THE ASSOCIATIONS BETWEEN FOOTBALL EXPOSURE, CONCUSSION HISTORY AND PLAYING POSITION ON CEREBRAL WHITE MATTER INTEGRITY AND NEUROCOGNITIVE PERFORMANCE

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ABSTRACT

ALLEN CHAMPAGNE: The associations between football exposure, concussion history and playing position on cerebral white matter integrity and neurocognitive performance
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Diffusion tensor imaging (DTI) has emerged as an important tool for quantitative analysis of white matter (WM) integrity following sport-related concussions. The purpose of this research was to investigate the variances in WM integrity (defined by fractional anisotropy (FA) and medial diffusivity (MD)) and neurocognitive performances in retired college and professional football athletes based on concussion history, duration of playing career, and playing position. MRI scans and neurocognitive test scores from 32 former college and 31 former age-matched professional players (avg age=58.5 SD=3.7) were compared. A permuted, voxel-wise 3x2 ANOVA was performed on the WM skeleton to investigate the main and interaction effects on WM integrity. Threshold-free cluster enhancement (TFCE) was used to identify clusters of significantly different FA or MD and post-hoc univariate analyses were used to determine the direction of interaction effects. A priori alpha level was set at 0.05 after correction for multiple comparisons. Differences in FA were observed in 3 clusters in the forceps minor and genu of the corpus callosum for the concussion by position interaction. Post-hoc analysis of the peak voxels within each cluster revealed consistently lower FA in non-speed athletes with 3+ concussions as compared to those with 0-1 concussions (Cohen's d: 0.89, 0.95, and 1.29; $P<0.05$). No clear differences in neurocognitive abilities were identified. Our results suggest a history of multiple concussions is associated with lower FA in former non-speed position players compared to speed players, particularly in frontal white matter tracts. We did not observe main effects of football exposure, suggesting that without concussive injuries, added football exposure does not account for variances in white matter integrity and neurocognitive abilities. A limitation of these results is the lack of a control group without history of football participation.

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CHAPTER I: INTRODUCTION

1.1 Goal of the work

The purpose of this research is to investigate the long-term effects of concussive traumatic brain injuries, football exposure and playing position on white matter integrity and neurocognitive performances in former professional and college football athletes. It is intended, via the use of diffusion tensor imaging and neurocognitive testing, to determine the degree of neuroanatomical and neurocognitive variance between those two populations based on concussion history, duration of playing career, and playing position. Conclusions about the clinical use of brain-imaging techniques to assess concussive injuries and the predictability of the structural integrity changes are expected from this project.

1.2 Background and Significance

The field of traumatic brain injuries (TBI) has attracted a lot of attention in the past few years with the fast growth of the branch of research as well as the increasing number of players that are suffering of post-injury, long-term neurocognitive deficits. This has become a public health concern, even more so with the recent evidence suggesting earlier onset Alzheimer's and increased prevalence of memory problems and dementia [1-3]. This work is therefore an extension of the current research projects that are conducted on traumatic brain injuries and hopes to provide clarifications on the structural differences in cerebral white matter of retired football players. More research on concussions has established relationships with high risk of long-term memory deficits and it is for those reasons that this project is significant for this field of research. Hopes are geared towards better identifying the long-term damaging effects of these injuries, and their neuroanatomical underpinnings, in former college and professional football athletes. This work will also potentially improve our understanding of the correlation between the sub-clinical neurocognitive deficits observed in players and the number of concussions they have sustained during their career along with total exposure hours and playing position. Looking further down the path, our findings may allow for more accurate prediction and understanding of the clinical significance of regional neuroanatomical changes shown on neuroimaging tests. To date, no similar studies have collected longitudinal multimodal imaging in individuals with a

history of concussions, different levels of football exposure and positions with the aim of investigating the contribution of these potential predispositions to differences in white matter integrity and cognitive aptitudes over time. As such, the proposed study represents the first of its kind.

Our research group already explored the field of neuroimaging by utilizing event-based functional magnetic resonance imaging (fMRI) to examine long-term differences in functional activity during memory tasks in former athletes who have sustained sport-related concussions [4]. In that study, former football players reporting multiple sport-related concussions (i.e., three or more) were compared to players who reported zero or one concussion during a memory paradigm, which examined item memory (i.e., memory for the particular elements of an event) and relational memory (i.e., memory for the relationships between elements). Behaviorally, it was observed that concussion history did not significantly affect behavioral performance, as individuals in the LOW (0-1) and HIGH (≥3) concussion groups had equivalent performance on both memory tasks. Despite demonstrating equivalent behavioral performances, the two groups of former players showed different neural recruitment patterns during relational memory retrieval, suggesting that multiple concussions may be associated with functional inefficiencies in the relational memory network. Additionally, the number of prior concussions was significantly correlated with functional activity in a number of brain regions, including the medial temporal globe and inferior parietal lobe. Such results provide important insights in understanding the long-term functional repercussions of sustaining multiple sports-related concussive injuries. As mentioned above, the proposed work will be a continuation of the current findings on sport related concussive injuries and exposure to repetitive head impacts with the use of diffusion tensor imaging to further investigate the differences in cerebral white matter integrity.

1.3 Specific Aims/Research Questions

1.3a Primary aim - Investigate the integrity of the white matter tracts in National Football League (COL+NFL) and college (COL) football retired players with respect to concussion history, football exposure, and playing position.

1. Are there significant differences between the integrity of the white matter tracts, defined by fractional anisotropy (FA) and medial diffusivity (MD), observed in diffusion tensor images of National Football League and college football retired players based on years of football exposure, position played and concussion history?

    (1) Variable analysis:

        I. Independent: Years of football exposure, playing position and concussion history.

        II. Dependent: The integrity of the white matter tracts (defined by FA and MD).

    (2) Hypotheses:

        I. Research hypothesis: We hypothesize that significant differences in white matter integrity, more specifically, decreased FA and increased MD, will be observed in the high concussion history group and the speed position group.

        II. Statistical hypothesis:

            1. Null (H_o): There will be no significant differences between white matter integrity across the groups.

            2. Alternate (H_A): There will be significant differences in white matter integrity across groups; more specifically speed players of the high concussion history group will show greater degree of white matter alterations compared to all other groups.

1.3b Secondary aim: Investigate the effects of concussion history, football exposure and playing position on neurocognitive performances of National Football League and college football retired players.

1. Are there significant differences between the performances of National Football League and college retired football players on the selected neurocognitive battery tests based on football exposure, playing position and concussion history?
    (1) Variable analysis:
        I. Independent: Years of football exposure, playing position and concussion history.
        II. Dependent: Performance on selected neurocognitive battery tests.
    (2) Hypotheses:
        I. Research hypothesis: We hypothesize that the high concussion history group will demonstrate greater deficits in neurocognitive tests performance compared to the other groups.
        II. Statistical hypothesis:
            1. Null (H_o): There will be no significant differences in performance on the neurocognitive testing across the groups.
            2. Alternate (H_A): There will be significant differences in performance on the neurocognitive testing between the groups. More specifically, the high concussion group will perform worse than all other groups.

1.3c Exploratory aim: Investigate the relationship between neurocognitive performances of National Football League and college football retired players and observed structural differences in white matter integrity.

1. If there are significant differences between the performances on the neurocognitive tests, do the observed differences in cerebral white matter integrity (from DTI) correlate with cognitive domains where athletes have shown performance deficits?
    (1) Variable analysis:
        I. Independent: The structural differences in white matter integrity (expected decrease in FA and increase in MD) observed from years of football exposure, position played and concussion history.
        II. Dependent: The performance on neurocognitive tests.
    (2) Hypotheses:
        I. Research hypothesis: We hypothesize that there will be a correlation between regions of significant differences in white matter integrity (decreased FA and increased MD) and deficits in neurocognitive test performances involving such damaged regions.
        II. Statistical hypothesis:
            1. Null (H_o): There will be no significant correlation between the performances on neurocognitive tests and region-specific structural differences in white matter integrity (decreased FA and increased MD).
            2. Alternate (H_A): There will be significant correlations between the performances on neurocognitive tests and region-specific structural differences in white matter integrity (decreased FA and increased MD).

1.4 Operational Definitions

Concussion: Functional brain injuries resulting from rapid direct or indirect biomechanical forces applied to the head that may cause tissue alterations and lead to physiological and neurological complications [5].

Concussion History: Subjects will be split into two groups: A 'LOW' concussion history group, with players who have reported none or a single concussion injury (0-1), and a 'HIGH' concussion history group, with players who have reported three or more concussive injuries (≥3).

Football Exposure: Determined by the subjects' number of years of football played and the weighted number of hours from both practices and games. Subjects will be divided into two groups: Players who have played college football only ('COL') and players who have played both college and professional (NFL) football ('COL+NFL').

Sub-concussive head impact: Repeated head trauma, none-concussive (that does not result in a concussion), that may also contribute to the development of neurodegenerative diseases such as CTE [6].

White matter integrity differences: Linear rearrangement of the axonal cytoskeleton caused from stretch-induced axonal damage (swelling, disconnection, retraction) within the cerebral white matter detected and quantified by diffusion tensor imaging [7].

1.5 Delimitations

1. This study recruitment was delimited by the fact that data was already collected, which restricted what could be done and future analyses. The previous recruiting team selected all selecting variables.
2. Participants are aged between 50-65 years (N=63) and consist of 32 former college players and 31 National Football League retired players.
3. The previously recruited population of college and professional retired players limits our future positional analysis.
4. Subjects were randomly assigned an order of testing conditions in order to control for procedural bias within the order of test taking.
5. No cognitively impaired subjects were included in the study in order to provide insightful results form the neurocognitive performance tests.
6. LOW concussion group was defined with individual who have suffered none or a single concussive injury.
7. HIGH concussion group was defined with individual who have suffered of three or more concussive injuries.

1.6 Assumptions and Limitations

1. Assumption: The data collected by the DTI and fMRI technicians is valid and reliable.
2. Assumption: The scanner used to collect the DTI and fMRI images were properly adjusted and functional.
3. Assumption: Subjects were honest in answering all questions and in adhering to the inclusive and exclusive criteria required for this study.
4. Assumption: DTI analyzing tools used in this study, such as TBSS, are reliable and accurate.
5. Assumption: Subjects gave maximal effort when preforming their neurocognitive tests.
6. Limitation: No baselines were used to compare the potential changes in white matter integrity and neurocognitive performances.
7. Limitation: Subjects may have experienced learning curve practice effects as a result of repetitive testing and/or from previous exposure to concussion assessment tools.

1.7 Direction of the thesis

Comparisons of white matter integrity and neurocognitive performances will be used to infer conclusions about the neuroanatomical and neurological long-term effects of traumatic brain injuries and repetitive head traumas sustained by football players. Such could potentially provide meaningful information about the effects of region-specific white matter structural changes and their impact on tested cognitive functions.

CHAPTER II: LITERATURE REVIEW

2.1 Introduction

Defined as a complex pathophysiological process induced by rapid biomechanical forces applied directly or indirectly to the head, concussions are functional brain injuries that result in tissue alterations, which lead to physiological and neurological complications [5]. In 2011, the Centers for Disease Control estimated that 1.6 to 3.8 million concussions occurred in sports and recreational activities [8]. Such is still an under-estimate, although it includes an expected number of individuals who suffered of concussive injuries and did not seek medical assistance. American football, among all other sports played in the US, has the greatest number of traumatic brain injuries, but also the largest participation rates [9]. Because of its popularity and such high rates, traumatic brain injuries sustained in football players deserve deeper examination in order to allow coaches, athletic trainers and physicians to better assess and manage head injuries.

The recent boom in research on sport-related concussive injuries has allowed the field of sports medicine to improve its knowledge of the psychological and neurocognitive effects of those injuries, which has led to many positive changes both on and off the football fields. The improvements in the ways physicians and athletic trainers manage concussed athletes is an indication that the findings provided by research are being put into good use. Consequences of this increase in public knowledge has also led public and private organizations as well as state legislatures to implement management protocols of sport related concussions [10]. Along with this, raised awareness on concussions has affected other areas of sports, such as equipment design and rule revisions. Such have shown to be effective when for example looking at the recent decline in the number of concussions in the National Football League (NFL) following the changes in kickoff rules [11].

With the recent development of the literature on the long-term symptomatic effects of concussions, concerning trends in alterations of cerebral tissues integrity and connectivity have emerged from the use of brain imaging techniques as potential brain biomarkers. Such have become a major focus in the field of sport concussion research with hopes to improve our ability to identify and understand those injuries. It is still unclear whether repetitive subconcussive

impacts to the head or actual concussions sustained over a football career weighs more into exposing players to long-term health issues like working memory problems, neuropsychiatric conditions and diffuse axonal injuries in the white matter (WM), which has recently been found to be a common pathological feature in the retired population of former football players who have suffered of traumatic brain injuries [12]. It is therefore anticipated that through the use of techniques like functional magnetic resonance imaging (fMRI) and diffusion tensor imaging (DTI), researchers will be able to deepen their understanding of the acute and chronic effects of concussions at a neuroanatomical and structural level. Such improvements would be clinically significant by allowing physicians and other healthcare professionals to better educate their players on the risks that they face in the long term after sustaining concussive injuries. Additionally, such findings may lead to supplementary changes in rules by the governing bodies in order to further protect the players' health.

Recent work in the field of brain traumatic injury research has looked at differences in working memory performance in retired National Football League (NFL) and college players based on their concussion history and football exposure using blood-oxygen level dependent (BOLD) signal changes from fMRI analyzes. Findings showed that concussion history accounted for significant differences in neural activation patterns [4]. To better understand the effects of concussion history, position played and added football exposure on the neuroanatomical structural changes in football players, this study will further analyze a population of NFL and college retired football players with the use of DTI, which will allow the potential identification of changes in white matter structures and axonal integrity. Integrity of the white matter tracts, mainly defined by fractional anisotropy (FA) and medial diffusivity (MD), will be analyzed through voxel-wise analysis called tract-based spatial statistics (TBSS). Such changes may provide further information about the contribution of repetitive head impacts and concussive injuries along with the significance of player's position on the long-term health of college and professional football players.

2.2 Neuroanatomy

2.2a General Overview of the Cerebral Cortex

The brain is the main structure affected by concussive injuries [13]. Fueled by the cerebrovascular blood supply, originating from internal carotid and vertebral arteries, the main regions of the brain are the medulla, pons, cerebellum, midbrain, thalamus, basal ganglia and cerebral cortex. Separated by sulci, the cerebral cortex is divided into five main lobes: the Frontal, the Parietal, the Temporal, the Occipital and the insular lobes. Previous work on head injuries has found significant changes in regions of white and gray matter (GM) volumes, where patients with history of concussions showed cerebral tissue atrophy [14]. This global atrophy was observed with overall changes in the brains' volume along with more specific affected areas such as the anterior cingulate and the left cingulate gyrus isthmus. Such observations may suggest vulnerability of the frontal regions of the brain to long-term structural changes, which functionally, is mainly responsible for cognitive and motor functions. Regional changes are thought to affect athletes' abilities to accomplish certain cortical tasks such as motor planning and cognitive processing [15, 16]. Further changes in neurocognitive functions such as speed of information processing, attention, memory, and reaction time have also been related to traumatic brain injuries [17]. Additionally, evidences have linked concussive injuries and neurocognitive functions deficits to neural substrates such as alterations in distributed network connections within the cortex [18].

As we investigate the functional and structural impacts of concussions in adult former college and professional football players, it is important to keep in mind the natural process of aging. Such contributes to changes in the brain experienced throughout a lifetime, and can therefore affect one's ability to accomplish certain tasks, as he or she gets older. For instance, it is known that structural changes such as regional atrophy, reduced synaptic density and decreased neural plasticity are common in aging individuals [19]. Functionally, declines in information processing speed, reasoning skills and memory are also expected in adults, especially in later years [19]. This is important in that generalization of research findings on the effects of traumatic brain injuries with respect to specific age groups need to be carefully argued.

2.2b The regional functionality of the Cerebral Cortex

To understand the symptomatology of sport related concussions and their neurological effects on the cognitive abilities of athletes, it is important to take a more detailed look at the way our brain is topographically divided, and at how cortical functions are related to the cerebral anatomy. At the microarchitectural level, cerebral tissues consist of gray and the white matter; gray matter is primarily comprised of neuronal cell bodies, whereas white matter consists largely of neuron extensions referred to as axons [20]. Major pathways of the white matter are called tracts (e.g. the corpus callosum is the white matter tract connecting the two cerebral hemispheres). At a macroarchitectural level, the division of the brain into lobes is subdivided into main regions (i.e. the Brodman's areas), which are known to be involved with a number of functions.

The frontal lobe is of great interest to this study, as it appears to be commonly affected in post-concussed athletes [14]. It is divided into the motor, the premotor and the prefrontal (PFC) regions. The motor cortex is principally made of the precentral gyrus, which forms neurons of the corticobulbospinal (corticospinal) tract responsible for the control of motor movements. Further projections of the motor region extend to the basal ganglia and from there into the thalamic and subcortical nuclei for more specific motor coordination and planning [21]. The premotor cortex, localized anteriorly to the precentral gyrus, occupies parts of Brodman's area (BA) 6 and 8, which have been subdivided into the premotor and supplementary motor cortices. Working in association with the parietal association cortex, the premotor cortex also projects axons into the corticospinal path and is principally responsible for motor planning [21]. Lastly, divided into three main regions, the prefrontal cortex is mostly involved with planning, organizing, executing and selecting behaviors along with having major functional associations with working memory and learning. The dorsolateral prefrontal region (DLPFC) is mainly responsible for reasoning, problem solving and the sequencing of behaviors. Patients with affected DLPFC may show slower learning rate and poor memory as well as difficulties in retrieving temporal sequence of occurred events. The orbitofrontal cortex (OFC) is involved with behavioral inhibition, emotional regulation and olfaction. As expected, deficits in this area are usually accompanied with disorganized behaviors and abnormal emotional responses. The third and last region of the prefrontal cortex, the medial frontal/anterior cingulate cortex, is mainly

associated with attention, behavior regulation and motivation, and social cognition. Damaged in this area are usually accompanied with impaired explicit memory and lack of behavioral self-initiation [21].

The complex neural interconnections of those areas within the brain are key to proper functionality of the nervous system. These interactions can be structurally and functionally delineated based on white matter tracts projections, and concurrent regional activation, respectively. As shown by previous studies in this field, the neural recruitment process is not localized. For instance, neural mechanisms responsible for working memory functions involve complicated networks between regions like the prefrontal cortex and the rest of the brain, which are necessary for the conservation of normal behaviors [22]. Furthermore, concussions have recently been associated with functional inefficiencies in episodic memory network and different neural recruitment patterns. These findings are thought to be caused by changes in white matter integrity as a result of multiple sport-related concussions [23]. Such outcomes are the foundations that drive the continuation of research about the neuronal changes following concussions.

2.2c The limbic system

Recent interest in studying the effects of TBI on the limbic system has been triggered by the growing number of victims of chronic traumatic encephalopathy (CTE). CTE is a neurodegenerative disease and is thought to be caused in part by repetitive head trauma, including concussions. The medial structures of the limbic system, including the amygdala, the mammillary bodies and the hippocampus seem to be most severely damaged in CTE victims [24]. Made of a number of different structures from the telencephalon, diencephalon and mesencephalon, the limbic system provides support in cognitive functions such as emotions, long-term memory, behavior and motivation. More specifically, the hippocampus was identified as the main region responsible for functions of spatial orientation and temporal integration of learning [25]. Essential to this topic, alterations in the structure of the limbic system deserve more specific attention as recent work showed that mild traumatic brain injuries (mTBI) were associated with acute loss of microstructural integrity of gray and white matter regions of limbic-subcortical structures as measured by declines in fractional anisotropy (FA) [26]. Further findings also showed that changes in emotional states of concussed athletes, such as depressed

moods, reflected pathophysiology that was consistent with limbic-frontal neural activation changes [27]. Furthermore, those symptoms of depression experienced by patients following head trauma seem to closely share neural mechanisms found in diagnosed major depressive disorder (MDD) [27].

In vivo exploration of the brain using diffusion tensor imaging (DTI), which allows for identification of changes in white matter integrity, has revealed that symptomatic patients suffering of mTBI show declines in FA of limbic regions such the bilateral sub-genual and perigenual anterior cingulated cortex, the bilateral posterior cingulate cortex, the bilateral amygdala and the parahippocampal gyrus [26]. Other findings, comparing a control and a mild depression symptom group, have also targeted differences of gray matter concentration in the medial frontal and temporal regions along with the left parahippocampal gyrus [27]. These post-concussive regional changes in the limbic system may provide partial evidence to help explain changes in emotional behaviors experienced by patients who suffer and have suffered of traumatic brain injuries [27].

2.3 Concussions

2.3a Neurophysiology of Concussions

Concussions have been established as one of the most common form of traumatic brain injuries (TBI) across the world [28]. It is important, however, to note that not all cases of mild TBI (mTBI) are concussive, meaning that some cases of mTBI do not result in concussions. One understands that the complex interrelated chemistry between the cellular and vascular changes following concussive impacts, caused by biomechanical sheering forces within the cranial vault, is responsible for the triggering of a multilayered chain of neuro-pathophysiological reactions in the brain tissues [28, 29]. Ionic shifts, abnormal energy metabolism, diminished cerebral flow, and abnormal neurotransmission have all been identified acutely following concussion. This sequence of events is known as the "neurometabolic cascade" of concussion and is initiated by the mechanical stretching and shearing of neural axons leading to deregulated influx of Ca^{2+} and efflux of K^{+} ions [29-31]. This shearing of axons is referred to as diffuse axonal injury (DAI) and is known to be proportional to the magnitude of the decelerating forces from impact [12, 29]. Enhancing the release of excitatory neurotransmitters like glutamate, which then binds to N-

methyl-D-aspartate (NMDA) receptors, the uncontrolled flux of ions induces further depolarization (with more influx of Ca^{2+} ions) and the suppression of neurons with glucose hypometabolism [29-31]. The increase activity of the active pumps working to restore the membrane ionic balance is energetically demanding. High glucose consumption, through increased glycolysis, causes more Ca^{2+} ions influx into the mitochondria of the neural cells, which eventually disrupts the oxidative metabolic and anaerobic glycolytic reactions of the cells leading to acidosis (decrease in pH) and edema (swelling) [29-31]. For several days following a concussive injury, levels of intracellular magnesium appear to decline substantially. Magnesium is used for production of adenosine-triphosphate (ATP), along with initiation of protein synthesis and balance of the cellular membrane potential [30].

As it will be seen later in this chapter, diffusion tensor imaging has been identified as a new tool to asses and identify changes in axonal integrity involved sports-related concussive and sub-concussive brain injuries [32]. Along with the triggering of neurophysiological reactions like the one described above, DAI have also shown to cause neurofilament compaction and microtubule disassembly. This disorganization, resulting from axons stretching, seems to trigger even more progressive disassembly of microtubules leading to axons breakage and axonal swelling [29]. Such micro-changes in the structural connectivity of the axons in the white matter can be quantified using DTI techniques. DTI tracts the water diffusion along the axonal parallel tract arrangement and provides a quantified analysis of the directionality along with changes in integrity of the white matter as a function of spatial locations [33-35]. Therefore, DTI is expected to become a significant structural analysis tool in the field of concussion research.

2.3b Biomechanics of Concussions

Recent advances in technology have provided innovative ways, such as helmet accelerometers, video analysis and reconstruction of in-games impacts and injuries, to study the biomechanical factors of head impacts in more natural settings [36]. As mentioned earlier in this chapter, concussive impacts in football are primarily related to head acceleration resulting from helmet to helmet, helmet to other body regions, or helmet to ground impacts (or collisions). Head accelerations cause the brain to physically collide with the inner lining of the skull, leading to brain tissue alterations [37]. Reconstructions and studies of game collisions have established that impacts to the facemask and to the front of the head, as well as to the top of the helmet and the

sides were most common in leading to concussive injuries [36]. As discussed above, such impacts transmit strain and shear forces through the white matter tracts causing alterations of the axonal integrity [29]. Symptoms most often reported by players following a concussion are headache, balance/dizziness, slowing down of mental reasoning, concentration, sensitivity to light and memory loss, which anatomically match the association areas of the brain related to the frontal lobe [3, 38].

More findings in this field have failed to identify a specific threshold force at which a certain impacts would guarantee a concussive injury. Although one would expect that a greater impact would increase the likelihood of concussions, work by Mihalik and al. [39] reports that it is not actually the case. The collected data showed that only 0.35% of studied impacts with forces greater than 80 g resulted in concussions.

Finally, another interesting aspect of the biomechanical factors of concussions is the positional analysis. Findings have shown differences in the head impact magnitude and likelihood of concussions among player positions. Linebackers (LB) and offensive linemen (OL) tend to be the players who are most likely to sustain a concussion in college [39]. When comparing college division I and high school players however, other reports found that compared to linemen (non-skill), skill positions including quarterbacks (QB), running backs (RB), wide receivers (WR), cornerbacks (CB) and safeties (ST) are more likely to sustain higher magnitude impacts to the head but just not as often [40]. More positional differences were reported by another study that showed that player's position affected both the head impact frequencies and locations [41]. Furthermore, it was found that impacts to the front of the helmet were most common for all positions except QBs, with OLs leading the group with highest percentage (~45%) [41]. Another work looking at the peak linear accelerations found more differences in the magnitude of impact location based on positions. RBs and LBs, among all players, tend to have the greatest peak linear acceleration for front impacts, which was significantly greater than OLs (29%) and WRs (57%). QBs seem to sustain the greatest linear acceleration to the back of the head, although the differences to other positions were not statistically significant [42]. Such results propose that magnitude, frequency and location are all critical measures of head impact exposure. This leaves some openings with regards to positional analysis within the literature and this project will

examine differences in white matter structural differences and neurocognitive performances between retired speed and non-speed players (both college and professional).

2.3c Anatomical vulnerability of cerebral tissues

Because of the nature of head injuries, some regions of the brain tend to be more commonly affected than others. While only reported as preliminary evidence, findings from a small group of concussed athletes have shown increased neuropsychological deficits in athletes who had sustained head injuries following impacts to the crown of the head [39]. This provides an interesting bridge between the biomechanical factors of concussive impacts and the vulnerability of cerebral brain tissues. Further work in this field has also confirmed that post-concussive symptoms are associated with regional changes in brain activations [43].

Anatomical analysis of concussive symptoms can reveal more information about specific affected areas of the brain resulting from head injuries. For instance, cognitive processing and speed, verbal fluency, and memory problems are commonly associated with injuries to certain parts of the frontal lobe, which in acute injuries, seems to be resolved, in most part, after a certain period of time (~7 days) [44]. On the other hand, damages to the temporal lobe seem to result in significant long-term deficits in memory and language. More specifically, demonstrated using imaging techniques, abnormalities in the bilateral parts of medial temporal lobe (MTL) seem to correlate with decreased memory [45]. Additionally, damages to the left side of the temporal lobe seem to be linked with language deficits, and may even causes aphasia in adults [46]. Other changes in subcortical areas of the brain such as the hypothalamus, the cerebellum and the basal ganglia have also been linked to specific symptoms reported by concussed young patients [44]. Such findings have been extrapolated and used to explain similar symptoms reported by older athletes. Functional disruption of the hypothalamus due to the shearing forces of head impacts [47] has been related to a number of dysfunctions such as autonomic system deregulations [48], altered circadian cycles and sleep disturbance [49], thermoregulation abnormalities[50] and even sexual dysfunctions[51]. Likewise, vulnerability of the cerebellum, as a result of acute head injuries, was identified by balance and motor organization problems [52], while dysfunctions of the basal ganglia, although underreported (~ 2-3%), were observed through altered integration of motor-sensory-cognitive functions [53].

Recent development of brain imaging techniques such as DTI have shown that contact sport athletes are exposed to changes in fractional anisotropy (FA) and medial diffusivity (MD) in the cerebral white matter [33]. Although not limited to the following, certain cerebral regions such as the corpus callosum, the external capsule, the inferior fronto-occipital and corticospinal tracts, the inferior fronto-occipital fasciculus, as well as smaller regions of the superior/posterior corona radiata have shown consistent structural changes from TBI under DTI analysis [33]. Furthermore, changes in axonal structures and white matter connectivity not only resulted from concussive impacts but also from repeated hits to the head sustained by contact athletes, which is now known to contribute to the cumulative long term changes in the integrity of the white matter observed in football players [54]. Although this topic will be revisited later in this chapter, further understanding of such microstructural changes in the integrity of the white matter is expected to provide more information about the vulnerability and changes in functionality of cerebral tissues following concussions.

2.4 Symptomatology of Concussions

2.4a Short-term neurological symptoms

Acute symptoms of concussive injuries such as headache, dizziness, concentration problems, fatigue and memory loss have already been mentioned in this chapter. An important distinction that needs to be made with regards to sport-related concussions is that acute loss of consciousness is not usually seen in athletes, and that post-traumatic amnesia is most often very brief [30]. Furthermore, it is also known that overall symptom duration is typically 3.5 days and that most athletes reach a full asymptomatic point within a week following their head injury. This is known as the recovery period [3]. Even if an athlete is asymptomatic, it does not imply full recovery. There may be persistent microstructural changes in cerebral tissues, which are important when discussing the long-term effects of concussions.

Athletes who have sustained a concussion are more likely to suffer a subsequent concussive injury than players with no history of concussion [3]. Furthermore, athletes with previous history of concussion tend to experience longer recovery periods before returning to play [3]. It also appears that in addition to being more sensitive to future impacts, “concussed” brain cells are, within a certain time frame, under a particular state of vulnerability. Sustaining a second

concussive impact within that time frame, which would normally be considered non-lethal for the athlete, could in fact cause irreversible cell damages leading to drastic unpredicted increase in intracranial hypertension and death. The phenomenon is known as the "second impact syndrome" and has involved mainly sport-related activities [28].

Acute molecular changes within the brain have also been identified with concussive injuries. Following the acceleration-deceleration forces applied to the brain, beta amyloid and tau proteins tend to accumulate on neuronal bodies within hours of the injury [55]. Tau is a normal axonal protein that promotes microtubule assembly and stability, and exists in six different isoforms each containing potential levels of phosphorylation. This is clinically significant since hyper-phosphorylated tau proteins are known to impair axonal transport disrupting neuronal and synaptic functions. Hyper-phosphorylated tau is a hallmark feature of Alzheimer's disease (AD) [29]. Additional findings have shown that the APOE gene plays a role in the pathophysiological response to head injuries [56]. Following head injuries in animal studies, the APOE-mediated lipid transport system is stimulated [56]. Greater deposition of beta-amyloid proteins were observed with possession of the APOE e4 following traumatic brain injuries [57] and in AD brains [58]. As it will be discussed later, such information has led researchers to look into former athlete's prevalence to neuropsychiatric disease and the contribution of previous head injury exposure.

2.4b Long-term neurological symptoms

The long-term repercussions of concussive injuries on retired football athletes and the correlations between neurocognitive dysfunctions and the integrity of white matter in the brain have garnered considerable attention recently. Findings have revealed potential late-life memory problems and risk of mild cognitive impairment along with potential early onsets of AD [1]. Retired professional players with a history of concussion showed declined mental health and cognitive functioning along with higher rates of memory problems. Additional dementia-related symptoms were also hypothesized to be linked with concussion history and repetitive impacts to the head sustained by football players [1].

Such findings can be challenged by the body changes accompanied with the normal process of aging. Neurocognitive declines with age are due to natural degeneration of neural structures and

functions [59]. Research on aging former NFL players compared to healthy controls found that additional cognitive deficits such as difficulties in naming and word finding were more common in the retired athletes [60]. Such deficits are thought to be associated with abnormal changes in white matter integrity, presence of deep white matter lesions and alterations of regional cerebral blood flow [60]. This leaves questions on whether or not football exposure and/or concussion history plays a major role in causing retired NFL players to experience greater cognitive impairments or depression as they age when compared to the general population.

2.4c Prevalence to neurological and neuropsychiatric conditions

The increased risks of neurological and neuropsychiatric conditions as a result of head injuries is a controversial topic of research as scientists debate on whether or not CTE is a distinct pathology [2]. First witnessed in a 51-year-old boxer suffering from delayed posttraumatic dementia with AD pathological changes [61], the search for the long-term neurodegenerative effects of concussive injuries is still incomplete. Recent imaging research has shown that athletes suffering from depression symptoms as a result of concussive injuries display reduced activation of the DLPFC and striatum, and "attenuated deactivation" of the MTL and frontal regions [27]. These findings indicated losses of gray matter in these cerebral regions. Additionally, neurodegenerative mortality in retired NFL players is three times higher than the general population in the United States, and that it is even greater (four times) more specifically for two major neurodegenerative conditions: Alzheimer disease (AD) and amyotrophic lateral sclerosis (ALS) [2]. Although unable to determine the cause of those correlations, the research seems to suggest that having a history of concussions is associated with an increased risk of neurologic disorders.

Another neurodegenerative disease of interest in this field is known as chronic traumatic encephalopathy (CTE), which has attracted rising attention from the recent suicides of former NFL players [60]. CTE is thought to be caused, at least in part, by repetitive brain trauma including both concussive and sub-concussive impacts. Symptoms of CTE are posited to be memory impairment, depression and suicidality, apathy, poor impulse control and dementia. Regions of the brain most commonly and severely damaged in CTE patients autopsies are the cerebral cortex and medial structures of the limbic system including the amygdala, the mammillary bodies and the hippocampus [24]. Note the similarities with the region-specific

vulnerability of cerebral tissues discussed in section 2.3c. AD, ALS and CTE patients often share similar symptoms, which has led researchers in this field to hypothesized that some causes of death from AD and ALS, as reported on the death certificates, may have actually been caused by CTE, which is still not recognized as a distinct cause of death in the current ICD (International Classification of Disease) revisions [2]. Hopes are that advances in DTI techniques will deepen our understanding of the interrelations between the degradation of white matter and cognitive dysfunctions along with the prevalence to neurodegenerative conditions observed in retired football players.

2.5 Neurocognitive Testing and Concussions

Research on the relationship between concussion and neurocognitive performance in college football players have found significant associations between history of concussions and long-term deficits of certain cerebral domains [62]. More specifically, domains of executive functioning, speed of information processing and areas associated with self reported symptoms were identified in individuals who have sustained two or more concussions. This is significant for this research since concussion history is a main variable in looking at white matter integrity of former football players. This work will look for deficits in neurocognitive outcomes in retired players and a potential coincidence with alterations in white matter integrity from the DTI analyzes. Future findings may allow for new combinations of tests, such as specific neurocognitive and neuroimaging examinations, in order to better predict regional anatomical structural changes and network connectivity alterations as a result of concussive injuries along with better understanding their effects on the cognitive state of injured/recovered athletes [17].

2.6 Neuroimaging and traumatic brain injuries

2.6a Functional magnetic resonance imaging (fMRI)

2.6a(1) Description and overview

The use of functional magnetic resonance imaging to investigate the neurocognitive deficits of concussive injuries is an innovative extension to the current research conducted on concussions and will continue to be improved in years to come. Numerous studies [38, 43, 63] have established that functional neuroimaging has great potential to serve as a future biomarker for severity of concussive injuries and TBIs. fMRI tracks blood oxygenation level changes,

which is an indirect measure of neuronal activity [64]. Cognitive tasks and associated stimuli can be presented during a scanning session to determine neuronal activity in response to the task. A typical experimental paradigm used for fMRI analysis of working memory is the N-back task, which involves identification and location of verbal and nonverbal stimuli. Consistent activation of the frontal and parietal cortical regions have been shown using this test [65]. Advantages of fMRI in clinical neuroscience are that it is noninvasive, relatively widely available in both medical and university settings, and has high spatial resolution [66].

The use of neuroimaging techniques has been explored clinically to assess cognitive alterations. Such deficits in cognitive performances were observed on tasks associated with memory, naming and word finding in aging former NFL players when compared to non-depressed and healthy subject groups [60]. The use of fMRI has also recently been used to map human functional networks, which has confirmed hypotheses about the changing functionality of the brain as adults age. Aging is associated with changes in properties of the brain functional networks, which confirmed previous findings on the differences of "default-mode networks" between children and adults [67]. Such findings suggest that the natural process of aging along with the maturation of the brain may account for quantifiable changes in neurological functions and network topology. These findings should therefore be considered in this current study while carrying DTI analyses, as they could account for some of the predicted changes in white matter integrity and structural connectivity of former college and professional players.

2.6a(2) Abnormal activation patterns related to concussions

The clinical significance of fMRI analysis in studying the effects of concussions is constantly growing as researchers expand their knowledge in this field. For instance, recent findings examining the relationship between symptoms severity and neural functioning in concussed athletes have suggested that during the recovery period following an acute head injury, patients suffering from increased concussive symptoms also required additional cognitive resources to complete cognitive tasks [38]. Furthermore, it was shown that concussed subjects show abnormal activation patterns up to two months following the acute injury. Specific regions of higher activity were identified to be the left and right DLPFC, which lasted for two months, and the left inferior parietal area, which lasted up to two weeks post-injury [63]. More findings reported by another research group, showed that although former NFL alumni did not show

significant differences in their performance on computerized tests of executive functions, when compared to a control group, their fMRI analysis revealed "pronounced abnormalities in functional activation within the dorsal frontoparietal network" [68]. There appears to be a trend where the underlying functional and structural neurological changes are common long-term consequences of repetitive traumatic brain injuries. More importantly, those changes do not seem to be reflected in neurocognitive performances, which indicates the need for imaging analyses when managing concussed athletes. It was also suggested that this "compensatory mechanism" and over-activation of the brain was in place to counter the structural changes associated with repetitive concussive impacts [68].

The number of previous concussions was identified as a potential factor for differences in neuro-functional activity with regards to episodic memory. Although performing similarly to an age-matched adult group behaviorally, fMRI data analysis of former NFL players showed that high concussion group (three or more) exhibited different patterns of activation with relational memory processing (memory for the relationships between elements) [23]. As proposed by the researchers, these findings suggests that repeated number of concussive injuries and neurological trauma may be a factor in causing inefficient changes in neural recruitment, which seems to have an effect on memory tasks in the long term. Further findings on this topic are consistent with these conclusions showing that concussion history may contribute to greater differences in neural recruitment during working memory task performance when compared to weighted football exposure in both college and NFL retired players [4].

2.6b Diffusion Tensor imaging (DTI)

2.6b(1) Description and overview

As previously discussed in this chapter, diffuse axonal injury (DAI) is neural microstructural damage from traumatic brain injury resulting from shearing forces applied to nerves fibers [33, 69]. Recently, diffusion tensor imaging (DTI) has been identified as an important noninvasive tool allowing quantitative analysis of white matter changes following concussive injuries [70-72] as well as repetitive exposure to head impacts [54]. Cerebral white matter is made up of axons and their associated microtubules and myelin sheaths, which are oriented in a tract parallel arrangement. This organization of the white matter allows for anisotropic water diffusion, with greater diffusion along the axons as opposed to any other

directions. This unique property of white matter is exploited by DTI analysis to tract and quantify three-dimensional directionality and integrity as a function of spatial location [33-35]. Structural connectivity patterns can be derived from whiter matter tractography where trajectories of fibers of interest can be three-dimensionally reconstructed and segmented using continuous tracking algorithm [35, 73]. The two most frequently used DTI markers used by this field of research are trace and anisotropy of the diffusion tensor [35]. The diffusion tensor uses a symmetric 3x3 matrix, which represent the 3D properties of the diffusion of water molecules based on a Gaussian model [74]. Three pairs of orthogonal eigenvectors (ε_1, ε_2, ε_3) can be calculated from each diffusion tensor of imaging voxels using matrix diagonalization [34]. Such vectors are ordered by the magnitude of their eigenvalues ($\lambda_1>\lambda_2>\lambda_3$) and represent the direction and magnitude of diffusivity within biological tissues; ε_1, the largest diffusion magnitude in each voxel, is the dominant fiber direction [34]. The spatial arrangement of the diffusion tensor is visualized using the ellipsoid model, which is defined by the three eigenvectors as seen in Figure 1 [35].

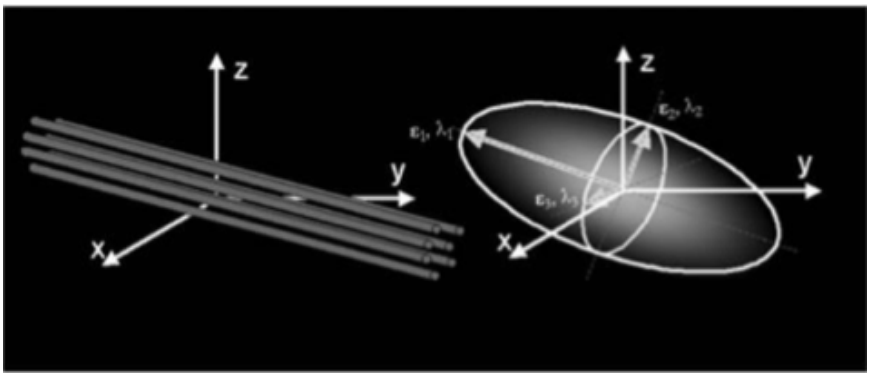

Figure 2.1: The three dimensional ellipsoid model used for diffusion tensor

Various magnitudes of eigenvalues change the shape of the ellipsoid. The trace of the diffusion tensor (Tr) is the sum of the diagonal elements (D) of the diffusion tensor. It is a measure of the magnitude of diffusion and is rotationally invariant [35]. The apparent diffusion coefficient (also called mean diffusivity or MD) acts as an averaged index of water molecules mobility and diffusivity. It is obtained by dividing the trace by three (MD=Tr/3), which is equivalent to the three-eigenvector values of the ellipsoid model:

$$MD = {(Tr)}/{3} = {(\lambda_1 + \lambda_2 + \lambda_3)}/{3}$$

Changes in MD are suggested to correlate pathologically with myelin loss and/or axonal injuries [35]. Fractional anisotropy (FA) on the other hand is more commonly reported as an indicator for white mater integrity. Diffusion anisotropy measures the degree to which the diffusivities are function of the diffusion-weighted encoding directions [35]. FA is the most widely used measure of anisotropy and is described by the formula below [35]:

$$FA = \sqrt{\frac{(\lambda_1 - MD)^2 + (\lambda_2 - MD)^2 + (\lambda_3 - MD)^2}{2(\lambda_1{}^2 + \lambda_2{}^2 + \lambda_3{}^2)}}$$

When diffusion is isotropic, with diffusivity equal in all directions ($\lambda_1=\lambda_2=\lambda_3$), the ellipsoid is reduced to a sphere. Abnormalities and deficits in the white matter such as loss and destructive lesions of axons have values approaching 0. Contrarily, if diffusion is anisotropic, the highly directional white matter has a FA value approaching 1 (1 being completely anisotropic diffusion), which defines the theoretical FA range between 0 and 1 based on the three eigenvalues of the ellipsoid [35]. Figure 2 provides a visual for the various shapes of the ellipsoid based on relative magnitudes of eigenvalues [34]:

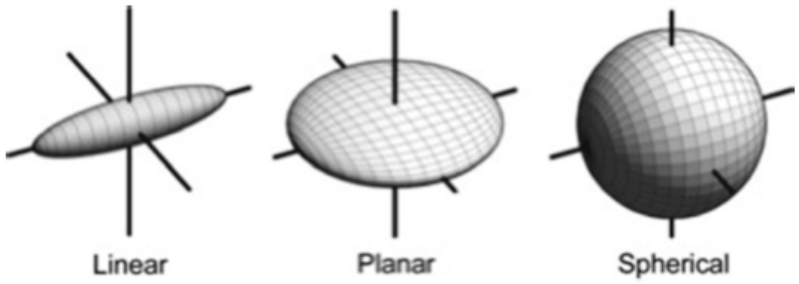

Figure 2.2: Diffusion tensor ellipsoid shapes with various magnitudes of eigenvalues

Further work in DTI analysis of axonal injuries in sport related concussion has suggested that MD may be more sensitive in detecting smaller structural whiter matter abnormalities when compared to FA [71]. Because of their complementary nature, we plan to make use of both measures in our study of the white matter integrity of retired college and NFL players. Such analyses will be correlated with concussion history, position played and football exposure as well as neurocognitive deficits from cognitive performance tests.

It is important to note that studies have disagreed concerning the direction of changes in FA and MD in early phase of recovery following concussive injuries [69]. While some reported increases in FA and decreases in MD [75, 76], other works reported opposite trends with decreased FA and increased MD [77, 78]. Acknowledging the fact that this current work will be focusing on long-term effects of concussive impacts and repetitive head injuries, those conflicting results are less of interest. What is more important, and also consistent with research, is that individuals with persistent cognitive impairment in later phases of recovery showed decreased FA [79], and increased MD [80], which are the methodological guidelines that will be used to conduct our DTI analyses. An additional note about previous DTI findings in neurology is that region-specific analysis of DTI differences have been proposed on verbal and memory tasks performed by populations suffering from neuropsychiatric diseases such as schizophrenia or parkinsonism, among others [16]. Cerebral areas such as the uncinate fasciculus, anterior corona radiata, forceps minor, and superior longitudinal fasciculus seem to show meaningful differences on DTI analyzes.

Final comments about the general overview of DTI regard the tract-based spatial statistics (TBSS) that will be used to conduct voxel-wise analysis on the cerebral white matter tracts [81]. Under the assumption that maximum FA is found at the center of the white matter tracts, TBSS generates a three-dimensional skeleton of the white matter using a non-linear FA volumes registration tool (FNIRT) which transforms all subjects' FA into a common space (MNI) [82]. These averaged spatial arrangement allow for statistical voxel-wise analysis of the differences in white matter integrity [34, 71] between the subdivided groups.

2.6b(2) Tractography and specific networks of interest

The next step in studying the cerebral white matter using DTI is known as diffusion tensor tractography (DTT) and involves the specific localization of three-dimensional neural networks. Such pathways reveal information about the anatomical relations tracts trajectory of white matter in the brain [34, 83]. Although cellular networks undergo dynamic functional and structural changes as they age, questions remain about the effects of concussive injuries and repetitive head traumas sustained in sports such as football. The degree of such changes in functional and structural connectivity are suggested to be highly predictive of cognitive alterations [67], which promises a lot for the future of neuroscience. For instance, recent DTI

network analysis of individuals with sport-related concussions reported lack of structural integrity of the left temporal lobe, which typically host association and projection fiber tracts running anterior-posterior and superior-inferior within the white matter [71]. Affected regions included the anterior-posterior oriented long associations fibers from the inferior fronto-occipital fasciculus, which connects the frontal and occipital lobes, as well as from the inferior longitudinal fasciculus, connecting the temporal and occipital lobes. Such findings, and others with similar conclusions [32], correlate with neurobehavioral impairments experienced by individuals post-injury. Further work on white matter anatomy determined that only association and commissural fibers were involved in cortico-cortical networks, whereas projection fibers were typically involved in connecting the cortex to non-cortical structures such as the brainstem and the thalamus [83].

2.6b(3) Concordance with fMRI readings

The brain's structural and functional connectivity seem to be intimately related, sharing different correlations with neurocognitive deficits observed in post-traumatically brain-injured patients. Structural disconnections measured using DTI were found to correlate with traumatic brain injury severity, which provided evidences for an underlying relationship between the gravity of the initial injury and long-term white matter alterations [84].

Although findings have shown that efficiency of communication between different regions of the default mode network are consistent with underlying axonal arrangement and integrity [84], other work on traumatic brain injuries has failed to match the presence of functional abnormalities, obtained thought fMRI BOLD analysis, with decreased FA values, observed via DTI [16], and vice-versa. In other words, significant changes in fMRI activation patterns, showing over-activation and compensatory mechanisms during spatial memory tasks of certain cerebral regions, did not match TBSS analysis displaying no significant changes between the injured and control groups at $p < 0.05$. Additionally, common fMRI regions of interest in TBI individuals (visual cortex, parietal cortex, right dorsal-lateral prefrontal cortex, and right hippocampus) did not seem to show significant changes in FA and number of fibers [16]. Such findings may propose that fMRI and DTI are sensitive to different cortical changes resulting from head traumas, although this doesn't rule out the possibility for future correlations.

Considering that both fMRI and DTI contribute to our understanding of diffused axonal injuries resulting from traumatic brain trauma, it will be important to continue to look for such relationships linking the two methods in order to advance our knowledge in this field.

2.6c Limitations and Complications

Both fMRI and DTI techniques have their own limitations based on the type of signal that is collected for analyses. For instance, although functional neuroimaging offers insights on neuroanatomical functionality, the selection of task paradigms, in addition to the alterable brain-behavior plasticity limits the breath of individual study conclusions and comparability of results in the literature [15]. More limitations on fMRI reports include its dependency on metabolic demands of neuronal activity, which itself depends on the density of capillary networks and their locations. Such interdependence is suggested to limit the spatial resolution and quantitative comparisons of different areas of the cortex, which differ in cerebrovascular supplies and therefore their ability to be detected by hemodynamic-based signal [85]. Highly vascularized area such as the frontal cortex, which will be the focus of this work, should reveal relatively high spatial resolution reducing this limitation. DTI is limited by factors such as thermal and physiological image noises along with misregistration errors in images resulting from eddy currents and head motions at the time of the capture. Further limitations in DTI, although inevitable considering the type of analysis conducted, are the averaging of tissues such as gray matter, white matter and cerebrovascular fluid into large voxels and the many cerebral areas known to contain significant amount of fiber crossing. Such considerable fiber crossing within one voxel hinders one's ability to correctly identify white mater abnormalities since the micro-alterations are usually undetected [35].

Controversies and challenges with the use of neuroimaging techniques in concussion research are also predicted to be encountered in the clinical management of injured players [86] along with the future use of these techniques for diagnosis of concussive injuries and identification of recovery periods. Concussions are categorized as functional injuries, not structural, which makes difficult their diagnosis and the establishment of future guidelines for recovery periods [43]. On-field procedures currently in place to deal with injured (or potentially injured) players are often challenged by the physiological differences between them and their ability to sustain different degrees of impacts, which also needs to be considered as a future continued challenge. Other

limitations to this research are the age and target groups that have been mentioned in earlier studies, which usually vary between college and professional athletes, and others.

2.7 Conclusion and Direction for future Research

In summary, as mentioned earlier, the neurological effects of concussions have been intensely studied as well as the biomechanics of impacts leading to these injuries. The recovery periods of injured athletes at all levels, from high school to professional as well as the neurocognitive ways to test for these traumatic brain injuries have also been deeply investigated. The next step in the research is to deepen our understanding of the relationship between anatomical and neurocognitive changes occurring from these brain injuries, which will allow us to find ways to better prevent them, in addition to improve our treatments. Recently, various procedures have been put into place with specific focus on behavioral changes of playing athletes as well as rule changes, which have allowed experts to observe a significant reduction in the rate of concussive injuries. The use of brain imaging techniques, like the fMRI and DTI, will allow experts in field to further improve their understanding of concussions as well as to develop more specific and accurate methods to diagnose injured athletes.

This proposed project has the potential to advance the science on the long-term effects of repetitive head trauma. Studying differences in white matter integrity between former college and professional football players will expand studies already published on the topic. If differences in brain structural and functional abnormalities can be explained by different levels of head impact exposure, recommendations may be made regarding dose limits on career exposure to head trauma. This will also provide an opportunity to further investigate the effects of added exposure to head impacts, sustained by different positions, on neurocognitive functions as well as to more precisely identify the additional risks involved with playing professional football. It will also potentially improve our understanding of the correlation between the long-term symptoms reported by players and the number of concussions they have sustained during their career. Our findings may finally allow for less expensive tests, such as specific neurocognitive test batteries, to predict regional neuroanatomical outcomes shown on expensive neuroimaging tests, based on the correlations that will be drawn from the data.

CHAPTER III: METHODOLOGY

3.1 Subjects

3.1a Participants

Recruited participants in this study compiled a total of thirty-two former collegiate football players who played a minimum of three years of college football ("COL"; M=58.63; sd=3.663; all male) and thirty-one former National Football League (NFL) players who played a minimum of five seasons of professional football ("COL+NFL"; M age=58.12; sd=3.74; all male). An additional former professional football player from the high concussive group underwent all neurocognitive and genotyping assessments but had to be removed from the dataset because of his invalid magnetic resonance imaging results. All participants were given written informed consent prior to participating in the study, which were in accordance with the requirements of the Institutional Review Board at the University of North Carolina at Chapel Hill.

3.1b Recruitment

The survey and interview data that was used to conduct this study has already been collected by research teams of the Matthew Alan Gfeller Sport-Related Traumatic Brain Injury Research Center and the Biomedical Research Imaging Center of the University of North Carolina at Chapel Hill (UNC-CH). Retired professional football players were recruited from a database of approximately 3000, which has been developed in conjunction with the National Football League Players' Association. The database includes results from self-reported general health survey, which contains information about the retired players' playing career (exposure), medical history (including neurological conditions such as depression, Alzheimer's disease, and Parkinson's disease, among others), history of concussive injuries (including number of concussions in high school, college, and professional football playing careers), and general health status. An additional questionnaire that focused on memory and issues related to mild cognitive impairment was also sent to the selected subjects. This questionnaire was also sent to spouses and/or a close relative to confirm or further identify any memory problems exhibited by the retiree. From a selected group that fulfilled our inclusive and exclusive criteria (and based upon concussion history), retired players were selected and contacted by telephone regarding their willingness to participate in the study.

Former collegiate football players were recruited from the North Carolina, Virginia, and South Carolina regions surrounding Chapel Hill, NC. A sufficient pool of subjects was extracted to match with the recruited retired NFL players. The sport information directors and athletic trainers of Division I football programs within this tri-state region were contacted to identify graduates of those respective programs who lived in these areas. Identified individuals were then sent a letter from the research team inquiring if they would be willing to participate in the research study. Interested participants were then further assessed on the same screening instruments and then invited to campus for further studies. Former college and professional football players were matched on age, education level, and position played. The following inclusive and exclusive criteria were determined prior to recruitment of the subjects.

3.1c Inclusion Criteria

- Must be a retired American football player at either the college or professional level.
- Must be 50 years of age and older.
- Subject stratified in the college group were required to have played a minimum of 3 years of college football.
- Subject stratified in the professional group were required to have played a minimum of 5 years in the NFL (non-kicker/punter).
- Subjects stratified in the HIGH concussion history group were required to have a sustained and reported three or more concussions (≥3) during their football career.
- All subjects had recent self-reported memory problems.
- Report of below average response on at least two of the following four questions on the mild cognitive impairment survey instrument: Questions 1, 2, 3 and 7. In cases where both the retiree and the intimate partner or close relative returned a survey, the average of the two scores on the four previous highlighted questions was used.

3.1d Exclusion Criteria

- Former kickers or punters.
- Self-reported history of stroke or any central nervous system disease (e.g. multiple sclerosis, amyotrophic lateral sclerosis).

- Conditions unsafe for magnetic resonance scanning, such as cardiac pacemaker, epicardial pacemaker leads, cochlear implants or claustrophobia.
- Functional Activities Questionnaire (FAQ) score below 8 (indicative of dementia).

3.1e Stratification

3.1e(1) Concussion history and football exposure

Selected subjects were subdivided into four groups based on both concussion history and football exposure. The following groups are defined below with their respective inclusion criteria:

1) (COL+NFL)-LOW: A sample of 16 retired professional football players.
   - Minimum of 5 years of NFL experience;
   - Mild memory impairment reported and confirmed on MCI survey;
   - Reported none or single concussion (0-1) injury during professional football career.

2) (COL+NFL)-HIGH: A sample of 15 retired professional football players.
   - Minimum of 5 years of NFL experience;
   - Mild memory impairment reported and confirmed on MCI survey;
   - Reported 3 or more (≥3) concussions during professional football career

3) (COL)-LOW: A sample of 16 former collegiate football players.
   - Minimum of 3 years of collegiate experience;
   - Mild memory impairment reported and confirmed on MCI survey;
   - Reported none or single concussion (0-1) during collegiate football career.

4) (COL)-HIGH: A sample of 16 former collegiate football players.
   - Minimum of 3 years of collegiate experience;
   - Mild memory impairment reported and confirmed on MCI survey;
   - Reported 3 or more (≥3) concussions during collegiate football career.

Table 3.1: Football exposure and concussion history

		Football Exposure	
		COL	COL+NFL
Concussion History	LOW (0-1)	16	16
	HIGH (≥3)	16	15

3.1e(2) Positional

This project also plans to stratify subjects based on primary football positions played in college and in the professional football league. Such stratification is inspired from a recent study on neurodegenerative causes of death in NFL players by Lehman et al. published in 2012 [2]. The recruited players will be sub-divided into two positional groups: 'speed' (Quarterbacks, Running backs, Halfbacks, Fullbacks, Wide Receivers, Thigh ends, Defensive backs, Safeties, Linebackers) and 'non-speed' (All Offensive and Defensive linemen). Table 3.2 below summarizes the sub-division:

Table 3.2: Participants' football playing position

Stratified Groups	College: COL (n=32)	NFL: COL+NFL (n=31)	Total
Speed (QB, RB, HB, FB, WR, TE, DB, ST, LB)	16	19	35
Non-speed (All OL and DL)	16	12	28
Total	32	31	63

The next table (Table 3.3) summarizes all three variables of interest and the stratification used to conduct the three-way ANOVA.

Table 3.3: Stratification by football exposure, concussion history and playing position

		Football Exposure		
Concussion History	Position	N (COL)	N (COL+NFL)	N (Total)
LOW (0-1)	Non-speed	9	6	15
	Speed	7	10	17
	Total	16	16	32
HIGH (≥3)	Non-speed	7	6	12
	Speed	9	9	18
	Total	16	15	31

3.2 Measurements and Instrumentation

3.2a Concussion history

Concussion history was obtained from the data collected from the retirees' general health survey. Once selected subjects were finally invited to campus, the concussion history was further confirmed through interviewing the subjects and getting access to any available medical documentation provided by the subject. Following confirmation of the concussion history, each subjects was assigned to the HIGH and LOW concussion sub-groups. Permission to review any available pertinent clinical records was obtained from each retiree. In most cases, such records were hardcopy files stored by team physicians, neurologists, and athletic trainers. High ability to recall concussion history was observed in the subject and this because such injuries are significant to the athletes in influencing their ability to play. Data on concussive injuries other than those that occurred during the professional playing career were also collected. These include concussions from motor vehicle crashes, or from pre-professional play. We have noted that the data obtained on professional playing career concussions tend to be of high quality and this will form our main focus.

3.2b Contact Exposure Index

Research staff tracked subjects' football career histories from high school level continuing through college and all the way to professional career by conducting structured oral interviews. For each year of their football career, participants provided information about: primary position played (i.e., quarterback, offensive line, running back, defensive line, defensive back, linebacker, wide receiver, special teams); number of games in the pre-season, regular season, and post-season; percent of time that they played in games; number and length of contact practices. The exposure history variable was then created from this information [4].

The following formula was used to calculate the number of practice contact hours for each year:

> (# pre-season practice sessions/week * # pre-season weeks * # hours/pre-season practice session) + (# regular season practice sessions/week * # regular season weeks * # hours/regular season practice session) + (# post-season practice sessions/week * # post-season weeks * # hours/post-season practice session)

The number of games hours was then calculated using this following formula:

(# pre-season games * % of time active in pre-season games * 1 hour) + (# regular season games * % of time active in regular season games * 1 hour) + (# post-season games * % of time active in post-season games * 1 hour)

Finally, the sum of both the number of hours for practices and games per year throughout participants' career was used to create their total contact exposure [4].

3.2c Magnetic Resonance Imaging

3.2c(1) Structural Images

Magnetic resonance images were acquired using a Siemens Trim-Trio 3-T scanner. The subjects' heads were held in place in the scanner using cushions and a headrest. Following an initial localizing scan, T1 weighted images were obtained. Covering the entire brain with a voxel size of 1x1x1 mm^3, T1 weighted structural scans for anatomical visualization were obtained with the following scanner parameters: 160 slices at TR/TE/TH/=1750msec/4.38msec/1mm. DTI series of images were taken following the T1 scans (details below).

3.2c(2) Diffusion Tensor Imaging (DTI)

A spin echo diffusion tensor weighted sequence was used to acquire the images. A baseline image and 64 directional images were acquired at an isotropic resolution of 1x1x1 mm^3. The baseline image was taken without a diffusion gradient (b = 0) and the remaining six images were taken with b = 1000s/mm^2. As outlined below, tensor maps, fractional anisotropy and mean diffusivity will be the metrics of interest computed from the raw DTI data. All diffusion tensor images will be transferred to a workstation for data analysis.

MD is used as an averaged index of water molecules' mobility and diffusivity within white matter. It is obtained by dividing the trace (Tr) of the diffusion tensor in three (MD=Tr/3) in order to get the three-eigenvector values of the ellipsoid model used for diffusion tensor 3x3 spatial matrix [35].

Fractional anisotropic diffusion of water in the white matter tracts of the brain is commonly used in diffusion imaging to measure the directionality of the local tract structures [35]. FA theoretical range for tract directionality ranges from values within 0 and 1. FA values near 0 represent

highly isotropic white matter tracts whereas FA values approaching 1 signify highly directional white matter. Recalling from chapter 2, the FA formula is outlined below [35]:

$$FA = \sqrt{\frac{(\lambda_1 - MD)^2 + (\lambda_2 - MD)^2 + (\lambda_3 - MD)^2}{2(\lambda_1{}^2 + \lambda_2{}^2 + \lambda_3{}^2)}}$$

3.2d Neurocognitive Selected Test Battery

The selected neurocognitive test battery for this research project was carefully chosen in order to cover as many of the cognitive domains as possible. Find below a description of each selected test and an accompanied reasoning for their selection. Table 3.4 below summarizes the selected test battery along with more specific information about expected regions of interests affiliated with each test.

Delayed Verbal Memory Test (VBM) & Visual Memory Test (VIM) – Vital Signs includes parallel tests of verbal memory (word list learning) and visual memory (figure learning). The tests are virtually identical, but one uses words as target stimuli, while the other uses geometric shapes. The verbal memory test (VBM) is an adaptation of the Rey Auditory Verbal Learning Test. In the CNS Vital Signs version, fifteen words are presented, one by one, on the screen. A new word is presented every two seconds. The subject is asked to remember these words. Then, a list of thirty words is presented. The fifteen target words are mixed randomly among fifteen new words. When the subject recognizes a word from the original list, he or she presses the space bar. After this trial of thirty stimuli, the subject goes on to do the next six tests. At the end of the battery, about 20 minutes later, the fifteen target words appear again, mixed with 15 new non-target words. The Visual Memory Test (VIM) in CNS Vital Signs is based on the Rey Visual Design Learning Test; the latter is, in turn, a parallel to the Rey Auditory Verbal Learning Test, using geometric figures rather than words, and requiring the subject to draw the figures from memory. In CNS Vital Signs, the visual memory test is just like the verbal memory test. Fifteen geometric figures are presented; the subject has to identify those figures nested among fifteen new figures. Then, after five more tests, there is a delayed recognition trial. The VBM draws from a "reservoir" of 100 plus words selected from word-frequency Tables. The VIM draws from a reservoir of 60 simple geometric designs. The scoring is straightforward: correct

hits and correct passes, immediate and delayed. Correct responses from VBM and VIM are summed to generate a composite memory or memory domain score. The highest score one can attain is 120; the lowest is 60. Scores below 60 suggest willful exaggeration.

For this specific test battery, only the delayed memory score were used to conduct the statistical analysis since delayed forgetting seams to be more symptomatic in patient with history of traumatic brain injuries [87]. A corrected recognition score was generated for both the VERM and the VISM (VERM_cr and VISM_cr). The converted scores were obtained using the formulas below:

$$VERM\_delayed\_CR = [(verm\_delayed\_correct\_hits) - (15 - (verm\_delayed\_correct\_passes))]/15$$

$$VISM\_delayed\_CR = [(virm\_delayed\_correct\_hits) - (15 - (virm\_delayed\_correct\_passes))]/15$$

Symbol-Digit Coding (SDC) – Coding has been a component of the Wechsler Intelligence Scales since 1944 (Digit Symbol Substitution, DSST). The Symbol Digit Modalities Test (SDMT) is a variant of the Wechsler DSST, but the position of symbols and digits is reversed. The clinical and psychometric properties of the SDMT are similar to those of the DSST. Although the SDMT may be a "harder" test, and thus more sensitive to neurotoxicity, performance on the SDMT and the DSST are highly correlated. The SDC in CNS Vital Signs draws from a reservoir of 32 symbols. Each time the test is administered, the program randomly chooses eight new symbols to match to the eight digits. Scoring is the number of correct responses generated in 2 minutes. The total of right and left taps from the FTT and total correct responses on the SDC generates a composite score for "psychomotor speed." Thus the SCD correct scores were selected in this battery for their ability to provide a cognitive measure of processing and fine motor speed.

The Shifting Attention Test – The Shifting Attention Test (SAT) measures the subject's ability to shift from one instruction set to another quickly and accurately. In the SAT test, subjects are instructed to match geometric objects either by shape or by color. Three figures appear on the screen, one on top and two on the bottom. The top figure is either a square or a circle. The bottom figures are a square and a circle. The figures are either red or blue; the colors are mixed randomly. The subject is asked to match one of the bottom figures to the top figure. The rules

change at random. For one presentation, the rule is to match the figures by shape, for another, by color. This goes on for 90 seconds. The goal is to make as many correct matches as one can in the time allowed. The scores generated by the SAT are: number correct, errors, and response time in milliseconds. There is not a precise parallel to the SAT in the compendium of conventional neuropsychological tests, although Trails B and the Wisconsin Cart Sort are considered to be tests of shifting attention. Thus, the correct SAT correct and response time scores were selected in this battery to provide a measure of complex attention, cognitive flexibility and executive functioning as well as reaction time.

Finger Tapping Test (FTT) – The FTT is one of the most commonly used tests in neuropsychology, because of its simplicity and reliability, and because it generates relevant data about fine motor control, which is based on motor speed as well as kinesthetic and visual-motor ability. The FTT is believed to be one of the most sensitive neurocognitive tests for determining brain impairment. In CNS Vital Signs, the FTT is a very simple test. Subjects are asked to press the Space Bar with their right index finger as many times as they can in 10 seconds. They do this once for practice, and then there are three test trials. The test is repeated with the left hand. In this selected test battery, the scores of the non-dominant hand were used to provide a measure of fine motor speed as more variations in those scores would be expected due less immunity to fatigue [88] and a slower tapping rate [89].

Non-verbal Reasoning Test (NVRT) – The Reasoning test is usually less than 5 minutes as those who are capable can respond much more quickly than the time-out allows. There are 15 presentations with 14 seconds response time. The test runs continuously for about 5 minutes. It consists of a series of puzzles, or visual analogies, similar to those in Raven's Progressive Matrices. The puzzles are progressively more difficult. The subject identifies the correct response from field possible answers by selecting a number to match the answer. The report captures correct and incorrect responses as well as the reaction time. This test measures ability of subject to understand and analyze visual information and solve problems using visual reasoning. The correct and reaction time scores were selected in this battery.

Table 3.4: Instrumentation, measurements and their roles

	Measure	Role
Concussion History	Self-Administered Questionnaire	Stratification tool
Contact Exposure Index	Exposure history including games and practices	Stratification tool
Playing Position	Self-Administered Questionnaire	Stratification tool
Diffusion-Tensor Imaging (DTI)	Fractional anisotropy (FA)	White matter integrity
	Mean diffusion (MD)	White matter integrity
CNS Vital signs tests	Delayed VERM and VISM corrected recognition score	Provides measure of verbal and visual memory.
	Symbol-Digit Coding (SDC)	Provides measure of psychomotor speed.
	The Shifting Performance Test (SAT)	Provides measure of subject's ability to shift from one instruction set to another quickly and accurately.
	Finger Tapping Test (FTT) – Non dominant hand	Provides measure of fine motor speed (kinesthetic and visual-motor ability).
	Non-verbal Reasoning test	Provides measure of subject's ability to understand and analyze visual information and solve problems using visual reasoning.

3.3 Testing Procedures

3.3a Setting

All subjects were assessed at UNC-Chapel Hill's Biomedical Imaging Research Center, and The Matthew Gfeller Sport-Related TBI Research Center. Study participants were evaluated as part of a two-day visit in which they were assessed for neurocognitive status, brain imaging findings, ApoE4 genotyping, and depressive status. A pre-visit telephone interview conducted by researchers at the Center for the Study of Retired Athletes established background demographics regarding their previous football exposure (involving both sub-concussive and concussive impacts) and prior medical history, including sport-related and non-sport-related concussions sustained during their career.

3.4 Data Analysis

3.4a Magnetic Resonance Imaging

3.4a(1) Preprocessing steps

All acquired neuroimaging data were processed under supervision of our image analysis expert, Dr. Shen, who has applied his techniques to study various brain diseases [90, 91]. All pre-processing steps were performed with the FMRIB Diffusion Tool Box (FDT; FMRIB Centre, University of Oxford, UK). The pre-processing steps for the neuroimaging analyses included (1) alignment of all modality images (including T1, and DTI) of the same subject by rigid registration [92], (2) removal of extra-cranial tissues (skull-stripping) [93], and (3) fitting of the diffusion tensors at each voxel of the corrected data to check for quality. The end outputs (the FA and L2 images) were carried into the TBSS protocol prior to the voxelwise analysis.

This diffusion dataset included 32 former College players and 31 former NFL players for a total of 63 retirees' 3D brain images. However, subjects NFLC_1141 (COL+NFL) and NFLC_2092 (COL) were removed from the imaging dataset because of missing and/or incomplete images, thus, reducing our imaging sample size to 61 total subjects (31 former College and 30 former NFL players). Each set of images encoded diffusion strength in various directions. The diffuse weighted image for each participant were created using *fslmerge*, which concatenated all NIFTI images files into a single 4D output. Prior to carrying the data analysis, the preprocessing procedure included aligning all the images with one another (registration) before estimating any

diffusion related measures: the diffusion tensor, the principal diffusion direction and fractional anisotropy [81]. Such alignment allowed for correction of the head motion (during the scanning session) and reduction of effects from the gradient coil eddy currents [94]. The registration process involved finding the best geometric alignment of all (volumetric brain) images to create an optimal alignment between a reference image (I^r) and a floating image (I^f). Such maximized similarities between the subjects' brains and allowed to identify regions with differences [82, 95]. The eddy current corrections were carried using the diffusion-weighted images (using *eddy_correct*). The output images from the correction were then skull-stripped using the brain extraction tool (BET). BET deletes non-brain tissues from the diffusion-weighted images and generates a binary brain mask (using *bet2*), which was used in the next pre-processing step [96]. BET was carried at a fractional intensity threshold of 0.16. A diffusion tensor model was then fitted at each voxels of the pre-processed output data, using *dtifit* (from the FDT fsl toolbox), which then created the output files (FA, L2) that were used in TBSS. Non-linear intra- and inter-modal brain image registration methods were implemented using a fully automated robust and accurate tool called FNIRT (under FSL fMRIB's non-Linear Image Registration Tool) [82, 95, 97].

3.4a(2) Tract-Based Spatial Statistics (TBSS)

The robust white matter registration and following voxel wise analyses were performed on skeleton voxels using Tract-Based Spatial Statistics method (TBSS; FMRIB Centre, University of Oxford, UK). Prior to starting the TBSS imaging processing steps, the output images from the pre-processing analysis needed to be reoriented (using *fslreorient2std*) to match the orientation of the standard template images (MNI152). The oriented images were then used in the TBSS scripts, which first organized the FA images. Second, TBSS ran the nonlinear registration and aligned every FA images to a 1x1x1mm standard space. The high resolution pre-determined target image (FMRI58_FA) was used for registration to which every subject's FA image was aligned. The following TBSS step applied the nonlinear transforms to all subjects in order to bring them into a common standard space and create a single 4D image. TBSS estimated a group mean FA skeleton representing the center of all fiber bundles (mean of a collection of white matter neurons following a similar anatomical path locally) that are common to the subjects from the dataset [81]. The mean FA skeleton image was created by aligning all FA images onto the skeleton and applying the "thinning" function (a non-maximum-suppression

perpendicular to the local tract structure). Next, a threshold function (>0.2) was applied to the mean FA skeleton map to remove areas of low mean FA and high inter-subject variability [81]. Each subjects' aligned FA image was then projected onto the skeleton by filling the skeleton with FA values from the nearest relevant tract center. Such projection was done for each skeleton voxels by searching the maximum value in the subjects' FA image perpendicular to the local skeleton structure [81].

The MD images were created through FSL's non-FA images TBSS protocol using the second eigenvalue (L2) images created above (recommended option). Following the non-linear and skeletonisation stages, each subjects' vectors were projected onto the mean MD skeleton. The statistical analyses comprised under the FSL comprehensive library of analysis tools were then carried across the stratified groups on the skeleton-space FA and MD data.

3.4a(3) Voxelwise Statistical Analysis

A 3-factors between groups multivariate ANOVA was carried for the voxelwise analysis (2(COL, COL+NFL) x 2(Low, High) x 2(Nonspeed, Speed)) to investigate the main and interaction effects of all three fixed variables (Concussion history, football exposure, and playing position) [98]. This work used "randomise" to conduct its statistical analysis of DTI images [99] and investigate the presence of white matter integrity differences between the different history/exposure groups and the positional groups. Randomise is FSL's tool (FMRIB Centre, University of Oxford, UK) for nonparametric permutation inference on neuroimaging data [100]. Randomisation (at 500 permutations) method is used for inference thresholding on statistic maps when the null distribution is unknown using standard GLM (general linear models) design setup [99]. The null distribution is unknown because the noise does not follow a normal distribution or because the non-standard statistics are used to summarize the data [100]. Randomise produces a test statistic image and sets of p-value images, which allows to determine if the observed differences between the groups is large enough to reject the null hypotheses. The results from the permutation testing were followed with correction for multiple comparisons using the threshold-free cluster enhancement (TFCE) approach of randomise.

The TFCE-based test is rigorous and aims to enhance areas of signal (clusters) that show spatial contiguity without depending on hard-threshold-based clustering [101]. TFCE takes the initial

raw statistical neuroimage and runs it through an algorithm that should enhance the intensity within cluster-like regions more than background (noise) regions [101]. Such process produces an output image that is not intrinsically clustered/thresholded, but in which voxel-wise values represent the amount of cluster-like spatial support. It is hoped that following TFCE enhancements, optimized thresholding will be better discriminated between background noise and spatially-extended signal [101].

The final significant results ($p<0.05$; corrected) were superimposed on the normalized group FA (based of FMRI58_FA standard template) and skeleton maps. The TBSS-fill feature of the FSL tool box was applied to all significant results to enhance the visualization of the clusters, which has been commonly used in previous studies [71, 102]. The white-matter tracts highlighted by the voxelwise analysis were identified using the ICBM-DTI-81 white-matter labels atlas and the JHU white-matter tractography atlas, both provided by Johns Hopkins University [103-105]. The ICBM-DTI-81 white-matter atlas was created by averaging 81 diffusion MRI tensors maps by hand segmentation. It contains a total of 48 white matter tract labels. The JHU white-matter tractography atlas was obtained by averaging probabilistic tractography data collected by 28 normal subjects and contains a total of 20 structures.

3.4b Neurocognitive Tests

3.4b(1) Statistical Analysis

A 2(COL, COL+NFL) x 2(Low, High) x 2(Nonspeed, Speed) three-way multivariate ANOVA analysis was carried to compare the effects of all three variables (football exposure, concussion history and position) on the players' neurocognitive performance. Due to missing scores for two specific participants, the sample sizes were re-adjusted for each test. The variations in sample size for each neurocognitive test are summarized in Table 3.5. Subject NFLC_1569 had scores of 0.00 for the VERM_Delayed_CR, VISM_Delayed_CR, and FTT_taps_NONDominant-hand. Subject NFLC_2560 had a score of 0.00 for the SCD test. Such were therefore deleted from their respective sample in order to run the three-way ANOVA analysis. The Table 3.6 below summarizes the statistical analysis plan for both the neuroimaging data and the neurocognitive tests.

Table 3.5: Adjusted sample sizes used for 3-way ANOVA by tasks

*Specific sample size from which NFLC_1569 scores were removed due to incompletion.
**Specific sample size from which NFLC_2560 scores were removed due to incompletion.

Neurocognitive Test	N (COL)	N (COL+NFL)	N (Total)
Delay Verbal Memory	32	*30	62
Delay Visual Memory	32	*30	62
Symbol Digit Coding	**31	31	62
Shifting Attention Task	32	31	63
Finger Tapping Test	32	*30	62
Nonverbal Reasoning Task	32	31	63

Table 3.6: Data analysis plan

Focus	Research Questions	Variable Analysis	Statistical Method
Primary Aim	Are there significant differences between the integrity of the white matter tracts (defined by fractional anisotropy and medial diffusivity) observed in diffusion tensor images of National Football League and college football retired players based on years of football exposure, position played and concussion history?	IV: Years of football exposure, playing position and concussion history. DV: The integrity of the white matter tracts (defined by FA and MD).	Three-way ANOVA (with randomise non-parametric analysis and TFCE correction for multiple comparisons) for history, exposure and position for both FA and MD.
Secondary Aim	Are there significant differences between the performance of National Football League and college retired football players on the neurocognitive testing based on football exposure, position played and concussion history?	IV: Years of football exposure, playing position and concussion history. DV: Performance on neurocognitive tests.	Three-way ANOVA for history, exposure and position on each selected neurocognitive tests.
Exploratory Aim	If there are differences between the performances on the cognitive tests, do the observed changes in cerebral white matter integrity (from DTI) correlate with the domain of the cognitive tests where athletes have shown deficits?	IV: The structural changes in white matter integrity (expected decrease in FA and increase in MD) observed from years of football exposure, playing position and concussion history. DV: The performance on neurocognitive cognitive tests.	Multivariate regressions

CHAPTER IV: RESULTS

The purpose of this study was to investigate the differences in white matter integrity and neurocognitive performances of former college and NFL football players based on reported concussion history (HIGH vs. LOW), football exposure (COL vs. COL+NFL) and primary playing position (Non-speed vs. Speed). The results are summarized in the sections below. Demographics for our study participants are presented in Table 4.1. Variables are stratified by the concussion history, football exposure and playing position variables used to conduct the data analyses.

4.1 Magnetic Resonance Imaging: Diffuse Tensor Imaging (Aim 1)

4.1a Structural Differences In Fractional Anisotropy (FA)

The three-factor multivariate ANOVA (Analysis of Variance) design measuring between-group interaction and main effects of concussion history, football exposure and position, with TFCE corrections, only showed significant interaction between the ***concussion history*** and ***position*** variables (with the apriori p value set at $p<0.05$). There were no significant main effects (alone) for concussion history, football exposure, or playing position. A total of three significant clusters were identified from the randomise TFCE interaction. The voxel peak intensities (for x, y and z coordinates in mm) and the p values for each cluster are summarized in Table 4.2. It is important to note that weighed exposure hours for the position and concussion groups interaction were balanced (Table 4.3). Thus, contact exposure is not a co-variable to this interaction. The mean FA peak voxel in all three significant clusters, stratified by playing position and concussion history, can be found in Table 4.4. It is also important to note that not every voxel within each defined cluster is significant. The rigorous TFCE randomise analysis isolates probability clusters within which at least one (or more) FA voxel is significant. Our structural analysis using the FSL atlas toolbox reveals that the significant differences in FA based on differences in concussion history and position played are located in the frontal white-matter of the brain, in both hemispheres. More specifically, differences in FA were concentrated towards the forceps minor [103-105] white matter structure. Figure 4.1 shows all three significant clusters: cluster 1 (A, D and G), cluster 2 (B, E and H) and cluster 3 (C, F and I). When overlapping the significant clusters over the white matter maps, provided by JHU DTI-based

white-matter tractography atlas (see Section 3.4a3), the bilateral forceps minor (cluster 1 and 2; light blue) is primarily identified (Figure 4.2). Cluster 3 was reported unclassified by the atlas although the closest white-matter structure is likely to be the forceps minor.

In order to ensure the significance of the concussion history and position interaction, the mean FA peak voxel from all three clusters (Table 4.4) was analyzed in a 2(High, Low) x 2(Nonspeed, Speed) two ways ANOVA. The interaction was confirmed to be significant in all three clusters as seen in Table 4.5 (cluster 1 ($p=0.01$), cluster 2 ($p=0.001$) and cluster 3 ($p=0.01$)).

To further investigate the direction of the differences in FA from the concussion history and position interaction, a set of univariate ANOVA post-hoc analyses were conducted. Table 4.6 summarizes the post-hoc analysis when controlling for the position variable (Non-speed vs. Speed). The analysis showed that non-speed players in the LOW concussion (0-1) group had significantly higher FA than non-speed players in the HIGH concussion group (≥3) for all three clusters (cluster 1 ($p=0.02$), cluster 2 ($p=0.03$) and cluster 3 ($p=0.002$)). Contrary to this finding, the same analysis revealed no significant differences in two of the three clusters for speed players, while significantly higher FA was observed in the HIGH concussion group for cluster 2 ($p=0.01$).

The second univariate ANOVA post-hoc analysis controlled for concussion history, and is summarized in Table 4.7. When analyzing the HIGH concussion group (≥3), only cluster 1 ($p=0.04$) showed significant results with speed players having significantly higher FA than non-speed players. On the other hand, the analysis for the LOW (0-1) concussion group showed significant results trending in the opposite direction, with non-speed players having significantly higher FA in both clusters 2 ($p=0.001$) and cluster 3 (0.04) compared to the speed position. Figure 4.3 provides a visual representation of the post-hos analyses and the significant interaction between the concussion history and position variables on FA for all three clusters (1-3).

4.1b Structural Differences In Medial Diffusivity (MD)

No main effects or interactions for differences in MD between concussion history, football exposure and playing position survived the randomise three-factor multivariate ANOVA with TFCE corrections. Thus, no apparent significant differences in MD could be identified between all three variables.

4.2 Neurocognitive Test Performances (Aim 2)

A three-factor multivariate ANOVA measuring interaction effects and between-group main effects only revealed significant differences in neurocognitive performance of the shifting attention task between all three group variables ($F=5.14$, $p=0.03$). Table 4.8 summarizes the interaction and main effects of exposure, concussion history and position variables (with the apriori p value set at $p<0.05$). Given the 3-way interactions for the shifting attention test, a series of two-factor post-hoc analyses were conducted in order to identify where significant differences existed among the three variables. Table 4.9 summarizes the means (and standard deviation) shifting attention score results used to carry the post-hoc analyses. As presented in Table 4.10, there were no further interaction effects for the shifting attention test between concussion history and position played for the college group (COL: $F=1.449$, $p=0.24$). On the other hand, the NFL group showed significant differences between the ***concussion history*** and ***position*** variables (COL+NFL: $F=6$, $p=0.02$). Such interaction was further investigated using the mean shifting attention test scores in Table 4.9. There was no difference in performance between the speed (45.00 ± 5.87) and non-speed (47.33 ± 4.32) position groups with LOW (0-1) concussion history. Contrastingly, those in the COL+NFL HIGH group with ≥ 3 concussions and who played a speed position showed significantly better shifting attention scores (52.13 ± 5.33) compared to the non-speed group (42.50 ± 9.12). Figures 4.4 and 4.5 provide a visual representation of the post-hoc analyses described above.

4.3 The Relationship Between Structural White Matter Differences And Neurocognitive Performances (Exploratory Aim)

The assessment of the potential relationship between differences in cerebral white matter integrity and neurocognitive performances was dependent on the fact that significant variations were to be found with respect to both independent variables. Given that no clear differences in neurocognitive performances between concussion history, football exposure and position were identified (section 4.2), this exploratory aim was not further investigated.

CHAPTER V: DISCUSSION AND CONCLUSION

5.1 Discussion

The use of brain imaging has recently been introduced to the field of concussion research in order to further investigate the neuro-structural and functional repercussions of such injury to the head. This study aimed to better understand the effects of concussion history, football exposure and playing position on white matter integrity and neurocognitive performances in a random sample of former collegiate and professional football players. One of the most important finding of our study was that a novel interaction effect between concussion history and playing position was found with respect to differences in white matter integrity (FA). Additionally, our findings from both the neurocognitive and the imaging analyses showed no main effects or interactions of added football exposure in the retired athletes. Our findings seem to suggest that without concussive injuries, the added football exposure (and therefore increased number of sub-concussive impacts to the head) does not result in significant differences in white matter integrity or neurocognitive abilities. To a certain degree, such results challenge the current perceived idea that added exposure is harmful to football athletes. For instance, previous work in this field by Bazarian and al. [54] suggested that college football athletes with no history of concussion experienced greater differences in FA and MD compared to non-athlete students. Although our study does not provide a comparison to a control group, our findings do not show differences in FA or MD based on added football exposure. Other work published by Lehman and al. [2] proposed that professional football players may be at increased risks of death from neurodegenerative diseases, which again is now being challenged by our recent findings since comparisons between white matter tracts based of football exposure revealed no significant differences between college and professional players. However, we did not study neurodegenerative diseases in this cohort and therefore, our findings may not be directly comparable.

Overall, our results showed no main effects of concussion history, football exposure or playing position on neurocognitive performances, which all appeared to be normal with respect to one another. The observed three-way interaction in the shifting attention scores (Table 4.8) revealed that COL+NFL speed players, within the HIGH concussion group, scored significantly higher

than COL+NFL non-speed players of the same group. The large effect size of such interaction (d=1.29) supports the significant interaction suggesting that shifting attention abilities in players with a high history of concussions may vary across positions. Such results differ from what was originally hypothesized in this study in that a high concussion history was predicted to result in the greatest neurocognitive deficits.

The findings of the imaging analyses (FA and MD) were consistent with most of the findings from the neurocognitive analyses in that no main effects of concussion history, football exposure or playing position were found. More interestingly, a two-way interaction between concussion history and playing position (Table 4.2) showed significant differences in fractional anisotropy whereas no variations in white matter were associated with differences in football exposure. When further analyzing the interaction between concussion history and playing position, football exposure was confirmed to be equivalent and non-significantly different across all groups (Table 4.3). In the post-hoc analyses, it was found that concussion history accounted for consistent differences in white matter integrity within non-speed players. In fact, non-speed players with LOW concussion history (0-1) were found to have greater white matter integrity (increased FA) compared to non-speed players with HIGH concussion history (≥3). Such findings suggest that concussion history may play a larger role in non-speed players, with regards to structural changes in the white matter. Other less consistent findings within the positional univariate post-hoc imaging analysis were also observed with speed players in the HIGH concussion history group having higher FA then the LOW concussion group. Further inconsistences with differences in white matter integrity were also observed in the univariate post-hoc concussion history analysis where HIGH concussion groups players showed greater FA in the speed players whereas LOW concussions groups showed greater FA in non-speed players. Given the inconsistence of those results, emphasis is put on the differences in white matter integrity observed in non-speed players, which showed to be significant in all three defined clusters resulting from the rigorous randomise TFCE voxel-wise analysis. Such results differ from what was initially hypothesized in that greater white matter variations, based on concussion history, were expected in speed players since they typically build of more momentum before the impact [36]. Interestingly however, the findings from the imaging analyses were consistent with the general findings from the neurocognitive analyses in that increases in football exposure, which

indirectly correlates with increases in sustained sub-concussive impacts, may not be as damaging structurally as originally believed. Indeed, no differences in white matter integrity were associated with differences in football exposure. The fact that both neurocognitive performances and structural imaging analyses have showed no significant differences based on added years of football is an important finding, which seems to suggest that the current perceptions on the perceived detrimental effects of increased football exposure may need to be reevaluated. In the current literature, chronic traumatic encephalopathy (CTE) observed in deceased former football players is thought to be caused, at least in part, by repetitive brain traumas including both concussive and sub-concussive impacts to the head [60]. Findings from this study seem to refute, to some degree, such notion and re-emphasize that increases in sub-concussive impacts from prolonged football exposure may not be as substantial as a factor contributing to neurocognitive and neuro-structural changes.

As previously mentioned, the initial intention of investigating neurocognitive performances and white matter integrity in former retired athletes was to find cognitive impairments and structural differences between players based on three major variables that are likely to define football players' exposure to head injuries. We hypothesized that a main effect of concussion history would explain significant differences in both neurocognitive functions and structural integrity of the white matter. Our findings do not support our hypotheses. Furthermore, it is unclear as to why differences in shifting attention between non-speed and speed positions were only observed in the HIGH concussion group. No significant differences between the HIGH and LOW concussion groups were observed, which limits clinical meanings of such findings. Given that the difference in shifting attention is mainly driven by the playing position, it could be argued that such variance in attention is the result of typical position specific athletic and cognitive abilities required to perform in the National Football League. It may be that speed players, given the required skills and physical demands of such positions, are advantaged and therefore naturally selected for higher shifting attention abilities, compared to non-speed players, which may be driving the direction of this significant difference.

Although, the imaging results may not be as easily interpreted as initially hoped, one consistent finding from the voxel-wise analysis is the differences in white matter integrity located towards

the frontal region of the brain. More specifically, the forceps minor seems to be the primary damaged structure (see Figure 4.2). Such is made of commissural fibers, which connects the lateral and medial surfaces of the frontal lobes crossing the midline at the genu of the corpus callosum. Although not yet fully understood, the forceps minor has been linked to processing speed functions [106]. As previously discussed, diffuse axonal injuries resulting in white matter changes are hypothesized to originate from neural microstructural damages in the brain resulting from shearing forces applied to the nerve fibers following concussive impacts to the head [33, 69]. As reported by previous work published by Crisco and al. [41], the highest percentage of impacts in collegiate football players occurred to the front of the helmet. Although these findings may be limited to our study since they do not quantify head impact locations in NFL players, the validation that most impacts are sustained towards the front of the helmet allows for interesting speculations about the potential effects of continuous shearing forces applied to the frontal lobe. Two-dimensional biomechanical studies of frontal head impact injuries have shown that impact strains may be inducing tension forces that are distributed throughout the anterior region of the longitudinal fissure [107]. The anatomical location of the forceps minor, which crosses the midline of the genu and is therefore split by the anterior section of the fissure, may expose it to greater structural damages. Indeed, repeated application of the shearing forces, following concussive impacts, may cause tissue strain within the forceps minor and therefore expose non-speed players to increased risks for axonal damages resulting in structural white matter abnormalities. Previous work by Tremblay and al. [108] compared white matter tract abnormalities between age-matched retired collegiate hockey and American football athletes with history of sports-related concussions to a control group of athletes who had no prior history of concussion. When compared to controls, the concussion group had significant decreased FA in the hemispheric fibers of the corpus callosum and the forceps minor, as well as decreased FA in the intra-hemispheric association fibers of the right the right inferior longitudinal fasciculus, the right inferior fronto-occipital fasciculus and bilateral superior longitudinal fasciculi. Further signs of significantly low FA were also found, by the same group, in the projection fibers of the anterior limb of the right internal capsule, the right external capsule, as well as the right corona radiata. Although our cohort did not compare changes in FA to a control group with no history of concussive injuries, our findings do not seem to agree with the findings from the Temblay group since significant differences in FA were only observed within the forceps minor. Furthermore,

the same group reported similar region-specific abnormal increases in MD within the inter-hemispheric fibres, the association fibres and the projections fibres whereas our study showed no significant differences in mean diffusivity across all three variables. Preceding work on traumatic brain injuries has also failed to match the presence of functional abnormalities, obtained through fMRI BOLD analysis, with changes in white matter integrities, observed via DTI [16]. However, our findings regarding the insignificant effects of added football exposure are in agreement with previous work done by this team using functional MRI measures. Such study presented that concussion history, and not football exposure, was related to abnormal functional activation patterns in episodic memory [4]. Other previous work on functional neuroimaging has also proposed that multiple concussions may be associated with functional inefficiencies in memory network [23]. Although there may not be a direct accepted correlation between long-term functional and structural changes in the brain following a history of concussive injuries, our study of retired collegiate and professional football players, along with previous findings, surely seem to suggest that concussion history (and playing position) play a larger role with respect to long-term damages of the white matter and neural recruitment patterns when compared to added football exposure.

5.2 Limitations

Our study is not without limitations. The recruitment of study participants relied on a general health questionnaire of self-reported data. The estimates of concussion history and football exposure were both assessed using retrospective interviewing of the former athletes at the time of their visit. Although there may be some inaccuracies in memory with respect to recalling the total number of sustained concussions and history of football exposure, data from our group suggests that such inaccuracies are limited. Indeed, football players actually tend to be accurate and consistent when reporting history of previous concussion injuries [109]. This study also estimated weighted contact exposure from high school through professional careers and so did not take into account football participation prior to high school. Such early time period should be considered in future studies given that players at that level may use improper tackling techniques, which could affect early neural and functional development. The weighted total hours of contact exposure defined football exposure in our study and was designed to help our

understanding of the cumulative effects of sustaining repetitive subconcussive impacts to the head.

Our findings are further limited to collegiate and professional football players as no control group (non-football players) was used to compare the effects of football exposure and concussion history. Although the LOW COL group served as our 'control' for low exposure and low concussion history, players within that group still sustained a great amount of head impacts [39] during their college careers even though such may have been sub-concussive. Furthermore, there is also potential for those players with low history of concussion to have greater natural tolerance for sustaining impacts to the head without resulting in concussive injuries. White matter damages following traumatic brain injuries in the general population have been identified from previous studies [110, 111], which shows that such changes are not limited to collegiate and professional football athletes.

The findings from the imaging analysis are limited by certain technological restraints of the field of human brain imaging. Primarily, it is important to note that TBSS evaluates diffusion behavior on a voxel-wise basis for fractional anisotropy skeleton of major white matter tracts, which restrict its findings to the center of the white matter pathways. Furthermore, due to the complexity of diffusion tensor imaging as a brain imaging technique, TBSS analysis of voxels with crossing bundles are hard to interpret since they create an oblate, planar shaped tensor, which misdirects the white matter fibers. Such may limit TBSS's sensitivity in locating potential differences in FA and MD [81, 105]. Consequently, DTI data analysis is strongly limited in fibers with crossing or kissing white matter. With that being said, it should be noted that the forceps minor structure outlined in this cohort is well defined within the JHU white matter tractography atlas [103-105] and should therefore not be affected by such limitation.

5.3 Conclusion

The results of this study are important to the current literature on long-term effects of concussive injuries, as it is the first of its kind to look at the interacting effects of football exposure, concussion history and playing position on neurocognitive performances and structural white matter differences. Specifically, it highlights a novel interaction between concussion

history and playing position in football players with respect to differences in fractional anisotropy. Secondly, it challenges a number of recent findings such as work presented by Lehman and al. [2], suggesting that added football exposure may expose players to greater risks of neurodegenerative diseases. Although our findings are limited, it appears that a higher concussion history and playing a non-speed position account for some degree of decreased white matter integrity. The effect of such an interaction is not yet fully understood and should be further investigated in future studies. More importantly, such outcomes should to be enough to argue for the continuation of current behavior modification interventions in football, which aim to reinforce proper hitting and tackling techniques in football players. The purpose of such biomechanical adjustments is to reduce the incidence of sport related concussive injuries, which may in turn be effective in lessening future structural white matter abnormalities in football athletes. Future research in this field should further investigate the effects of player's position on neurocognitive functions as well as potential functional and structural abnormalities within specific brain regions. Understanding how different football athletes, competing at different positions, may be exposed to different risks of head injuries will allow us to better comprehend patterns of reported long-term deficits as well as to develop innovative coaching techniques to further protect the players' health.

Table 4.1: Demographics (mean and standard deviation)

	COL (n=32)				COL+NFL (n=31)			
	LOW (0-1)		HIGH (≥3)		LOW (0-1)		HIGH (≥3)	
	Non-Speed (n=9)	Speed (n=7)	Non-Speed (n=7)	Speed (n=9)	Non-Speed (n=6)	Speed (n=10)	Non-Speed (n=6)	Speed (n=9)
Age	58.67 (5.7)	59.86 (2.5)	58.57 (3.7)	57.89 (3.4)	57.33 (4.5)	59.60 (3.4)	58.67 (5.7)	57.33 (1.9
Height (inches)	74.00 (2.0)	72.71 (2.1)	75.00 (1.5)	72.33 (1.8)	76.83 (1.7)	73.90 (2.8)	75.50 (1.6)	74.00 (2.5)
Weight (lbs)	239.22 (22.8)	217.14 (39.5)	257.43 (17.3)	228.67 (34.4)	259.67 (22.8)	238.50 (34.0)	256.33 (22.8)	234.22 (24.9)
Number of Concussions	0.67 (0.5)	0.57 (0.5)	8.71 (9.9)	6.00 (3.4)	0.50 (0.5)	0.40 (0.5)	4.8 (1.7)	8.44 (8.7)
Years of Football Played	8.33 (0.5)	7.68 (0.5)	8.00 (0.8)	8.11 (0.6)	18.42 (2.5)	17.05 (2.6)	17.33 (5.5)	17.83 (2.6)

Table 4.2: 2x2x2 ANOVA significant cluster locations from randomise TFCE analysis (concussion history-position). Significance p value set at $p<0.05$.

		Voxel Peak Intensity (mm)					
Cluster #	p value	X	Y	Z	Number of Voxels	Hemisphere	Primary Structure
1	**0.044**	-17	30	18	98	Left	Forceps Minor
2	**0.036**	-20	47	6	460	Left	Forceps Minor
3	**0.036**	12	33	-15	1041	Right	Frontal White Matter

Table 4.3: Mean weighted contact exposure hours (standard deviation) for post-hoc imaging analyses

Position	Concussion Group	N	Weighted Contact Hours
Non-speed (n=27)	LOW (0-1)	14	2257.30 (1622.5)
	HIGH (≥3)	13	2233.91 (1409.5)
Speed (n=34)	LOW (0-1)	16	2100.14 (1334.1)
	HIGH (≥3)	18	2105.99 (1454.2)

*Total of 61 subjects in the imaging analysis since players NFLC_1141 and NFLC_2092 were removed from the dataset due to incomplete imaging data.

Table 4.4: Mean FA voxel (standard deviation) for position-concussion history significant clusters from TFCE analysis

			Mean FA peak voxel		
Position	Concussion Group	N	Cluster 1	Cluster 2	Cluster 3
Non-speed (n=27)	LOW (0-1)	14	0.501 (0.036)	0.427 (0.053)	0.452 (0.037)
	HIGH (≥3)	13	0.459 (0.051)	0.376 (0.061)	0.402(0.040)
Speed (n=34)	LOW (0-1)	16	0.470 (0.060)	0.357 (0.047)	0.420 (0.043)
	HIGH (≥3)	18	0.499 (0.051)	0.404 (0.054)	0.423 (0.032)

Table 4.5: Post-hoc 2x2 ANOVA F-values (p-values) for main effects and interactions of concussion history-position. Significance p value set at $p<0.05$.

	Concussion History (C)	Position (P)	CxP
FA Peak voxel Cluster 1	0.023 (0.63)	0.14 (0.71)	**7.42 (0.01)**
FA Peak voxel Cluster 2	0.03 (0.87)	2.24 (0.14)	**12.51 (0.001)**
FA Peak voxel Cluster 3	5.73 (0.02)*	0.24 (0.62)	**7.27 (0.01)**

*Although this specific peak voxel may be significant, the randomise TFCE analysis did not show a significant main effect of concussion history on targeted cluster 3.

Table 4.6: Post-hoc univariate analysis F-values (p-values) for concussion history-position interaction and direction of significant FA differences. Significance p value set at $p<0.05$.

	Non-speed		Speed	
	F value (p value)	Mean FA peak voxel interaction	F value (p value)	Mean FA peak voxel interaction
Cluster 1	**6.17 (0.02)**	LOW > HIGH	2.37 (0.13)	No difference
Cluster 2	**5.45 (0.03)**	LOW > HIGH	**7.13 (0.01)**	LOW < HIGH
Cluster 3	**11.46 (0.002)**	LOW > HIGH	0.05 (0.82)	No difference

Table 4.7: Post-hoc univariate analysis F-values (p-values) for position played-concussion history interaction and direction of significant FA differences. Significance p value set at $p<0.05$.

	HIGH (≥3)		LOW (0-1)	
	F value (p value)	Mean FA peak voxel interaction	F value (p value)	Mean FA peak voxel interaction
Cluster 1	**4.76 (0.04)**	Speed > Non-speed	2.79 (0.11)	No difference
Cluster 2	1.84 (0.19)	No difference	**14.69 (0.001)**	Speed < Non-speed
Cluster 3	2.75 (0.11)	No difference	**4.53 (0.04)**	Speed < Non-speed

Table 4.8: 2x2x2 ANOVA F-values (p-values) for the main effects and interaction of exposure, concussion history and position variables. Significance p value set at $p<0.05$.

Neurocognitive Task	Exposure (E)	Concussion History (C)	Position (P)	ExC	ExP	CxP	ExCxP
Delay Verbal Memory Corrected Recognition	0.03 (0.86)	1.88 (0.18)	0.48 (0.49)	0.85 (0.36)	0.42 (0.52)	0.04 (0.85)	0.14 (0.71)
Delay Visual Memory Corrected Recognition	0.07 (0.79)	0.00 (0.99)	1.46 (0.23)	0.25 (0.62)	1.00 (0.32)	0.03 (0.86)	1.00 (0.32)
Symbol Digit Coding Accuracy	0.28 (0.60)	1.56 (0.22)	0.00 (0.98)	0.00 (0.98)	0.06 (0.80)	1.99 (0.16)	1.78 (0.19)
Shifting Attention Task Number Correct	4.63 (0.04)	0.00 (0.99)	1.11 (0.30)	0.28 (0.60)	0.41 (0.52)	0.26 (0.61)	**5.14 (0.03)**
Shifting Attention Task Correct Reaction Time	0.85 (0.36)	0.75 (0.39)	0.95 (0.33)	0.04 (0.84)	2.31 (0.14)	0.09 (0.77)	0.48 (0.49)
Finger Tapping Test Non-dominant Hand	1.46 (0.23)	0.47 (0.50)	2.38 (0.13)	0.60 (0.44)	0.15 (0.71)	0.19 (0.66)	2.21 (0.14)
Nonverbal Reasoning Task Number Correct	2.80 (0.10)	0.22 (0.64)	0.08 (0.77)	0.95 (0.33)	0.63 (0.43)	0.59 (0.45)	0.24 (0.62)
Nonverbal Reasoning Task Correct Reaction Time	0.52 (0.48)	0.00 (0.96)	0.16 (0.69)	0.71 (0.40)	0.33 (0.57)	2.58 (0.11)	0.29 (0.59)

Table 4.9: SAT mean scores (standard deviations) from 3-way ANOVA analysis

Position	Concussion Group	Football Exposure	
		COL	COL+NFL
Non-speed	LOW (0-1)	40.33 (13.42)	47.33 (4.32)
	HIGH (≥3)	43.00 (10.00)	**42.50 (9.12)**
Speed	LOW (0-1)	45.00 (3.51)	45.00 (5.87)
	HIGH (≥3)	40.11 (8.42)	**52.13 (5.33)**

Table 4.10: Post-hoc 2x2 ANOVA F-values (p-values) of SAT scores 3-way interaction between exposure, position and concussion history (ExPxC). Significance p value set at $p<0.05$.

Exposure Group	Concussion History (C)	Position (P)	PxC
COL only	0.05 (0.83)	0.02 (0.88)	1.449 (0.24)
COL+NFL only	0.12 (0.73)	2.06 (0.16)	**6 (0.02)**

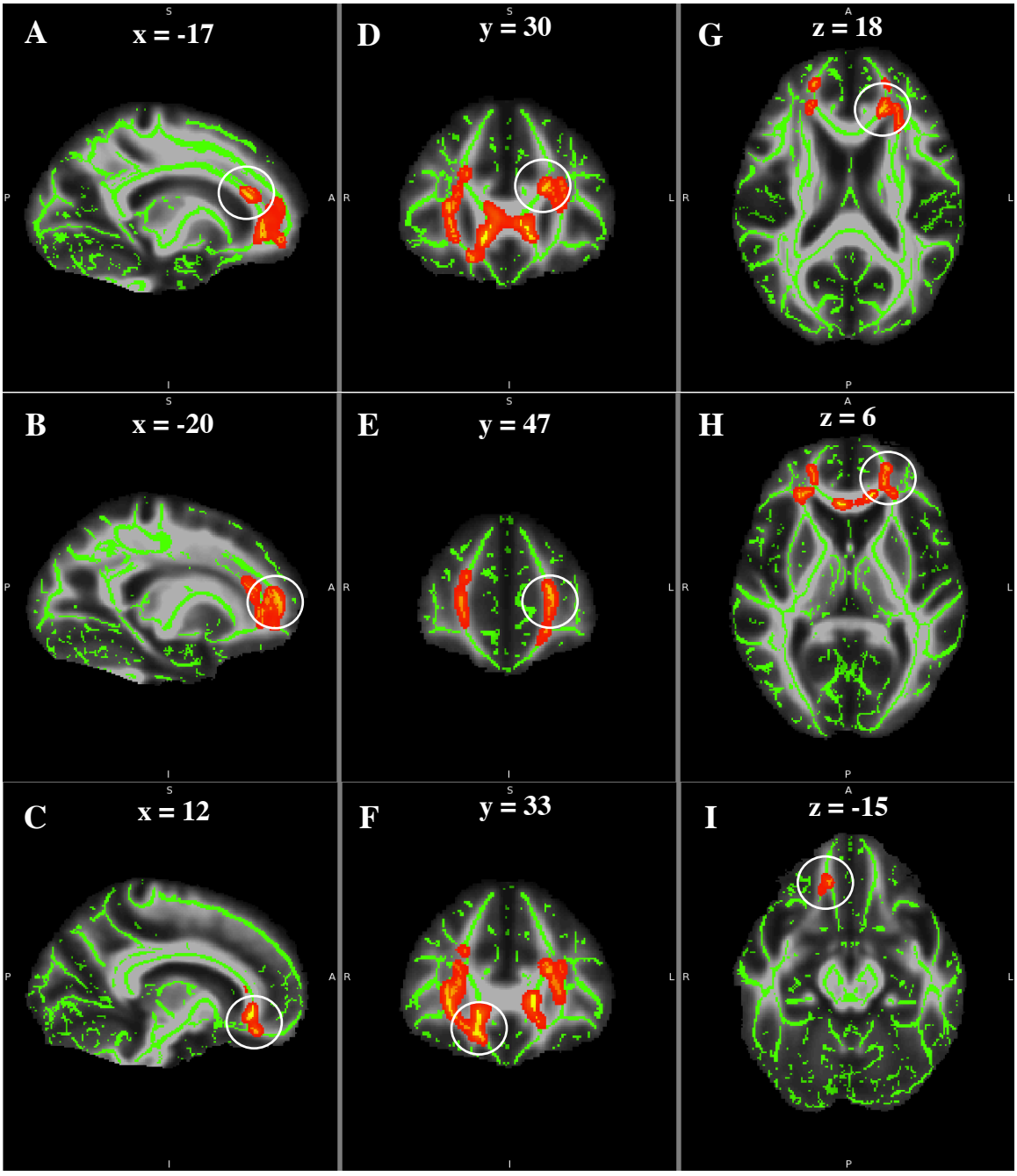

Figure 4.1: Diffuse differences in fractional anisotropy (FA) interaction between concussion history and playing position variables

Sagittal (A,B and C), coronal (D, E and F), and axial (G, H and I) slices from the TBSS voxelwise analysis contrasting the FA maps. The TBSS filled FA skeleton contrasts are overlaid over the mean FA skeleton (green) and the mean FA template (based of FMRI58_FA 1mm standard template). The results are thresholded at p<0.05, and corrected for multiple comparisons (from TFCE analysis). Clusters 1 (A, D and G), cluster 2 (B, E and H) and cluster 3 (C, F and I) are displayed.

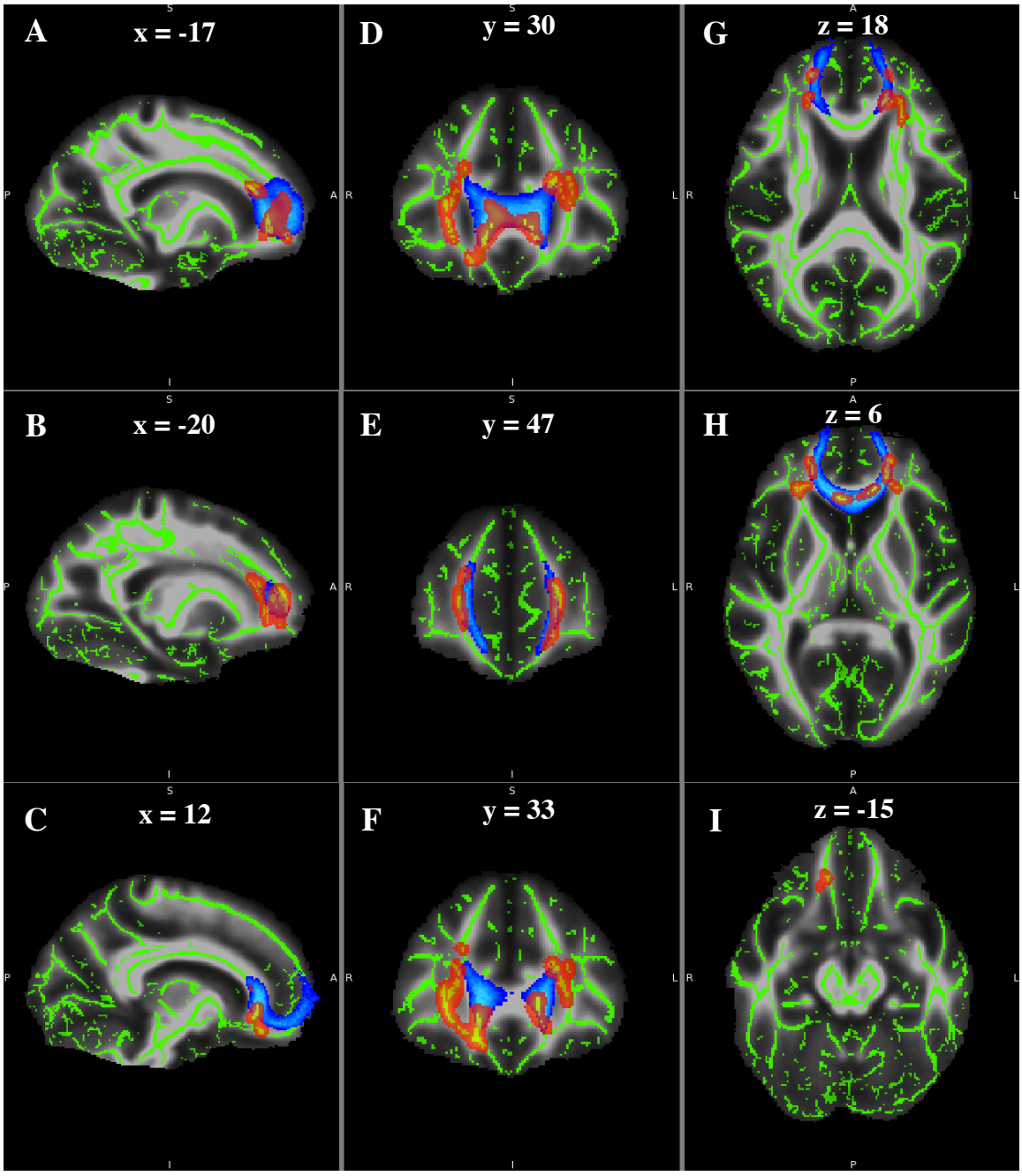

Figure 4.2: Diffuse differences in fractional anisotropy (FA) interaction between concussion history and playing position variables overlaid on forceps minor (light blue)

Sagittal (A,B and C), coronal (D, E and F), and axial (G, H and I) slices from Figure 4.1. The TBSS filled FA skeleton contrasts are overlaid over the identified primary structure from the JHU white matter tractography atlas [103-105]: forceps minor (light blue). The results are thresholded at p<0.05, and corrected for multiple comparisons (from TFCE analysis). Clusters 1 (A, D and G), cluster 2 (B, E and H) and cluster 3 (C, F and I) are displayed.

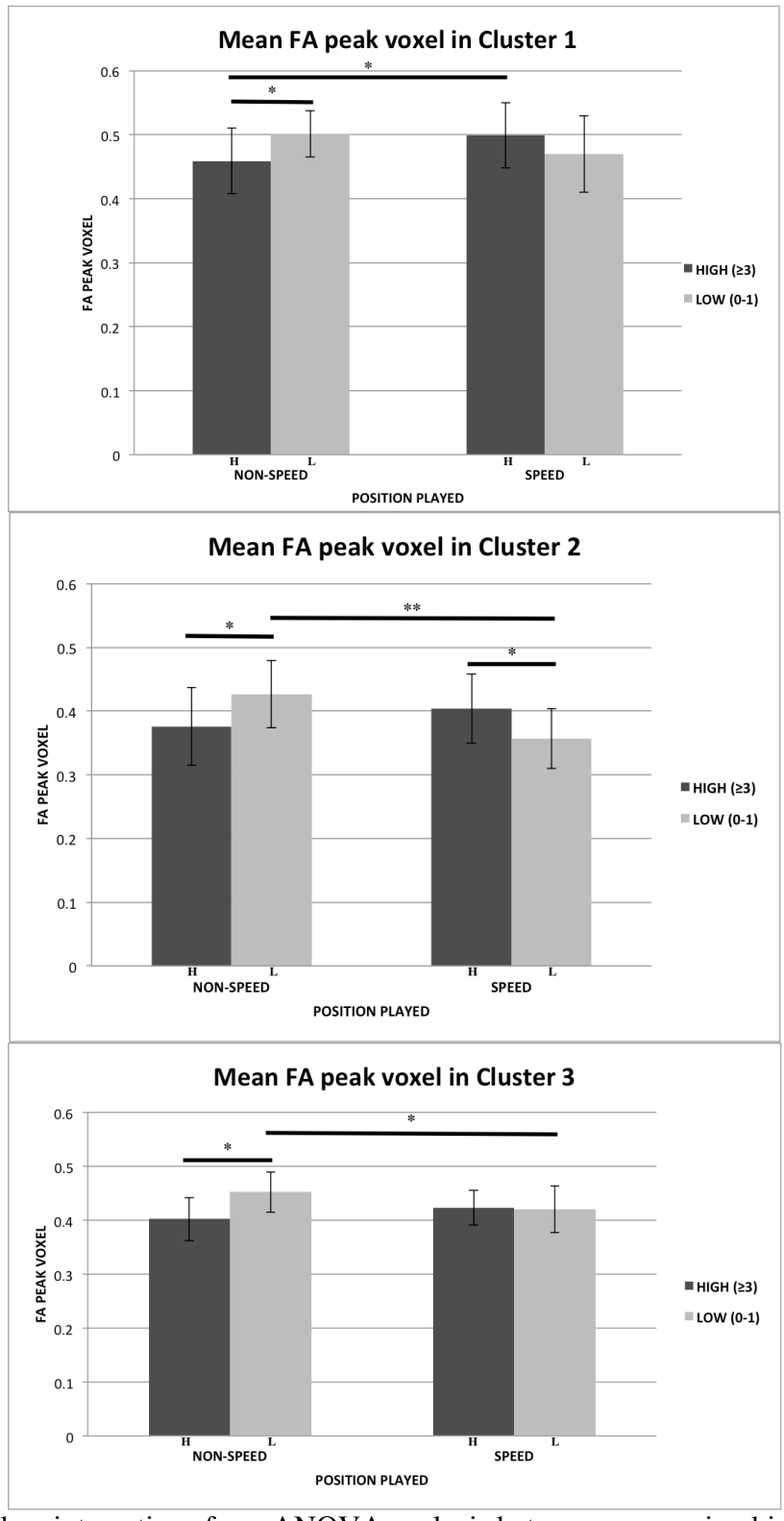

Figure 4.3: Post-hoc interactions from ANOVA analysis between concussion history and playing position variables in clusters (1-3) defined by TFCE analysis

** = p<0.05. ** = p<0.001.*

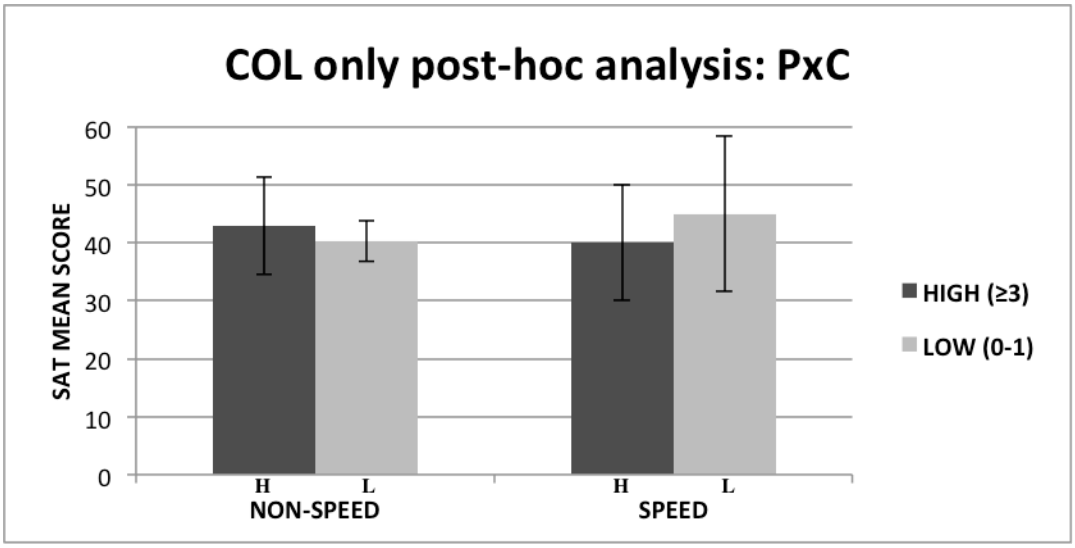

Figure 4.4: Post-hoc 2-way (playing position and concussion history) ANOVA for mean SAT scores in COL only exposure group

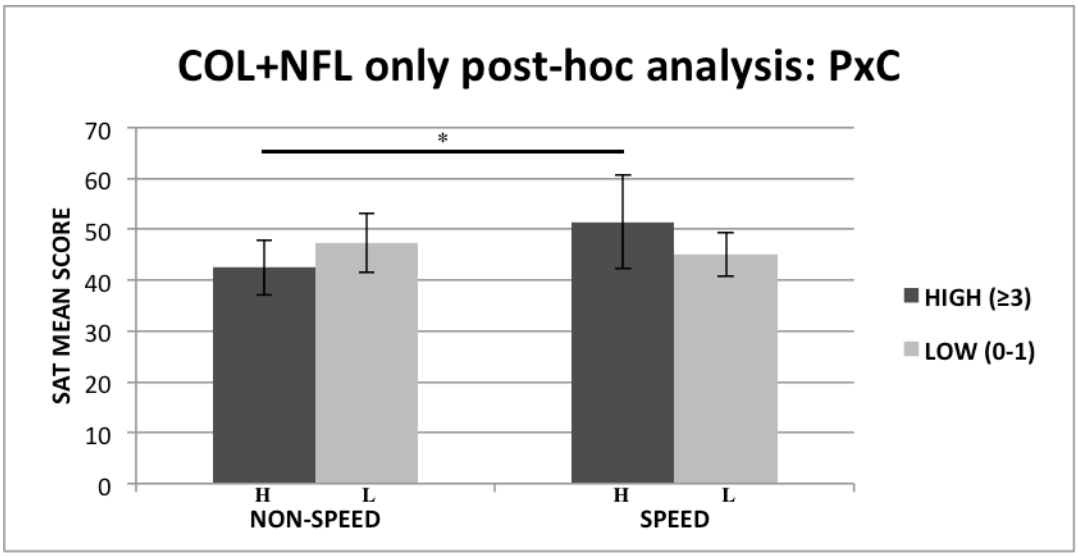

Figure 4.5: Post-hoc 2-way (playing position and concussion history) ANOVA for mean SAT scores in COL+NFL only exposure group

* = $p<0.05$.

REFERENCES

1. Guskiewicz, K., et al., *Association between recurrent concussion and late-life cognitiive impairment in retired professional football players*. Neurosurgery, 2005. 57: p. 719-726.
2. Lehman, E., et al., *Neurodegenerative causes of death among retired National Football League players*. Neurology, 2012. 79: p. 1970-1974.
3. Guskiewicz, K., et al., *Cumulative effects associated with recurrent concussion in Collegiate football players*. Journal of American Medical Association, 2003. 290(19): p. 2549-2555.
4. Varangis, E., et al., *Effects of concussion history and football exposure on working memory performance in retired National Football League and college football players*. (In review), 2013.
5. McCrory, P., et al., *Consensus statement on concussion in sport: the 4th International Conference on Concussion in Sport held in Zurich, November 2012*. Journal of Sports Medicine, 2013. 47: p. 250-258.
6. Greenwald, R.M., et al., *Head impact severity measures for evaluating mild traumatic brain injury risk exposure*. Neurosurgery, 2008. 62: p. 789-798.
7. Buki, A. and J. Povlishock, *All roads lead to disconnection? - Traumatic axonal injury revisited*. Acta Neurochirurgica, 2006. 148: p. 181-193.
8. Langlois, J., W. Rutland-Brown, and M. Wald, *The epidemiology and impact of traumatic brain injury: a brief overview*. Journal of Head Trauma Rehabilitation, 2006. 21(5): p. 375-8.
9. Daneshvar, D., et al., *The Epidemiology of Sport-Related Concussion*. Clinical Sports Medicine, 2011. 30(1): p. 1-17.
10. Panczykowski, D. and J. Pardini, *The multidisciplinary concussion management program*. Prog Neurol Surg., 2014. 28: p. 195-212.
11. Press, A. *Concussions decline after change to kickoff rule*. 2012.
12. Johnson, V., W. Stewart, and D. Smith, *Axonal pathology in traumatic brain injury*. Experimental Neurology, 2013. 246: p. 35-43.
13. Strominger, N.L., *Cerebral Cortex*, in *Noback's Human Nervous System, Seventh Edition: Structure and Function*, S.S.B. Media, Editor. 2012, Springer Science+Business Media: New York. p. 429-451.
14. Zhou, Y., et al., *Mild Traumatic Brain Injury: Longitudinal Regional Brain Volume Changes*. Radiology, 2013. 267(3): p. 880-890.
15. Levine, B., et al., *In Vivo Characterization of Traumatic Brain Injury Neuropathology with Structural and Functional Neuroimaging*. Journal of Neurotrauma, 2006. 23(10): p. 1396-1411.
16. Zhang, K., et al., *Are functional deficits in concussed individuals consistent with white matter structural alterations: combined FMRI & DTI study*. National Institute of Health, 2010. 204(1): p. 57-70.
17. Majerske, C., et al., *Concussion in Sports: Postconcussive Activity Levels, Symptoms, and Neurocognitive Performance*. Journal of Athletic Training, 2008. 43(3): p. 265-274.
18. Zhang, K., et al., *Default Mode Network in Concussed Individuals in Response to the YMCA Physical Stress Test*. Journal of Neurotrauma, 2012. 29: p. 756-765.

19. Dams-O'Connor, K. and A. Yi, *Psychosocial functioning in older adults with Traumatic Brain Injury*. NeuroRehabilitation, 2013. 32: p. 267-273.
20. Swanson, L., *Basic Principles of Nervous System Organization*, in *Neuroscience in the 21st Century*, D.W. Pfaff, Editor. 2013, Springer Science+Business Media p. 1255-1288.
21. Schoenberg, M., P. Mars, and A. Lerner, *Neuroanatomy Primer: Structure and Function of the Human Nervous System*, in *The Little Black Book of Neuropsychology: A Syndrome-Based Approach* 2011, Springer; 2011 edition. p. 59-126.
22. D'Esposito, M., *From cognitive to neural models of working memory*. Philosophical Transactions of the Royal Society, 2007. 362: p. 761-772.
23. Ford, J., K. Giovanello, and K. Guskiewicz, *Episodic Memory in Former Professional Football Players with a History of Concussion:*

An Event-Related Functional Neuroimaging Study. Journal of Neurotrauma, 2013. 30: p. 1683-1701.
24. Baugh, C., et al., *Chronic traumatic encephalopathy: neurodegeneration following repetitive concussive and subconcussive brain trauma*. Brain Imaging and Behavior, 2012. 6: p. 244-254.
25. Freeman, W., *The limbic action-perception cycle controlling goal-directed animal behavior*, 2002: Department of Molecular and Cell Biology University of California. p. 2249-2254.
26. Zhu, Y., et al., *Loss of Microstructural Integrity in the Limbic-Subcortical Networks for Acute Symptomatic Traumatic Brain Injury*. BioMed Research International, 2014. 2014: p. 1-7.
27. Chen, J.-K., et al., *Neural Substrates of Symptoms of Depression Following Concussion in Male Athletes With Persisting Postconcussion Symptoms*. Arch Gen Psychiatry, 2008. 65(1): p. 81-89.
28. Signoretti, S., et al., *The Pathophysiology of Concussion*. The American Academy of Physical Medicine and Rehabilitation, 2011. 3(10S2): p. S359-S368.
29. Blennow, K., J. Hardy, and H. Zetterberg, *The Neuropathology and Neurobiology of Traumatic Brain Injury*. Neuron, 2012. 76: p. 886-899.
30. Iverson, G., *Sport-Related Concussion*, in *The Little Black Book of Neuropsychology: A Syndrome-Based Approach*. 2011, Springer Science+Business Media. p. 721-744.
31. Gaetz, M., *The neurophysiology of brain injury*. Clinical Neurophysiology, 2004. 115: p. 4-18.
32. Pal, D., et al., *Diffusion tensor tractography indices in patients with frontal lobe injury and its correlation with neuropsychological tests*. Clinical Neurology and Neurosurgery, 2012. 114: p. 564-571.
33. Gajawelli, N., et al., *Neuroimaging Changes in the Brain in Contact versus Noncontact Sport Athletes Using Diffusion Tensor Imaging*. World Neurosurgery, 2013. 80(6): p. 824-828.
34. Correia, S. and A. Gongvatana, *Diffusion-Tensor Imaging and Behavioral Medicine*, in *Brain Imaging in Behavioral Medicine and Clinical Neuroscience*, S.S.B. Media, Editor. 2011, LLC: Providence, RI, USA. p. 49-66.
35. Alexander, A., et al., *Diffusion Tensor Imaging of the Brain*. National Institute of Health, 2007. 4(3): p. 316-329.
36. Pellman, E., et al., *Concussion in professional football: reconstruction of game impacts and injuries*. Neurosurgery, 2003. 53: p. 799-814.

37. Barth, J., et al., *Acceleration-Deceleration Sport-Related Concussion: The Gravity of It All*. Journal of Athletic Training 2001. 36(3): p. 253-256.
38. Pardini, J., et al., *Postconcussive symptoms are associated with compensatory cortical recruitment during a working memory task*. Neurosurgery, 2010. 67(4): p. 1020-1028.
39. Mihalik, J., et al., *Measurment of head impacts in collegiate football players: An investigation of positional and event-type differences*. Neurosurgery, 2007. 61(6): p. 1229-1235.
40. Schnebel, B., et al., *In Vivo study of head impacts in football: A comparison of national collegiate athletic association division I versus high school players*. Neurosurgery, 2007. 60: p. 490-496.
41. Crisco, J., et al., *Frequency and Location of Head Impact Exposures in Individual Collegiate Football Players*. Journal of Athletic Training, 2010. 45(6): p. 549-559.
42. Crisco, J., et al., *Magnitude of Head Impact Exposures in Individual Collegiate Football Players*. Journal of Applied Biomechanics, 2012. 28: p. 174-183.
43. Lovell, M., et al., *Functional brain abnormalities are related to clinical recovert and time to return-to-play in athletes*. Neurosurgery, 2007. 61(2): p. 352-360.
44. Toledo, E., et al., *The young brain and concussion: Imaging as a biomarker for diagnosis and prognosis*. Neuroscience and Biobehavioral Reviews, 2012. 36: p. 1510-1531.
45. Umile, E., et al., *Dynamic imaging in mild traumatic brain injury: support for the theory of medial temportal vulnerability*. . Arch. Phys. Med. Rehabil., 2002. 83: p. 1506-1513.
46. Junque, C., *Neuropsychological sequelae of head injury*. Rev. Neurol., 1999. 28: p. 423-429.
47. Behan, L.A., et al., *Neuroendocrine disorders after traumatic brain injury*. J. Neurol. Neurosurg. Psychiatry, 2008. 79: p. 753-759.
48. Baguley, I., et al., *The incidence of dysautonomia and its relationship with autonomic arousal following traumatic brain injury*. . Brain Inj., 2007. 21: p. 1175-1181.
49. Ouellet, M.C., J. Savard, and C.M. Morin, *Insomnia following traumatic brain injury: a review*. . Neurorehabil. Neural Repair, 2004. 18: p. 187-198.
50. De Anti, A., G. Gasperini, and M. Rossini, *Paroxysmal episodic hypothalamic instability with hypothermia after traumatic brain injury*. . Brain Inj. , 2005. 19: p. 1277-1283.
51. Elliott, M.L. and L.S. Biever, *Head injury and sexual dysfunction*. . Brain Inj. , 1996. 10: p. 703–717.
52. Geurts, A.C., J.A. Knoop, and J. Van Limbeek, *Is postural control associated with mental functioning in the persistent postconcussion syndrome?* Arch. Phys. Med. Rehabil., 1999. 80: p. 144-149.
53. Macpherson, P., et al., *The significance of traumatic haematoma in the region of the basal ganglia*. . J. Neurol. Neurosurg. Psychiatry, 1896. 49: p. 29-34.
54. Bazarian, J., et al., *Persistent, long-term cerebral white matter changes after sport-related repetitive head impacts*. PLOS ONE, 2014. 9(4): p. 1-12.
55. Hamberger, A., et al., *Redistribution of neurofilaments and accumulation of beta-amyloid protein after brain injury by rotational accel- eration of the head*. J Neurotrauma 2003. 20: p. 169-178.
56. Poirer, J., *Apolipoprotein E in animal models of CNS injury and in Alzheimer's disease*. . Trends Neurosci. , 1994. 17: p. 525-530.

57. Roberts, G., et al., *Beta amyloid protein deposition in the brain after severe head injury: implications for the pathogenesis of Alzheimer's disease*. J Neurol Neurosurg Psychiatr., 1994. 57: p. 419-425.
58. Nicoll, J., G. Roberts, and D. Graham, *Apolipoprotein E epsilon4 allele is associated with deposition of amyloid Beta-protein following head injury*. Nat Med., 1995. 1: p. 135-137.
59. Park, D. and P. Reuter-Lorenz, *The Adaptive Brain: Aging and Neurocognitive Scaffolding*. Annu Rev Psychol., 2009. 60: p. 173-196.
60. Hart, J., et al., *Neuroimaging of Cognitive Dysfunction and Depression in Aging Retired National Football League Players*. Journal of American Medical Association, 2013. 70(3): p. 326-335.
61. Omalu, B., et al., *Chronic traumatic encephalopathy in a National Football League player*. Neurosurgery, 2005. 57(1): p. 128-134.
62. Collins, M., et al., *Relationship between concussion and neuropsychological performance in college football players*. Journal of American Medical Association, 1999. 282(10): p. 964-970.
63. Dettwiler, A., et al., *Persistent Differences in Patterns of Brain Activation after Sports-Related Concussion: A Longitudinal Functional Magnetic Resonance Imaging Study*. Journal of Neurotrauma, 2014. 31: p. 180-188.
64. Ellemberg, D., et al., *Advances in Sport Concussion Assessment: From Behavioral to Brain Imaging Measures*. Journal of Neurotrauma, 2009. 26: p. 2365-2382.
65. Owen, A., et al., *N-Back Working Memory Paradigm: A Meta-Analysis of Normative Functional Neuroimaging Studies*. Human Brain Mapping, 2005. 25: p. 46-59.
66. Cohen, R. and L. Sweet, *Brain Imaging in Behavioral Medicine and Clinical Neuroscience: Synthesis*, in *Brain Imaging in Behavioral Medicine and Clinical Neuroscience*, S.S.B. Media, Editor. 2011, LLC: School of Brown University, Providence. p. 383-393.
67. Bullmore, E. and O. Sporns, *Complex brain networks: graph theoretical analysis of structural and functional systems*. Nature, 2009. 10.
68. Hampshire, A., A. MacDonald, and A. Owen, *Hypoconnectivity and Hyperfrontality in Retired American Football Players*. Nature, 2013. 3(2972): p. 1-8.
69. Shenton, M., et al., *A Review of Magnetic Resonance Imaging and Diffusion Tensor Imaging Findings in Mild Traumatic Brain Injury*. National Institute of Health, 2012. 6(2): p. 137-192.
70. Brun, C., et al., *Acute and chronic changes in diffusivity measures after sports concussion*. Journal of Neurotrauma, 2011. 28(10): p. 2049-2059.
71. Cubon, V., et al., *A Diffusion Tensor Imaging Study on the White Matter Skeleton in Individuals with Sports-Related Concussion*. Journal of Neurotrauma, 2011. 28: p. 189-201.
72. Bazarian, J., et al., *Subject-Specific Changes in Brain White Matter on Diffusion Tensor Imaging After Sports-Related Concussion*. National Institute of Health, 2011. 30(2): p. 171-180.
73. Mori, S., et al., *Three-dimensional tracking of axonal projections in the brain by magnetic resonance imaging*. . Ann Neurol 1999. 45: p. 265–269.
74. Zhang, J., M. Aggarwal, and S. Mori, *Structural insights into the rodent CNS via diffusion tensor imaging*. Trends in Neurosciences, 2012. 35(7): p. 412-421.

75. Bazarian, J., et al., *Diffusion tensor imaging detects clinically important axonal damage after mild traumatic brain injury: A pilot study*. Journal of Neurotrauma, 2007. 24: p. 1447-1459.
76. Mayer, A., et al., *A prospective diffusion tensor imaging study in mild traumatic brain injury*. Neurology, 2010. 74: p. 643-650.
77. Arfanakis, K., et al., *Mapping functionally related regions of brain with functional connectivity MR imaging*. American Journal of Neuroradiology , 2002. 21(9): p. 1636-1644.
78. Inglese, M., et al., *Diffuse axonal injury in mild traumatic brain injury: a diffusion tensor imaging study* . Journal of Neurosurgery, 2012. 116(6): p. 298-303.
79. Niogi, S., et al., *Extent of microstructural white matter injury in postconcussive syndrome correlates with impaired cognitive reaction time: A 3T diffusion tensor imaging study of mild traumatic brain injury*. Journal of Neuroradiology, 2008. 29: p. 967-973.
80. Lipton, M., et al., *Multifocal white matter ultrastructural abnormalities in mild traumatic brain injury with cognitive disability: A voxel-wise analysis of dif- fusion tensor imaging*. Journal of Neurotrauma, 2008. 25: p. 1335-1342.
81. Smith, S.M., et al., *Tract-based spatial statistics: Voxelwise analysis of multi-subject diffusion data*. NeuroImage, 2006. 31: p. 1487-1505.
82. Jenkinson, M., et al., *Improved optimization for the robust and accurate linear registration and motion correction of brain images*. NeuroImage, 2002. 17(2): p. 825-841.
83. Gong, G., et al., *Mapping Anatomical Connectivity Patterns of Human Cerebral Cortex Using In Vivo Diffusion Tensor Imaging Tractography*. Cerebral Cortex, 2009. 19: p. 524-536.
84. Tang, C., et al., *Diffuse Disconnectivity in tBi: a resting state fMri anD Dti stuDy*. National Institute of Health, 2012. 3(1): p. 9-14.
85. Harrison, R., et al., *Blood capillary distribution correlates with hemodynamic-based functional imaging in cerebral cortex*. Cerebral Cortex, 2002. 12: p. 225-233.
86. Slobounov, S., et al., *Concussion in athletics: ongoing clinical and brain imaging research controversies*. Brain Imaging and Behavior, 2012. 6: p. 224-243.
87. Donders, J., *Memory functioning after traumatic brain injuryin children*. Brain Injury, 1993. 7(5): p. 431-437.
88. Wells, F.L., *Normal performance in the tapping test: before and during practice, with special reference to fatigue phenomenon*. The American Journal of Psychology, 1908. 19(4): p. 437-483.
89. Hubel, K., et al., *COMPUTERIZED MEASURES OF FINGER TAPPING: EFFECTS OF HAND DOMINANCE, AGE, AND SEX*. Perceptual & Motor Skills: Motor Skills & Ergonomics, 2013. 116(3): p. 1-24.
90. Davatzikos, C., et al., *Whole-brain morphometric study of schizophrenia revealing a spatially complex set of focal abnormalities*. Arch Gen Psychiatry, 2005. 62(11): p. 1218-27.
91. Fan, Y., et al., *Diagnosis of brain abnormality using both structural and functional MR images*. Conf Proc IEEE Eng Med Biol Soc, 2006. Suppl: p. 6585-8.
92. Smith, S.M., et al., *Advances in functional and structural MR image analysis and implementation as FSL*. Neuroimage, 2004. 23 Suppl 1: p. S208-19.

93. Smith, S.M., *Fast robust automated brain extraction*. Hum Brain Mapp, 2002. 17(3): p. 143-55.
94. Horsfield, M., *Mapping eddy current induced field for the correction of diffusion weighted echo planar images*. . Magnetic Resonance Imaging, 1999. 17: p. 1335-1345.
95. Jenkinson, M. and S. Smith, *A global optimisation method for robust affine registration of brain images*. Medical Image Analysis, 2001. 5: p. 143-156.
96. Smith, S., *Fast robust automated brain extraction*. Human Brain Mapping., 2002. 17(3): p. 143-155.
97. Andersson, J., M. Jenkinson, and S. Smith, *Non-linear registration, aka spatial normalisation*. FMRIB Technial Report TR07JA2, 2010.
98. Jenkinson, M., *ANOVA: 3-factors 2-levels (3-way between-subjects ANOVA)*, 2014, GLM.
99. Winkler, A.M., et al., *Permutation inference for the general linear model*. NeuroImage, 2014. 92: p. 381-397.
100. Behrens, T., et al. *Randomise*. 2004-2014 [cited 2014 10/14]; Available from: http://fsl.fmrib.ox.ac.uk/fsl/fslwiki/Randomise.
101. Smith, S. and T. Nichols, *Threshold-free cluster enhancement: addressing problems of smoothing, threshold dependence and localisation in cluster inference*. . NeuroImage, 2009. 44(1): p. 83-98.
102. L, Z., et al., *White matter integrity in mild cognitive impairment: a tract-based spatial statistics study*. NeuroImage, 2010. 53(1): p. 16-25.
103. Mori, S., et al., *MRI Atlas of Human White Matter*., ed. E. Science. 2005, Elsevier, Amsterdam, The Netherlands.
104. Wakana, S., et al., *Reproducibility of quantitative tractography methods applied to cerebral white matter*. NeuroImage, 2007. 36(3): p. 630-644.
105. Hua, K., et al., *Tract probability maps in stereotaxic spaces: analysis of white matter anatomy and tract-specific quantification*. NeuroImage, 2007. 39(1): p. 336-347.
106. Duering, M., et al., *Strategic role of frontal white matter tracts in vascular cognitive impairment: a voxel-based lesion-symptom mapping study in CADASIL*. Brain, 2011. 134: p. 2366-2375.
107. Gilchrist, M., D. O'Donoghue, and T.J. Horgan, *A two-dimensional analysis of the biomechanics of frontal and occipital head impact injuries*. International Journal of Crashworthiness, 2001. 6(2): p. 253-262.
108. Tremblay, S., et al., *Diffuse white matter tract abnormalities in clinically normal ageing retired athletes with a history of sports-related concussions*. Brain, 2014: p. 1-15.
109. Kerr, Z., S. Marshall, and K. Guskiewicz, *Med Sci Sports Exerc*. Medicine and sc, 2012. 44(3): p. 377-382.
110. Kinnunin, K., et al., *White matter damage and cognitive impairment after traumatic brain injury*. Brain, 2010: p. 1-15.
111. Palacios, E., et al., *Diffusion tensor imaging differences relate to memory deficits in diffuse traumatic brain injury*. BMC Neurology, 2011: p. 11-24.