our results. Our results are in agreement with a previous systematic review which found that T2DM was associated with reduced cognition in later life among memory, executive function, and global cognition across almost all 17 included studies; although statistical significance was not apparent in all studies (855). In the context of an ageing population, intervention strategies at early and critical timepoints such as midlife are warranted to reduce the risk of later life cognitive decline among those with T2DM.

The majority of studies in the present review found no discernible relationship between T2DM and cognition at midlife. Those studies that did report a negative relationship between midlife T2DM and cognition were in the cognitive domains of memory, executive function, and global cognition. Some of our results are in line with previous meta-analysis across 24 studies which found memory and executive function were negatively impacted by T2DM (856). These findings were however of small-to-moderate effect sizes and study population aged 50-85 years differed to our population of 40-65 years. There is however previous systematic evidence of a positive association between T2DM and cognition reported with less cognitive decline from 5 of 13 studies; although changes in cognitive function were evidently small being less than a 3-point difference on the Mini-mental state examination (MMSE) over a 6-12 month follow-up (783). We found most studies reported no relationship between FBG or HbA_1c and cognition mainly among memory, attention, executive function, psychomotor speed, and global cognition. These results are in line with previous systematic evidence reporting close to 80% of cross sectional studies found no relation between FBG and cognition (857). The limited number of studies that did report a significant relationship in their review were inconsistent with negative, positive, and bell-shaped associations reported in less than 10% of included studies. We also report that HbA1c levels at midlife did not affect memory, attention, executive function, and global cognition in more than half of included studies. These findings are similar to previous evidence where 53% of cross-sectional studies had no association between HbA1c concentrations and cognitive function across memory, executive function, and attention (857). In all, a better understanding of the long-term effects of metabolic correlates of midlife T2DM will enable more insightful conclusions to be drawn about the cognitive decline trajectories with age and the impact metabolic disease states confer to its development for suitable intervention and management strategies to be put in place.

Our meta-analysis found that memory, executive function, and global cognition, but not attention, at midlife were negatively impacted by T2DM. These findings contradict the qualitative findings that T2DM does not affect cognition at midlife across most studies. This is highly significant given that the negative relationships among the select cognitive domains at midlife appear to parallel our later life results. Recent meta-analysis found that those with diabetes and experiencing depression exhibited greater declines in memory, executive function, and global cognition, and no significant differences were found for attention (858). However, the study population included all those aged ≥18 years and had a clinical diagnosis of depression. To our knowledge our meta-analytic findings are the first to analyse cognition and T2DM at midlife. What strengthens our ability to conduct a meta-analysis is the stringent search criteria and defined population. Previous systematic evidence on the topic of the diabetes-cognition relationship has been hindered by the heterogeneity in study populations, cognitive outcomes, and broad search strategy. The three select cognitive domains of memory, executive function, and global cognition are negatively impacted at midlife and appear to continue to decline into later life. Our results thus support previous evidence that those with T2DM are at an increased risk of accelerated cognitive decline and dementia with a higher propensity for subtle changes in cognition with age (453, 454, 859). There are multiple cross-sectional and longitudinal investigations in support of this relationship however the underlying cause(s) remains unclear (860, 861). Nonetheless, it is from midlife onwards when the moment cognitive ageing is supported by the presence of T2DM among select domains that remains to be clearly identified.

7.6. Limitations

The present review has several limitations. Firstly, the methods of test administration varied considerably across all studies. This lack of consistency reduced cross-study comparison. This was also accompanied by a large variation in follow-up times among longitudinal studies, limiting our ability to fully represent the relationship between midlife diabetes and later life cognition. Second, the inclusion of cofounding variables such as age, hypertension, dyslipidaemia, and educational attainment limited our ability to extract data for quantitative analysis. Thirdly, the generalisability of results may be hindered by differences in the number of males and females across studies as well as ethnic and racial disparities. Fourthly, our meta-analysis was limited as several non-standardised neurocognitive tests were employed and raw data was not included. The meta-analysis must therefore be considered a sub-analysis of all included studies in the present review and a measure of performance decrements

rather than clinical significance. Finally, all cognitive tests were grouped by cognitive domain, while most tests of cognition will in fact utilise several cognitive domains.

7.7. Conclusion

The risk of diabetes-related cognitive dysfunction with an ageing population globally is of current and future concern. T2DM and associated cognitive decline pose a significant burden and hurdles to patients and global healthcare alike. A negative relationship between midlife T2DM and cognitive function were reported for memory, executive function, and global cognition at both midlife and later life. Despite qualitative findings suggesting no relationship across a proportion of studies, findings from our meta-analysis suggest a negative effect on memory, executive function, and global cognition. Cognition must be considered as multidimensional, with each of these cognitive domains making an independent contribution to health-related quality of life, such that these three domains work in unison to manage daily living and behaviour including medication management, exercise, and self-care (862-864). Given the select cognitive domains negatively affected by the presence of T2DM at later life and midlife, future studies should aim to uncover the neuropathogenesis surrounding the relationship between T2DM and cognition. Personalised intervention strategies during this vulnerable period of ageing would allow targeted and clinically founded management strategies to promote healthy cognitive ageing among those with T2DM.

7.8. Question(s) Raised

- What is the relationship between significant CV risk factors like cholesterol on cognition which remains poorly understood and was not included in the most recent Lancet Commission on dementia prevention, intervention, and care?

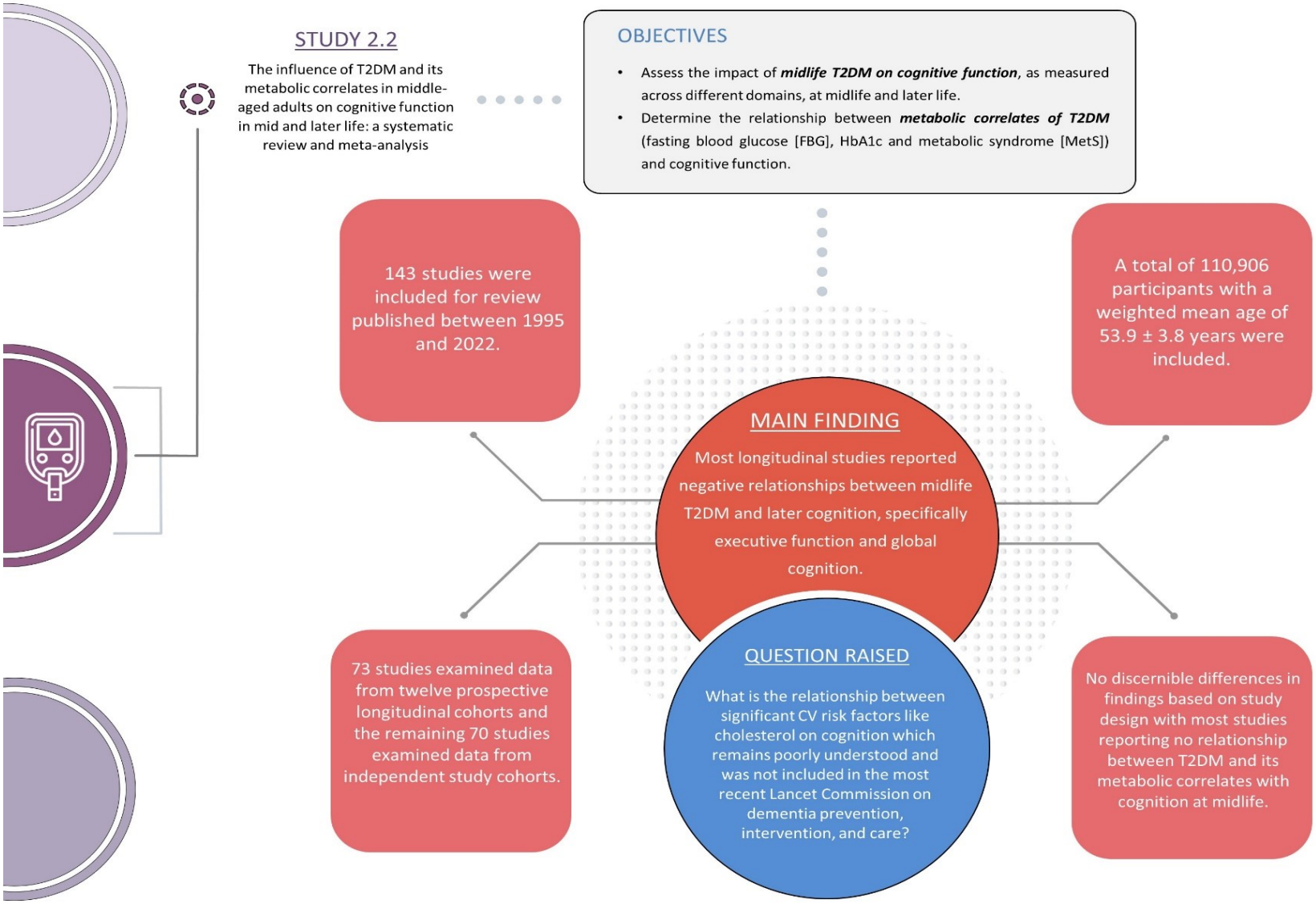

Figure 7.7. *Chapter Seven – Study 2.2 Summary.*

8

Chapter Eight:

The influence of Cholesterol and its Sub-Components in Middle-Aged Adults on Cognitive Function in Mid and Later Life: A Systematic Review

8.1. Highlights

- 103 studies published between 1995 and 2022 were included.
- 195,210 participants with a weighted mean age of 62.6 ± 5 years were included.
- Forty-five studies examined subsets of data from six prospective longitudinal cohorts with the remaining 58 studies examined data from individual study cohorts.
- There was inconsistent reporting on the relationship between midlife cholesterol metrics and later-life cognitive function.
- Based on study design and study quality there were no discernible differences in the relationship between cholesterol metrics and measures of cognition at midlife.

8.2. Introduction

With an increasing and aging population, the number of people globally aged 60 years and over is expected to double by 2050, bringing with it an elevated risk and susceptibility to Alzheimer's Disease (AD) and cognitive decline (865, 866). Several risk factors of cardiovascular disease, such as diabetes, obesity, and hypertension, have been strongly linked to lower cognitive function with advancing age (430, 867, 868). However, the relationship between cholesterol and cognitive function remains less understood. The recent Lancet Commission on dementia prevention, intervention, and care suggests that up to 40% of all dementia cases are linked to modifiable risk factors (414). Although cholesterol and dyslipidaemia were not included, the presence of dyslipidaemia and high cholesterol at midlife have been linked to reduced cognitive function in later life (869). With the global public health challenge of dementia and cognitive impairment (433) and no current treatments available, identification of all modifiable risk factors are critical for prevention.

Cholesterol is required for healthy neuronal structure and metabolic activity but has been linked to the development of neurodegenerative disorders associated with cognitive decline (436, 870, 871). The synthesis of cholesterol in the central nervous system primarily occurs through the mevalonate pathway and is vital for creating the myelin sheath that insulates neurons and speeds up nerve signals. Total cholesterol (TC), encompassing both high-density lipoprotein (HDL) and low-density lipoprotein (LDL), plays a pivotal role in the interplay between cardiovascular (CV) health and cognition. While

HDL cholesterol, often known as "good cholesterol," contributes to CV well-being by aiding in the removal of LDL cholesterol from the bloodstream and reducing inflammation and oxidative stress, LDL cholesterol, or "bad cholesterol," is relevant due to its association with atherosclerosis and vascular dysfunction. Elevated LDL cholesterol levels can lead to reduced blood flow to the brain, potentially increasing the risk of vascular dementia and impacting cognitive function. Furthermore, high LDL cholesterol has been linked to conditions like Alzheimer's disease and increased oxidative stress, which may contribute to cognitive impairment. The management of cholesterol levels, encompassing both HDL and LDL, is integral to promoting cognitive health, particularly for individuals at risk of CV-related cognitive impairments. However, the rate of cognitive decline among select domains with age can be viewed as a surrogate marker of metabolic dysregulation and neural mechanics in tandem with the pace of change in TC (265). Lipid composition within cell membranes can lead to domain-specific and dependent changes in age-related, non-pathological cognitive decline and neuronal survival (872, 873). Specifically at midlife, deterioration and reduced functional capacity of the longitudinal frontotemporal fibres impacting executive function have been linked to high levels of TC (874). Pathogenic risk factors induced by high cholesterol during midlife can lead to metabolic dysregulation (416, 875) inducing possible cognitive decline (876). Dyslipidaemia acts as a contributing force in the development of neuropathological states from midlife onwards where higher levels of TC is predictive of reduced cognitive function and capacity (440) and correlated with a 27% increased risk of dementia in later life (439). Conversely, a moderate decrease in TC from midlife onwards is correlated with a 3.5-fold increase in the risk of cognitive impairment after more than two-decades suggestive of a protective effect on cognitive status and function (877). The impact of TC on age-related cognitive decline and neural trajectories is more pronounced in midlife (447, 448). However, the evidence of TC as a significant risk factor for cognitive decline remains unsettled with LDL versus HDL exerting different effects on cognitive status. Current available research is conflicting and unclear citing high TC during midlife is a significant vascular risk factor and predictor for early onset dementia and cognitive decline (428, 438, 439, 877-879), while others have reported no effect (878) or even a positive effect on cognitive function (880, 881).

While the relationship between levels of cholesterol, its subcomponents (LDL, HDL, and triglycerides) and cognitive function has been extensively studied, findings are inconsistent. Currently there is no clear consensus as to whether intervention and management of cholesterol at midlife reduces risk of cognitive decline, despite some evidence indicating potential benefits of management across the adult lifespan (403-405). Therefore, the primary aim of this review was to systematically

determine the relationship between measures of cholesterol, triglycerides, and metabolic disorders of cholesterol measured at midlife and cognitive function in later-life. The secondary aim was to investigate the relationship between measures of cholesterol at midlife and cognitive function at midlife.

8.3. Methods

This review is a sub-analysis of the overall registered review on CV risk factors and cognitive health; see **Chapter Two Section 2.5.** for further details.

8.3.1. Search Strategy

The online databases EMBASE, MEDLINE, PubMed, Web of Science, and CINAHL from their inception until May 2022 were searched using "*cholesterol*" and a combination of key words and Mesh terms for cognition, midlife, and later life. The full search strategy is outlined in the accompanying **Appendices.** There were no language or date constraints applied. **Figure 8.1** depicts the search methodology's step-by-step approach.

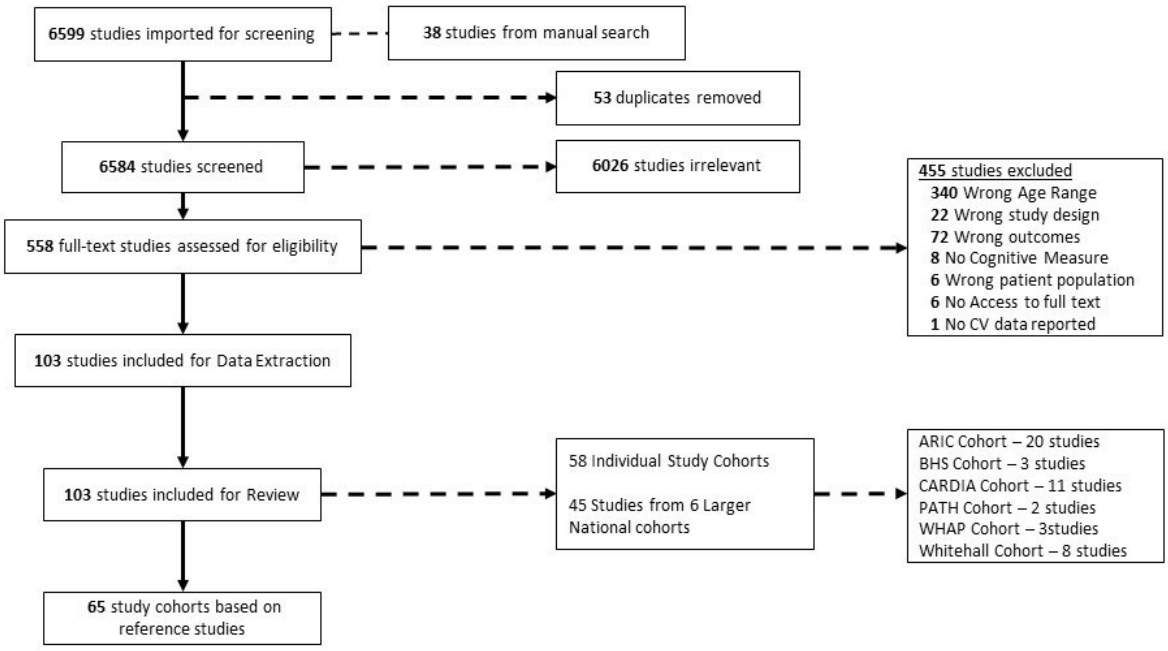

Figure 8.1. *Flow chart of the study selection process.*

8.3.2. Eligibility Criteria

Studies deemed eligible included those with human participants, cholesterol metrics at *midlife* (adults aged between 40-65 years, defined by World Health Organisation [WHO]), later life (adults aged >65-years old, defined by WHO), or both to establish the longitudinal relationships between midlife cholesterol and later life cognition. Cognition was divided into several domains, including but not limited to memory, attention, executive function, global cognition, and intelligence (see **Table 8.2 and Supplemental Table 8.3**). For full details of the eligibility criteria please see **Chapter Two Section 2.5.2**.

8.3.3. Data Extraction

Data extraction was carried out in accordance with the STROBE guidelines (546) using Endnote version 20 and Microsoft Excel. Studies were assigned a reference number and data including study aims, participant characteristics, measures of cognition and cholesterol alongside relevant outcome data as group means, standard deviation (SD), standard error (SE), statistical significance, and precision estimates were extracted. For multiple articles identified from a single study, preference

was given to the most recent publication with the longest follow-up period or the most comprehensive reporting of relevant data. Where data provided in the study was not sufficient, authors were contacted for further information. Cognitive outcomes were grouped according to cognitive each domain where applicable, and data was extracted accordingly (see **Table 8.2**.).

8.3.4. Risk of Bias and Methodological Assessment

The Appraisal Tool for Cross Sectional Research (AXIS) was used to assess the methodological quality of the included studies (547). As seen in Chapters Six and Seven and the work of McHugh and colleagues (484), the study's quality was rated as either low, moderate, or high. For full details of the eligibility criteria please see **Chapter Two Section 2.5.4**.

8.3.5. Statistical Analysis

Descriptive analysis of cholesterol-related metrics was calculated, including dyslipidaemia, hypercholesterolemia, and lipid-lowering medication use. The weighted mean of all cognitive measures (cognitive-specific domains and accompanying neuropsychological tests), TC, HDL-C, LDL-C, and triglyceride (TG) levels were calculated from all included studies and extracted data. Qualitative analysis was used to assess the associations between midlife cholesterol and cognition at later life and midlife according to each cognitive domain; positive, negative, or neutral. A meta-analysis was not deemed possible based on the extracted data.

8.4. Results

8.4.1. Literature Search

Figure 8.1 displays an overview of the study selection. In all, 6,599 records were found during the initial search accompanied by 38 records from a manual search. Following removal of duplicates and title and abstract screening, 558 complete texts were examined. As a result, 455 studies were excluded for reasons outlined in **Figure 8.1**. The authors of six papers were approached for full-text access but were later eliminated owing to a lack of response. A total of 103 studies published between 1995 and 2022 were included, 58 studies from individual study cohorts and 45 studies from larger

national cohorts resulting in 6 reference studies, resultant in 65 study cohorts included for demographics and weighted means of cholesterol and cognitive metrics.

8.4.2. Methodological and Risk of Bias Assessment

Overall, studies were deemed of moderate-to-high quality; with 19 low-quality, 40 moderate-quality, and 44 high-quality studies. Unfavourable commonalities emerged across studies for several categories, specifically failure to justify their sample size (n = 89), to classify non-responders (n = 88), to provide information regarding non-responders (n = 94), and to provide a clear assessment of statistical significance (n = 33).

8.4.3. Study Characteristics

Of the 103 studies included, 87 included both males and females, 9 assessed males, and 7 assessed females only. Forty-five studies examined subsets of data from six prospective longitudinal cohorts (see **Supplementary Material**). The remaining 58 studies examined data from individual study cohorts. Studies were conducted in 26 countries, including United States (n= 45), the United Kingdom (n= 12), China (n= 4), Australia (n= 6), and Italy (n= 3).

8.4.4. Participant Characteristics

A total of 195,210 participants with a weighted mean age of 62.6 ± 5 years were included. The mean weighted BMI was 30.8 ± 4.9 kg/m2. The average height and weight were 168.5 ± 0.8 cm and 79.3 ± 8.3 kg, respectively. Studies that reported by sex (n= 57), included 78,458 men and 80,009 females. There was no discernible difference in age (57.7 ± 4.6 vs. 55.7 ± 3.9 years) or BMI (30.9 ± 2.4 vs. 30.4 ± 3.4 kg/m2), with males having a higher mean weight (88.4 vs. 78.6 kg) than females. The average height was not available by sex.

8.4.5. Total Cholesterol, HDL-C, LDL-C, Triglycerides, and Medication Use

The weighted values for TC were 179.5 ± 34.4 (n= 55), HDL-C was 43.1 ± 13.7 (n= 43), LDL-C was 94.4 ± 26.9 (n= 26) and triglycerides was 84.7 ± 46.4 (n= 29). Dyslipidaemia and hypercholesterolemia were reported in 5,610 and 18,560 participants across 8 and 11 studies, respectively (see **Table 8.2**).

In studies that reported by sex, males had higher mean weighted values for TC (179 ± 31.1 vs. 138.6 ± 24.8), HDL-C (49.1 ± 11.9 vs. 42.2 ± 10.4) and triglycerides (179.04 ± 31.1 vs 138.6 ± 24.8). Females were found to have an LDL-C value of 67.2 ± 16.3 and no values were available for males. A higher proportion of females were reported with dyslipidaemia and hypercholesterolemia than males (1,066 vs. 894 and 902 vs. 894). Conversely, a higher number of males were reported using lipid/cholesterol lowering medication than females (523 vs. 430) (see **Table 8.2** and **Figure 8.2.**).

8.4.6. Associations between Cholesterol Metrics at Midlife and Measures of Cognition in Later Life

Of the 6 longitudinal study cohorts, 4 included measures of midlife cholesterol and later life cognition. A negative relationship was reported in a single study between executive function and TC, LDL-C, HDL-C and triglycerides; between memory and TC and triglycerides and between global cognition and TC, triglycerides (732). The remaining three studies found no relationship with cholesterol metrics at midlife and later life cognition (457, 733, 882).

Of the 58 individual study cohorts, 6 reported on the relationship between midlife cholesterol and later life cognitive function. Three studies reported negative associations between TC and cognitive measures (memory, executive function, processing speed, and global cognition) (416, 437, 883, 884) and for HDL-C, LDL-C and triglycerides and cognitive measures (memory, attention, global cognition, spatial ability, and perceptual speed) (437, 884, 885). A positive association between TC, HDL-C, and hypercholesterolemia at midlife and cognitive measures, including executive function, processing speed, global cognition, and verbal learning and memory was reported in three studies (437, 832, 883).

In summary, there was inconsistent reporting on the relationship between midlife cholesterol metrics and later-life cognitive function across included studies.

Table 8.1. *Summary of longitudinal studies with negative or null relationship between cholesterol and cognitive measures at later life.*

Author	Year	Setting	Study Quality	Cognitive Variables	Relationship
An et al.	2019	Multicentre prospective, longitudinal	Moderate	Memory, attention, executive function, processing speed, and global cognition	TC: - (Executive Function, processing speed, global cognition) HDL-C: - (Global Cognition); + (Executive function, processing speed) LDL-C: - (Memory, attention, global cognition)
Cherbuin et al.	2009	Prospective, longitudinal	High	Global cognition	Lipid Lowering medication: - (Global cognition)
Henderson et al.	2003	Longitudinal	Moderate	Memory	TC, LDL-C: - (Memory)
Kaffashian et al.	2013	Prospective	High	Memory, executive function, attention, global cognition, inductive reasoning	0
Kivipelto et al.	2001	Prospective and cross-sectional analysis of population-based, longitudinal	High	Memory, attention, executive function, global cognition	TC: - (Global cognition)
Nunley et al.	2017	Prospective, observational	High	Memory, attention, executive function, global cognition, intelligence and psychomotor speed	Lipid-lowering Medication: - (Memory and psychomotor speed)
Power et al.	2017	Prospective	High	Memory, executive function, global cognition	TC, TG: - (Memory, executive function, Global cognition) HDL, LDL: - (Executive function)
Reynolds et al.	2011	Longitudinal, population-based	Moderate	Memory, global cognition, perceptual speed, verbal and spatial ability	TG: - (Spatial ability, perceptual speed, and general cognition – Females)
Szoeke et al.	2019	Longitudinal	High	Global Cognition	0
Tuligenga et al.	2014	Prospective, longitudinal	Moderate	Memory, executive function, and inductive reasoning	0
Wendell et al.	2014	Prospective	High	Memory, attention, executive function, global cognition, and visuospatial ability	TC: - (Memory); + (Memory, executive function, global cognition)
Yang et al.	2018	Prospective	Moderate	Psychomotor speed, attention, executive function, memory	Hypercholesterolemia: + (verbal learning and memory)

0, no association; -, negative association; +, positive association

8.4.7. Associations between Total Cholesterol, Associated Metrics, and Cognitive Function at Midlife

Table 8.2 details mean pooled outcomes for all midlife cognitive measures used across included studies and associated cholesterol metrics.

Of all included studies, most reported no relationship (n = 81, 78.6%) between cholesterol metrics at midlife and cognitive measures at midlife, including memory, attention, executive function, global cognition, inductive reasoning, intelligence, psychomotor speed, and visuospatial organisation. Those that reported no relationship were highest across select cognitive domains in the case of **TC** (Memory: n = 40, 85.1%; Attention: n = 17, 94.1%; Executive function: : n = 38, 71.1%; Global Cognition: n = 23, 69.6%), **HDL-C** (Memory: n = 34, 89.5%; Attention: n = 17, 94.4%; Executive function: : n = 34, 81.2%; Global Cognition: n = 20, 95.2%), **LDL-C** (Memory: n = 18, 78.3%; Attention: n = 11, 84.6%; Executive function: : n = 19, 86.4%; Global Cognition: n = 13, 92.9%), **TG** (Memory: n = 22, 88%; Attention: n = 12, 92.3%; Executive function: : n = 22, 91.7%; Global Cognition: n = 14, 82.3%), **hypercholesterolemia** (Memory: n = 9, 69.2%; Attention: n = 5, 100%; Executive function: : n = 10, 76.9%; Global Cognition: n = 8, 88.9%), **lipid lowering medication** (Memory: n = 11, 91.7%; Global Cognition: n = 5, 83.3%), and **dyslipidaemia** (Executive function: n = 4, 80%; Global Cognition: n = 3, 50%). All remaining studies reported negative relationships and were far less substantial; see **Figure 8.2.** below.

Based on study design (individual cohorts vs. large cohorts) and study quality (low vs. moderate vs. high) there were no discernible differences in the relationship between cholesterol metrics and measures of cognition (see **Tables 8.7. and Supplemental Tables 8.4. and 8.5.**).

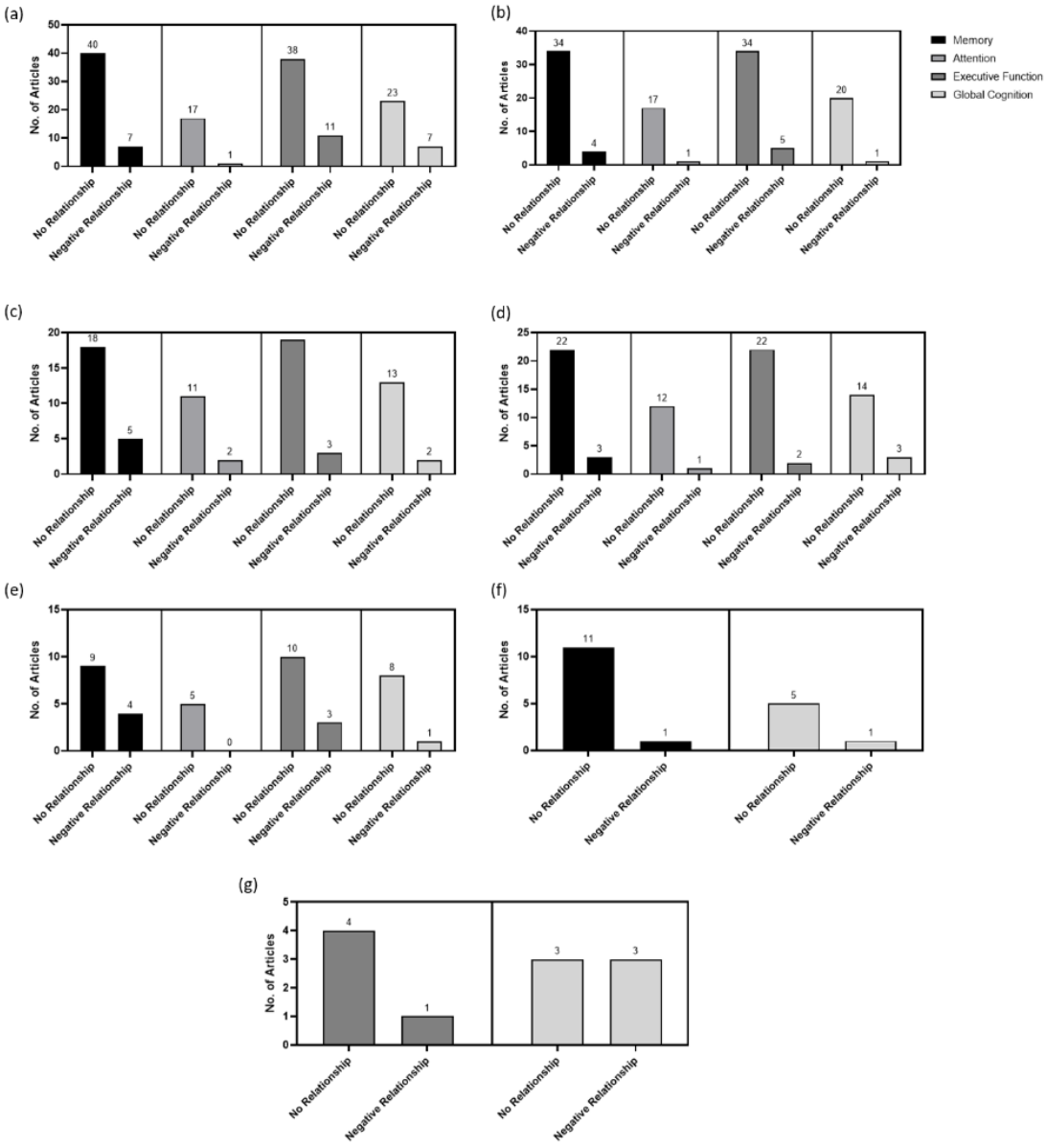

Figure 8.2. *The Null and Negative relationships between cholesterol, its sub-components, and disorders with cognitive domains of memory, attention, executive function, and global cognition at midlife*. **(a)** TC, **(b)** HDL-C, **(c)** LDL-C, **(d)** TG, **(e)** Hypercholerolemia, **(f)** Lipid-lowering medication, and **(g)** Dyslipidemia.

Table 8.2. *Summary table of weighted average for all cognitive measures and associated cholesterol metrics at baseline (i.e., midlife).*

Cognitive Variable	No. of Studies (n =)	Weighted Average (Mean ±SD)	Age (Mean ±SD; Years)	TC (mg/dL)	HDL-C (mg/dL)	LDL-C (mg/dL)	TG (mg/dL)
Memory *Verbal Memory*	*Total*: n= 10 ***Immediate***: n= 1, ***Delayed***: n= 3, ***EBM***: n= 1, ***RAVLT (Immediate & Delayed recall):*** n= 1, ***RAVLT (Learning Score):*** n= 1, ***RAVLT (Summary Metric):*** n= 3 ***SRT***: n= 1, ***ROCF (Immediate & Delayed recall):*** n= 1, ***CERAD (Immediate & Delayed):*** n= 1, ***CVLT (Immediate & Delayed):*** n= 1	***Immediate:*** Total = 14.5 ***Delayed:*** Total = 6.9 ± 1.5 ***EBM***: Total = 10 ***RAVLT (Immediate & Delayed recall):*** Total = 7.1 ± 2.7, 7.1 ± 2.8 ***RAVLT (Learning Score):*** Total = 36.8 ± 8.3 ***RAVLT (Summary metric):*** Total = 8.9 ± 3.1 ***SRT:*** Total = 34.3 ***ROCF (Immediate & Delayed recall):*** 16.1 ± 7.6, 14.9 ± 7.8 ***CERAD (Immediate & Delayed):*** Total = 7.2 ± 1.1, 7.7 ± 1.5 ***CVLT (Immediate & Delayed):*** Total = 8.8 ± 2.1, 8.8 ± 3.2	Total = 50.9 ± 4.7, Males = 56.6 ± 7.1, Females = 56.2 ± 7.1	Total = 102.8 ± 17.9, Males = 104.4 ± 18.2, Females = 104.6 ± 18.9 ***Delayed:*** Total = 81.2, Males = 104.4 ± 18.2, Females = 104.6 ± 18.9 ***Immediate:*** Total = 81.2 ***EBM:*** Not available ***RAVLT (Summary metric):*** Total = 90.6 ± 17.3 ***RAVLT (Immediate & Delayed recall, Learning Score):*** Total = 219.4 ± 39.7 ***SRT:*** Total = 106.4 ± 19.4 ***ROCF (Immediate & Delayed recall):*** Total = 219.4 ± 39.7 ***CERAD (Immediate & Delayed):*** Total = 104.9 ± 18.9 ***CVLT (Immediate & Delayed):*** Total = 104.9 ± 18.9	Total = 27.8 ± 6.6 ***Delayed:*** Total = 22.3 ***Immediate:*** Total = 22.3 ***EBM:*** Not available ***RAVLT (Summary metric):*** Total = 34.8 ± 9.5 ***RAVLT (Immediate & Delayed recall, Learning Score):*** Total = 45.3 ± 6.6 ***SRT:*** Total = 23.2 ± 5.4 ***ROCF (Immediate & Delayed recall):*** Total = 45.3 ± 6.6 ***CERAD (Immediate & Delayed):*** Total = 27.9 ± 7.56 ***CVLT (Immediate & Delayed):*** Total = 27.9 ± 7.56	Total = 74.7 ± 16.8 ***Delayed:*** Not available ***Immediate:*** Total = 49.86 ***EBM:*** Not available ***RAVLT (Summary metric):*** Total = 80.9 ± 12.1 ***RAVLT (Immediate & Delayed recall, Learning Score):*** Total = 146.6 ± 35.2 ***SRT:*** Total = 72.9 ± 18.2 ***ROCF (Immediate & Delayed recall):*** Total = 146.6 ± 35.2 ***CERAD (Immediate & Delayed):*** Not available ***CVLT (Immediate & Delayed):*** Not available	Total = 31.2 ± 16.6 ***Delayed:*** Not available ***Immediate:*** Total = 27 ***EBM:*** Not available ***RAVLT (Summary metric):*** Total = 20.9 ± 0.5 ***RAVLT (Immediate & Delayed recall, Learning Score):*** Total = 184.7 ± 104.7 ***SRT:*** Total = 23.6 ± 14.7 ***ROCF (Immediate & Delayed recall):*** Total = 184.7 ± 104.7 ***CERAD (Immediate & Delayed):*** Total = 23.9 ± 15.3 ***CVLT (Immediate & Delayed):*** Total = 23.9 ± 15.3
Episodic Memory	*Total:* n= 2	*Total* = 5.4 ± 0.2	Total = 49.8 ± 8, Males = 50.3 ± 8, Females = 51 ± 8.1	Total = 118.7 ± 14.3	Not available	Not available	Not available
Semantic Memory	*Total*: n= 1	*Total* = 15.6 ± 2.9 Males = 15.2 ± 3.0, Females = 15.9 ± 2.8	Total = 50.7 ± 8, Males = 50.3 ± 8, Females = 51 ± 8.1	Not available	Not available	Not available	Not available
Working Memory	*Total:* n= 16 ***DSST:*** n= 5 ***CMS Score:*** Total: n= 1 ***DSB Test***: n= 8 ***MIS (MoCA):*** n= 1 ***VRT***: n= 1 ***LMT:*** n = 2	***DSST:*** Total = 45.5 ± 8.5 ***CMS Score:*** Total = 76.6 ± 12.9 ***DSB Test:*** Total = 6.2 ± 1.9 ***MIS (MoCA):*** Total = 12.7 ± 2.4 ***VRT:*** Total = 11.3 ***LMT:*** Total = 10.1	Total = 52.02 ± 4.4	Total = 89.4 ± 16.4 ***DSST:*** Total = 79.3 ± 18.4, Males = 111.96 ± 19.3, Females = 124.05 ± 25.4 ***CMS Score:*** 81.1 ± 11.6 ***DSB Test:*** Total = 144.7 ± 23.6 ***MIS (MoCA):*** Not available ***VRT:*** Total = 106.4 ± 19.4 ***LMT:*** Total = 83.9 ± 0.7	Total = 26.93 ± 5.6 ***DSST:*** Total = 28.3 ± 8.7, Males = 23.8 ± 6.3, Females = 28.9 ± 7.7 ***CMS Score:*** 47.16 ± 13.3 ***DSB Test:*** Total = 108.01 ± 21.6 ***MIS (MoCA):*** Not available ***VRT:*** Total = 23.2 ± 5.4 ***LMT:*** 22.9	Total = 67.9 ± 13.0 ***DSST:*** Not available ***CMS Score:*** Total = 47.2 ± 13.3 ***DSB Test:*** Total = 108.01 ± 21.6 ***MIS (MoCA):*** Not available ***VRT:*** Total = 72.9 ± 18.2 ***LMT:*** 49.9	Total = 30.1 ± 13.2, Males = 30.06 **± 17.5**, Females = 26.8 **± 14.2** ***DSST:*** Total = 28.08 ± 15.84, Males = 30.06 ± 17.5, Females = 26.8 ± 14.2 ***CMS Score:*** Total = 28.9 ± 14.1 ***DSB Test:*** Total = 88.7 ± 43.6 ***MIS (MoCA):*** Not available ***VRT:*** Total = 23.6 ± 14.7

							LMT: Total = 26.3
Attention	*Total:* n= 9 ***TMT-A:*** n= 5 ***CRT***: n= 3 ***SiRT:*** n= 2 ***DSF Test:*** n= 5 ***AI (MoCA):*** n = 1	***TMT-A:*** Total = 42.2 ± 30.9 ***CRT:*** Total = 783.1 ± 167.5 ***SiRT:*** Total = 250.9 ± 56.7 ***DSF Test:*** Total = 9.4 ± 1.5 ***AI (MoCA):*** 16.3 ± 1.8	Total = 49.9 ± 4.6, Males = 48.1 ± 5.2, Females = 48.1 ± 5.5	Total = 109.6 ± 16.9 ***TMT-A:*** Total = 142.1 ± 22.9 ***CRT:*** Total = 123.8 ± 23.1 ***SiRT:*** Total = 101.8 ± 19.3 ***DSF Test:*** Total = 117.8 ± 14.3 ***AI (MoCA):*** Not available	Total = 29.4 ± 5.3 ***TMT-A:*** Total = 39.3 ± 7.1 ***CRT:*** Total = 32.02 ± 7.3 ***SiRT:*** Total = 28.9 ± 6.7 ***DSF Test:*** Total = 29.9 ± 3.3 ***AI (MoCA):*** Not available	Total = 70.7 ± 14.2 ***TMT-A:*** Total = 117.1 ± 23.9 ***CRT:*** Total = 62.8 ± 16.02 ***SiRT:*** Total = 62.8 ± 16.02 ***DSF Test:*** Total = 81.8 ± 11.6 ***AI (MoCA):*** Not available	Total = 41.9 ± 12.9 ***TMT-A:*** Total = 88.7 ± 43.6 ***CRT:*** Total = 51.5 ± 20.1 ***SiRT:*** Total = 20.3 ± 0.3 ***DSF Test:*** Total = 65.3 ± 26.5 ***AI (MoCA):*** Not available
Intelligence	*Total:* n= 5 ***IQ***: n= 3 ***MR:*** n= 1 ***AFQT:*** n = 1	***IQ:*** Total = 102.3 ± 10.6 ***MR:*** Total = 18.1 ***AFQT:*** Total = 61.8 ± 0.9	Total = 54.8 ± 3.9	Total = 82.7 ± 12.03 ***IQ:*** Total = 94.7 ± 15.5 ***MR:*** Not available ***AFQT:*** Not available	Total = 27.3 ± 6.7 ***IQ:*** Total = 49.9 ± 13.1 ***MR:*** Not available ***AFQT:*** Not available	Total = 53.6 ± 14.9 ***IQ:*** Total = 126.6 ± 32.3 ***MR:*** Not available ***AFQT:*** Not available	Total = 40.2 ± 19.2 ***IQ:*** Total = 168.6 ± 77.3 ***MR:*** Not available ***AFQT:*** Not available
Executive Function *Letter Cancellation*	*Total:* n= 2 ***LSST***: n= 1 ***LCCS***: n= 1	***LSST:*** Total = 282 ***LCCS:*** Total = 50 ± 7.3	Total = 52.7 ± 2.6	*Total* = 102.4 ± 21.6 ***LSST:*** Total = 102.4 ± 21.6 ***LCCS:*** Not available	Total = 40.7 ± 10.6 ***LSST:*** Total = 29.2 ± 7.4 ***LCCS:*** Total = 64.7 ± 17.1	Total = 85.1 ± 21.9 ***LSST:*** Total = 63.4 ± 18 ***LCCS:*** Total = 128.9 ± 29.7	Total = 43.5 ± 16.9 ***LSST:*** Total = 19.8 ***LCCS:*** Total = 91.8 ± 51.4
Verbal Fluency	*Total:* n= 6 ***WFT***: n= 4 ***VIS (MoCA):*** n= 1 ***BeDT:*** n= 1 ***BuDT:*** n= 1	***WFT:*** Total = 31.5 ± 8.4 ***VIS (MoCA):*** Total = 6.48 ± 0.92 ***BeDT:*** Male = 12, Female: 12 ***BuDT:*** Male = 7, Female: 6	Total = 54.2 ± 5.6, Male = 56.6 ± 7.1, Female = 56.2 ± 7.1	Total = 148.1 ± 29.5, Male = 104.4 ± 18.8, Female = 104.6 ± 18.9 ***WFT:*** Total = 148.1 ± 29.5, Male = 104.4 ± 18.8, Female = 104.6 ± 18.9 ***VIS (MoCA):*** Not available ***BeDT:*** Not available ***BuDT:*** Not available	Total = 43.3 ± 13.5 ***WFT:*** Total = 43.3 ± 13.5 ***VIS (MoCA):*** Not available ***BeDT:*** Not available ***BuDT:*** Not available	Total = 72.9 ± 18.2 ***WFT:*** Total = 72.9 ± 18.2 ***VIS (MoCA):*** Not available ***BeDT:*** Not available ***BuDT:*** Not available	Total = 23.6 ± 14.7 ***WFT:*** Total = 23.6 ± 14.7 ***VIS (MoCA):*** Not available ***BeDT:*** Not available ***BuDT:*** Not available
Processing Speed	*Total*: n= 14 ***TMT-B:*** n= 8 ***STIT:*** n= 3 ***WMT:*** n= 1 ***CES***: n= 2 ***RVP (CANTAB):*** n= 1 ***SCWT:*** n= 1 ***EIS (MoCA):*** n= 1 ***LT***: n= 1	***TMT-B:*** Total = 94.4 ± 5.5 ***STIT:*** Total = 42.9 ± 1.5 ***WMT:*** Total = 2.3 ± 1.1 ***CES:*** Total = 48.9 ± 0.1 ***RVP (CANTAB):*** Total = 0.9 ***SCWT:*** Total = 19.1 ***EIS (MoCA):*** Total = 11.6 ± 1.4 ***LT:*** Male = 39.8 ± 17.8, Female = 45.5 ± 26.6	Total = 53.03 ± 4.7, Male = 56.6 ± 7.1, Female = 53.2 ± 4.8	Total = 92.8 ± 18.2, Male = 104.4 ± 18.8, Female = 104.4 ± 18.5 ***TMT-B:*** Total = 89.5 ± 18.8, Female = 104.04 ± 17.1 ***STIT:*** Total = 214.9 ± 38.9, Male = 104.4 ± 18.8, Female = 104.6 ± 18.9 ***WMT:*** Total = 200.05 ± 25.6 ***CES:*** Total = 121.3 ± 12.6 ***RVP (CANTAB):*** Total = 99 ***SCWT:*** Male = 104.4 ± 18.2, Female: 104.6 ± 18.9 ***EIS (MoCA):*** Not available ***LT:*** Not available	Total = 24.8 ± 5.8, Female = 28.1 ± 7.6 ***TMT-B:*** Total = 24.7 ± 5.8, Female = 28.1 ± 7.6 ***STIT:*** Total = 45.3 ± 10.01 ***WMT:*** Total = 49.5 ± 7.6 ***CES:*** Not available ***RVP (CANTAB):*** Not available ***SCWT:*** Not available ***EIS (MoCA):*** Not available ***LT:*** Not available	Total = 77.1 ± 19.2, Female = 67.5 ± 16.4 ***TMT-B:*** Total = 76.7 ± 19.1, Female = 67.5 ± 16.4 ***STIT:*** Total = 146.6 ± 35.2 ***WMT:*** Total = 127.9 ± 28.3 ***CES:*** Not available ***RVP (CANTAB):*** Not available ***SCWT:*** Not available ***EIS (MoCA):*** Not available ***LT:*** Not available	Total = 32.9 ± 19.04 ***TMT-B:*** Total = 32.4 ± 18.9 ***STIT:*** Total = 184.7 ± 104.7 ***WMT:*** Total = 113.3 ± 31.4 ***CES:*** Not available ***RVP (CANTAB):*** Not available ***SCWT:*** Not available ***EIS (MoCA):*** Not available ***LT:*** Not available
Global Cognition	*Total*: n= 22 ***MMSE:*** n= 14 ***MoCA:*** n= 7 ***IQCODE:*** n= 1 ***CAMCOG:*** n= 1 ***NART:*** n= 1	***MMSE***: Total = 27.7 ± 0.7 ***MoCA***: Total = 25.3 ± 3.1, Male = 25 ± 2.9, Females = 25.5 ± 2.9 ***IQCODE***: Total = 43.4 ± 3.01 ***CAMCOG:*** Total = 90 ***NART:*** Total = 28	Total = 54.6 ± 5.3, Males = 63.9 ± 0.7, Females = 63.9 ± 0.6	Total = 98.3 ± 14.1 ***MMSE:*** Total = 103.6 ± 15.9 ***MoCA:*** Total = 86.3 ± 9.7 ***IQCODE:*** Total = 200.3 ± 35.21 ***CAMCOG***: Not available	Total = 28.3 ± 6.7, Males = 22.9 ± 3.9, Females = 27.8 ± 5.3 ***MMSE:*** Total = 24.4 ± 6.4 ***MoCA:*** Total = 25.7 ± 4.6 ***IQCODE:*** Total = 200.3 ± 35.2	Total = 63.4 ± 12.6 ***MMSE:*** Total = 69.7 ± 17.9 ***MoCA:*** Total = 49.6 ± 0.9 ***IQCODE:*** Not available ***CAMCOG:*** Not available ***NART:*** Not available	Total = 26.1 ± 8.2, Males = 29.6 ± 11.2, Females = 25.8 ± 9.9 ***MMSE:*** Total = 25.8 ± 9.9 ***MoCA:*** Total = 26.3,

	IST: n= 1 ***BPP:*** n= 1 ***ACE:*** n= 1 ***HRS-CS:*** n= 1 ***GCA:*** n = 1	***IST:*** Total = 32.4 ***BPP:*** Total = 46.9 ***ACE:*** Total = 94.9 ***HRS-CS:*** Total = 14.31 ± 4.06, Male = 14.2 ± 4.15, Female = 14.44 ± 3.96 ***GCA:*** Male = 53.1 ± 10.7, Female = 51.7 ± 10.5		***NART:*** Not available ***IST***: Total = 99 ***BPP:*** Total = 99 ***ACE:*** Total = 99 ***HRS-CS:*** Not available ***GCA:*** Total = 119.2 ± 23.9, Males = 111.9 ± 19.3, Females = 124.02 ± 25.4	***CAMCOG****:* Total = 23.4 ***NART:*** Total = 23.4 ***IST:*** Not available ***BPP:*** Not available ***ACE:*** Not available ***HRS-CS:*** Not available ***GCA:*** Total = 26.8 ± 7.6, Males = 23.8 ± 6.3, Females = 28.9 ± 7.7	***IST:*** Not available ***BPP:*** Not available ***ACE:*** Not available ***HRS-CS:*** Not available ***GCA:*** Not available	Males = 28.8, Females = 23.4 ***IQCODE:*** Not available ***CAMCOG:*** Total = 27 ***NART:*** Total = 27 ***IST:*** Not available ***BPP:*** Not available ***ACE****:* Not available ***HRS-CS****:* Not available ***GCA:*** Total = 28.1 ± 15.8, Males = 30.1 ± 17.5, Females = 25.8 ± 9.9
Inductive Reasoning	*Total:* n= 2	***AH-4:*** Total = 73.1, Male = 49.2 ± 9.5, Female = 42.9 ± 11.6	Total = 52.8, Males = 55.1 ± 5.9, Females = 55.3 ± 5.9	Male= 227.5 ± 39.1, Female = 230.9 ± 41.3	Total = 23.4 ± 10.3, Male= 53 ± 13.2, Female = 65 ± 16.6	Not available	Total = 27
Psychomotor Speed	*Total:* n= 5	***SDMT:*** Total = 43.4 ± 1.7, Female = 50.5	Total = 48.4 ± 2.6, Females = 50.1 ± 2.6	Total = 89.02 ± 4.4, Female = 104.04 ± 17.1	Total = 24.5 ± 0.3, Female = 28.08 ± 0.4	Total = 55.5 ± 4.1, Female = 67.5 ± 16.4	Total = 26.5 ± 0.4
Visuospatial Organisation	*Total:* n= 4 ***BDT:*** n= 1 ***VIS MoCA:*** n= 1 ***Vr-D:*** n = 1 ***SCS:*** n = 1	***BDT:*** Total = 25.6 ± 0.6 ***VIS MoCA:*** Total = 6.48 ± 0.92 ***Vr-D:*** Total = 8.62 ± 3.24 ***SCS:*** Male = 53.6 ± 10.7, Female = 49.2 ± 9.9	Total = 46.5 ± 8.7	Total = 115.5 ± 20.4, Male = 111.9 ± 19.3, Female = 124.02 ± 25.4 ***BDT:*** Total = 96.8 ± 1.9 ***VIS MoCA:*** Not available ***Vr-D:*** Not available ***SCS:*** Total = 119.2 ± 23.9, Male = 111.9 ± 19.3, Female = 124.02 ± 25.4	Total = 26.9 ± 6.5, Male = 23.8 ± 6.3, Female = 28.9 ± 7.7 ***BDT:*** Total = 27.6 ± 1.5 ***VIS MoCA:*** Not available ***Vr-D:*** Not available ***SCS:*** Total = 26.8 ± 7.6, Male = 23.8 ± 6.3, Female = 28.9 ± 7.7	Total = 58.9 ± 1.4 ***BDT:*** Total = 63.02 ± 1.5 ***VIS MoCA:*** Not available ***Vr-D:*** Total = 48.2 ± 1.3 ***SCS:*** Not available	Total = 27.5 ± 13.6 ***BDT:*** Total = 24.3 ± 2.2 ***VIS MoCA:*** Not available ***Vr-D:*** Not available ***SCS:*** Total = 28.1 ± 15.8, Male = 30.1 ± 17.5, Female = 26.8 ± 14.2
Temporal Orientation	*Total: n = 2*	*Total = 6.6* ± 0.9	Total = 54.5 ± 7.1	Total = 207.1 ± 61.5	Not available	Not available	Not available

Abbreviations: *ACE*, Addenbrooke's cognitive examination, *AH-4*, Alice Heim 4-I, *BDT:* Block Design Test; *BeDT:* Benson Delay Test; *BNT:* Boston Naming Test; *BP*, Blood Pressure, *BPP*, Børge Priens Prøve, *BuDT: Buschke Delay Test; CAMCOG*, Cambridge Cognition Examination, *CANTAB*, Cambridge Neuropsychological Test Automated Battery, *CDT:* Clock Drawing Test; *CERAD*, Consortium to Establish a Registry for Alzheimer's Disease, *CMS*, Chinese Clinical Memory Scale, *CRT:* Choice Reaction Time; *CVLT*, California Verbal Learning Test *DSB*, Digit Span Backwards, *CES:* Composite Executive Score; *DSF*, Digit Span Forward, *DSST*, Digit Symbol Substitution Test, *EBM:* East Boston Memory Test; *EIS*, Executive Index Score, *GCA,* General Cognitive Ability; *HRS-CS*, U.S. Health and Retirement Study Composite Score, *IST*, Intelligenz-Struktur-Test, *IQCODE*, Informant Questionnaire on Cognitive Decline in the Elderly, *IQ*, Intelligence Quotient, *LCCS:* Letter Cancellation Composite Score; *LSST*, Letter Search Speed Test, *LT:* Labyrinth Test*; McNS:* McNair Survey; *MINT*, Multilingual Naming Test, *MIS:* Memory Index Score; *MoCA*, Montreal Cognitive Assessment, *MMSE*, Mini-Mental State Exam, *MR:* Mental Rotation Test; *MVT:* Mill Hill Vocabulary Test; *NART*, National Adult Reading Test, *PFT: Phonemic Fluency Test; RAVLT*, Rey Auditory Verbal Learning

Test, *ROCF*, Rey–Osterreith complex figure, *RVP:* Rapid Visual Processing*; SCWT:* Stroop Colour Word Test; *SDMT*, Symbol Digits Modalities Test, *SCS:* Spatial Composite Score; *SFT:* Semantic Fluency Test; *SRT:* Selective Reminding Test; *SiRT*: Simple Reaction Time; *STIT:* Stroop Test (Interference Time)*; STW:* Spot the Word Test; *TMT-A*, Trail making Test Part A, *TMT-B*, Trail making Test Part B, *TrB-A*, Trail making Test Difference between Part B and A, *VIS*, Visuospatial Index Score, *VRT:* Visual Reproduction Test; *VSS:* Visual Search Speed; *WFT:* Word Fluency Test; *WDS:* WAIS-IV Digit Sequencing*; WAIS*, Wechsler Adult Intelligence Scale; *WMT:* Word Matching Test*; 5-CMT:* -Choice Movement Test.

Table 8.3. *Summary of weighted cholesterol metrics across included studies (reference studies applied).*

	TC	No Studies (n=)	No. Participants (n=)	HDLC	No Studies (n=)	No. Participants (n=)	LDLC	No Studies (n=)	No. Participants (n=)	TG)	No Studies (n=)	No. Participants (n=)
Total	179.5 ± 34.4	55	176,438	43.1 ± 13.7	43	108,811	94.4 ± 26.9	26	38,829	84.7 ± 46.4	29	61,340
Male	179.0 ± 31.1		5,610	49.1 ± 11.9		3,861	-	-	-	29.6 ± 11.2		523
Female	138.6 ± 24.8		5,265	42.2 ± 10.4		3,381	67.2 ± 16.3		1,416	23.6 ± 12.5		1,621

Table 8.4. *Summary of hypercholesterolremia, dyslipidemia, and lipid-cholesterol lowering medication across included studies (reference studies applied).*

	Hypercholesterolemia (n=)	No Studies (n=)	Dyslipidemia (n=)	No Studies (n=)	Lipid/Cholesterol Lowering Medication (n=)	No Studies (n=)
Total	18,560	11	5,610	8	6,542	16
Male	894		894		523	
Female	902		1,066		430	

Table 8.5. *Summary of negative relationships between cholesterol metrics and cognition at midlife by study design and quality.*

Study Design	Memory	Attention	Executive Function	Global Cognition	Psychomotor Speed	Inductive Reasoning	Intelligence	Visuospatial Organisation
TC								
Individual Study Cohorts	(440, 747, 841, 886, 887)	(887)	(440, 747, 883, 885)	(883)	-	-	-	-
Longitudinal Study Cohorts	(732, 888)	-	(732, 764, 882, 888-891)	(400, 732, 844, 882, 888, 891)	-	(882, 891)	-	-
Study Quality (n=)	Low: 1 Moderate: 5 High: 1	Low: - Moderate: 1 High: -	Low: 1 Moderate: 7 High: 3	Low: 1 Moderate: 3 High: 3	Low: - Moderate: - High: -	Low: - Moderate: 1 High: 1	Low: - Moderate: - High: -	Low: - Moderate: - High: -
HDL-C								
Individual Study Cohorts	(745, 747)	(745)	(747)	(745)	-	-	-	**(745)**
Longitudinal Study Cohorts	(892, 893)	-	(732, 892-894)	-	-	(893)	-	-
Study Quality (n=)	Low: - Moderate: 2 High: 2	Low: - Moderate: - High: 1	Low: - Moderate: 2 High: 3	Low: - Moderate: - High: 1	Low: - Moderate: - High: -	Low: - Moderate: 1 High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: 1
LDL-C								
Individual Study Cohorts	(745, 750, 884)	(745, 750)	(750)	(745)	(750)	-	-	**(745)**
Longitudinal Study Cohorts	(895, 896)	-	(732, 894)	-	-	-	-	-
Study Quality (n=)	Low: 1 Moderate: 3 High: 1	Low: - Moderate: 1 High: 1	Low: - Moderate: 1 High: 2	Low: - Moderate: - High: 1	Low: - Moderate: 1 High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: 1
TG								
Individual Study Cohorts	(745)	(745)	-	(745, 885)	-	-	-	**(745)**
Longitudinal Study Cohorts	(732, 892)	-	(732, 892)	(732)	-	-	-	-
Study Quality (n=)	Low: - Moderate: - High: 3	Low: - Moderate: - High: 1	Low: - Moderate: - High: 2	Low: - Moderate: 1 High: 2	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: 1
Hypercholesterolemia								
Individual Study Cohorts	(832, 897)	-	(897)	-	-	-	-	-
Longitudinal Study Cohorts	(898, 899)	-	(898, 899)	(899)	-	-	-	-
Study Quality (n=)	Low: 2 Moderate: 1 High: 1	Low: - Moderate: - High: -	Low: 2 Moderate: - High: 1	Low: 1 Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -
Lipid Lowering Medication								
Individual Study Cohorts	(440)	-	-	-	(440)	-	-	-
Longitudinal Study Cohorts	-	-	-	(900)	-	-	-	-

Study Quality (n=)	Low: 1 Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: 1	Low: 1 Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -
Dyslipidaemia								
Individual Study Cohorts	-	-	(755)	(749, 755, 836)	-	-	-	-
Longitudinal Study Cohorts	-	-	-	-	-	-	-	-
Study Quality (n=)	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: 1	Low: - Moderate: 2 High: 1	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -	Low: - Moderate: - High: -

8.5. Discussion

This review aimed to provide a better understanding of the relationship between cholesterol, its sub-components, associated metabolic disorders at midlife and their impact on cognitive function in both later life and midlife. Using qualitative analysis, we found inconsistent reporting on the relationship between midlife cholesterol and later life cognition. Of the longitudinal cohort studies 75% reported no identifiable relationship and of the individual cohort studies 50% of studies reported negative relationship and 50% reported positive relationship between midlife cholesterol metrics and all cognitive domains in later life. Those that reported a negative relationship on cognitive function included memory, executive function, global cognition, and psychomotor speed. Overall, there was high levels of inconsistency in reporting and no clear consensus to draw. Midlife cholesterol metrics and associated disorders predominantly produced no relationship with midlife cognition as reported in almost 80% of included studies across the domains of memory, attention, executive function, and global cognition. The inconsistency of findings in the present review was irrespective of study design or study quality.

Our results showcase an inconsistent relationship between midlife cholesterol and later life cognition with most studies reporting no relationship, specifically among select domains of memory, executive function, and global cognition. The assumption that elevated peripheral cholesterol at midlife can impair several domains of cognition over time remains controversial with suggested non-linear relationships (883, 901, 902). Similar to our findings, several studies have demonstrated that although midlife TC increases the risk of cognitive decline and AD (415, 416), others report null or negative findings (878, 903, 904). Our results partly support previous systematic evidence where TC levels were seen to increase up to late midlife peaking at around 60-years and subsequently decline thereafter (869). In contrast, our findings negate earlier meta-analytic evidence of 18 studies where increased TC and LDL-C at midlife heightens the risk for future cognitive decline (905). Those studies in our review reporting a negative relationship among several cognitive domains in later life affected by midlife cholesterol were evident among memory, executive function, and global cognition. These findings indicate potential for regional cognitive deterioration among a subset of the general ageing population which may exacerbate the onset of disease progression and predict future cognitive decline in later life (402, 447, 448, 730, 900, 906-910). As the brain ages, cognitive performance can become altered with a reduction in brain cholesterol catabolism leading to structural and functional neurological deficiencies among different brain regions at varying rates (911-913). Any alteration to

central homeostatic cholesterol concentration may lead to an increase in amyloid beta plaque formation, synaptic dysfunction, impaired neuron morphology, neurodegeneration, and subsequent cognitive decline (914-918). The inconsistency surrounding the cholesterol-cognition relationship in our review is previously seen in the Baltimore Longitudinal Study of Aging where elevated TC among middle-aged and young-old participants linked to cognitive decline, and conversely lower cholesterol levels among the old-old population was correlated with cognitive decline (883). Yet to be established is whether TC or its individual sub-components alone significantly influence this connection, given the primarily indirect role of LDL cholesterol in cognition, associated with cardiovascular health. While maintaining higher levels of HDL cholesterol is generally seen as advantageous for overall health, it represents only a part of the multifaceted factors impacting cognitive function. Notably, the direct correlation between HDL cholesterol and cognition has not been as extensively researched as the connection established between LDL cholesterol and cognitive health. Future research is ultimately required to better unravel the temporal and functional contributions of cholesterol and its metabolites across the adult lifespan.

The relationship between midlife LDL-C and TG with later life cognition were conflicting. Most studies cited no relationship with later life cognition. However, a select number of cognitive functions were in fact negatively affected by midlife LDL-C and TG including memory, attention, global cognition, spatial ability, and perceptual speed. LDL-C is a widely accepted risk factor for CVD (919). Previous reports suggest LDL-C levels may contribute towards the formation of cortical plaques and fibrillary tangles which in turn can increase the rate of cognitive decline associated with dementia (920, 921). However, increased levels of LDL-C have been reported to both increase and decrease structural and functional connectivity among select cognitive networks and even increase GM volume of the frontal cortex associated with executive function and global cognition (922-925). This pattern of conflicting evidence as seen in our review may be accounted for by the omission of relevant data or lack thereof regarding pathological and clinical signs of cholesterol-induced cognitive dysfunction with age.

Midlife HDL-C is previously found to affect functional cognitive status decades later albeit limited to a select few studies among the literature in memory, attention, and global cognition (419, 926). High levels of HDL-C are inversely linked with mild cognitive impairment and age-related cognitive decline (927), although there is contrary evidence of no association between HDL-C and cognition (906). Our review highlights the protective role of midlife HDL-C, and indeed hypercholesterolemia and TC to some extent, in the preservation of cognitive function in later life among executive function,

processing speed, global cognition, and verbal learning and memory (437, 832, 883). Low levels of HDL-C are linked with reduced cognitive function and diminished bilateral anterior temporal and temporo-occipital GM volume, cerebral regions presenting with neurodegeneration (928). The deteriorations in cognition were expressed as a function of visuospatial memory, such that the authors speculate that these deficits "*may be an early sign of deleterious anterior or lateral temporal lobe changes and that deficient HDL cholesterol levels may play a role in this subtle cognitive change*" (928). Notably, TC gradually decreases from midlife into later years for most individuals, reflecting metabolic dysfunction and cognitive decline (877). This cholesterol-related metabolic dysregulation is associated with brain pathology that contributes to cognitive decline but falls short of meeting AD or dementia diagnostic criteria (876). It's essential to consider that the use of statins by almost a third of adults over 65 years old can influence the rate of cognitive decline associated with TC levels (929). In line with our findings, elevated HDL-C in later middle age and early later life is protective against functional cognitive decline up to 2-decades later and positively correlates with memory performance among older adults with subjective memory complaints (930-933). Higher levels of HDL-C at midlife are protective and positively associated with several regions of memory, executive function, and global cognition (928). This may incur a synergistic and functional protective capacity towards cognition with advancing age as seen in our review. Previous evidence cites only elevated TC as the contributor towards AD and cognitive decline with all remaining cholesterol markers remaining within normal range supporting a bidirectional relationship associative of age-related cognitive decline and increased midlife TC (440, 730, 877, 905, 908). However, prior literature suggests that low HDL-C in midlife and a further decrease over a 5-year follow-up was a significant risk factor for decline in short-term verbal memory function as seen in the Whitehall II study (934). Nevertheless, more recent evidence suggests that compared with very low levels of HDL-C at midlife, elevated HDL-C lessens the risk by almost half for the development of mild-cognitive impairment and dementia almost two-decades later (935). HDL-C is known to regulate intracellular cholesterol homeostasis and amyloid protein metabolism centrally leading to the prevention of Aβ-induced endothelial inflammation and Aβ-plaque deposits and thus the possible prevention of cognitive decline and dementia (936-938). Thus, there may be a defining threshold of midlife HDL-C towards the prevention later life cognitive decline where early intervention and behavioural strategies may mitigate cognitive impairment across a range of sub-domains. These levels of inconsistency surrounding the impact of midlife cholesterol on cognition across the adult lifespan is vastly apparent and a cautionary tale of interpretation.

Epidemiological and pathological investigations have proposed that midlife modifiable cerebrovascular risk factors such as high cholesterol can lead to cognitive decline although with some controversy (415-419). Our results support this inconsistency with both negative and no relationships identified between midlife cognition and cholesterol metrics, although most favoured a null relationship. Although positive relationships at midlife were also evident for select cognitive outcomes of memory, executive function, and global cognition (437, 883, 885). Initial evidence suggests that there is a slow yet progressive decrease in central lipid content beginning from the second and third decades of life and accelerating after the eighth decade (872, 913, 939, 940), which may reflect the inconsistency in the present review. Two main regions that can become significantly impacted by age are the Inferior temporal cortex and cingulate gyrus due to lipoxidative damage and associated inflammation which could lead to alterations in memory and attention (941). It appears that from midlife into later life both memory and executive function are negatively impacted by midlife cholesterol. In relation to cognition and domain specific function, a host of neurotransmitter receptors and their corresponding post-synaptic components are inherently tied with cholesterol-imbued lipid rafts such that defined levels of cholesterol are required for the release and maintenance of $GABA_A$ for example (942). This may in part explain how the biological decreases in cholesterol over time create low cholesterol in later life leading to cognitive impairment versus the contribution of high cholesterol in midlife (883). A range of cognitive functions such as working memory, psychomotor speed, verbal fluency and episodic memory develop at a steady rate from childhood into adolescence, peak in young adulthood and begin a steady decline from midlife into later life (441, 442). This advanced rate of cognitive decline from midlife onwards may therefore become altered by fluctuating rates of cholesterol levels and metabolism which may even be protective in some cases. Intervention and prevention strategies early in life are therefore warranted to halt if not slow down signs of future cognitive impairment for the preservation of brain health with age.

8.6. Limitations

Several limitations arose throughout the review process. Firstly, there was significant heterogeneity across all studies due to reporting of cognitive test data and length in follow-up. Second, due to the lack of comparative groups and raw data provided within studies a meta-analysis was not possible. Qualitative associations do not imply causation therefore limiting our understanding of the underlying temporal and biological catalysts of cholesterol, its sub-components, and cognition.

Thirdly, we did not factor in the known contribution of select genes. For example, the APOe4 gene is associated with LDL levels and the risk of cognitive decline and dementia from midlife onwards. Fourth, we did not consider the contribution of additional CV risk factors alongside cholesterol which few have previously analysed (417, 943). Fourth, we did not consider the variance of ages throughout midlife at which plasma cholesterol metrics were obtained. Many studies did not include whether blood samples and cognitive testing was conducted on the same day, and some studies reported taking a single blood sample versus those that had multiple over several timepoints throughout midlife. Having this differentiation would enable a better understanding of the temporal contributions of cholesterol across midlife towards cognition with ageing given the varying rates of decline in biological cholesterol and domain specific cognitive functions. Finally, our understanding of the weighted contribution of each cholesterol sub-component and the mechanisms driving the protective effects and cognitive deteriorations with age were difficult to extrapolate in the context of the literature as our understanding of the underlying mechanisms remains to be established.

8.7. Conclusion

CV health is essential in the maintenance of brain health and cognitive status such that the contributing role of cholesterol should be considered a preventable and interventional component of healthy ageing. This key CV risk factor has been linked with age-related cognitive decline and may hold the key to targeted and accountable intervention strategies (944). The present review highlights the inconsistent relationship between cholesterol at midlife and cognition at both midlife and later life. Currently the evidence appears inadequate to support the implementation of suitable interventions to delay and prevent cognitive decline. There is a need to increase our understanding of biological and temporal lipid profiles pertaining to midlife as a critical period for prevention of cognitive decline. Management strategies should focus on CV risk factors such as hypertension and diabetes where the stronger scientific evidence exists towards the negative effect on several domains of cognition across the adult lifespan.

8.8. Question(s) Raised

- Is there an identifiable relationship between the CV health profile and cognitive function of a middle-aged population?
- Is there an effect of lifetime sporting exposure and concussion history on CV and brain health?
- Do PA levels confer any benefit or protection towards health and wellbeing at midlife among those who previously participated in amateur sport?

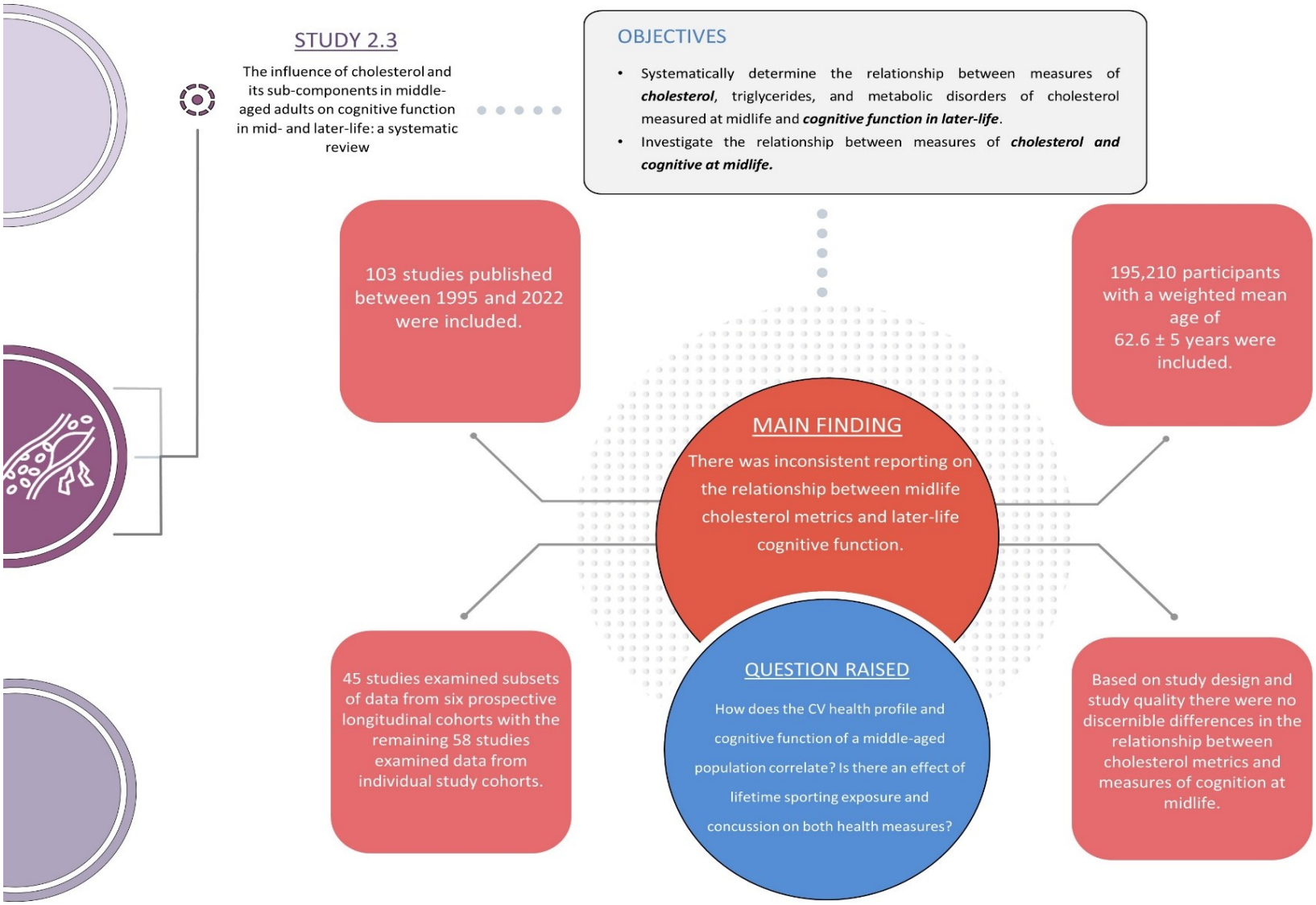

Figure 8.3. *Chapter Eight – Study 2.3 Summary.*

9

Chapter Nine:

An Assessment of Neurocognitive Function and Cardiovascular Health Profiles in Middle-Aged Community Sporting Athletes: A Cross-Sectional Pilot Study

9.1. Highlights

- Males reported significantly higher levels of SBP and DBP than females with 31.25% of males classified as hypertensive.
- The mean number of years of lifetime sporting exposure was significantly higher among males.
- Lifetime sporting exposure was significantly and positively correlated with SBP and depression, and time since ceasing regular sporting activity was correlated with metrics of somatisation and mental health and negatively correlated overall mental wellbeing.
- No significant correlations were found indicating a relationship between cognition and perceptual accuracy during the SIFI task.
- Similar patterns of susceptibility to the SIFI task were seen across all older participants in comparison to those of younger adults in **Chapter Five**.

9.2. Introduction

Health has been defined by the WHO as "*a state of complete physical, mental and social wellbeing and not merely the absence of disease or infirmity*" (865). Regular physical activity (PA) is associated with a plethora of health benefits, both physical and mental, which include increased lifespan, as well as the prevention or delay of chronic diseases. It can be assumed that consistent participation in sport should confer health benefits throughout life. Despite the known ability of regular PA to reduce adiposity, improve the presence of metabolic risk factors and delay the onset of cognitive decline, much of the literature to date has focused on the negative associations found with long-term participation in sport including a propensity for high levels of injury like concussion (3, 945, 946). There is much literature exploring the significant impact on cognition, mental health, and cardiovascular (CV) profiles among former professional athletes. Similarly, most evidence from the general population relies on data examining the correlation between PA patterns and cognitive function among geriatric populations while studies at midlife remain to be validated. While there exists a substantial amount of evidence concerning the immediate and short-term effects of sport and PA on cognition and CV function, there is a notable scarcity of knowledge regarding the long-term impact of engaging in sport and PA during earlier stages of adulthood for preserving cognitive functions and overall health among the ageing population.

Current and available data exhibit variations in the age periods during which lifetime sporting exposure and PA have been previously evaluated, along with differences in the respective follow-up periods. There are three broad life periods that can be classified among sporting athletes both professional and recreational: '*early life'* (while participating in sport and exposed to injury); '*mid-life'* (retired from sport and no longer exposed to injury); and '*late life'* (when symptoms might emerge), with mid-life being the focus of identifying and characterising brain health (947). The available evidence on the impact of sport and PA suggests that its positive effects on cognitive function may vary depending on specific cognitive domains. Therefore, it is essential to adopt a differentiated perspective while examining neurocognitive measures specific to each domain to gain a comprehensive understanding of the long-term effects. For example, sports-related concussion (SRC) has significant long-term health implications, including neurological, cognitive, and CV dysfunction (396, 397, 948). The effects of repetitive head impacts (RHI) accumulate over years of sports participation which may predispose individuals to neurocognitive dysfunction in later life (508). Over 10% of the adult population aged 45 and older report perceived cognitive decline to some degree (949). Among retired athletes, subjective cognitive complaints and cognitive decline increase the likelihood of objective cognitive decline in older age (950). However, studies on the long-term health effects of contact sports are often limited to specific populations including male college- or middle-aged former collision sport athletes, making comparisons by sex and sporting type difficult (51, 951-954). Participation in contact sports may also increase the risk of cardiovascular disease (CVD), such as hypertension, arterial stiffness and thicker carotid intima-media, in midlife and beyond (483, 484). Monitoring CV risks alongside cognition and mental health is therefore necessary for effective player welfare programs and healthy ageing among the general population.

In the last decade, there has been a surge in research examining the relationship between CVD with past and present PA levels, as it is a significant predictor of later-life cognitive functioning and vascular dementia. The American Heart Association (AHA)/ American College of Cardiology (ACC) has defined ideal CV health as (955):

> *"The absence of clinically manifest CVD together with the simultaneous presence of optimal levels of all 7 metrics, including not smoking and having a healthy diet pattern, sufficient physical activity, normal body weight, and normal levels of total cholesterol, blood pressure, and fasting blood glucose, in the absence of drug treatment" (Benjamin et al., 2019; pp.70).*

Increased incidence rates of cardiometabolic syndrome and metabolic syndrome have been observed among both current and retired NFL athletes (956, 957). NFL players historically have a higher prevalence of pre-hypertension and hypertension in comparison to the general population (958). Specifically, there is a higher rate of hypertension among those with higher BMI levels linked to increased levels of CVD risk factors (958). The ever-increasing size of professional athletes in contact sports and its relationship to CV health confers an admirable concern for future health which also extends to the general population. Research has recognised PA as a highly promising lifestyle factor that can be modified to prevent age-related cognitive decline and reduce the risk of CVD's. Interestingly, both cognitive performance decline and issues with CV health may begin as early as midlife, rather than being solely confined to old age. As individuals age, their cognitive function in mid-to-later life appears to rely on maintaining a healthy heart-brain relationship in tandem with continuous engagement in sport despite the risk of injury like concussion. The current state of research warrants better insight as to whether the risks of RHI exposure in sport for example are offset by the benefits of PA and if chronicity of lifetime PA confers health benefits in later life among recreational athletes.

Evidence supporting the independent effects of PA during early- and mid-adulthood implies the presence of multiple critical periods for intervention. Identifying independent effects would contribute to the refinement of predictive models of cognitive decline and CV health with dementia risk, as current models primarily associate physical inactivity in later life with an increased risk of dementia. While lifetime PA has been linked with better physical health outcomes in later life, it is unclear whether sustained positive effects in cognitive function, CV and mental health also occur (3, 482, 483). The long-term welfare of ageing athletes thus raises significant unease as it is unclear whether participation in contact and collision-based sports is linked to poorer brain and CV health in later life (72, 953, 959). Research exploring the advantageous effects of PA, encompassing outcomes that reflect the biological foundation for sustaining brain plasticity, cognitive function, and CV health, has generated significant interest whereby it is recognised that early detection and intervention at midlife may be critical to the preservation of long-term health. Prior investigations into cognitive and CV outcomes have predominantly focused on studying PA during either a distinct and isolated period (*e.g., early or mid-adulthood*) or continuous periods (*e.g., early through mid-adulthood*) across the general population. As a result, there remains a gap in understanding the distinct associations between the effects of contact sport and duration of lifetime PA during different stages of adulthood and independent influence on health outcomes.

The combined effect of SRC and ageing is a salient issue that requires interdisciplinary research and collaboration. Further exploration into the development of cognitive decline, mental well-being, and CV health in community-level athletes will provide valuable insight into the mechanisms behind these long-term consequences and lead to new strategies for prevention and management. Therefore, the primary aim of this study was to determine the relationship between concussion history and previous sports participation with cognitive function among a cohort of previous community level athletes at midlife. The secondary aim was to determine whether participant reported outcomes in midlife in those with varying levels of concussion exposure and sports/exercise participation levels were associated with better or worse self-reported health profiles. It was hypothesised that those with an extensive exposure rate to concussion and were physically inactive would have poorer outcomes and cognition compared to those who were physically active and had little to no concussion history.

9.3. Methods

9.3.1. Study Design

The current cross-sectional and explorative study was conducted as part of **Study Three**. See **Chapter Two, Section 2.6.** for full details of methodology including detailed information regarding participants, recruitment, eligibility criteria, assessment protocol and ethical approval.

9.3.2. Participants

A convenience sample of older community athletes were recruited to take part in this study conducted onsite at Trinity College Dublin. Those participants agreeing to take part were then contacted by the research team to schedule a testing time that was convenient for the participant. Participation in the study was completely voluntary and participants were informed that they could withdraw from the study at any time. Written informed consent was gained by all participants prior to testing. Recruitment for this study commenced in May 2023 and concluded in June 2023.

9.3.3. Sociodemographic Outcomes

Information including age, sex, height, weight, marital status, level of education, and self-report of CV risk factors and cognitive difficulties was gathered. Sport-specific information including lifetime sporting exposure, time since ceasing regular sporting activity, and previous sport played was obtained. Measures of CV health including resting heart rate (HR) and BP measurement were also acquired to determine the relationship with cognitive function (see **Chapter Two - Section 2.6.6.1a** for full details of assessment).

9.3.3a Concussion History, Knowledge, & Attitudes

Concussion history information was collected from each participant using the Michigan Traumatic Brain Injury Identification Method (see **Appendix 1.7.**). Before completing the questionnaire, participants were provided with a written explanation of the concussion definition based on the latest guidelines, specifically the 5th Consensus Statement on Concussion in Sport. The assessment of concussion history was subsequently utilised to investigate the association between sports-related concussions and performance on the SIFI test. In addition, concussion reporting behaviour, knowledge, attitudes, and education was obtained by a short survey based on previously published work (519-521). See **Chapter Two Section 2.4.2b** for full details.

9.3.4. Cognitive Assessments

9.3.4a The Montreal Cognitive Assessment (MoCA)

The Montreal Cognitive Assessment (MoCA) was developed as a concise screening tool to identify Mild Cognitive Impairment, see **Appendix 3.2**. It takes approximately 10 minutes to administer and is scored on a scale of 30 points. The MoCA is a validated paper-based assessment that evaluates overall cognitive functioning. A score of 26 or higher is considered normal. The MoCA covers various cognitive domains including orientation, language functions (verbal fluency, naming, and sentence repetition), attention (number repetition, subtraction, and tapping), and visuospatial functions (clock drawing and Necker cube reproduction). It is widely utilised worldwide and has been translated into 36 languages and dialects. The test and instructions are freely accessible on the official MoCA website (www.mocatest.org) without the need for permission in clinical or educational settings.

9.3.4b The Sound Induced Flash Illusion (SIFI) Test

The SIFI Task was employed to examine the efficiency of cross-modal MSI, which involves the processing of information from two sensory modalities. The task assesses MSI by measuring susceptibility to the audio-visual fission illusion, where a single flash is perceived as two flashes due to simultaneous auditory beeps. The task includes trials with unisensory visual and auditory conditions to control for any visual or hearing impairments. Congruent trials consist of one or two synchronized flashes or beeps, while multisensory incongruent trials involve the presentation of a secondary auditory beep either preceding or following the first beep at different time delays of 230ms, 150ms, and 70ms before (A-V/A) and after (V/A-A) the visual flash. These time delays are known as negative and positive stimulus onset asynchronies (SOAs), respectively. These incongruent trials are also referred to as '*illusory trials'*. A detailed description of the SIFI experiment can be found in **Chapter Two - Section 2.4.3a**.

9.3.5. Clinical Questionnaires

All questionnaires were answered by self-report pertaining to physical, mental, and general health. All eligible participants were asked about domains including cognitive functioning, alcohol use, sleep, pain, exercise and physical activity levels and mental health. The questionnaires were designed to examine aspects other than concussion history that may affect long-term brain health. All were straightforward in their method of administration. The following health-related questionnaires, previously described in **Chapter Two** were completed:

- The International Physical Activity Questionnaire (IPAQ) Short Form *(see **Section 2.4.2c**, Appendix 1.3.)*
- Patient Health Questionnaire (PHQ-9) *(see **Section 2.4.2f**, Appendix 1.4.)*
- Generalised Anxiety Disorder Assessment (GAD-7) *(see **Section 2.4.2e**, Appendix 1.5.)*
- Short Form 12 (SF-12) Health Survey *(see **Section 2.4.2g**, Appendix 1.6.)*
- Brief Symptom Inventory (BSI) *(see **Section 2.4.2d**, Appendix 1.8.)*
- Pain Disability Index (PDI) *(see **Section 2.6.6.2a**, Appendix 3.3.)*
- The Pittsburgh Sleep Quality Index (PSQI) *(see **Section 2.6.6.2b**, Appendix 3.4.)*
- The Alcohol Use Disorders Identification Test (AUDIT) *(see **Section 2.6.6.2c**, Appendix 3.5.)*

9.4. Statistical Analysis

All data was tested for normality using Kolmogorov-Smirnoff test and for homogeneity of variance using Levene's test. Comparisons were made based on sex, previous sporting cohort (contact/non-contact), concussion history (yes/no), and PA levels. On all demographic data, an independent samples t-test, one-way ANOVA, or a Mann-Whitney U-test was used where appropriate to compare all groups. A pairwise comparison approach and Bonferroni correction for multiple comparisons were used to follow up on significant results. A multiple linear regression analysis was done using the enter method to evaluate the connection between sporting career duration, time since ceasing regular sporting activity and patient reported outcomes with several known covariates such as age, sex, and concussion history and number of previous concussions. The test assumptions of observation independence, linearity, homoscedasticity, multicollinearity, and normal distribution of residuals were all verified. Similar to our analysis in **Study One**, we can appreciate considerable heterogeneity in the response patterns when undertaking the SIFI task, and therefore it is not unreasonable to expect the probability θ_{tj} to be specific to a subgroup exhibiting a particular response pattern. To account for this heterogeneity we once again introduced a mechanism for stratifying participants by response pattern through introduction of a class label for individual i and defining stratum specific probabilities θ_{tkj} , $k = 1, \ldots, K$.

$$[c_i \mid \pi, K] \quad \sim \quad \text{Categorical } (K, \pi)$$

$$[y_{tij} \mid c_i = k, \theta] \quad \sim \quad \text{Binomial } (r, \theta_{tkj})$$

where $\boldsymbol{\theta}$ denotes all the θ_{tkj}. Letting the strata labels c_i act as an individual level effect, correlating a participant's observations over time, we assume that y_{tij} are independent of $y_{t'ij}$ for $t' \neq t$ and all i, j conditional on c_i. We also assume local independence, so that conditional on c_i, y_{tij} is independent of $y_{tij'}$, $j' \neq j$. Our definition of agreement is to judge agreement within stratum k if:

$$\theta_{1kj} = \theta_{2kj} = \cdots = \theta_{Tkj} = \theta_{kj} \qquad j = 1, \ldots,$$

i.e., the probability of success within the stratum is the same across all time points for all *m* SOA conditions. Given that the SIFI task was only undertaken once by this sample of older community athletes, our Bayesian hierarchical model of agreement and subsequent multinomial regression was not suitable to accurately predict SIFI behaviour from baseline variables. Therefore, an isolated analysis of the six SOA conditions was undertaken by Two-way ANOVA to better understand the influence of sex, sporting cohort, and skill type as covariates on perceptual ability.

A p-value of <0.05 indicated statistical significance for all tests. Effect sizes were described using as either eta-squared for unadjusted models or partial eta squared for adjusted models and interpreted as small = 0.01; medium = 0.06; large = 0.14 (602). All analyses were performed in SPSS version 28 (SPSS Inc, Armonk, NY), and graphs created in Graphpad Prism (GraphPad Software version 8, San Diego, CA).

9.4. Results

9.4.1. Participant Characteristics

Twenty-three older community level athletes (16 Males, age: 59.41 ± 7.51 years; 7 Females, age: 55.29 ± 7.16 years) were included in the present study. A power analysis for participant recruitment was not performed for the present prospective study as it is exploratory in nature. A summary of results across the study population is provided in **Table 9.1.** below.

Mean BMI was significantly higher among males (28.43 ± 3.59 $Kg.m^2$) than females (22.24 ± 2.59 $Kg.m^2$; p = 0.001). Seven males had a BMI classification of overweight compared to only one female, while four males were classified as obese. The remaining eleven participants were in the healthy BMI and no individual was classified as underweight. Males and females did not differ in most demographic characteristics such as age, marital status, level of education, and PA levels. Most participants were classified as having moderate-to-high levels of PA. No significant differences were exhibited between males and females across walking, moderate, vigorous, and total METS; see **Table 9.1.** for full details.

The SF-12 revealed no significant difference between males and females on physical and overall health-related-quality of life. The level of self-reported pain and disability did not statistically differ between males and females (p>0.05). Using the AUDIT to examine present level of alcohol consumption where hazardous alcohol use is defined by a score of ≥ 8, no significant difference was found between males and females, although four males reported a score ≥ 8 (range: 8-29). The

Pittsburgh Sleep Quality Index revealed no significant differences between males and females ($p>0.05$). Although two females and twelve males reported a score >5 indicative of poor sleep. Self-reported memory problems were expressed only by one female participant. No statistically significant differences in cognition assessed by the MoCA between males and females were found ($p = 0.494$).

There was a higher number of males self-reporting medication use for control of both BP and cholesterol. At rest, heart rate was not statistically different between males (64.42 ± 7.67 bpm) and females (62.33 ± 6.09 bpm, $p = 0.533$). Males reported statistically higher levels of SBP (137.56 ± 16.16 mmHg) and DBP (85.88 ± 10.47 mmHg) than females (SBP: 112.86 ± 16.98, $p = 0.003$; DBP: 73.00 ± 7.68, $p = 0.008$). The majority of females resided in the optimal category for both SBP and DBP ESC categorisation, with only one female participant considered in the high-normal category. Two males were categorised as optimal, two in the normal range, and two in the high-normal range. The remaining four males were classified as Grade 1 (n = 2), Grade 2 (n = 2) and Grade 3 hypertensive (n = 1) for both SBP and SBP; see **Table 9.2.** for full details.

Females were found to only have participated in non-contact sports (n = 7), however, an equal number of males were found to have previously played both contact (n = 8) and non-contact (n = 8) sports. The mean number of years of lifetime sporting exposure was statistically higher among males (35.83 ± 17.59 yrs) than females (19.43 ± 15.14; $p = 0.045$), although time since ceasing regular sporting activity did not statistically differ ($p>0.05$). Four participants, all of whom were male, reported one or more previous concussions whilst the remaining 19 participants reported no history of prior concussion. No significant differences were found between in both concussion knowledge and attitude scores between males and females (both $p>0.05$). Full details can be found in **Table 9.3.** and **Table 9.4.** below.

Table 9.1. *Participant Characteristics.*

	Female (n=7)	*Male (n=16)*	*Total (n=23)*	*p-value*
Age (yrs; mean ± SD)	55.29 ± 7.16	59.41 ± 7.51	58.15 ± 7.49	0.23
Height (m; mean ± SD)	1.64 ± 0.09	1.73 ± 0.08	1.70 ± 0.09	**0.03***
Weight (Kg; mean ± SD)	60.29 ± 10.09	85.00 ±10.67	77.48 ± 15.51	**0.00*****
BMI (Kg.m^2; mean ± SD; Category, n=)	22.24 ± 2.59	28.43 ± 3.59	26.55 ± 4.37	**0.001*****
Underweight	-	-	-	
Healthy Weight	6	5	11	
Overweight	1	7	8	
Obese	0	4	4	
Marital Status (n=)				
Single/Never Married	-	2	2	
Married/Engaged	6	13	19	
Separated	-	-	-	
Divorced	1	1	2	
Widowed	-	-	-	
Education (n=)				
Secondary School Education Only	3	5	8	
At least one year of university but no degree	2	0	2	
University Graduate	-	5	5	
Master's Graduate	-	4	4	
Higher Degree	2	2	4	
Smoking Status (n=)				
Never	*5*	*11*	*16*	
Past	*2*	*5*	*7*	
Current	-	-	-	
Cholesterol Medication (n=)				
Yes	1	6	7	
No	6	10	16	
BP Medication (n=)				
Yes	0	3	3	
No	7	13	20	
Memory Problems: Self-report (n=)				
Yes	1	0	1	
No	6	16	22	
MoCA (mean ± SD)	25.86 ± 3.39	26.56 ± 1.55	26.35 ± 2.21	0.494
Alcohol Usage (Score: 0-20)	2.29 ± 0.76	5.56 ± 7.49	4.57 ± 6.39	0.267
Pain/Disability (Score: 0-10)	1.14 ± 2.27	5.06 ± 9.05	3.87 ± 7.79	0.277
Sleep Quality (Score: 0-21)	4.86 ± 4.45	7.75 ± 3.91	6.87 ± 4.42	0.132
PA Levels (METs; mean ± SD)				
Vigorous	1134.43 ± 1097.32	1382.00 ± 1727.23	1305.74 ± 1541.55	0.729
Moderate	808.71 ± 971.71	626.25 ± 1176.36	680.87 ± 1099.17	0.728
Walking	803.79 ± 575.43	1775.81 ± 1308.86	1479.98 ± 1211.39	0.076
Total	2741.00 ± 1730.30	3784.06 ± 1867.15	3466.61 ± 1853.19	0.222
Category (n=)				
Low	2	0	2	
Moderate	2	5	7	

High	3	11	14	
Sporting Cohort (n=)				
Contact	0	8	8	
Non-Contact	7	8	15	
Sporting Type (n=)				
Open Skill	1	8	9	
Closed Skill	6	8	14	
Lifetime Sporting Exposure (yrs)	19.43 ± 15.14	35.83 ± 17.59	30.83 ± 18.24	**0.045***
Time Since Ceasing Regular Sporting Activity (yrs)	6.86 ± 12.81	2.56 ± 4.75	3.89 ± 8.01	0.246

Table 9.2. *CV Health Profile and Surrogate Measures of Cardiorespiratory Fitness.*

	Female (n=7)	***Male (n=16)***	***Total (n=23)***	***p-value***
SBP (mmHg; mean ± SD)	112.86 ± 16.98	137.56 ± 16.16	130.04 ± 19.79	**0.003****
DBP (mmHg; mean ± SD)	73.00 ± 7.68	85.88 ± 10.47	81.86 ± 11.29	**0.008****
SBP Classification (n=)				
Optimal (<120 mm Hg)	5	2	7	
Normal (120-129 mm Hg)	0	7	7	
High Normal (130-139 mm Hg)	1	2	3	
Grade 1 Hypertension (140-159 mm Hg)	0	2	2	
Grade 2 Hypertension (160-179 mm Hg)	0	1	1	
Grade 3 Hypertension (≥180 mm Hg)	0	1	1	
DBP Classification (n=)				
Optimal (<80 mm Hg)	3	6	9	
Normal (80-84 mm Hg)	0	7	7	
High Normal (85-90 mm Hg)	1	2	3	
Grade 1 Hypertension (90-99 mm Hg)	0	2	2	
Grade 2 Hypertension (100-109 mm Hg)	0	1	1	
Grade 3 Hypertension (≥110 mm Hg)	0	1	1	
Resting Heart Rate (bpm)	62.33 ± 6.09	64.42 ± 7.67	63.78 ± 7.16	0.533

Table 9.3. *Concussion History.*

	Female (n=7)	*Male (n=16)*	*Total (n=23)*
Concussion History			
Yes	0	4	4
No	7	12	19
No. Previous Concussions	0	14	14
One Previous Concussion	0	1	1
Multiple Previous Concussions	0	3	3

Table 9.4. *Concussion Knowledge Score, Item Frequency of Correct Responses, and Concussion Attitude Score.*

	Female (n=7)	Male (n=16)	Total (n=23)	p-value
Concussion Knowledge Score (mean no. correct responses ± SD)	9.86 ± 1.46	9.19 ± 2.11	9.39 ± 1.92	0.455
Concussion Attitude Score (mean % who agree/strongly agree ± SD)	67.86 ± 4.67	66.31 ± 5.78	66.78 ± 5.41	0.541
Recognition of Signs & Symptoms (n=)				
Headache (True)	7	16	23	
Sensitivity to Light (True)	7	11	18	
Difficulty Remembering (True)	6	12	18	
In a "fog" (True)	7	12	19	
Difficulty Concentrating (True)	7	13	20	
Dizziness (True)	5	16	21	
Hives (False)	7	16	23	
Difficulty Speaking (False)	3	5	8	
Arthritis (False)	7	16	23	
Panic attacks (False)	6	12	18	
Drowsiness (False)	0	5	5	
Weight gain (False)	7	16	23	
Feeling Slowed down (False)	2	9	11	
Reduced breathing rate (False)	7	15	22	
Excessive studying (False)	7	16	23	
Hair loss (False)	7	16	23	
General Concussion Knowledge (n=)				
There is a possible risk of death if a second concussion occurs before the first one has healed (True)	4	7	11	
In order to be diagnosed with a concussion, you have to be knocked out (False)	7	16	23	
A concussion can only occur if there is a direct hit to the head (False)	3	13	16	
Symptoms of a concussion can last several weeks (True)	2	6	8	
Concussions can sometimes lead to emotional disruptions (True)	6	14	20	
There is rarely a risk to long-term health and well-being from multiple concussions (False)	5	15	20	
Attitude Towards Concussion (n=)				
I would continue playing a sport while also having a headache that resulted from a minor concussion (Disagree)	7	13	20	
I feel that concussions are less important than other injuries (Disagree)	6	16	22	
I feel that a player has a responsibility to return to a game even if it means playing while still experiencing symptoms of a concussion (Disagree)	7	15	22	
I feel that a player who is knocked unconscious should be taken to the emergency room (Agree)	6	16	22	
I feel that Coach A made the right decision to keep Player R out of the game (Agree)	7	16	23	
I feel that Player H should tell his coach about the symptoms (Agree)	7	16	23	

***Note:** Some but not all questions from the questionnaire were chosen as representative figures for general concussion knowledge and attitudes towards concussion of the chosen sub-population.*

****Note:** Figures for attitudes towards concussion (n=) represents the sum of those who strongly agree/agree and strongly disagree/disagree which was question dependent.*

Table 9.5. *Prevalence of Symptoms of Physical and Mental Health Disorders.*

	Female (n=7)	*Male (n=16)*	*Total (n=23)*	*p-value*
PHQ-9	2.29 ± 2.49	2.50 ± 2.58	2.43 ± 2.50	0.855
BSI-18				
GSI	51.47 ± 12.68	49.36 ± 8.99	50.00 ± 10.00	0.652
SOM	53.18 ± 16.47	48.61 ± 5.59	50.00 ± 10.00	0.324
DEP	47.59 ± 3.19	51.05 ± 11.78	50.00 ± 10.00	0.459
ANX	51.93 ± 12.56	49.16 ± 9.01	50.00 ± 10.00	0.553
SF-12				
PCS-12	43.95 ± 4.77	44.68 ± 4.46	44.46 ± 4.46	0.727
MCS-12	48.52 ± 5.39	45.97 ± 5.96	46.75 ± 5.79	0.343
GAD-7	4.86 ± 3.63	3.63 ± 4.22	4.00 ± 4.01	0.511

***Abbreviations:** PHQ-9: *Patient Health Questionnaire*, BSI-18: *Brief Symptom Inventory*, SF-12: *Short Form Health Survey*, GAD-7: *Generalised Anxiety Disorder Assessment*

***Significance is at the <0.05 level (two-tailed)*

Table 9.6. *Bivariate Correlations (Pearson Correlation) between Anthropometric, CV Health, PA Levels with Lifetime Sporting Exposure and Time Since Ceasing Regular Sporting Activity.*

		Lifetime Sporting Exposure	Time Since Ceasing Regular Sporting Activity	Age	BMI	RHR	SBP	DBP	Total MetS
Lifetime Sporting Exposure	*Pearson Correlation*								
	Sig. (2-tailed)	--							
	n	23							
Time Since Ceasing Regular Sporting Activity	*Pearson Correlation*	-.335	--						
	Sig. (2-tailed)	.118							
	n	23	23						
Age	*Pearson Correlation*	.374	-.012	--					
	Sig. (2-tailed)	.079	.958						
	n	23	23	23					
BMI	*Pearson Correlation*	.242	.089	.269	--				
	Sig. (2-tailed)	.266	.686	.214					
	n	23	23	23	23				
RHR	*Pearson Correlation*	-.081	.292	.109	.262	--			
	Sig. (2-tailed)	.712	.177	.621	.227				
	n	23	23	23	23	23			
SBP	*Pearson Correlation*	**.504***	.000	.033	.404	.092	--		
	Sig. (2-tailed)	.014	1.000	.880	.056	.676			
	n	23	23	23	23	23	23		
DBP	*Pearson Correlation*	.306	.006	-.021	.359	.308	**.826****	--	
	Sig. (2-tailed)	.156	.978	.923	.093	.152	.000		
	n	23	23	23	23	23	23	23	
Total MetS	*Pearson Correlation*	.151	**-.563****	-.009	.005	-.186	-.057	.113	--
	Sig. (2-tailed)	.490	.005	.966	.984	.395	.797	.608	
	n	23	23	23	23	23	23	23	23

***** *Correlation is significant at the 0.05 level (2-tailed).*

****** *Correlation is significant at the 0.01 level (2-tailed).*

Table 9.7. *Bivariate Correlations (Pearson Correlation) between Mental Health Indicators and Sporting Metrics.*

		Lifetime Sporting Exposure	Time Since Ceasing Regular Sporting Activity	Number of Concussions	PHQ-9	GAD-7	SOM	DEP	ANX	GSI	PDI	AUDIT	PSQI	PCS-12	MCS-12
Lifetime Sporting Exposure	*Pearson Correlation*														
	Sig. (2-tailed)	--													
	n	23													
Time Since Ceasing Regular Sporting Activity	*Pearson Correlation*	-0.335	--												
	Sig. (2-tailed)	0.118													
	n	23	23												
Number of Concussions	*Pearson Correlation*	0.221	0.163	--											
	Sig. (2-tailed)	0.310	0.457												
	n	23	23	23											
PHQ-9	*Pearson Correlation*	-0.080	0.286	-0.118	--										
	Sig. (2-tailed)	0.717	0.185	0.593											
	n	23	23	23	23										
GAD-7	*Pearson Correlation*	-0.230	0.122	0.276	0.009	--									
	Sig. (2-tailed)	0.291	0.580	0.202	0.967										
	n	23	23	23	23	23									
SOM	*Pearson Correlation*	-0.230	**.754****	-0.011	**.451***	0.179	--								
	Sig. (2-tailed)	0.292	0.000	0.962	0.031	0.415									
	n	23	23	23	23	23	23								
DEP	*Pearson Correlation*	**.497***	-0.134	0.093	**.494***	-0.290	0.183	--							
	Sig. (2-tailed)	0.016	0.542	0.673	0.017	0.180	0.402								
	n	23	23	23	23	23	23	23							
ANX	*Pearson Correlation*	-0.037	0.384	-0.258	**.567****	0.161	**.789****	0.377	--						
	Sig. (2-tailed)	0.866	0.070	0.235	0.005	0.463	0.000	0.076							

	n	23	23	23	23	23	23	23	23						
GSI	*Pearson Correlation*	0.049	**.496***	-0.044	**.617****	0.037	**.878****	**.603****	**.892****	--					
	Sig. (2-tailed)	0.824	0.016	0.842	0.002	0.865	0.000	0.002	0.000						
	n	23	23	23	23	23	23	23	23	23					
PDI	*Pearson Correlation*	0.341	-0.059	-0.012	0.071	-0.177	0.004	0.245	0.225	0.161	--				
	Sig. (2-tailed)	0.111	0.791	0.957	0.749	0.418	0.987	0.261	0.303	0.462					
	n	23	23	23	23	23	23	23	23	23	23				
AUDIT	*Pearson Correlation*	0.022	-0.024	-0.077	0.257	0.160	0.073	-0.151	0.151	0.027	0.164	--			
	Sig. (2-tailed)	0.919	0.913	0.726	0.236	0.467	0.742	0.490	0.493	0.904	0.454				
	n	23	23	23	23	23	23	23	23	23	23	23			
PSQI	*Pearson Correlation*	-0.035	0.255	-0.057	**.468***	**.429***	**.443***	0.198	**.512***	**.481***	0.173	0.275	--		
	Sig. (2-tailed)	0.873	0.241	0.797	0.024	0.041	0.034	0.365	0.012	0.020	0.430	0.203			
	n	23	23	23	23	23	23	23	23	23	23	23	23		
PCS-12	*Pearson Correlation*	0.356	-0.225	-0.155	0.083	-0.075	0.247	**.461***	**.461***	**.455***	-0.167	-0.137	0.148	--	
	Sig. (2-tailed)	0.096	0.302	0.481	0.705	0.735	0.256	0.027	0.027	0.029	0.447	0.535	0.500		
	n	23	23	23	23	23	23	23	23	23	23	23	23	23	
MCS-12	*Pearson Correlation*	-0.025	**-.497***	-0.269	**-.589****	-0.187	**-.680****	-0.342	**-.495***	**-.670****	-0.044	-0.317	**-.540****	-0.184	--
	Sig. (2-tailed)	0.911	0.016	0.214	0.003	0.394	0.000	0.111	0.016	0.000	0.842	0.141	0.008	0.400	
	n	23	23	23	23	23	23	23	23	23	23	23	23	23	23

***** Correlation is significant at the 0.05 level (2-tailed).*

****** Correlation is significant at the 0.01 level (2-tailed).*

9.4.2. Association between Sporting Metrics, PA Levels, Physical Well-Being, and Mental Health

Lifetime sporting exposure was significantly and positively correlated with SBP, proposing that prolonged engagement in sport is associated with a moderate increase in SBP ($r = 0.504$, $p = 0.014$). The total amount of PA was negatively and moderately correlated with time since ceasing regular sporting activity indicating those who have been retired from sport longer have reduced PA levels ($r = 0.563$, $p = 0.005$); see **Table 9.6**. Lifetime sporting exposure was also moderately correlated with depression ($r = 0.497$, $p = 0.016$), and time since ceasing regular sporting activity was correlated with metrics of somatization and mental health ($r = 0.754$, $p = 0.00$; GSI: $r = 0.496$, $p = 0.016$) and also negatively correlated overall mental wellbeing (MCS-12: $r = -0.497$, $p = 0.016$); see **Table 9.7**. No significant correlations were observed between total PA levels and all metrics of mental and physical well-being; see **Table 9.8** below for further details.

Table 9.8. *Bivariate Correlations (Pearson Correlation) between PA Levels and Mental Health Metrics.*

		PHQ-9	GAD-7	SOM	DEP	ANX	GSI	MCS-12	PCS-12
Total MeTS	*Pearson Correlation*	-0.090	0.258	-0.264	-0.057	-0.042	-0.187	0.282	0.060
	Sig. (2-tailed)	0.682	0.235	0.224	0.796	0.850	0.392	0.193	0.787
	n	23	23	23	23	23	23	23	23

9.4.3. Multisensory Integration Efficiency

Figure 9.1. shows the strata detected for all older participants (n = 23), with an optimal value of $\hat{k} = 3$ (***Stratum 1***: n = 14, ***Stratum 2***: n = 3, ***Stratum 3***: n = 6). The smaller number of strata here was expected due to the lesser sample size of this cohort of older individuals. What is striking are the patterns of susceptibility across the present cohort which resemble that of our findings in **Chapter Five**, where there appears to be high, moderate, and low performers. There was no indication of possible 'learners' provided those in the present sample only participated in a single testing session with one SIFI test completed.

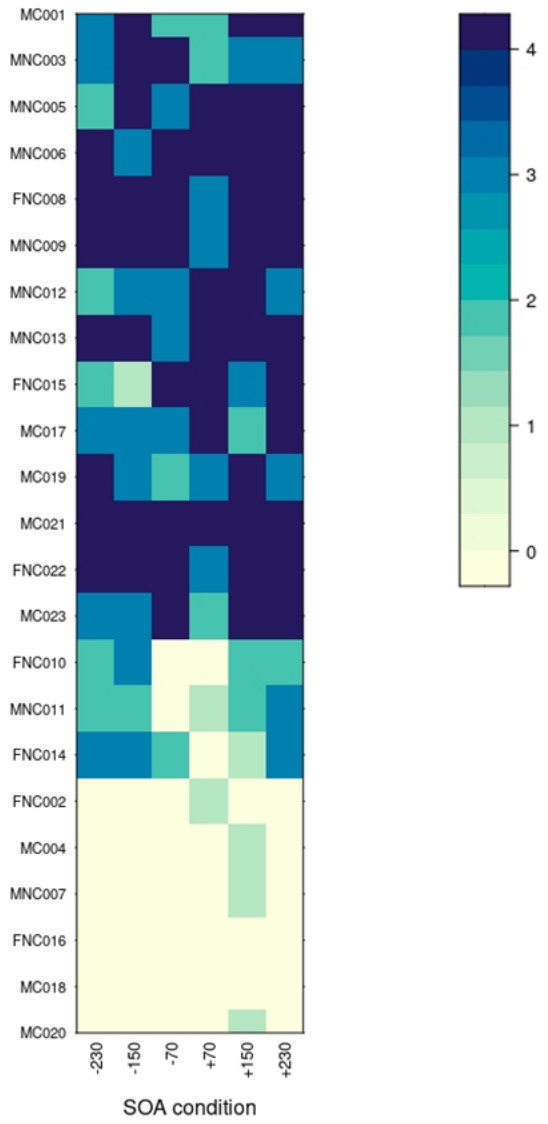

Figure 9.1. *Strata of Older Community Athletes for Baseline SIFI Performance.*

Although it can be seen from **Figure 9.2.** below that males perform with higher levels of perceptual accuracy across a wide range of temporal asynchronies than females, an isolated analysis by Two-way ANOVA of the individual SIFI conditions; in line with our rationale from **Study One – Chapter Four**; revealed no significant differences between males and females ($p>0.05$). Surprisingly, given our findings from **Study One**, those in non-contact sports performed consistently better across all SOA conditions. However, these results were not statistically significant ($p>0.05$); see **Figure 9.3.**

below. Moreover, across all six SOA conditions of the SIFI, there were no noteworthy or significant distinctions observed ($p>0.05$) between individuals participating in Open Skill sports versus Closed Skill sports or those with and without a history of concussion; see **Figure 9.4** and **9.5** below.

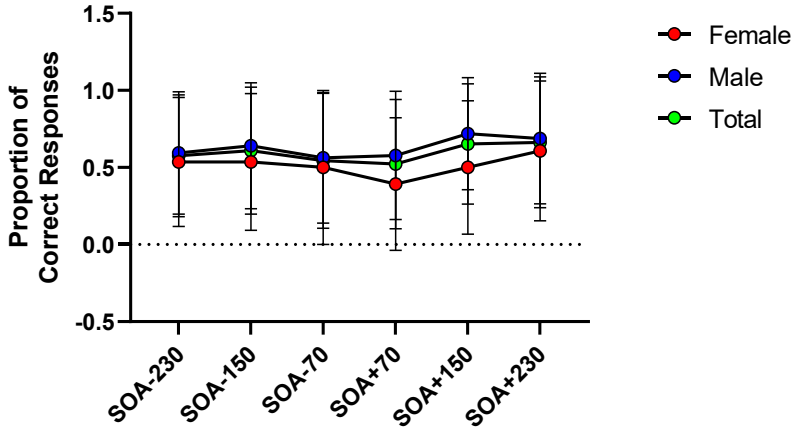

Figure 9.2. *Proportion of correct responses across SOA conditions by Sex.* No significant differences were exhibited between males and females across all six SOA conditions.

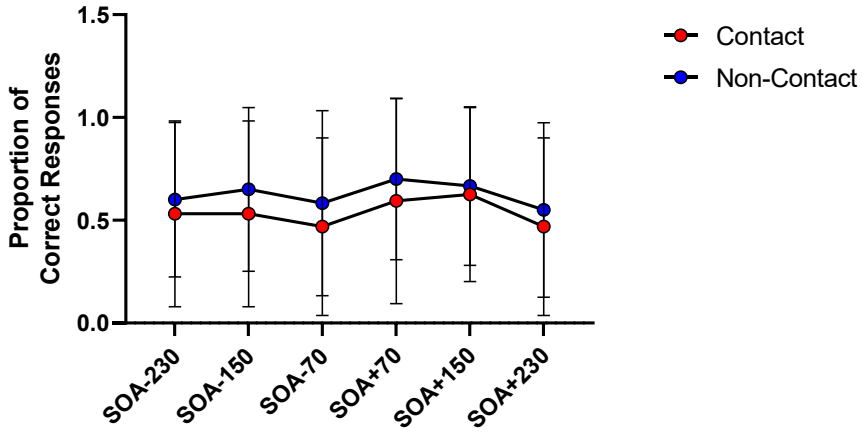

Figure 9.3. *Proportion of correct responses across SOA conditions by Sporting Cohort.* No significant differences were exhibited between those in Contact versus Non-Contact sports across all six SOA conditions.

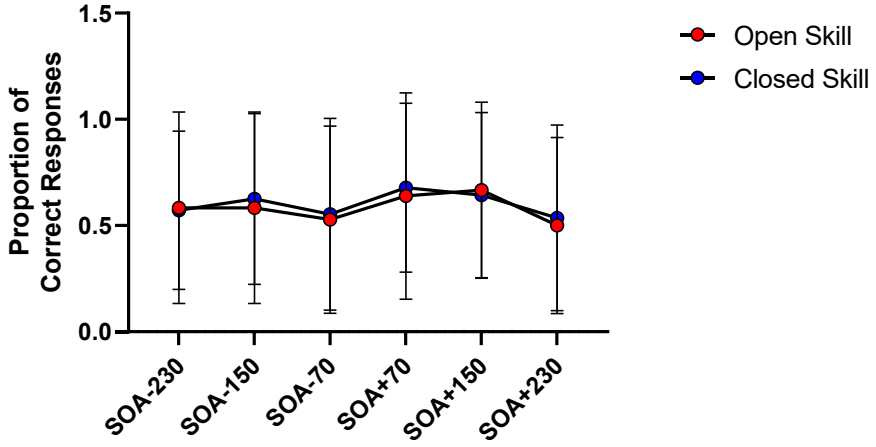

Figure 9.4. *Proportion of correct responses across SOA conditions by Sporting Type.* No significant differences were exhibited between those in Open Skill versus Closed Skill sports across all six SOA conditions.

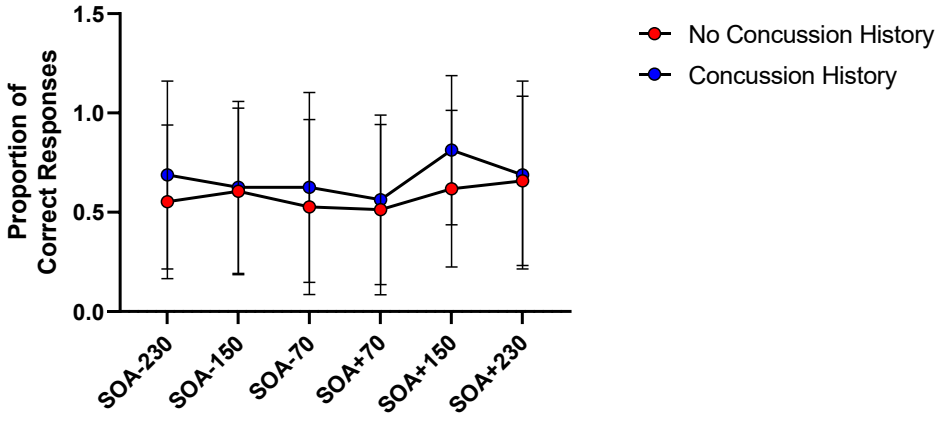

Figure 9.5. *Proportion of correct responses across SOA conditions by Concussion History.* No significant differences were exhibited between those with and without a history of concussion across all six SOA conditions.

9.4.4. Cognitive-Perceptual Correlates

Based on the outcomes from the SIFI analyses in this thesis, the existing evidence of sex differences, and the known age-related decline of global cognition, the individual A-V/A and V/A-A SOAs were employed to explore potential relationships with global cognitive functioning on the MoCA using Pearson's Correlation (r) across the entire cohort and by sex. No significant correlations were found by sex or sporting cohort indicating that there was no relationship between cognition and the

select cognitive domain of MSI. However, there is evidence from this analysis to suggest that better cognitive function may be correlated with greater accuracy on the SIFI task across the shortest SOA conditions as they approach statistical significance (SOA-70: p = 0.06; SOA+70: p = 0.08); see **Table 9.9.** below for further information. Moreover, no significant correlation was found between cognitive MoCA scores with previous concussion (p = 0.122), PA level (p = 0.179), or lifetime sporting exposure (p = 0.172) which was unexpected given our hypothesis and previous evidence; see **Table 9.10.** below.

Table 9.9. *Bivariate Correlations (Pearson Correlation) between SIFI incongruent trials and Cognition (MoCA).*

		SOA-230	SOA-150	SOA-70	SOA+70	SOA+150	SOA+230
MoCA (Total)	*Pearson Correlation*	0.307	0.132	0.396	0.372	0.292	0.313
	Sig. (2-tailed)	0.154	0.550	0.062	0.080	0.176	0.146
	n	23	23	23	23	23	23
MoCA (Female)	*Pearson Correlation*	0.239	-0.052	0.640	0.589	0.199	0.392
	Sig. (2-tailed)	0.606	0.913	0.122	0.164	0.669	0.385
	n	7	7	7	7	7	7
MoCA (Male)	*Pearson Correlation*	0.397	0.289	0.172	0.186	0.359	0.261
	Sig. (2-tailed)	0.128	0.278	0.525	0.490	0.172	0.329
	n	16	16	16	16	16	16

Table 9.10. *Bivariate Correlations (Pearson Correlation) between Cognition (MoCA), Previous Concussion, PA Level, or Lifetime Sporting Exposure.*

		MoCA	Number of Concussions	Total MeTS	Lifetime Sporting Exposure
MoCA	*Pearson Correlation*				
	Sig. (2-tailed)	--			
	n	23			
Number of Concussions	*Pearson Correlation*	0.332	--		
	Sig. (2-tailed)	0.122			
	n	23	23		
Total MeTS	*Pearson Correlation*	0.291	0.080	--	
	Sig. (2-tailed)	0.179	0.715		
	n	23	23	23	
Lifetime Sporting Exposure	*Pearson Correlation*	0.295	0.221	0.151	--
	Sig. (2-tailed)	0.172	0.310	0.490	
	n	23	23	23	23

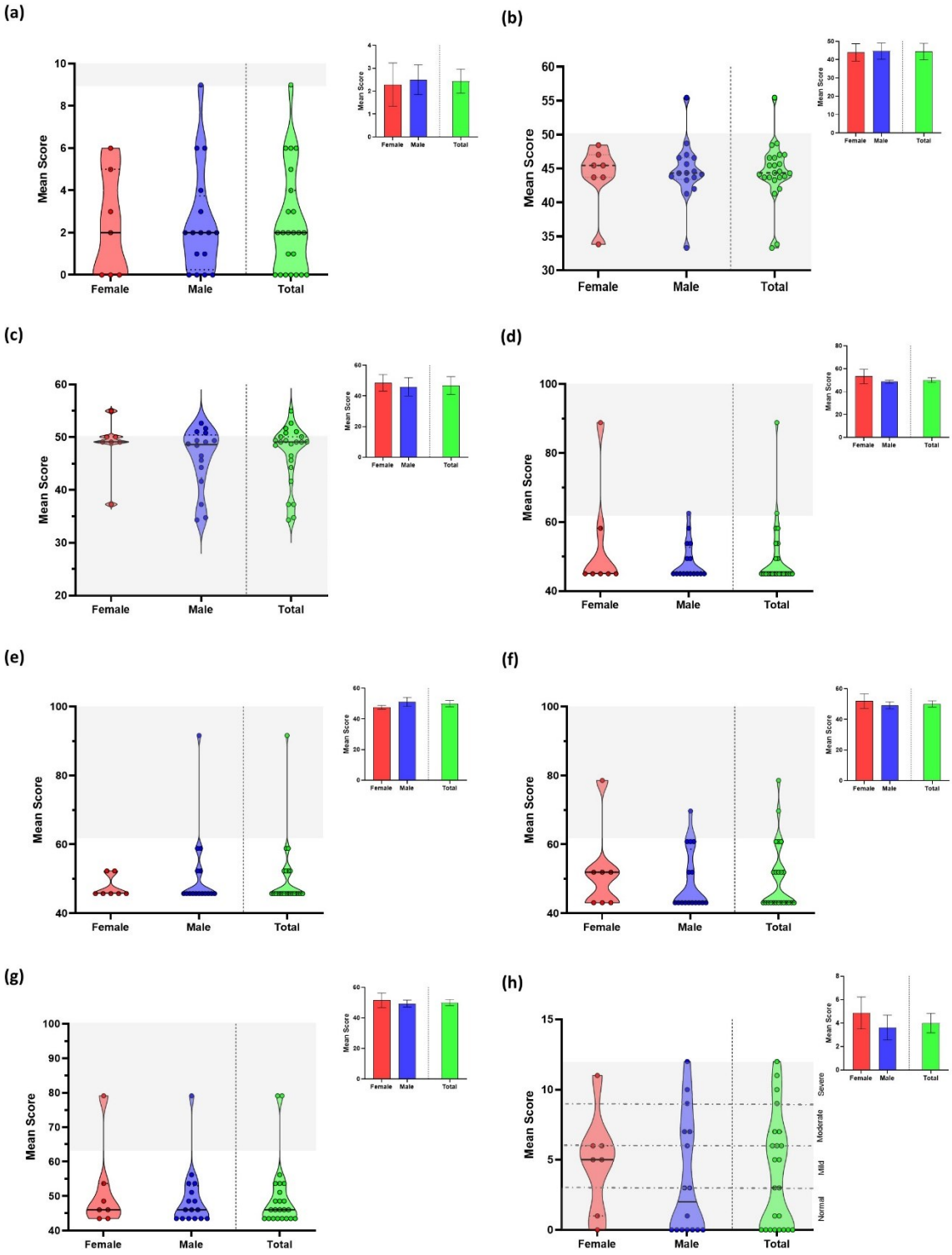

Figure 9.6. *Patient Reported Mental Health Outcomes by Sex*. Abnormal scores are highlighted in grey. **(a)** PHQ-9, **(b)** PCS-12, **(c)** MCS-12, **(d)** SOM, **(e)** DEP, **(f)** ANX, **(g)** GSI, **(h)** GAD-7.

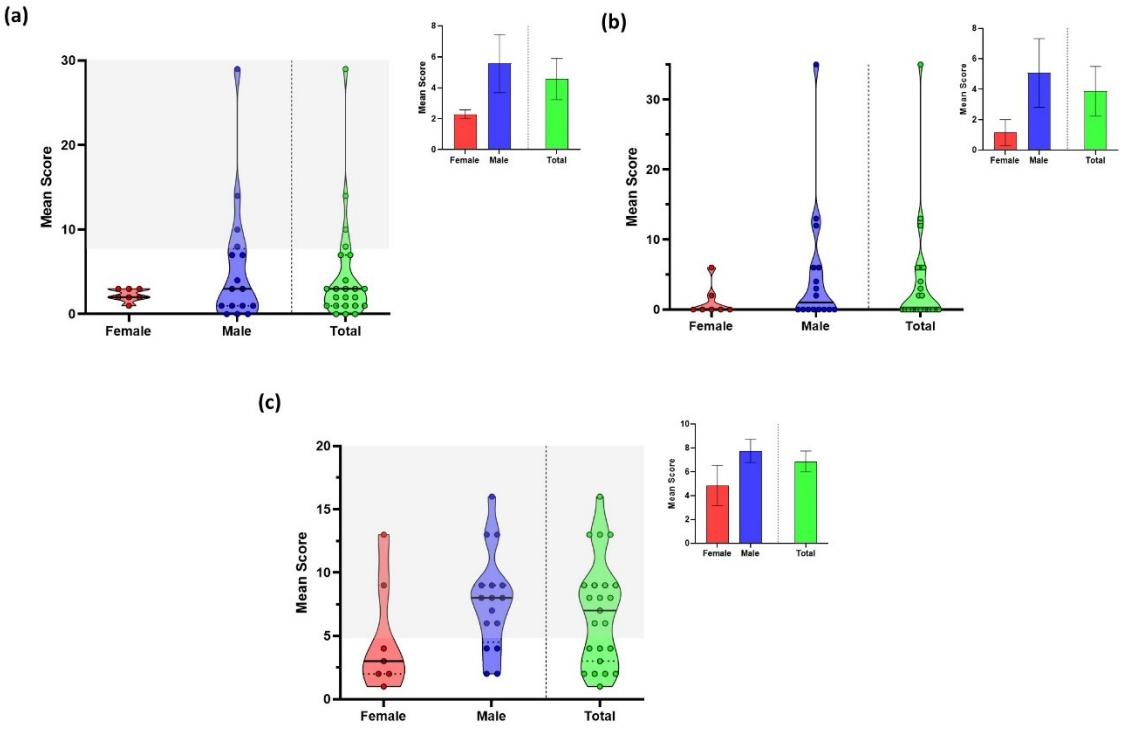

Figure 9.7. *Patient Reported Characteristics by Sex*. Abnormal scores are highlighted in grey. **(a)** AUDIT, **(b)** Pain Disability Index, **(c)** Pittsburgh Sleep Quality Index.

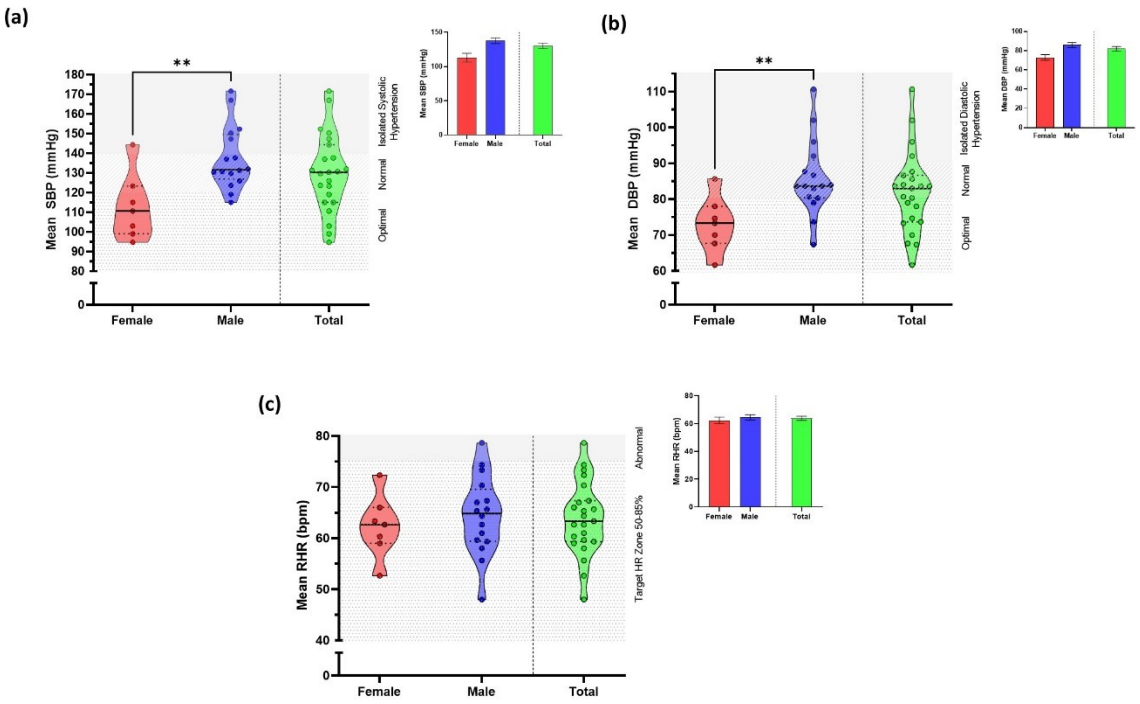

Figure 9.8. *Cardiovascular Health Metrics of Cardiorespiratory Fitness*. Abnormal scores are highlighted in grey. **(a)** Systolic BP, **(b)** Diastolic BP, **(c)** Resting HR.

9.5. Discussion

The aim of this chapter was to describe the general health status and cognitive function ability of older community level athletes at midlife. A higher proportion of males were classified as overweight-obese and self-reporting medication use for control of both BP and cholesterol, with 25% of males classified as being hypertensive. Lifetime sporting exposure was correlated with depression symptoms and measurements of SBP, although not linked to engagement in exercise itself but rather the influence of exercise in excess and at high levels of training intensity that were not tailored for the individual which may have led to maladaptive CV changes over time. However, the participants' age did not lead to this outcome, as no significant correlations between BP metrics, mental health, and age as a covariate were observed. No significant differences in cognition were exhibited between males and females by the MoCA, although females presented with an average score below the clinical cutoff indicating a possible increased likelihood of mild cognitive impairment. Susceptibility patterns to the SIFI illusory conditions in this older cohort resemble those found in **Chapter Five**, showing high, moderate, and low performers. Although sex was not a significant predictor of perceptual accuracy, males displayed reduced susceptibility across a wider range of temporal asynchronies. Intriguingly, non-contact sports consistently outperformed contact sports across all SOA conditions, and no significant distinctions emerged between Open Skill and Closed Skill sports despite **Study One's** contrasting findings. Moreover, unexpectedly but in line with the results from **Study One**, there were no observed differences between those with and without a history of concussion. Just over a fifth of the population reported one or more previous concussion(s), however all were sustained by males. Of note, 17.39% of the population indicated hazardous alcohol use and 60.87% indicated poor sleep quality. This study provides insight into the physical, CV and brain health among older community athletes with the unique comparison to previous literature from middle aged retired professional athletes.

Athletes possess finely tuned psychomotor skills involving quick visuomotor reaction time and stimulus inhibition. These skills result from neuroplastic adaptations due to motor skill acquisition and repetition, along with higher cognitive processing. Efficient motor responses, sustained attention, and quick reactions are essential in time-sensitive situations. However, former elite rugby players have been found to perform worse on processing speed tests when compared to non-contact sport athletes (392). The present study's results did not align with previous findings, as no significant differences in cognitive ability were observed between individuals who played contact vs. non-contact sports. Open

skill athletes, mainly in contact-based field sports, consistently need strong visuospatial awareness to tactically manipulate their environment tactically. In contrast, closed skill sports require less developed visuospatial skills. Concussions and/or repetitive head impacts (RHIs) are more common in contact-based sports, which may lead to an accelerated age-related cognitive decline around sporting retirement. The results suggest a slight deviation towards mild cognitive impairment (MCI), especially among females, as indicated by the MoCA. However, these differences were not significant, and comparisons based on sporting type and cohort also yielded non-significant results. These findings make it difficult to draw conclusions about cognitive function among community athletes, but research from retired professional athletes with a history of concussion and RHI may provide better insight.

Low cognitive reserve and older age increase the risk of MCI after mTBI (960, 961). Some retired NFL athletes with multiple concussions exhibit a relatively high level of cognitive reserve (962). However, retired NFL players are at a higher risk for MCI in their sixth decade of life, especially with a history of more than 3 concussions during their NFL career, increasing the likelihood of MCI by 5-fold (963, 964). Despite a long sporting career and exposure to concussive and sub-concussive events, those players who are cognitively normal at midlife "may not reflective of the highly exposed former professional football player population as a whole" (962) which may also apply to the present study cohort provided no significant differences in perceptual ability were observed between those with and without concussion history. Cognitive reserve, rather than CTE, likely contributes to the development of MCI in retired athletes. Higher cognitive reserve may provide individualised resilience to age-related neuropathological changes through synaptic plasticity via BDNF expression, regulating cognitive stimulation (965-967). However, the neural mechanisms underlying cognitive reserve are not fully understood. Factors like age, exercise, education, and stress contribute to population-specific diversity in cognitive resilience, particularly during critical time points for intervention like midlife, warranting further research, especially considering previous contact sports exposure.

Comparing literature on cognitive performance among contact sporting athletes and non-contact controls is challenging due to study design variability and selective cohorts. Previous studies have found inconclusive results which are shown to vary with age in both male and female athletes (503, 968-970). Cognitive deficits have been linked to structural changes in neural architecture, including WM alterations of the CC, fornix, internal capsule, arcuate, and uncinate fasciculi and neuroplastic changes with ageing, resulting in reduced memory and attention performance (971). While some

evidence suggests that contact sports are associated with worse neurobehavioural outcomes, other studies in fact suggest no discernible difference in brain health and cognition in those who participated in contact sports during high school or in retired rugby league players with a history of concussions in line with our present findings (959, 972). Although recent meta-analysis shows significant alterations in verbal memory and attention in retired athletes with a history of SRC (973), the relationship between contact sports participation and cognitive decline particularly with age remains poorly understood (974) and requires alternative metrics of cognitive function like the SIFI.

Ageing is associated with a decline in cognitive ability, and RHIs are believed to accelerate this deterioration. MSI also declines with age, affecting perception, behaviour, and decision-making (102). Older individuals are more susceptible to the illusion across various temporal asynchronies, while younger adults show sensitivity mainly at shorter intervals (102, 270). University students with recent concussion history were less effective at evaluating multimodal A-V occurrences (615). However, the impact of lifelong concussion history on age-related MSI decline in older athletes during midlife remains uncertain. The present study has provided novel insight into the use of the SIFI among a sample of older, community athletes to ascertain the level of perceptual performance in relation to self-reported concussion history and a lifetime exposure to sport. Our results indicate sex, concussion history, and sporting type/cohort had no statistically significant impact in MSI capabilities. Although it can be seen from **Figures 9.2. and 9.3.** that males and those in non-contact sports showed enhanced perceptual abilities, the small sample size and balance of males-to-females may have influenced the results and requires further investigation. The SIFI neurocognitive test may enhance the sensitivity of cognitive function and brain health assessments (975-977). Open skill athletes on team-based field sports are consistently required to process the visuospatial conditions around them and react appropriately making them less susceptible to the illusion of the SIFI, although our results from **Study Three** were in opposition. Information processing across multiple senses is classified and prioritised by a hierarchy of importance known as 'sensory weighting'. We found that susceptibility to the illusion increased as the onset delay decreased, but no inherent differences were exhibited based on sporting type. While it was assumed that a career in a dynamic sport setting would provide players with a higher capacity to integrate and interpret many sensory streams, the incongruent multisensory condition results indicate no learned performance benefits during the SIFI. Nonetheless, due to the lack of normative data and the novelty of the assessment, further exploration is required in the context of previous sporting exposure with potential for head impacts.

Contact sports increase the risk of exposure to sub-concussive and concussive-inducing events which can lead to neurodegeneration and cognitive dysfunction (963, 964). Former contact sport athletes show reduced brain volumes with age, particularly in the hippocampus, amygdala, and cingulate gyrus, leading to cognitive impairments in psychomotor speed and executive function (978, 979). Even at the collegiate level, contact sport athletes exhibit changes in fractional anisotropy and mean diffusivity metrics of WM brain structure that can persist for months after the last sporting contact (980, 981). These brain changes suggest possible damage without a clinical diagnosis of concussion. Concussions can induce long-term neurophysiological deficits and γ-aminobutyric acid (GABA) receptor abnormalities, impacting intracortical inhibition, neuroplasticity, and motor learning for up to 3 decades after injury (982). Older adults' reduced MSI efficiency may yet be due to an age-related decline in GABA, which could also contribute to poorer MSI after concussion (615). GABA levels decline in the brain's frontal and parietal regions by around 5% per decade and may lead to decreased inhibition at both perceptual and cognitive levels as we age. This inhibition deficit is associated with chronic stress, specifically allostatic load (AL), which refers to the cumulative physiological strain caused by prolonged exposure to stressors (983). AL accelerates aging and increases susceptibility to age-related diseases. Recent evidence suggests that at midlife SIFI performance was linked to AL, where individuals with high AL scores demonstrated heightened susceptibility to SIFI at longer SOAs (984). Longitudinal evaluation across two testing waves indicated a similar trend, where exceptionally high AL scores were associated with increased susceptibility to SIFI at longer SOAs. The statistical models validated the previously observed relationship between age and SIFI accuracy, implying that age influences SIFI performance. However, this suggests that while age-related differences in SIFI susceptibility might be linked to AL, they are not solely attributable to AL disparities.

The SIFI as a measure of A-V integration may be sensitive to previous concussion history and identify minor alterations in perceptual capacity. Studies have shown that retired contact sport athletes often exhibit reductions in neuropsychological assessments of cognitive function (482, 979, 985). However, some research has found no significant correlation between the number of years playing sport and neuropsychological metrics (964, 986). The results from our study are in line with the latter that lifetime sporting exposure and concussion history had no impact on cognitive function. Recent investigations support these findings, suggesting no significant linear relationship between cognitive performance outcomes and concussion history or years in the NFL (162). Age may therefore be the significant factor as older age is associated with increased cognitive deficits in both longitudinal

and cross-sectional studies (987, 988). Normal age-related cognitive decline follows a curvilinear trend in old age, leading to a loss of specific cognitive functions (989). Studies on retired athletes have shown reduced rates of mild cognitive disorder, with no correlation between self-reported concussions and cognitive function (990). However, other studies have suggested that over half of retired professional rugby players aged 49-55 years were cognitively impaired (991). Our work, along with Wise and Barnett-Cowan (615), suggests that A-V perception tasks may serve as sensitive markers for detecting brain function changes caused by concussion. Decreased MSI efficiency may impact decision-making and behaviour, characteristic of long-term effects of recurrent head trauma. A recent study identified five clinical domains associated with sporting retirement, including neurocognitive impairment, CVD, and chronic pain (992, 993). Global cognitive function impairment is reported by nearly one-third of players, with older age having the highest number of afflictions in mid-to-late life. Diabetes, hypertension, and dementia are significantly correlated with global cognition, and neurocognitive afflictions are linked to a higher risk of cognitive impairment (993). Cumulative, multiple afflictions may further impair global cognitive function among retired athletes, highlighting concerns for overall health and in particular CV health.

The results from our study suggest that, similar to retired professional athletes, older community athletes at midlife from the general population with a high BMI had an increased prevalence and severity of CVD risk factors, specifically high BP. Sufficient evidence exists to show that increased body weight and body fat are linked to a higher risk of chronic diseases, such as hypertension, and CVD, alongside cognitive deterioration. Retired middle-aged NFL players with obesity exhibit worse neuropsychological test results compared to players with healthy weight, likely due to higher BMI and CV risk factors (994, 995). However, the strength of association between neuropsychological measures with BMI and alcohol use is not well-defined in the literature (996). Age-related weight gain of 0.0272 kg and BMI increase of 0.051 units may further exacerbate the effects of obesity on brain function in retired contact sport athletes (98), which appears similar among community-level athletes. Older or retired competitive athletes are susceptible to CVD despite lifelong engagement in sports, indicating that an athletic heart can include pathological changes. A recent systematic review indicated that current sportspeople, particularly those in contact sports, had a higher incidence of obesity (BMI >30 kg/m^2) and increased hypertension rates (ranging from 13.8-53%) associated with body size, leading to the conclusion that exercise and participation in professional sport may not fully protect athletes, especially larger ones, from developing various CVD risk factors (483). The defining

threshold beyond which the benefits of PA are lost and become maladaptive is however yet to be established.

Cardiac adaptation in athletes, characterised by changes in coronary artery blood flow and structural remodelling, may have potential pathological effects, especially considering long sporting careers (997). The clinical overlap between exercise induced cardiac remodelling (EICR) and cardiac pathology, referred to as the "grey zone," remains unclear, and factors such as obesity and isometric training can contribute to considerable cardiac changes on a structural level that is dose-dependent with potential implications for coronary artery disease and hypertension (998, 999). Contact-based team sports, like Rugby and American football, place significant demands on the heart and vasculature, leading to increased BP, arterial stiffness, and concentric left ventricular hypertrophy (LVH), which may have negative long-term effects on health particularly among obese athletes creating higher risk of early mortality before the age of 50 (1000-1002). Moreover, arterial stiffness, which is more prevalent in strength-trained athletes compared to endurance-trained athletes or controls (1003-1005), plays a significant role in the development of hypertension, LVH, and atherosclerosis, preceding hypertension onset and predicting fatal CV events independently of classical risk factors (1005, 1006). Further investigations are needed to understand these cardiac adaptations in athletes and their impact on the general population alongside several health-related factors including pain, mental health, alcohol use.

The prevalence of musculoskeletal injuries including joint and ligament injuries, along with previous fractures and dislocations, are quite common among those with a history in contact sports which can be secondary to osteoarthritis and chronic pain (1007, 1008). This level of chronic pain in former athletes can present itself as a barrier to engagement in PA associated with a sedentary lifestyle and risk of overweight/obesity alongside several CV risk factors. Our results show very low levels of pain, although higher among males, PA levels were significantly correlated with time since ceasing regular sporting activity, although were not inherently correlated with PCS-12 scores. Older athletes at midlife coupled with a termination in their sporting career and reduction in exercise levels can elevate their CV risk profiles compared to non-athletes on metrics including BMI, BP, cholesterol, and glucose/insulin (1009, 1010) as seen with higher rates of hypertension among male participants in this study. The beneficial effects of PA are not restricted to physical health but transfer to improved psychological measures of mental health and psychosocial functioning translating to a positive impact

on mood states (1011, 1012). However, the cessation of continuous exercise will bring about increases in anxiety and depression-like behaviour.

Depression has become one of the most cited psychiatric disorders globally affecting more than 300 million people equating to 4.4% of the world's population (1013). Mental health problems are present in one in three adults throughout their lifetime (1014), but little to no research is current available on the psychological well-being of older community athletes at midlife. Our results demonstrated that lifetime sporting exposure was moderately correlated with depression, and time since ceasing sporting activity was correlated with metrics of somatisation and mental health, and negatively correlated overall mental wellbeing. The relationship between exercise and mental health is complex, and the optimal types, durations, and frequencies of exercise for reducing mental health burden are not yet fully understood. PA has been linked to numerous benefits for the brain and heart, with a meta-analysis of 33 studies reporting a reduction in CV and all-cause mortality by up to 50% with increasing levels of PA (1015). Our results indicated no significant relationship between total PA levels and all recorded metrics of mental health. However, there is concern for athletes engaging in excessive exercise for prolonged periods, which can lead to a U-shaped or reverse J-shaped dose-response relationship with long-term exposure to high levels of PA, CV-related death, and mental health status (1016-1018). A greater amount of exercise may not always be the most efficient way of addressing mental health burdens as among elite athletes the onset of issues surrounding mental health tends to overlap with years of sporting participation (1019) in line with our present findings.

Sport and exercise engagement have been found to be effective treatments for depression, alongside antidepressant medication, with higher levels of self-reported PA associated with fewer depressive symptoms (1020, 1021), which may support our present findings. It is yet important to acknowledge the link between depression and a history of concussion among former athletes. Previous research has demonstrated a frequency-response relationship between concussion history and depression, showing that retired NFL players and collegiate athletes with a history of concussions are more prone to experiencing depressive symptoms (1022, 1023). A study by Hunzinger et al. investigated the relationship between contact/collision sports exposure, RHIs, PA levels, and patient-reported outcomes among young to middle-aged adults (1024). Among 113 participants, RHI exposure rates were not correlated with physical and mental health outcomes at midlife in those who were physically active. However, those who were not physically active had worse outcomes. This suggests that regular PA may have an adaptive and protective effect on physical and mental well-

being, potentially mitigating some of the negative effects associated with a history of RHI exposure. These findings suggest that maintaining an active lifestyle through regular exercise can have positive effects on overall well-being, while a lack of PA may lead to various health-related issues, non-communicable diseases, and unhealthy lifestyle choices as seen in our present analysis.

Similar to the general population, older athletes also face potential psychological challenges stemming from life stressors, identity crises, and substance abuse. . Prevalence of alcohol misuse using the AUDIT screening tool among the general population is estimated at 18% (567, 1025). Our results indicated isolated incidences of alcohol abuse particularly among males. Retirement from competitive and even amateur sport poses a difficult transitioning period for athletes due to lack of structure and identity. There are several overlapping common and sports-specific stressors that impact former players and the general population such as sleep disturbance, alcohol misuse, and pain. Based on our results, a high level of alcohol misuse and dependency coincided with poor sleep quality. Previous systematic evidence has reported that two-thirds of research on retired athletes in contact sport included alcohol screening in their report (482), and former rugby players reported significantly greater alcohol consumption in comparison with controls. The authors clearly state that given the prevalence rates of self-reported sleeping disturbance, unhealthy nutrition, smoking, and adverse alcohol behaviour among former rugby players, former players should be screened for health-related quality of life and general wellbeing in association with symptoms of common mental health disorders. Consequently, many potential contributory factors to poorer mental and physical health may also extend to older community athletes.

9.6. Limitations

This study has several limitations. First, there may be selection bias due to voluntary participation and multiple sources of contact, potentially affecting the participant cohort based on personal interest, especially among those with a history of concussion. Second, the cross-sectional nature of the concussion knowledge and attitudes survey relied on participants' recall, introducing potential bias and inaccuracy. Additionally, a standardised definition of concussion and concussion education was not provided, which may impact the interpretation of results. Third, individuals with neurophysiological dysfunction might have avoided contact sports involvement or declined to participate in the study, while healthy individuals continued to participate, leading to potential bias. Fourth, all questionnaires relied on self-reporting, which may limit the accuracy and precision needed

for outcome metrics. Fifth, though the groups were not age- and sex-matched, there were no significant differences found in age or sex categorisation between groups. Sixth, the study did not measure blood samples for brain and heart health markers, such as BDNF and VCAM, which hindered exploration of their associations with cognitive and health outcomes. Finally, although we did consider career duration, which plays a significant role in measuring concussion history and exposure rates backed by the National Institute of Neurological Disorders and Stroke Consensus Diagnostic Criteria for Traumatic Encephalopathy Syndrome (NINDS TES) Consensus Statement of RHI exposure as the only modifiable risk factor, it potentially overlooks factors like sporting type, playing position, level of play, and lifetime volume of play which should be accounted for in future research.

9.7. Conclusion

Results from this study indicate that modifiable risk factors including overweight and obesity, and hypertension in older community-level athletes are of considerable concern given their association with long term health outcomes in later life. Community-level athletes at midlife were found to be vulnerable to substantial prevalence of mental health symptoms and disorders. It appears that despite more than five decades of sporting exposure, cognitive function and perceptual ability remained intact and were not affected by sex, concussion history, sporting cohort, or skill type. Given that similar findings have been seen among retired professional athletes, the introduction of appropriate detraining protocols should become standard practice across all levels of sporting participation. An active alliance by coaching staff and health professionals is required to educate those in sport of the long-term health risks associated with sporting exposure and how to mitigate against declines in physical and mental health in conjunction with cognitive function.

9.8. Question(s) Raised

- Would the co-assessment of biomarkers of CV and brain health have added more in-depth insight into the present findings?
- Would the influence of hormones, especially among females at midlife or having experienced menopause, influence CV health profiles and cognitive function?
- What are the potential cognitive and CV resilience factors at play which could benefit or harm those who report lifelong sporting engagement?

- Does cardiorespiratory fitness and PA levels significantly impact overall health with advancing age?
- Is there a delicate interplay between the levels of PA and years of sporting exposure influencing mental health profiles of those among general population having ceased engagement in competitive sporting activities?

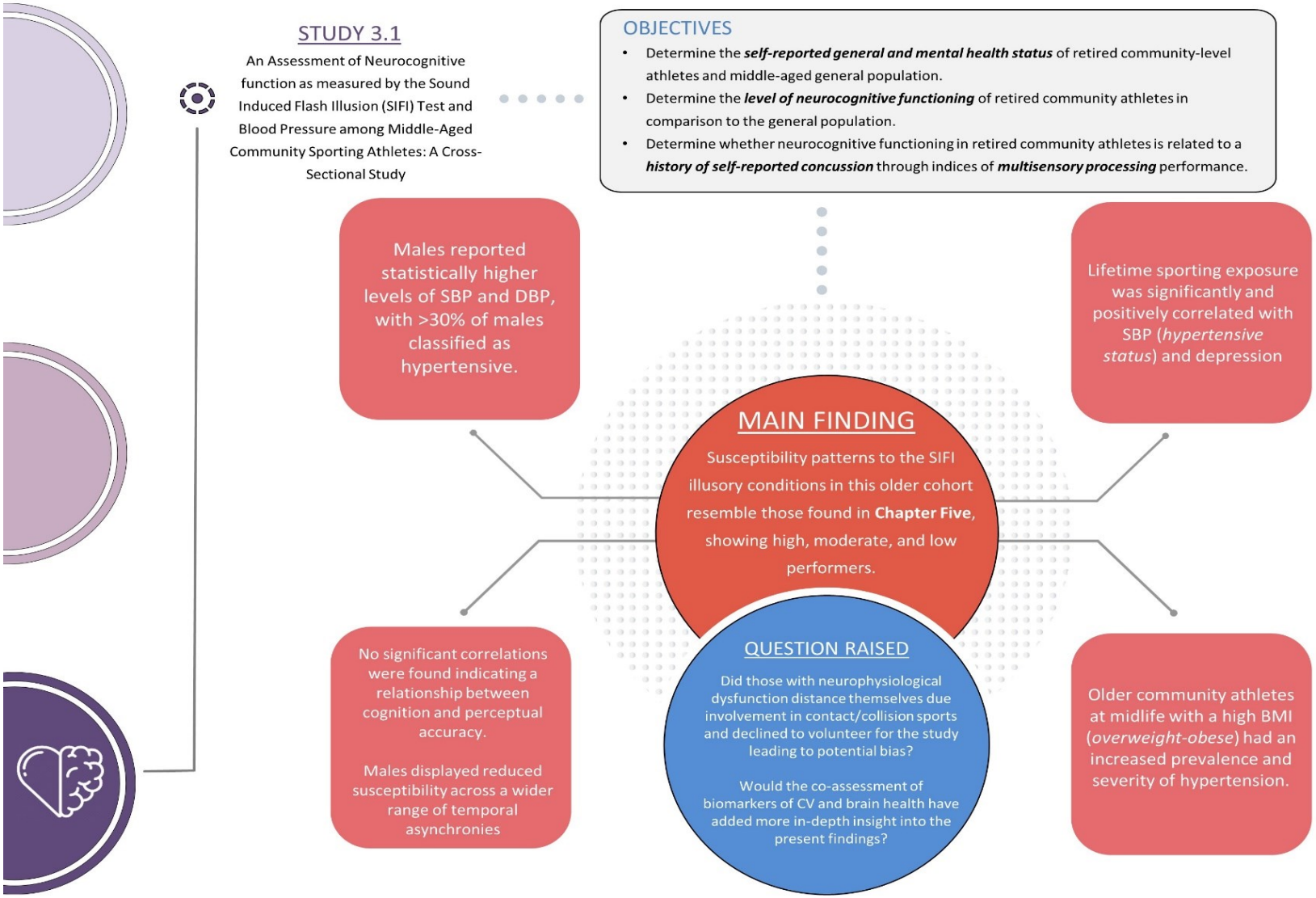

Figure 9.9. *Chapter Nine – Study 3.1 Summary*.

10

Chapter Ten:

Discussion

10.1. Summary of Findings

This study aimed to investigate the relationships connecting perceptual performance, brain health, and cardiovascular (CV) profiles among both recreational athletes and non-athletes, bridging the divide between sensory integration, cognitive function, and physical well-being. The significance of both brain and CV health is increasingly apparent in sports at all levels and across all domains, none more so than in collision sports such as rugby, where body mass of players has increased over recent decades. Professional status in sports raises concerns about CV health for both current and retired athletes, given that cumulative head amd body impacts sustained during training and competition from opponents, teammates, or their sporting environment might unpredictably influence CV well-being in tandem with the neurocognitive decline seen with normal aging. Recognising the role of CV health in athletes of all ages becomes crucial for safeguarding brain health in later life, encompassing both active and retired sports participants. This led to the development of three distinct yet intertwined studies included in the present body of work: **(i)** Investigating the associative link between cognitive function, as assessed by a task designed to test MSI ability, and sporting expertise in the presence of acute exercise; **(ii)** Analysing the impact of CV risk factors at midlife and their effect on cognitive function in later life; and **(iii)** Examining the brain health and CV health profiles of recreational, community-level athletes at midlife.

The pilot study in **Chapter Four** of **Study One** which explored the effect of multiple covariates on cognition indicated similar perceptual accuracy across illusory conditions for participants with and without concussion history. Females were more susceptible to the fission illusion across a wide range of stimulus onset ansynchrony's (SOA's) which was also evident from the results of **Study** Three exploring the impact of lifetime sporting exposure of CV and cognitive health; but neither sex nor concussion history could explain perceptual performance variance. Contact sports participants had higher MSI levels, which contrasted with what we found in **Study Three** among those at midlife, but this did not significantly impact perceptual differences. Our follow-up study exploring more precise covariates to better understand perceptual performance among young adults in third level education confirmed the results of the pilot and that of **Chapter Nine** such that males outperformed females across all SOA conditions. Most notably open skill sports participants showed significantly better performance in larger temporal asynchrony conditions compared to closed skill sports, but this did not hold true for those in midlife in **Study Three**. In particular, the results of our stratified analysis in **Chapter Four** showed that there was a small group

of participants who experienced a learning effect associated with cumulative exposure to the SIFI test. Interestingly, moderate, and high-intensity exercise had no effect on perceptual performance as evidenced in **Chapter Five** where we explored the effect of acute exercise bouts on perceptual cognitive performance, which has implications for the applicability of the SIFI as an immediate sideline and prognostic assessment tool in a sport setting. Moreover, the SIFI was shown to have a significant level of agreement and reliability across three separate testing sessions giving credence to its utility. Our results suggest the SIFI is unaffected by physiological performance changes, making its versatility in sports-related concussion (SRC) diagnosis and prognosis more pragmatic. However, there may be several reasons conferring this resilience which should be explored further and could not be fully explained using the variables implemented in the present research.

Study Two involved three systematic reviews to explore the link between midlife CV risk factors (hypertension, diabetes, and cholesterol) and cognitive function at mid and later life. The results were mixed and inconsistent across cognitive domains with memory, executive function, and global cognition showing a notable negative relationship with several metrics and metabolic correlates of the chosen CV risk factors. Most longitudinal studies from **Chapters Six** and **Seven** supported a negative association between midlife hypertension and T2DM and cognition in later life. However, in **Chapter Eight** the relationship with cholesterol was inconsistent, with some studies reporting positive and others negative associations. Age appears to play a significant role in this cognitive decline, reflecting structural and functional maladaptation over time that are both specific and yet not physiologically unique to each CV risk factor. These findings formed the premise for the final investigation of this thesis.

The findings from **Study Three- Chapter Nine** indicate significant differences between males and females in BMI, with males having a significantly higher BMI being classified as overweight and obese. Males reported higher medication use for controlling BP and cholesterol and had higher resting HR, SBP, and DBP in line with our findings from **Study Two – Chapters Six and Eight**. Females participated only in non-contact sports, while males had experience in both contact and non-contact sports, accompanied by a higher lifetime sporting exposure. Lifetime sporting exposure was positively correlated with SBP and depression but negatively correlated with total PA and mental health metrics. There was no significant relationship between cognitive functioning and sporting experience which was unexpected given the results of **Study One** and previous literature. Moreover, the analysis of cognitive performance based on the MoCA did not reveal noteworthy disparities

between males and females. However, females had an average score below the clinical threshold, implying a potential elevated risk of MCI. In the context of the SIFI illusion-related conditions, susceptibility patterns among those at midlife mirror those detailed in **Study One - Chapters Four and Five**, characterised by individuals with high, moderate, and low performance levels. While sex did not emerge as a substantial predictor of perceptual accuracy, in contrast to **Study One**, it is intriguing to note that males displayed diminished susceptibility across a broader spectrum of temporal asynchronies. Notably, non-contact sports consistently outperformed contact sports across various SOA conditions, and no significant disparities were observed between open closed skill sports and those with and without concussion history indicating a disparity in results compared to **Study One**. Overall, these findings highlight important sex differences in BMI and health-related measures with advancing age, while suggesting that sporting engagement and PA play a role in BP and mental health outcomes even from young adulthood.

10.2. Perceptual Cognitive Training

Our results provide evidence that enhanced MSI capabilities may be conferred through the types of perceptual training experienced throughout a sporting career, irrespective of previous concussion history, from young adulthood (**Study One**) which may however dissipate with age (**Study Three**). Our results from **Study One** indicate those in open skill sports or who are younger in age were less susceptible to the SIFI illusory conditions, whereas those in non-contact sports at midlife in **Study Three** were in fact seen to have higher levels of performance during the SIFI task. Athletes who engage in competitive training and competitions from an early age demonstrate heightened levels of executive and sensory function in support of **Chapter Four's** findings (166, 362). Contrary to our hypothesis, contact sport athletes, with higher exposure rates to possible concussive events, develop the highest level of decision-making, attentional, and memory-related skills due to the competitive and dynamic nature of their sport (166). Multisensory training most likely leads to neural modifications in the prefrontal cortex, enhancing perceptual processing, attention, and spatial awareness for optimal performance (631, 1026). Athletes use prior experience and memory to understand sensory information, leading to better perceptual judgments and motor responses (1027). Sufficient perceptual cognitive training can lead to sporting expertise through the brain's plasticity for adaptation (231, 1028), but appear to decline with advancing age as evidenced by our cohort of midlife community athletes in **Study Three**. Although not directly recorded in **Study One and Three**, our results

suggest a performance enhancement based on sporting modality and prior perceptual training which is most evident in young adulthood.

Sports-specific training in dynamic environments is highly reliant on modulating stimulus characteristics from multiple sensory inputs, as is the case of the SIFI. Within the follow-up study of **Chapter Four**, those in open skill sports performed significantly better in SOA conditions with a larger temporal asynchrony compared to closed skill sports; although this did not hold true for older athletes in midlife (**Chapter Nine**). Spatial and temporal modulations of distant-dependent methods can alter the size of the TBW and shift the *point of subjective simultaneity* (PSS) leading to heightened MSI abilities (118, 128, 1029, 1030). These neurobehavioral enhancements could arise from neuroplastic mechanisms that reorganise the sensory cortical maps and perceptual abilities of expert athletes from an early age, also observed in professional musicians (161). Expert athletes demonstrate superior perceptual accuracy and decision-making under various conditions, emphasising the positive impact of sporting expertise on perceptual cognitive processing. (1031-1033). Sports experts utilise the sensory principle of MSI known as 'inverse effectiveness' where stimuli of weaker saliency provide the largest input when integrated simultaneously with others (225, 610). Higher-order attentional networks may mediate additional attentional resource allocation for the integration of A-V stimuli via the intraparietal sulcus when exposed to a higher level of possible distractors, leading to more fine-tuned perceptual and attentional abilities. The 'attentional resource load theory' supports our findings from **Study One**, suggesting that enhanced motor-readiness and anticipation in contact sports, primarily open-skill sports, lead to higher levels of MSI (247, 611, 612). These enhancements are linked to the creation of cognitive sporting models, anticipating future events, and using alternative patterns of play through different search strategies (201, 1034, 1035). Certain open skill sports with high levels of information processing can ultimately enhance cognitive functioning and perceptual expertise. This improvement is attributed to previous training exposure methods that closely replicate a multisensory, competitive sporting environment.

10.2.1. Representative Learning Design and the Influence of Perceptual History in the Ageing Athlete

Our results from **Chapter Four** provided evidence that contact and open skill sports displayed higher temporal and spatial perception of A-V sensory input, although these differences did not persist with advancing age as seen in **Chapter Nine**. Small-sided games

which emphasise open-skill learning, perceptual-motor coupling, and effective skill transfer have been successful in identifying sporting expertise (1036, 1037). Representative sport-specific training should consider visual and perceptual information, action fidelity, and performance achievement (1038). Reactive agility movement time, influenced by anticipation and perception (1039), was observed to be highest among athletes in open-skill sports in **Study One**, but appears to decline with age. Soccer-specific training programs in youth sporting athletes have proven to improve muscle mass and power, CV endurance, agility, and flexibility, leading to augmented visual and auditory reaction times coinciding with 'developmental spikes' of a non-linear nature with ageing and maturation (1040). The results of **Study One** can be seen to coincide with the influence of perceptual history influencing a subsequent and successful behavioural response.

Recent evidence suggests that visual perception is influenced by perceptual history (1041), showing a tendency to perceive a current stimulus resembling the preceding one, a bias known as 'serial dependence' (1042). The origin of this effect is debated, with interpretations ranging from basic sensory processes to higher-order memory and decision-making (1043). However, there is support that serial dependence is a perceptual phenomenon, as EEG studies demonstrate encoding of past information in response to a current stimulus in the primary visual cortex (682, 1044, 1045). In the context of sports, serial dependence may arise from early perceptual computations, beginning at the sensory recognition levels of vision and hearing in support of the findings in **Study One**. The impact however of long-term perceptual training on susceptibility and overall perceptual performance on the SIFI task remains uncertain. Previous research suggests that long-term perceptual training can significantly improve accuracy and response time (193). In **Study One**, participants' exposure to multiple tests prior to each session could be considered a form of training, potentially reducing susceptibility to the illusory conditions which was experienced by a small sample (n = 30) of our population. Despite this, our results demonstrate high levels of reliability and agreement levels with a large sample size, which is noteworthy given previous empirical studies with very small sample sizes have yielded insignificant results (106, 123). However, the precise neural underpinnings of susceptibility to the SIFI and its relationship with cognitive function and clinical disorders require further investigation to support current hypotheses and explore other inherent factors such as advancing age and biological sex.

10.3. MSI and Sensory Plasticity with Advancing Age

Impairments in sensory systems, especially in aging individuals, can have substantial effects, which may worsen with the deterioration of multiple systems. MSI capacity, believed to increase with age, particularly for congruent stimuli, compensates for declines in vision and hearing (124, 194, 1046). Detecting and intervening early in midlife for multisensory dysfunction could aid healthcare professionals in mitigating long-term impacts on cognition and brain health. The results from **Chapter Nine** indicate that MSI performance capabilities were not different between those who engaged in contact and non-contact sports, or between those in open versus closed skill sports. Males were seen to be less susceptible to the SIFI illusory conditions, but similar to our MoCA results on cognition in **Study Three**, previous history of sports involvement or concussion history did not impact MSI, suggesting that cognitive status in later life, as opposed to midlife, might be predictable by the process of aging.

By 2050, the global population of individuals aged 60 and above is estimated to reach approximately 2 billion. This demographic shift is anticipated to bring about a rise in age-related declines in vision, hearing, and temporal orientation. These declines have the potential to affect cognitive function and might influence the capacity for MSI, which is particularly relevant to discussions surrounding the changes in sensory plasticity as individuals age (1047, 1048). Spatially connected intrinsic brain volume changes and network reconfiguration peaks at midlife and declines significantly with age, indicating the need for interventions to maintain cognitive functions with senescence. Although not directly seen from our results, possibly due to the small sample size of the population, age-related changes in A-V acuity, as well as cognition, can impact the reliability and precision of A-V stimuli to undermine MSI efficiency (209, 1049, 1050). Cerebral aging involves increased recruitment of task-relevant brain regions to compensate for age-related cognitive deficits (1051, 1052). Neural network disruptions, functional dysfunction, and structural atrophy are associated with age-dependent cognitive deficits, leading to compensatory mechanisms that become more prevalent with advancing age (796, 1053). Posterior brain regions show structural decrease with age, compensated by functional hyperactivation of frontal regions in middle to later life (1054), which may in part explain the lack of significant findings in **Study Three**. A model of the age-associated effects termed ‘HAROLD’ (hemispheric asymmetry reduction in older adults) has been postulated to account compensatory effects in cognitive performance, including MSI in the elderly. This positive correlation between a lack of recruitment of region-specific areas of the brain for

a given task and the co-inhibition of non-relevant areas (263, 443) with task performance in the elderly indicates preserved cognitive functions like MSI (265). However, despite HAROLD's explanatory power, it cannot fully account for the increased MSI capacity observed in older age (610). Those with expert-level performance and lifelong experience demonstrate the ability to counteract age-related declines in cognitive function, particularly in auditory processing, which may be attributed to inherent predisposition or lifelong neuroplastic development of associated neural correlates (161, 1055, 1056). The structural preservation of cortices, safeguarding frontal regions from the effects of aging (1057), may extend to those with lifelong sports participation and developed expertise (1058, 1059) as older individuals demonstrate advanced MSI compared to their younger counterparts (94, 95, 1060). The observed preservation of cognitive function in this context explains the absence of statistically significant outcomes in **Chapter Nine**. Nevertheless, the limited scope of our sample size necessitates a more intricate exploration, as our initial theory aligning concussion history and sporting engagement with brain health, in accordance with the literature, lacks validation and requires more thorough investigation.

The significance of studying MSI is underscored by its potential benefits in old age but may be compromised by a history of concussion (245). SIFI performance in **Chapter Nine** suggests that more profound changes in SIFI performance may in fact arise in later in life Task-specific training in sports lead to focal changes in the brain among experts for a given task (632). However, these changes tend to dissipate with age, recruiting more brain regions for the chosen task (443), and this 'unlearning' of MSI could be accelerated in individuals with a history of concussion. The definition of susceptibility profiles in individuals at midlife, characterised by varying performance levels, underscores the significance of healthy cognitive aging monitoring, considering potential late-life conditions such as CTE. The consistent superior performance of non-contact sports across SOA conditions in **Chapter Nine**, despite the higher prevalence of concussions in contact sports, presents an unexpected outcome and is in opposition to our findings from **Study One**. Furthermore, although sex did not emerge as a prominent predictor of perceptual accuracy, the observation of males having reduced susceptibility to the SIFI is of considerable interest. These results collectively emphasise the critical role of cognitive evaluation and well-being maintenance across an athletic career, serving as a vital step toward mitigating the onset of disease states in later life.

10.3.1. Sex Differences in Cognitive Performance

Results from **Study One** found that males consistently outperformed females in perceptual performance across all SOA conditions, with reduced susceptibility to the illusion being strongest at wider SOAs. The significant difference between males and females in the comparative analysis was not supported by LCA and regression analysis in the pilot study. Nevertheless, our Bayesian modelling in the follow-up study indicated a trend towards males' higher perceptual abilities, which can also be seen in our results from **Study Three**. This is in line with recent meta-analysis which indicates male athletes have cognitive advantages in working memory and visuospatial ability compared to female athletes (1061-1063). The observed sexual dimorphism in 'cognitive strategies' may explain these sex-based differences. These strategies, allocentric versus egocentric, are influenced by memory, and each sex may adopt a behavioural strategy aligned with their information encoding and retrieval dimorphisms (1064, 1065). However, there is limited research on sex differences related to perceptual cognitive tasks and particularly with advancing age.

Recent evidence suggests that females have longer reaction times and reduced accuracy when assessing irrelevant and conflicting stimuli as seen in our results from **Chapters Four**, **Five**, **and Nine** (1066, 1067). The origins of these sexual dimorphisms in cognition and behaviour are subject to considerable debate, with discussions about whether they arise from divergent life histories or selective evolutionary pressures related to sex roles. For example, enhanced spatial processing may be advantageous for males, historically assuming roles of hunters and warriors, similar to modern sporting athletes (1068). It's important to note that these sex differences are based on averages and may not apply universally to all individuals. Furthermore, there are numerous cognitive domains where sex differences are not observed.

The apparent sex differences in cognitive performance in selective domains may be influenced by circulating sex hormones. Oestrogen has been shown to enhance dendritic spine development and functional synaptic plasticity in females, while also improving spatial memory and synaptic plasticity in males to a lesser extent (1069, 1070). Cognitive performance varies across the menstrual cycle, with women performing better in verbal memory when oestrogen levels are high and worse in spatial tests during the same periods. This suggests a possible link between high oestrogen levels and the development of working memory, encoding, and retrieval, with different brain regions being affected at distinct capacities dependent on age (1071-1073). Research, primarily on rodents, indicates that oestrogens can enhance higher-order cognitive functioning across the whole brain in both

males and females through various interconnected mechanisms involving multiple signalling pathways and synaptic formation and turnover (1074); *see* ***Figure 10.1.*** *below*. These effects are observed in brain regions relevant to higher-order cognition, such as the hippocampus and prefrontal regions, promoting the synthesis of neurotrophins and modulating cholinergic-dopaminergic neurotransmitter systems and GABAergic activity (1073, 1075). Oestrogen and progesterone are believed to collaborate in enhancing neuronal function by regulating synapse formation and elimination, enhancing synaptic transmission, and exerting neuroprotective effects (1076). The prevailing theory linking menstrual cycle phases to cognition proposes that hormonal fluctuations influence cognitive performance. The early follicular phase, characterised by low levels of oestrogen and progesterone, is associated with superior performance in spatial abilities typically favouring males. Conversely, phases with increased oestrogen and/or progesterone, like the late follicular or mid-luteal phases, are linked to enhanced cognitive abilities typically favouring females, including verbal fluency and verbal memory (1077, 1078). Although the phase of menstruation or onset of menopause was not recorded in **Study One and Three** respectively, the functional and mechanistic activity of hormones on cognition may support our understanding of sex differences in MSI capabilities, particularly among those athletes with lifelong sporting exposure.

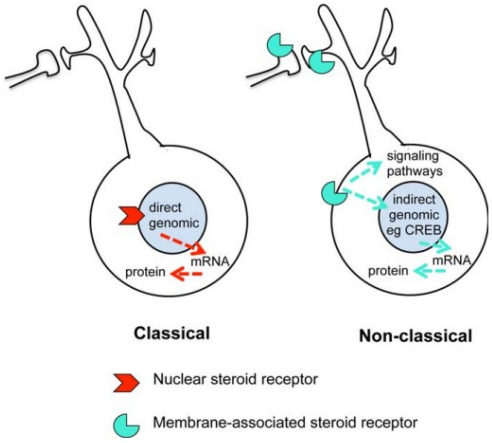

Figure 10.1. *Mechanisms of Steroid Hormone Action via Classical and Non-Classical Mechanisms. *Adapted from the work of McEwen et al. (1075).*

10.3.2. Biological Sex as an Influential Factor Linked to CV Risk Factors in Ageing

Age-related cognitive decline affects most domains of cognition, with executive function and working memory being more pronounced, especially in females (1079-1081).

Extensive research suggests that significant sex differences in general cognition are absent and is in line with our results of **Study Three**. Although not directly assessed in **Study Three**, sex differences in cognitive trajectories during healthy aging are diverse and linked to endocrine-related changes in sex hormone production (1082). Females experience a significant decline in oestrogen and progesterone during reproductive senescence, especially at critical time points like midlife. Testosterone levels in females decline gradually from the third or fourth decade of life onwards. In contrast, males experience a gradual decrease in testosterone accompanied by an increase in LH, FSH, and sex hormone-binding globulin causing a significant change in cognitive function (1083, 1084). Oestrogen crossing the BBB undergoes conversion into catechol oestrogen, which inhibits an enzyme responsible for dopamine degradation in the prefrontal cortex. Oestrogen may therefore modulate cognitive processes influenced by neurotransmitter release, including MSI and perception. Studies demonstrate that oestrogen particularly influences sex differences in dopaminergic-related cognitive functions, such as working memory and response inhibition employed during the SIFI test (1085-1087). This may in part explain the reduced perceptual ability of females at midlife in **Study Three** although not statistically significant, similar to what we observed in **Study One**. The impact of oestrogen on brain health extends to overall cognitive well-being, particularly in females with inherent links to CV risk factors and the onset of neurological diseases in later life.

The decline in ovarian hormones during menopause, coupled with the consequent loss of their neuroprotective effects, has been linked to the heightened vulnerability of females to AD. Neurodegeneration typically manifests in individuals after their third decade of life, and it becomes particularly noteworthy in those aged 71 and older. Among this older population, there is a notable sex-based disparity, with 16% of women developing AD as opposed to 11% of men (1088). This underscores the significance of considering biological sex as a pertinent factor when discussing how it is linked to CV risk factors in the aging process. For example, the regulation of cholesterol and lipid metabolism exhibits a sexual dimorphism, with elevated levels of circulating lipids associated with cognitive impairments. Our results from **Study Three** indicate that a higher proportion of males were prescribed cholesterol lowering medication, in line with our findings from **Chapter Eight**, which may be influenced by hormonal rates of secretion with increasing age. Oestrogen influences the secretase pathway within neurons and lipid metabolism, leading to a decrease in β-amyloid peptides production and influencing lipid content and lipid rafts on cell membranes, which may reduce the accumulation of Aβ in AD (1089). The reduction of

lipid biosynthesis due to oestrogen may have a potential protective effect, as cholesterol is the essential precursor to all sex hormones (294, 295) in support of **Chapter Nine and Eight's** results. These results show a difference between sexes in aging, and its relationship with midlife CV risk factors (1090, 1091). Colzato and Hommel stress that existing research on the influence of sex hormones on cognition has predominantly focused on demonstrating their effects rather than understanding the underlying mechanisms through which they modulate cognitive processes (1086). Research is required to better recognise the mechanisms behind cognitive sex differences and their relationship with identified CV risk factors in an aging society.

10.4. CV Health is associated with Advancing Age and Previous Sporting Exposure

10.4.1. The Influence of Possible Midlife Vascular Changes with CV Risk Factors on Cognitive Function

The study of CVD and associated risk factors among athletes has gained considerable attention. Many athletes from contact sports such as American Football show indications of obesity, including increases in SBP, fasting glucose, and body fat percentage (956, 1000, 1092). The results of **Study Three – Chapter Nine** indicate that a higher proportion of males were classified as overweight-obese, and similarly reported higher rates of BP and cholesterol medication use. The body composition and cardiovascular health of athletes, particularly those who have played in the National Football League, can undergo changes as they age. These changes are influenced by factors such as discontinuing regular sports activity and varying levels of physical activity. It's important to note that these alterations might not be immediately evident based on our findings from the general population during midlife (476). This underscores the importance of assessing how midlife vascular changes and CV risk factors can affect cognitive function among former athletes, raising concerns about their long-term well-being and the potential persistence of these risks despite their extensive exercise engagement.

Regular PA can improve CV health by reducing atherosclerotic risk factors like hypertension, dyslipidaemia, glucose intolerance, and abdominal adiposity. However, those in sport may still be vulnerable to CV risk factors despite their active lifestyles and still appear vulnerable to the harmful effects of cardiomyopathies and coronary risk factors with the danger of engaging in excessive exercise, or emergence of maladaptive physiological changes(992, 1093). Even though athletes benefit from regular exercise, identifying and

addressing these factors has the potential to decrease the occurrence of cardiac events among athletes. In **Study Three** we found that males reported statistically higher levels of SBP and DBP than females with 31.25% of males classified as hypertensive. CV risk factors, especially hypertension, are significant predictors of white matter hyperintensities (WMH) volume load and diminished frontoparietal white matter (WM) integrity leading to reductions in executive function as confirmed among those in midlife from the systematic results of **Chapter Six** (1094). Hypertension in midlife accompanied by alterations of WM in the frontal and parietal cortices, although not explored herein, may in part explain our results of reduced cognitive abilities in domains like executive function, attention, and working memory in line with previous evidence (422, 761, 762). Middle-aged to young adults might face a higher risk of premature pathological aging, particularly if they have a concussion history or lifelong engagement in contact sports. This could be a significant factor, as all males in **Study Three** had a background in contact sports. RHI and blows to the body in contact sports can affect the CV system, leading to arterial wall injury and calcification (485, 1095). It is therefore important to differentiate between exercise-induced cardiac remodelling (EICR) and CV pathology. EICR is the positive adaptation of the heart and vasculature to exercise catalysed by a dynamic relationship between neurohumoral and hemodynamic conditions experienced during exercise bouts, which include elevated levels of cardiac output and central aortic pressure (1096). Moreover, sex differences are apparent in EICR, with females often exhibiting less remodelling compared to males (1097) which may in part explain our findings in **Chapter Nine** although not directly assessed. Although the exact mechanisms for adaptation across a sporting career are varied and remain hypothetical, structural CV changes may be pivotal.

Arterial remodelling is a natural occurrence during aging, contributing to CVD risk (1098). Competitive sports' impact on the aortic root and ascending aorta is not fully understood, and whether exercise leads to positive structural adaptations remains uncertain (1099-1101). As individuals age, the aorta undergoes structural changes whereby the phenotypic expression of these adaptations can be skewed towards pathogenesis. Higher aortic stiffness in midlife for example is associated with WMH volume in later life with the potential to induce pathological features of AD and vascular dementia (781, 782), most notably found among retired athletes with a history of contact sports participation (488, 490, 492). Aged blood vessels exhibiting reduced compliance, elasticity, and increased stiffness lead to higher SBP and lower DBP which may align with our findings from **Study Three** of increased BP among males from contact sports (1102, 1103). These results from

older community athletes align with the suggestive early pathological aging at midlife seen in **Study Two – Chapter** Six. Our results support significant consequences of midlife hypertension in the time course and progression of domain specific cognitive impairment before the presentation of observable signs and symptoms which may only become noticeable in old age.

Deficits in later life among select cognitive domains, particularly working memory involving the right and left dorsolateral prefrontal cortex, and attention may be influenced by hypertension (769, 1104, 1105). Our results from **Study Two** show that higher-order processes like memory and attention, are more notably affected than others. Hypertension's temporal pathological changes may limit attentional capacity and inhibit the ability to suppress irrelevant stimuli, reducing cognitive efficiency as early as midlife evidenced by our systematic results in **Chapter Six**, and providing further support of results from **Chapter Nine**. Hypertension may exacerbate biological aging, negatively impacting the same three cognitive domains from midlife to later life, creating an age-dependent relationship (801, 803). However, our systematic review found an equivalent number of studies reporting both a negative and no association between midlife hypertension and cognitive decline (1106). While some studies show no relationship at midlife, uncontrolled hypertension can worsen age-related cognitive decline, making it crucial for appropriate management (430). The negative implications of hypertension however may not be identifiable through midlife cognitive testing. However, our results from **Chapter Nine**, in line with our systematic evidence from **Chapter Six** indicate significant variation in age-related cognitive trajectories and emphasise the importance of targeting modifiable CV risk factors. Focusing on midlife as a target for intervention could yield clinical benefit, but the timing and onset of pathological features and cognitive decline from midlife onwards require further investigation in the context of comorbidities.

10.4.2. The Presence of Co-existing CV Risk Factors Paves the Way for Future Cognitive Decline

Age-related cognitive decline has become a public health concern. It is the contribution of several traditional CV risk factors, to which athletes are not immune from, which may influence CV and cognitive health with age (1107). CV risk factors like hypertension and in particular T2DM catalyse pathogenic factors leading to accelerated vascular aging and cognitive decline (1108, 1109). Our findings among male contact athletes in **Chapter Nine**

may align with our systematic evidence from **Study Two- Chapter Eight** alongside recent evidence indicating that age-related decline in peripheral vascular health and factors such as insulin resistance at midlife predict cognitive decline in older adults (1110). T2DM can damage the CNS, precipitating neurodegenerative disease states (1111-1113). The underlying pathological feature contributing to cognitive decline and dementia is insulin resistance (402, 460, 1114, 1115). Insulin resistance at midlife is linked to senescent plaque buildup in the brain, a hallmark of AD (1116). Our findings from **Study Three** align with **Chapter Seven's** qualitative evidence that links midlife T2DM to later-life memory, executive function, and global cognition decline. However, three separate longitudinal studies reported inconsistent negative or null findings. This contradiction could be due to varied interindividual pathology and insulin resistance rates in T2DM. Recent meta-analyses suggest that increased levels of FPG, 2h-PG, HbA1c, and hypoglycaemia are related to a heightened risk of dementia (1117). Notably, a nonlinear positive link between FPG and cognitive disorder risk exists. Our results from **Chapter Seven** show no connection between FBG or HbA1c and cognition, such that midlife HbA1c levels did not significantly affect memory, attention, executive function, or global cognition in more than half of included studies similar to previous evidence (857). These distinct changes suggest a multifactorial treatment strategy for successful aging amidst inconsistent reports. Diabetes as a midlife risk factor for cognitive decline and dementia should therefore be considered in the context of co-factors such as hypertension and dyslipidaemia, which may lead to increased rates of cognitive decline (1118). For example, the coexistence of T2DM and hypertension can lead to stiffened cerebral arteries, increased pulsatile pressure resulting in age-related pathological features associated with dementia (1119). This may become more pronounced among older community athletes who are not adequately detrained to habituate to normal daily living conditions, leading to a rise in disease states and cognitive decline due to functional physiological and vascular disturbances.

The complex alterations in cerebral microvasculature contribute to age-related vascular cognitive decline, involving endothelial dysfunction negatively impacting CBF, neurovascular coupling, and 'Brain Vascular Reserve' (1108, 1109). RHI's can severely affect the brain-vascular reserve, causing reductions in local arteriolar vasodilation and CBF resulting in neurovascular dysfunction. While contact sporting exposure did not notably affect cognition in middle-aged athletes, the crucial factor influencing health on both functional and cellular levels might be lifetime sporting exposure as a proxy for consistent sub-concussive impacts in contact sports necessitating further investigation.

10.4.3. Underlying Processes Leading to Cognitive and CV Changes in the Ageing Athlete

Ageing and participation in team-based contact sports can increase the risk of endothelial dysfunction and hypertension. As individuals age, their vascular tissue experiences an increase in levels of p16, p21, phosphorylated p38, and double-stranded DNA breaks. Telomere length and telomerase activity are also believed to play a role where hypertension is associated with increased binding of p53 to the p21 promoter in arteries and telomere uncapping is twofold higher in hypertensive patients (1120-1122). The aortic expression of p16 is elevated in a preclinical model of hypertension, suggesting a feedback loop between cellular senescence and hypertension. However, in contrast to our BP results in **Study Three**, former middle-aged elite male rugby union players were found to have lower incidence of high BP and CV problems than the general population (481). The rugby participants had a mean sporting exposure level of more than 20 years and were predominantly amateur players (83.6%), whereas our cohort had an average sporting exposure level of >30 years. These results contradict earlier findings of increased BP in contact sporting athletes, which may be explained by the level of play achieved (collegiate vs. amateur vs. professional) (476). Endothelial activity ultimately declines with age due to decreased neo-angiogenic capability and altered sensitivity to microenvironmental cues. The ageing process is ultimately marked by a decline in cellular and tissue function and a diminished capacity to respond effectively to environmental challenges, leading to arterial dysfunction, CVD, and cognitive decline (1120-1122). Hence, further understanding the connection between age and exposure to CV risk factors at the cellular level could provide clarity on the significance of our findings within the context of PA and sporting history.

Senescent cells accumulate in arterial tissues, including atherosclerotic plaques, regardless of the presence of age-related vascular disorders (1120). Aging vessels often have medial calcification, along with age-related conditions like hypertension, dyslipidaemia, and DM, contributing to vascular remodelling and increasing the risk of atherosclerotic vascular diseases. Diagnosis of atherosclerotic coronary artery disease (CAD) is often confirmed through exercise stress testing, coronary imaging, and coronary artery calcium (CAC) scoring, which has high predictive capacity for CV-related morbidity and mortality (1123-1125). The relationship between PA and CAC is unclear (1126), with some studies reporting inverse or no relationships and others reporting U- or J-shaped relationships or even a positive relationship (1127-1131). Regular PA can reduce CVD development by improving CV risk factors, but a lifetime of sporting exposure may create a

higher CAC burden due to calcified plaques and undiagnosed traditional risk factors such as hypertension, lipid profiles, and blood glucose (1132). Our results from **Chapter Nine** show that as time passes since ceasing sporting engagement, overall PA decreases, while a higher lifetime involvement in sports is linked to higher SBP, which might indicate an underlying issue like CAC. However, more research is needed to confirm this in the context of additional CV risk factor pathology. For example, hyperglycaemia in DM leads to neuroinflammation, oxidative stress, and cellular dysfunction, contributing to cognitive decline. Hyperglycaemia known to affect cerebral microvascular endothelial cells and astrocytes, leads to intracellular glucose concentrations that catalyse cellular dysfunction, overproduction of ROS, and toxic abundance of advanced glycation end-products (AGE) with secondary activation and formation of receptor for advanced glycation end products (RAGE) (1133-1135). In addition, chronic hyperglycaemia leads to glucose deprivation and neurocognitive deficits due to reductions in endothelial glucose transporters (1136, 1137). These cellular processes may contribute to domain-specific rates of cognitive decline in midlife T2DM, necessitating further investigation, provided our findings from **Study Three** suggest that consistent, intense exercise throughout life could worsen CAD, regardless of typical risk factors, even among athletes with generally lower CAD risk profiles (1138).

10.5. Effects of Exercise on MSI Capabilities and Assessment of Cognition

Chapter Five's results reveal the SIFI's strong reliability within and between sessions, along with consistent agreement across baseline testing sessions from all six SOA conditions. Existing computerised batteries for determining the RTP status lack empirical evidence to support their validity and reliability. Studies suggest that lasting cognitive alterations may be observed several months-to-years post-concussion, particularly in executive functions like cognitive flexibility, working memory, inhibition, and interference (1139, 1140), providing the foundations for introducing the SIFI in sports assessment given its high levels of reliability. Engaging in PA while understanding perceptual information is essential in sports. Acute sessions of moderate-to-vigorous aerobic exercise in non-injured populations consistently improve cognition, especially executive functions with quicker reaction times and improved accuracy (514). However, combining cognitive assessment with exercise may lead to cognitive impairments in recently concussed athletes deemed ready to RTP, as well as in individuals with a history of concussion. The results of **Chapter Five** did not show apparent differences in perceptual performance pre-to-post exercise at moderate and high intensity. It is suggested that the lack of change in SIFI results may be

attributed to the interplay between theoretical constructs of performance and their neurobiological underpinnings.

The 'Cusp catastrophe model' suggests an inverted-U relationship between arousal and sports performance when cognitive anxiety is low (1141). However, high cognitive anxiety and physiological arousal induced by exercise diminish visual anticipation tracking performance, particularly in competitive environments (1142). Exercise at 90% of VO_2max and high cognitive anxiety during competition leads to reductions in visual discrimination and longer response times (1143). Interestingly, catastrophic performance decrements at the highest exercise intensity during our protocol did not translate into sensory and MSI cognitive performance in **Chapter Four** highlighting the SIFI's resilience. However, this may also be due to the inability to fully replicate the competitive component of sports despite reliable physiological and psychological measurements. Although not directly assessed in **Study One**, the competition for energy-rich resources for completion of physical and cognitive tasks plays a significant role in the exercise-cognition relationship which may better explain our results.

The 'Transient Hypofrontality Theory' (678) suggests exercise-induced cognitive enhancement as a result of competition for metabolic and neural resources between exercise and cognitive ability. High-intensity exercise reduces pre-frontal cortex functioning responsible for explicit learning, favouring automated tasks controlled by the brainstem (1144). This shift allocates resources to the brainstem over the pre-frontal cortex, affecting executive functions like inhibitory control and cognitive flexibility, leading to favoured activation in sensory and motor regions. However, the brain's fixed capacity for metabolic function, blood flow, and O_2 uptake may have contributed to the lack of change in SIFI performance between trials (1145). Studies using fNIRS have shown significant changes in cerebral hemodynamics in response to exercise in the prefrontal cortex (1146, 1147). High-intensity exercise can lead to hyperventilation, causing blood vessel constriction and a concomitant reduction in cerebral perfusion (1148, 1149), possibly resulting in cognitive decrements. Although such decrements were not observed in the study's results as cerebral perfusion was not measured, performance factors such as fitness levels may impact cognition.

Cognitive ability is more negatively affected in individuals with lower fitness levels due to a higher demand for metabolic resources during exercise, leading to depleted energy reserves for cognitive performance (514). In contrast, individuals with high fitness levels

experience positive effects on cognition after exercise, while those with moderate levels show no changes in behavioural performance (1150). **Study One and Three's** results showed that PA levels and previous sporting history did not affect the SIFI results, supporting the *'Life History Theory'*, which suggests that humans have limited energy resources, and energy allocation to one function may limit its use for others (1151). The relationship between O_2 levels and hemodynamic response in the prefrontal cortex during exercise affects central drive, fatigue, and cognitive performance during the SIFI (678, 1152, 1153). When O_2 levels in the prefrontal cortex reach a critical limit, neuronal and hemodynamic efficiency decline, and exercise stops. These changes depend on the intensity of the load and recovery phases (1154), which were considered in the ***Study Protocol 1.2*** (*see* ***Chapter Two*** *for methods*). A recent study showed that exercise above individual ventilatory threshold negatively impacted executive and perceptual performance (251), contrary to **Chapter Five's** results. Although there was an initial increase in blood oxygenation in the prefrontal cortex in support of the hypofrontality hypothesis, there was weak coupling between prefrontal cerebral oxygenation and cognitive performance (251). These findings, along with the results from **Chapter Four**, suggest that other physiological differentiators may be at play, conferring a level of robustness to SIFI performance in the presence of exercise.

Various factors, including exercise intensity, modality, and familiarity can modulate physiological arousal and affect cerebral neurotransmitter release. The proposed 'neuroendocrinological model' suggests that exercise can stimulate the release of noradrenaline and dopamine from the sympathetic adrenal system, affecting attention and neural information processing. Exercise-induced changes in background neural activity may influence processing speed but not performance accuracy, as supported by the **Study One's** results. Moderate-intensity exercise may lead to a decline in perceptual and cognitive performance through noradrenaline release from the locus coeruleus via the vagus nerve to the nucleus of the solitary tract (1155). However, recent research suggests that exercise can alternatively enhance cognition and attention through noradrenaline activity in the locus coeruleus-norepinephrine system (1156); *see* ***Figure 10.2.*** *below*. High-intensity exercise above 80% of VO_2max can alter perception and cognition through increased catecholamine concentrations, impacting cognitive functional networks in the dorsolateral prefrontal cortex (1155, 1157). The functional capacity of the prefrontal cortex is influenced by cerebral perfusion, which can be affected by age, exercise intensity, cognitive task difficulty, and history of concussion (372, 1158, 1159). Cognitive control measures of

switching and inhibition are found to be significantly correlated with CBF velocity in the middle cerebral artery, which is sensitive to changes in CO_2 and positively correlated with cognitive inhibitory control irrespective of age (1053, 1160). The relationship between cerebral hemodynamics and cognitive performance on tests like the SIFI, assessing the ability to ignore irrelevant stimuli, however remains to be established (1161). Nevertheless, the catecholamine influence of the neuroendocrinological model on exercise and cognition cannot be disregarded, as noradrenaline can enhance post-synaptic efficacy and sensitivity in hippocampal neurons and enhance the P3 amplitude (1162, 1163). Varying physiological modulators can significantly impact cognitive performance, depending on multiple energy pathways influenced by exercise intensity, where select proteins and metabolic by-products such as lactate may also alter perceptual performance.

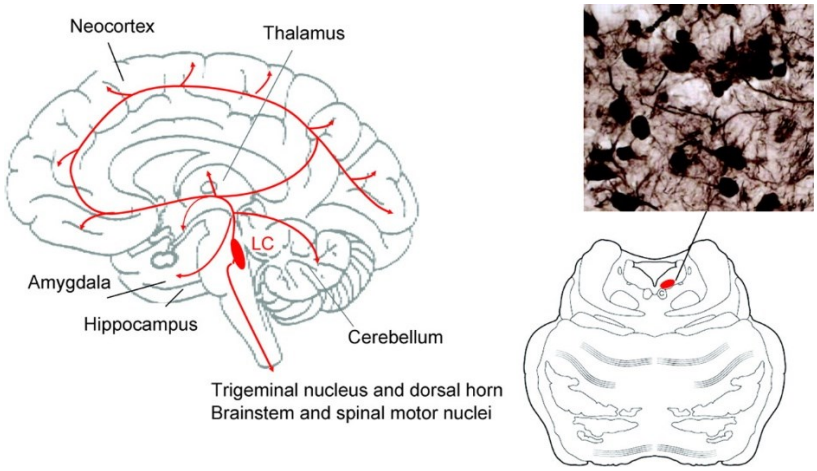

Figure 10.2. *The locus coeruleus norepinephrine system. *Adapted from the work of Benarroch (1164).*

Exercise can have various cognitive benefits, inducing neurogenesis, neuronal plasticity, and enhancing cognitive function through the expression of BDNF (686, 1165-1167). High-intensity exercise leads to peak blood lactate accumulation and circulating BDNF levels, potentially enhancing cognition (695, 1168). The 'astrocyte-neuronal lactate shuttle theory' suggests that lactate is released by astrocytes to provide metabolic support to neurons, and glutamate triggers anaerobic glycolysis in astrocytes, leading to lactate production (688, 1169). Lactate, known to cross the BBB via endothelial MCTs, can serve as an alternate CNS energy source, providing neuroprotection and promoting cognitive enhancements via synaptic plasticity transmission (1170). This occurs through BDNF/TRKB

signalling, activation of the SIRT1/PGC1a/FNDC5 pathway, and expression of plasticity-related genes like Arc, Zif268, and c-Fos in a lactate-dependent manner (687, 1171). Previous research links lactate to enhanced cognition and brain health in young adults, including improvements in visuospatial and declarative memory, as well as motor skill acquisition and retention (696, 1172-1174). Although, glutamate reductions have been observed in individuals with a history of concussion which may impact cognition post-recovery The brain prefers lactate over glucose when blood lactate levels rise above 2 mM, a threshold typically exceeded in high-intensity exercise protocols (686, 1175), as was seen in **Chapter Five**. It may be proposed from **Study One** that SIFI performance is unaffected following exercise, might be due to an increased ability to remove lactate and a reorganised consumption of energy substitutes to preserve cognition and perceptual performance. However, the role of the vasculature and distribution of blood flow as mentioned previously may be key to understanding **Study One's** results, particularly among those with a history of concussion. In all, the SIFI task shows a high level of reliability and versatility, boding well for its future implementation among athletes of all ages, which in turn would enable more insight into the relationship between key markers of cognitive and CV health related to a lifelong engagement in PA and sport.

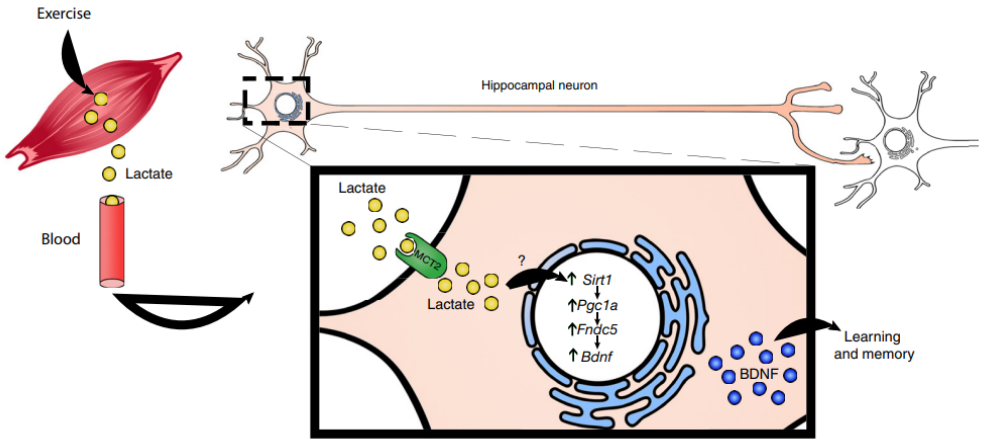

Figure 10.3. *A proposed model by which exercise induces BDNF expression in the hippocampus via lactate production. *Adapted from the work of El Hayek and colleagues* (1171).

10.5.1. Cardiorespiratory Fitness and PA Levels are a Key Indices of Brain and CV Health

There is consistent evidence that PA and exercise in mid-life is correlated with better global cognitive function in later life indicative of successful and healthy ageing (1176). Results from **Study Three** found moderate-to-high levels of PA were evident with vigorous activity linked to the highest number of recorded MET's. Although, no significant relationship was found between cognition and PA levels despite current evidence that regular exercise and PA can help reduce age-associated cognitive decline, and the degree of cognitive improvement is associated with cardiorespiratory fitness (CRF) and vascular ageing (646, 1177). Current data indicates a potential link between improved aerobic fitness levels and a favourable vascular profile, which may delay the onset of age-related cognitive impairment. Variability in cognitive trajectories is highly dependent on inter-individual factors influencing age-related and injury-exacerbated cognitive decline (1178) as seen throughout **Study Two**, particularly **Chapter Eight** surrounding cholesterol, which may explain the lack of significance found in **Chapter Nine**.

Ageing mediates significant alterations in cerebral cholesterol, leading to cognitive decline and neuropathology (915-918). Epidemiological studies similar to our results in **Chapter Eight** show conflicting evidence regarding midlife cholesterol suggesting ageing as a determining factor (439, 904). Although, there is an ongoing debate regarding the association between midlife lipids and dementia. Some evidence suggests that high levels of HDL-C and TC in midlife are key risk factors for AD and dementia in later life (875, 1179), while inconsistent findings indicate both high and low TC levels in midlife as predictors of cognitive decline and dementia emergence in later life in line with our findings (875, 904, 1179, 1180). Our systematic evidence from **Chapter Eight** suggests a protective role of midlife HDL-C, hypercholesterolemia, and TC to some extent in preserving cognitive function in later life (437, 832, 883). The rate of cognitive decline may serve as a surrogate marker of metabolic dysregulation in relation to cholesterol changes with age. For example, high baseline HDL-C in later middle age and early later life protects against cognitive decline up to 2 decades later, though some investigations found no relationship (930-932). Contradictory evidence suggests only elevated TC contributes to AD and cognitive decline, with other cholesterol markers remaining in the normal range (440, 730, 877, 905, 908). The relationship may therefore be bidirectional, with increased midlife TC leading to age-associated cognitive decline later in life which is of importance given our findings from males in **Chapter Nine**. This concept of 'reverse causation' begs the question as to when

and how cholesterol affects cognition across an ageing population. Longitudinal and prospective cohort studies show a decline in serum cholesterol from midlife onwards increases dementia risk with more than a three-decade follow-up, although the timing of lipid exposure in relation to cognitive decline remains uncertain (877-879). Follow-up intervals (i.e., *before or after the age of 65*) can therefore play a significant role in outcome variations and our understanding of cognitive decline, especially among those who have participated in lifelong exercise.

Long-term engagement in PA is considered beneficial for an aging population, helping to prevent age-related declines in vascular and cognitive function. However, there is growing evidence that excessive exercise may be detrimental to CV and cognitive health. Hippocrates once said, "*Everything in excess is opposed to nature*", but what is too much exercise in the wake of a sedentary society outside of the sporting world? Midlife individuals engaging in regular PA have enhanced peripheral endothelial function, higher CRF, and better cognitive function compared to sedentary counterparts (1181, 1182). Moderate and high-intensity exercise has been shown to reduce CV risk factors with age (1183-1185), but CV adaptation to exercise appears to be intensity-dependent (1186). It is important to note that engaging in very high doses of intense exercise training could have negative effects on indices of health (1187). Determining a precise numerical threshold for excessive exercise with negative health effects nonetheless remains challenging, as it varies among individuals due to factors such as age, fitness, and overall health status. Excessive exercise typically involves a sustained and substantial increase in exercise volume, intensity, or duration, leading to adverse health outcomes. These outcomes may encompass overtraining syndrome, injuries, hormonal imbalances, immune suppression, sleep disturbances, and other related health issues. However, the longer the length of training across the lifespan, the larger the achieved health benefits acutely on body mass, body fat, resting HR, VO_2max, TG's, and HDL-C (1188), which can dissipate over time following exercise cessation. Moderate intensity exercise is commonly used to treat CV risk factors, leading to enhanced endothelial function, reduced BP, and in particular insulin sensitivity (1189, 1190). These modifications account for 59% of the decrease in CVD, while other factors like increased vagal tone, vascular remodelling, and increased nitric oxide bioavailability contribute to the remaining 41% (1017, 1191). In contrast, recent evidence suggests high intensity exercise may offer equal or enhanced benefits for CV risk reduction and brain health (1192-1194). However, the effects of exercise discontinuity, or detraining, need to be considered, as rapid return to baseline values of cholesterol indices for example may occur after cessation

(1195-1197). High intensity exercise appears to have a more significant impact on lipid balance and body composition on a long-term basis compared to moderate intensity exercise, particularly among middle-aged and older individuals (1198, 1199). However, the dose-response curve between exercise and mortality varies, with smaller volumes of vigorous intensity exercise associated with maximum risk reduction (1191). Cholesterol and TG's found within lipid droplets can accumulate, causing cellular stress and cognitive decline (1200). Our results from **Chapter Eight** identified the relationship between cholesterol, its sub-components, and cognitive function in midlife and later life to be inconsistent. Epidemiological and pathological investigations have proposed that modifiable CV risk factors including, hypertension, DM, and high cholesterol lead to cognitive decline in later life although with some controversy in line with our review (415-419). Debates are ongoing whether the protective effects TC and LDL-C may in fact may outweigh the cognitive risks, except in the presence of other CV risk factors (417). Inconsistent findings suggest that the relationship between cholesterol and cognitive decline is influenced by age at measurement, follow-up duration, and other CV risk factors (417, 943). The effectiveness of lifestyle changes or lipid-lowering medications in preventing cognitive decline and dementia requires further research (1201). It is essential to understand the individual variability in responses to exercise and further investigate the impact of exercise intensity on CRF benefits and CV risk factors among an aging population (1202, 1203). Exploring physiological pathways behind exercise-induced neurocognitive capabilities could aid in developing appropriate therapies for combating age-related cognitive decline and extending to issues surrounding cognition and mental health in aging athletes from midlife onwards.

10.6. Cognitive Function of Ageing Athletes in Contact Sports

Objective investigations into the long-term cognitive health of retired community athletes with a history of concussion are scarce. Limited assessments have mostly concentrated on self-reported memory function, neglecting other domains like attention, perception, visuospatial, and psychomotor functions (3, 482). Our findings from **Study Three** suggest that lifetime sporting exposure had no effect on cognition. Although no differences by sex were evident for cognition, MoCA scores approached the clinical cut-off for MCI. However, the design of **Study Three** provided a mere 'snapshot' of the population's current health status assessing exposure and outcomes without altering the participants' exposure status, where differentiating cause and effect from association can be challenging.

Cognition was not found to be correlated with PA level, concussion history or lifetime sporting exposure, possibly due to the small sample size, which is opposed to previous evidence and our initial hypothesis. Although the exact underlying mechanisms remain to be fully understood, there is the possibility that it is both structured and functional alterations at the cellular level with sporting exposure over time which may influence cognitive abilities to a greater extent dependent of competitive level of play and sporting type.

Athletes who participate in contact sports have an increased risk of cognitive dysfunction, mental health deterioration, and inter-regional connectivity deficits related to time since play when sporting retirement is reached (393, 1204, 1205). The severity and heterogeneity of concussion and mTBI make it difficult to understand the degeneration of WM tracts and atrophy of associated structures. Multiple studies have reported that athletes with a history of concussion, even sub-concussive impacts, in contact sports can result in significant negative effects on brain structure and function within a short period of time (1206). These changes in WM tracts can be correlated with impaired cognitive function, creating a state of axonal degeneration over time suggestive of neurodegeneration (25, 1207, 1208). Moreover, the long-term effects of concussion and RHI from collision-based, contact sports are evidenced structurally by cortical thinning of GM, and reduced volume of the hippocampus and amygdala in both active and retired contact sport athletes (962, 1209, 1210). These structural changes are strongly linked to functional impairments in cognition, with concussion history negatively impacting memory, and executive function, but not MSI capabilities (3, 172, 482). However, this was not the case in **Chapter Nine**, where both cognition and MSI were unaffected by previous sporting experience and concussion history, indicating that ageing may be a predictor of cognitive status in later rather than midlife.

Aging leads to harmful functional changes in cells and tissues, resulting in a decline in physiological function and increased risk of morbidity and mortality. Chronological aging involves cellular senescence, alterations in intercellular communication, mitochondrial dysfunction, and nutrient sensing disruptions. Cellular senescence is marked by chronic inflammation and the release of senescence-associated secretory phenotype factors, promoting tissue remodelling over time (1211). Senescent cells accumulating in the CNS can contribute to neurodegeneration and cognitive decline, exacerbating the aging process (1212, 1213). Chronological aging is associated with DNA damage, telomere shortening, oncogenic stress, and exposure to reactive oxygen species (ROS). These factors activate the

p53 pathway, crucial for maintaining genomic stability and coordinating DNA repair, cell cycle regulation, apoptosis, and cellular senescence. The p53 protein, known as the "guardian of the genome," also influences cell metabolism, autophagy, antioxidant defences, and angiogenesis, making it central to cellular senescence. Cellular senescence can occur through replicative senescence (telomere shortening) or stress-induced premature senescence (various stress signals). Chronic cellular stress, such as RHI and mTBI, can trigger premature senescence leading to neurodegeneration due to a dysfunctional DNA damage response (DDR) pathway. (1214). A study of professional contact sport athletes with a history of RHI found that more than 90% of mTBI cases had neurobehavioral and/or psychiatric symptoms, cognitive dysfunction and/or dementia, and early-stage CTE. Findings indicate that genes associated with DDR pathways and gene integrity protection were downregulated among individuals with a history of mTBI, suggesting that cellular senescence can significantly impact overall brain health and may be worsened by chronic stress like RHI and mTBI. (1214). While not examined in **Study Three**, the absence of notable cognitive changes in midlife could potentially be attributed to cellular aging and chronic stress. This suggests that clinical indications might manifest later in life, and a deeper comprehension of cognitive decline among community-level athletes as they age could be gained through cognitive-behavioural frameworks.

Cognitive reserve is a crucial factor predicting the age of onset of neurodegenerative diseases (1215). It is part of a dual reserve framework comprising active and passive reserves, which define thresholds of neural damage before clinical cognitive deficits occur. The passive reserve model involves neural substrates that determine a threshold of neural damage leading to cognitive deficits. The active reserve model suggests that individuals with higher cognitive reserve can delay the onset of MCI and incident dementia, recovering faster from neuropathological disruption by recruiting existing cognitive systems and compensatory coping mechanisms (965, 1216). Cross-sectional studies indicate that cognitive reserve is a modifiable risk factor for RHI exposure and CTE. However, these studies have limitations in capturing functional cognitive recovery over time (1217, 1218). Individuals with a history of concussions may use additional brain regions to compensate for persistent cognitive and neurological issues even after clinical recovery (1219). This compensation is associated with a diminished cognitive reserve, extending to those with suspected CTE (395-397), although did not appear to be expressed in our results from **Chapter Nine** possibly due to level of competitive play. For example, more than a third of retired NFL players aged 30-65 years have deficits across two or more cognitive domains.

Concussion history explained 9% of the variance in global cognition, whereas cognitive reserve accounts for 25% of the variance in cognition (1220). Research suggests that not only concussion history, but also sub-concussive RHI's may contribute to neurobehavioral and cognitive decline in former athletes (386, 389, 391). However, inconsistencies exist in the literature, with some studies finding associations between longer participation in professional sports and cognitive impairments independent of concussion history (391, 1204, 1221), while others report no significant associations (1222-1224). These discrepancies could be due to the measurement of RHI, often using years of participation in a sport as a proxy as was undertaken in **Study Three**. Recent research attempts to overcome this bias by using the Head Impact Exposure Estimate (HIEE), a verbal interview with athletes to quantitatively estimate head impacts over their career. The HIEE has been associated with worse cognitive performance and general psychological distress in former athletes (1225, 1226), and its use would enable more accurate insight surrounding lifetime sporting exposure which has been known to impact the mental state of many retired athletes.

10.7. Depression (Mental Health) is Correlated with Lifetime Sporting Exposure & PA Levels

The prevalence of mental disorders is highest among younger individuals, particularly those aged 16-34, with about 25% meeting clinical criteria for depression or other disorders (1227). **Study One's** results align with this, showing over half of all participants reporting some level of depression, with those who had experienced concussions reporting higher levels of anxiety and depression. While younger athletes make up a significant portion of the community and elite populations, there is limited knowledge about the frequency of mental disorder symptoms in older community athletic groups. Available data indicates that athletes may experience mental disorders, such as depression, at rates similar to the general population, but further clarification is needed on specific sporting sub-populations. Findings from **Study Three** indicate among older athletes at midlife there was a moderate correlation observed between the extent of lifetime engagement in sports and the presence of depression. Additionally, the duration since discontinuing consistent participation in sports displayed a correlation with indicators of somatisation and mental health, whereby an inverse correlation was observed in overall mental well-being. Despite the belief that athletes, due to their engagement in high levels of PA, might be less susceptible to mental

disorders like depression, the current evidence is insufficient to support this assumption. Our results from **Study Three** indicate that total levels of PA are not correlated with mental health scores at midlife regardless of moderate-to-high levels of PA recorded from the sample population. The prevalence of mental disorders among retired athletes remains controversial. Contributing factors such as exposure to concussion, sport-related stress, and biological sex differences could increase vulnerability to mental disorders, including overall higher depression rates.

Limited attention has been given to sex differences in mental health among athletic populations (1228), despite notable differences in the general population (1229, 1230). Male athletes may underreport symptoms due to socialisation and mental health stigma in sports (1231). Recent reports show women have higher rates of mental health issues (1232), consistent with **Study One's** findings although contrary to **Study Three**. No identifiable sex differences were observed indicating that other factors not presently analysed may be contributory. Our results from **Chapter Nine** indicate the duration of sports engagement throughout one's life showed a moderate correlation with the experience of depression and was further linked to PA levels. Moreover, the length of time since discontinuing regular sports participation was found to have a correlation with indicators of somatisation and mental health, with an additional negative correlation observed between this discontinuation period and overall mental well-being. These findings suggest that both the extent of lifetime sports involvement and the duration since stopping regular sports can impact various aspects of mental health, including depression, somatisation, and overall well-being which may begin as early as young adulthood. The results from **Study One** indicate among younger adult athletes, the number of previous concussions was significantly correlated with worse outcomes across metrics of somatisation, anxiety, and depression but may not imply a causative link between the relationship. Similarly, self-diagnosed depression correlates with the number of self-reported concussions in retired contact athletes at midlife (1023, 1233), but the link to depression risk is not always clinically significant with >85% of retired players report no depressive symptoms (1234). Our findings from **Chapter Nine** are in line with the latter as no significant correlation was observed between the number of previous concussions and metrics of mental health at midlife. Further research is necessary to include community-level athletes and enabling more generalised findings. This will facilitate the development of appropriate programs and interventions to support a smooth transition into sporting retirement at various ages and levels of play.

10.8. Future Research and Directions

There are multiple novel research questions resulting from this PhD project. Findings from **Study One - Chapter Five** suggest that exercise of moderate and high intensity had no effect on perceptual performance assessed using the SIFI. The potential impact of NVC and CBF on cognitive function and cerebral well-being, particularly within the context of prolonged exercise engagement and concussion history, has been previously highlighted (1235). The evaluation of NVC response within the posterior cerebral artery during visual tasks is a possible avenue for investigating the dynamics of the visual and auditory cortices. Prior investigations have shown consistent maintenance of NVC response after a season of contact sport engagement, compared with acute alterations observed in the wake of SRC (1236, 1237). Moreover, individuals with a history of concussion have consistently diminished frontotemporal CBF when compared to those without concussion history (1238). A prospective longitudinal approach would be useful in further understanding the cerebral activation patterns occurring during exercise in cohorts of athletes over time.

The study's cross-sectional design facilitated efficient data collection yet posed limitations on the establishment of causality due to single-instance measurements and potential biases related to participant selection and recall. To interpret the evolving nature of risk factors across a lifespan of sporting engagement and post-retirement phases, encompassing both amateur and professional athletes, longitudinal investigations are warranted. Objective data on concussion history would strengthen the results obtained in **Study One and Three** and should be included in any future study design where possible. Incorporating control groups with no contact sport history would be essential to ensure a robust evaluation of findings for accurate risk interpretation. **Study Three** employed the SIFI to identify subtle pre-clinical alterations in cognition among older community athletes with histories of concussion and prolonged sports involvement. Nonetheless, comprehensive investigations involving a larger cohort of retired athletes are required to determine clinical significance. Employing a longitudinal framework spanning intervals of 3, 5, and 10 years would enable an in-depth exploration of cognitive and CV function, body mass fluctuations, and other pertinent health-related parameters.

The current body of literature predominantly centres on brief, transient investigations focused on male cohorts immersed in contact sports. This underscores the necessity for analogous inquiries spanning diverse athletic disciplines, aimed at comprehensively understanding the long-term consequences of lifelong sporting engagement on subsequent health outcomes. As in **Study One**, no sample size calculation was performed for **Study**

Three and thus it may be underpowered. This introduces the potential influence of convenience sampling and selection bias. To prevent such limitations in forthcoming research, the prospective power calculations should be performed. Moreover, the incorporation of a larger control group consisting of retired elite athletes from non-contact sports would serve the dual purpose of reducing potential biases and enhancing the overall robustness of the investigation.

The participants enrolled in **Study Three** had a low prevalence of concurrent morbidities which would influence both vascular function and cognitive performance. Subsequent investigations should aim to ascertain the prospective predictive capacity of peripheral vascular health assessments within larger cohorts. The incorporation of an extensive composite peripheral vascular health metric, encompassing microvascular reactivity, arterial endothelial function, and vascular rigidity may pinpoint individuals with a susceptibility to VCID, allowing a greater exploration of the role of age-related vascular dysfunction in cognitive decline from mid to later life.

The insights derived from the findings of **Study Two** underline the importance of dissecting the interplay between CV risk factors; including hypertension, DM, and cholesterol levels; and cognitive performance in aging populations, particularly those with a history of engagement in sports. Our results suggest that memory, executive function, and global cognitive domains were susceptible to the negative influence of hypertension and T2DM during midlife, although the impact of cholesterol was unclear. The cellular abnormalities identified within retired rugby players during midlife, demonstrated by neurovascular decoupling, suggest potential links to the history of SRCs. This disruption of cerebral autoregulation may culminate in a decline in cerebral perfusion pressure, impair CBF, and perturb perivascular neuronal function. Consequently, this cascade of events could increase the vulnerability of frontal cortices and their associated functional networks to the process of cognitive decline that occurs with advancing age. Longitudinal investigations focused on aging athletes with a history of concussions, could reveal the role of cerebrovascular and hemodynamic shifts that contribute to the trajectory of cognitive deterioration.

Across all three studies, a coherent pattern of sexual dimorphism in cognitive domains has emerged, spanning the transition from early adulthood to midlife. Specifically, male participants consistently have superior perceptual performance in comparison to their female counterparts, a phenomenon consistently observed under varying SOA conditions

in **Studies One and Three**. While not within the immediate purview of this thesis, it is important to acknowledge the established influence of oestrogen on higher-order cognitive processes and GABAergic activity, particularly in relation to menstrual cycle phases. To advance our understanding, future investigations should delve into the intricate interplay between hormonal fluctuations, menopausal transitions, and cognitive function within the context of aging cohorts, especially considering the concomitant impact of prior concussion history.

10.9. Conclusion

This thesis offers valuable insights into the complex interplay between brain and heart health with age, particularly concerning the impact of SRC. Through extensive literature review and original data analysis, important findings have been revealed.

Study One found a significant association between increased susceptibility to the SIFI illusion and sex, with males outperforming females in all conditions. Open skill sports showed higher levels of MSI, possibly due to training and exposure, but many athletes in these sports are also engaged in contact-based sports with higher concussion rates. While **Study One** could not fully explain the relationship between MSI and previous sporting experience, future research is needed to identify sport-specific metrics to enhance MSI assessment interpretation among athletes. Additionally, the study did not find differences between those with and without a history of concussion as hypothesised. Large-scale, prospective longitudinal studies are necessary to confirm concussion effects both acutely and beyond the recovery period throughout a sporting career, allowing interventions for community-based athletes.

Study Two provided insights into the potential underlying mechanisms connecting brain health and heart function in midlife and beyond. Shared pathological pathways, including chronic inflammation, oxidative stress, and impaired autonomic regulation, were hypothesised across three distinct CV risk factors: hypertension, T2DM, and cholesterol. A notable relationship between CV risk factors and cognitive decline was found for memory, executive function, and global cognition, suggesting common physiological alterations contribute to adverse cognitive outcomes among select domains. Targeted interventions addressing these pathways at midlife may be crucial to protecting long-term brain health. Further investigation is needed to understand brain and CV health of both community and

elite athletes transitioning into sporting retirement, considering the extensive evidence on their CV risk profiles with advancing age.

The findings from **Study Three** demonstrated that no significant impact or long-term consequences of participation in contact sport on brain health were observed There was however an increased risk of CV events, as a higher proportion of males were classified as overweight-obese, hypertensive, and taking antihypertensive medication. These results underscore the critical importance of considering concussion history as a significant factor in assessing the health trajectory of athletes, both during their active years and in later life. Previous data has shown a progressive decline in cognitive function, as well as a higher incidence of CVD, among older individuals. Importantly, these age-related changes are exacerbated in individuals with a history of concussion, suggesting a cumulative effect of brain trauma on overall health outcomes. Our results found that midlife participants' susceptibility patterns to the SIFI echo those observed in **Study One - Chapter Five**, delineated by high, moderate, and low performance levels. While sex lacked substantial predictive power for perceptual accuracy, intriguingly, males have reduced susceptibility across a broader spectrum of temporal asynchronies. Notably, non-contact sports consistently outperformed contact sports across various SOA conditions, with no significant differences between open and closed skill sports, contrasting **Study One's** findings. These observations collectively emphasise the complex interplay between cognitive function, sports engagement, and physiological factors, setting the stage for future investigations into cognitive health within athletic contexts of the general population.

This thesis has significant implications for athletes, coaches, healthcare professionals, policymakers, and the general public, providing empirical evidence on the long-term impact of concussions on brain and heart health with age. It calls for comprehensive prevention strategies, improved diagnostics, and evidence-based treatments incorporating a more multi-dimensional model of care with clinical and cognitive assessment. The research advances our understanding of the brain-heart relationship and highlights the need for proactive measures to protect athletes and the general population. Future research efforts should focus on understanding underlying mechanisms and developing targeted interventions to promote healthy aging and quality of life for both athletes and non-athletes alike.