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Functional Connectivity Changes in the Default Mode Network in Moderate-to-Severe Traumatic Brain Injury

Emily J. Haight
The Graduate Center, City University of New York

FUNCTIONAL CONNECTIVITY CHANGES IN THE DEFAULT MODE NETWORK IN MODERATE-TO-SEVERE TRAUMATIC BRAIN INJURY

By

EMILY HAIGHT

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_____	_____
Date	Junghoon Kim
	Thesis Advisor
_____	_____
Date	Valerie Shafer
	Second Reader
_____	_____
Date	Tony Ro
	Program Director

THE CITY UNIVERSITY OF NEW YORK

ABSTRACT

FUNCTIONAL CONNECTIVITY CHANGES IN THE DEFAULT MODE NETWORK IN MODERATE-TO-SEVERE TRAUMATIC BRAIN INJURY

by

EMILY HAIGHT

Advisor: Junghoon J. Kim, Ph.D.

Traumatic brain injury (TBI) patients are known to have altered functional connectivity (FC), which has cognitive and behavioral significance and bears clinical implications. Previous literature has discovered a hyperconnectivity response to TBI, most notably in the default mode network (DMN). However, the exact pattern of changes in resting FC during the first year of recovery is unknown. We used resting-state functional magnetic resonance imaging to investigate longitudinal connectivity patterns in the DMN of 28 moderate-to-severe TBI patients as compared to 33 demographically matched healthy controls (HC). FC was assessed at 3, 6, and 12 months post-injury for patients using the posterior cingulate cortex (PCC) as the seed. The first aim of this study was to investigate the resting state FC response in the DMN in the TBI group compared to HC. The second aim involved the evolution of FC responses throughout the first year of recovery from neurotrauma in the patient group. We found in our cross-sectional analysis that patients showed hyperconnectivity responses in the right middle temporal lobe at 3 months post-injury, and hypoconnectivity in the left lateral occipital lobe at 12 months post-injury. However, when we conducted our longitudinal analysis and compared FC across the time points directly with each other, no areas showed a significant change within the TBI group. Our findings suggest a dynamic nature of FC alteration over the first year of recovery after TBI, by showing a hyperconnectivity in the subacute phase and a hypoconnectivity in posterior brain regions as patients reach a more chronic stage. The pattern of FC changes should be further extrapolated into cognitive and behavioral implications, as well as translated into mechanisms of recovery.

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1. Introduction

Traumatic brain injury (TBI) is a major cause of disability, with incident rates reaching over 200,000 per year in the United States alone (Centers for Disease Control and Prevention, 2020). TBI can affect those in an accident, a fall, a sports injury, or a blow to the head. Those affected require critical and immediate attention, and consequences of injury can be permanent and irreversible. The development of degenerative brain diseases, such as Alzheimer disease (AD) and chronic traumatic encephalopathy (CTE), has been connected to the widespread disruption of neural tissues and neuroinflammation- both of which are consequences of TBI.

Cognitive effects of TBI most often include memory impairment, slowed information processing, and attention deficits. In a meta-analytic review, Mathias and Wheaton (2007) described a range of attentional deficits persistent throughout the TBI population, involving selective and focused attention, attention span, and sustained attention. Beyond cognitive deficits, TBI can result in major behavioral issues, motor impairments, and emotional lability (Castellanos et al., 2011).

Cognitive and behavioral consequences of TBI may be related to the disruption of functional connectivity (FC) networks (Palacios et al., 2013). Functional connectivity describes the physiological relationship between remote brain areas, often measured in blood-oxygenation-level- dependent (BOLD) signal. Highly correlated BOLD signals suggest a strong functional relationship between regions, which can form an entire network of connectivity. Higher order cognitive functioning relies on the interactions between networks (Bullmore & Sporns, 2009) and the disruption to these networks may be responsible for memory and attention deficits after TBI (Sharp et al., 2014). Despite the varying pathologies of TBI, the focal and widespread structural damage resulting from TBI consistently contribute to altered FC with cognitive

implications. The overarching goal of our study is to better understand the pattern of FC changes during the first-year post -injury in patients with moderate-to-severe- TBI. Our efforts are expected to eventually lead to better understanding of the neural mechanisms underlying cognitive impairment and recovery in patients with TBI during the first year post-injury.

1.1. Mechanisms of Disruption of Functional Connectivity in Traumatic Brain Injury

FC disruptions are the result of structural damage from the primary injury. Both focal and diffuse damage can occur at the time of trauma as a direct result of mechanical force. Focal damage is caused on impact as the brain moves within the skull and bounces against the hard skull walls; this results in bruising, bleeding, and tissue loss. The frontal cortex most commonly and severely suffers focal damage in TBI due to the adjacent jagged orbit and sinus cavities. The impact on the orbital bones can result in serious lesions in the frontal lobe. Less damage may occur for the parietal and occipital lobes, which are encased by smoother skull surfaces; however, the impact of a gelatinous brain on one side of the skull will nearly always require the rebounding of the brain on the opposite side of impact, the mechanics of which results in both a coup and countercoup, respectively (Payne et al., 2021).

Focal injuries, including stroke, hematoma, or hemorrhage, cause structural damage to a distinct group of neurons. Focal damage disrupts the functional capabilities of those neurons, which may result in whole-brain consequences (Fujiwara et al., 2008). Gratton et al. (2012) describes focal brain lesions as triggering a widespread impact on the organization of brain networks. This type of injury can be relatively well detected using neuroimaging techniques like CT-scan or conventional structural MRI imaging.

TBI can result in a primary, and nearly universal disruption of brain structure after injury, called diffuse axonal injury (DAI) (Wu et al., 2004). DAI is a widespread disruption of white matter, which consists of long reaching fibers throughout the brain, called axons. Axons are important for transmitting signals to distant brain regions, and injury to these fibers (i.e., DAI) may be a major contributor to deviant patterns of FC in TBI. Traumatic or diffuse axonal injury can include cell death, axon swelling, cytoskeleton breakage, membrane failure, and other effects (Johnson et al., 2013). Diffuse injuries are more difficult to detect than are focal injuries. DAI continues to evolve even beyond the initial insult, the consequences of which have been linked to late neural degenerative disease. Greer et al. (2011) found that axonal shearing can be a catalyst for cascading biological effects, contributing to neural degeneration.

The widespread damage to white matter tracts (i.e., DAI) is believed to be caused by different neural tissue types accelerating and decelerating at varying rates, due to different tissue densities; tissue formations are stretched and sheared under these forces resulting in widespread structural damage. DAI results in diminished functional integrity of axons and can contribute to disrupted FC between brain locations. An example of how DAI effects FC includes damage to the primary signaling route from one center, or node, to another due to axonal shearing. Alternative routes of communication between brain regions may be utilized in order to compensate for this damage, however, secondary routes of communication can negatively affect processing speed and signal strength. Since FC is dependent upon signal strength, the effects of DAI can also be observed beyond structural deficits and also include changes in FC.

Consequences of the structural damage following TBI include the limited capacity to integrate and process information. The loss of gray matter in addition to the damage of white matter tracts limits the resources in the injured brain, which therefore interrupts the efficient flow

of information across the brain (Caeyenberghs et al., 2017). The cascading neural events described as diffuse axonal injury also leads to slowed processing speeds and cognitive exhaust (Wu et al., 2004).

1.2. Functional Connectivity in Traumatic Brain Injury

FC is defined as the correlation of brain activity, measured by BOLD signal changes, between spatially distinct brain regions. Strong FC may exist between two nodes even in the absence of strong structural connection between them (Eickhoff & Müller, 2015). This can be explained by the relay of information from one area to another, thereby driving a functional connection. Functional magnetic resonance imaging (fMRI) techniques can be used to identify which brain regions couple together; this procedure uses synchronicity in brain activity between regions to infer strength of FC between them (Eickhoff & Müller, 2015). The FC literature in TBI has revealed mixed results (Hillary et al., 2015); some research implicates hypoconnectivity in TBI patients, while other studies reported hyperconnectivity. These contradictory findings can be due to differences across studies in TBI severity, time post-injury, and type of FC explored (resting state FC versus task-based FC). However, a majority of the literature has supported hyperconnectivity after TBI as a result of sustained structural damage (Hillary et al., 2015).

Much of the previous FC literature has focused on task-induced changes using fMRI. Task-induced changes involve the use of cognitive, motor, and sensory tasks in both healthy (Biswal et al., 1995) and injured brains (Hampshire et al., 2013). Task-based fMRI identifies brain regions functionally involved with a task.

Task-based FC studies, however, can be limited by design challenges, including the accuracy of control tasks and performance equivalence between groups and sessions. Task-based FC may also focus more on magnitude of signal rather than network communication.

Additionally, targeting specific brain areas through task-based FC may overshadow whole brain consequences of TBI influenced brain changes (Hillary et al., 2011).

Resting state MRI is a meritorious method to investigate FC, considering the shortcomings of task-based fMRI. Resting state fMRI identifies functionally specialized intrinsic networks. Resting state connectivity studies investigate the brain during the absence of a task and offer some advantages over the task-induced fMRI. Due to the nature of the brain at rest, resting brain mechanics can better reveal the FC between brain networks than can task-based FC (Biswal et al., 1995). The resting brain consists of tiny fluctuations of BOLD signal changes which define the resting state FC (rsFC). BOLD signal fluctuations are of very low frequency (~0.1Hz) and reflect the covariance between voxels during the absence of a task (Hillary et al., 2011). Influential findings from Biswal et al. (1995) indicate that the manifestation of FC is dependent on small, temporally correlated- yet spatially distinct- fluctuations in activity. Although the main purpose of Biswal's (1995) study was to investigate the task-influenced changes in healthy brains during a motor task, they also discovered synchronized activity across certain brain during the resting state. Resting state FC studies now extend beyond the healthy brain and have been a major influence in TBI related research.

1.3. Default Mode Network in Traumatic Brain Injury

The Default Mode Network (DMN) has been a major system under investigation in the FC of TBI patients for several reasons. The DMN is a resting state network that is most active when the brain is 'at rest' or without a task, therefore its activation is easy to observe. The DMN is also relatively easy to investigate because of its highly metabolic network of coordinated activity both within and outside of the network (Sharp et al., 2014).

Several hubs within the DMN are densely connected among themselves, including the precuneus, posterior cingulate cortex (PCC), medial and lateral frontal cortices (Hillary & Grafman, 2017). These hubs are considered part of the 'rich-club' because of their highly integrated nature that direct global brain dynamics, as well as the transfer of information (Zamora-Lopez et al., 2010). The strength and dense connectivity of the nodes within the DMN make this network an optimal target for plasticity changes after neurotrauma. Finally, the DMN is comprised of many long coursing projections which are vulnerable to disruption from force trauma (Hayes et al., 2016).

The role of the DMN is complementary to other task-positive or goal directed networks, such as the salience network (SN). The SN is active in detecting salient stimuli, while the DMN is active in a daydream-like state. While one network is active the other is inhibited, and coordination of these kinds of complementary networks contribute to cognitive function efficacy measures (Sharp et al., 2014). Fox et al. (2007) and Kelly et al. (2008) both found support for the interplay between the DMN and task-positive networks' prediction of task performance- an ability disrupted in TBI. The DMN typically deactivates as task difficulty increases in healthy persons. The smooth timewise transition between the DMN and the salience network (SN) is necessary for efficient cognitive processing as exemplified in Bonnelle et al. (2012). The activation of the task-related network requires the deactivation or inhibition of the DMN, and vice versa; the DMN is active upon the inhibition of the salience network.

Abnormal activation of this transition mechanism has been associated with poorer cognitive function in both healthy and TBI individuals (Bonnelle et al., 2012). Abnormal functioning of this complementary system results in the slowed and deficient transition of networks, quantified by poorer cognitive processing. Likewise, increased modularity of the PCC

is positively correlated with cognitive function in TBI patients (Venkatesan et al., 2015). The disruption in transition between the DMN and the SN is reflected in inefficient processing and can result from DAI found in TBI.

The ventromedial prefrontal cortex (vmPFC) and the posterior cingulate cortex (PCC) are the core nodes of the DMN (Sharp et al., 2014), which interact with other network regions along the midline, lateral parietal cortex, and medial temporal lobes (Buckner et al., 2008). The regions within the DMN are also highly interconnected to distributed brain regions. In particular, the PCC hub is a densely connected node; Venkatesan et al. (2015) stated that its unique connectivity profile "may provide insight into the neural mechanisms underlying cognitive sequelae post injury." Densely interconnected hubs, such as the vmPFC and PCC within the DMN, form part of the brain's core structural network (Hagmann et al., 2008) and are hypothesized to have important cognitive functions (Buckner et al., 2008).

TBI may damage the DMN, which plays an important role in cognitive functions such as memory and attention. Damage to the DMN can be measured in terms of FC, and the observed deficits in memory and attention in persons with TBI could be explained by FC changes. Taken together, the cognitive implications of TBI, including memory and attention deficits, may result from altered FC within the DMN. Support for FC changes implicating changes in cognitive functioning is observed in Venkatesan et al., (2015). In this study, improvement in cognitive tasks throughout the first year of neurotrauma recovery coincided with changes in FC states in relation to the PCC (Venkatesan et al., 2015). Relatedly, Hillary and Grafman (2017) reviewed studies in which the magnitude of altered connectivity throughout the brain (not just the DMN) was linked to positive clinical outcome.

1.4. Hyperconnectivity Versus Hypoconnectivity During Resting State in Traumatic Brain Injury

A consensus among rsFC studies is in support of hyperconnectivity as a fundamental response to brain injury (Hillary et al., 2015; Venkatesan & Hillary, 2019; Sharp et al., 2011, Caeyenberghs et al., 2017). FC is a time series correlation between two nodes which describes the functional connectedness between them. Hyperconnectivity is an increase in the correlation between two nodes beyond what is expected in a healthy brain. The inference is that neural networks adjust connectivity patterns as a response to disruption. In the TBI damaged brain, a hyperconnectivity response is meant to compensate for structural shortcomings, with the purpose of maintaining meaningful responses to environmental demands. Hillary et al. (2011) found hyperconnectivity in the intrinsic, or resting state networks, in TBI patients at 3 months post-injury as compared to controls. One possibility for an increase in connectivity patterns after neurotrauma relies on the increased need for detour paths, which require an ongoing recruitment of local neurons (Hillary & Grafman, 2017). Another possibility for hyperconnectivity could be that it serves as a mechanism of enhanced centrality of major network hubs, like the PCC, to operate as a magnet of collateral connections during recovery (Hawellek et al., 2011).

It is likely that hyperconnectivity does not occur in isolation, but rather occurs as a compensatory response to differential loss throughout the network (Hillary et al., 2011). The compensatory response attempts to resolve network damage and resource loss, often resulting in increased path length. Increasing path length is a consequence of navigating around damaged structures to restore connectivity. Hyperconnectivity responses during a time of resource loss may seem counterintuitive, however neuro-recovery mechanisms push the remaining neurons to work harder in this environment to modulate levels of neural function to that of pre-injury activity. This mechanism thereby over-activates the connectivity networks. It is suggested that

this hyperconnectivity response is only available in environments with enough resources to compensate in this manner (Sharp et al., 2011). After a critical threshold of neural resource depletion is met, hyperconnectivity responses are no longer sustainable (Hillary et al., 2015).

The benefit of hyperconnectivity as a response to TBI is the attempt to maintain functioning in a compromised neural environment. A concern of hyperconnectivity as a response to TBI, however, includes the disproportionate shift of metabolic load on the network hubs (Hillary & Grafman, 2017). This shift of functional responsibility to other nodes and pathways may become exhausting and not sustainable long term.

Patterns of hyperconnectivity have most commonly been observed in major connectivity networks such as the DMN, which is most active during a state of rest (Hillary et al., 2015). However, research studies have been mixed in whether they observe hyper- versus hypo-connectivity after neurological disruptions, including TBI, as evident in Hillary's (2014) review. Thus, there is some controversy about hyper- and hypoconnectivity patterns following brain injury. Factors determining the pattern may depend on the network in question, the task or resting state, time post-injury, or the severity of TBI. For example, Hillary et al. (2015) found support for a hyperconnectivity response to brain injury in the DMN, while another study from the same group (Hillary et al., 2011) found evidence for a hypoconnectivity response in an 'external resting state' network following brain injury. Venkatesan et al. (2015) found somewhat conflicting results, which included generalized hyperconnectivity in the DMN after injury, in addition to diminished synchrony between particular nodes. The diminished synchrony between nodes may implicate a hypoconnectivity response in some brain areas. The divergent responses provide evidence of heterogeneity of whole-brain FC changes after TBI. These mixed results

could be indicative of differences in experimental design, the varying severity and phase of TBI, or varied patterns of neural injury in response to TBI.

Factors that may contribute to the hyper- and hypoconnectivity debate include task (e.g., active task versus resting state), severity of injury, and time post-injury. So far it is unclear whether these factors, or some combination of these factors, can definitively predict hyper- or hypoconnectivity in TBI, or whether other recovery mechanisms are also involved in FC patterns.

1.5. Longitudinal Resting State Functional Connectivity in Traumatic Brain Injury

There are only a few longitudinal studies focusing on rsFC in TBI, so the point at which FC profiles stabilize after injury is not yet known. These few studies have shown increased FC in target networks within the first 6 months post-injury but suggest a decrease in connectivity over a longer period of time. A major contribution to rsFC research in TBI is the research conducted by Frank Hillary and colleagues. Specifically, Hillary et al. (2011) investigated the FC in moderate-to-severe TBI during resting state across two time points, 3 and 6 months post-injury. They found increased FC in the DMN of TBI patients as compared to healthy controls (HC) during resting state. They also found increased connectivity from the two key seed regions, the medial prefrontal cortex (mPFC) and the posterior cingulate cortex (PCC) to other regions (i.e., hippocampus and insula), within the TBI group at 6 months post injury as compared to 3 months post-injury. Venkatesan et al. (2015) attempted to investigate stability of FC in TBI as compared to HC by studying FC changes over time, however the longitudinal study was of small sample size (n=13) and participants' time post-injury was rather heterogenous (0.5 years-2.42 years post-injury for chronic phase). Their investigation of connectivity profiles in TBI showed hyperconnectivity between the PCC and the dorsomedial prefrontal cortex in the subacute (3-

month) minus chronic phase TBI analysis, and hypoconnectivity between the PCC and the frontoparietal operculum and inferior parietal lobule in the chronic minus subacute phase TBI analysis. In their cross-sectional analysis of TBI patients compared to HC, hyperconnectivity patterns were revealed between the PCC and the anterior DMN. This pattern of hyperconnectivity was reported in the subacute phase, which gradually reduced in connectivity strength over time. We propose to study the longitudinal and cross-sectional pattern of FC in TBI patients compared to HC, similar to Venkatesan et al. (2015). However, we will employ a more homogenous cohort and include more frequent time points post-injury.

1.6. Current Study

The progression of FC changes following TBI is still unknown, and the lack of this critical knowledge limits understanding of the neurocognitive changes post-TBI. In this study, we examine the chronology of FC in the DMN of TBI patients at 3, 6, and 12 months after injury as compared to HC. Longitudinal data will provide evidence for the time course of recovery from brain injury in moderate-to-severe TBI.

A unique aspect about this study includes the three time points of data, as well as the homogeneity in time post-injury across all participants (see Table 1). An important outcome of this study encompasses the progression of FC within the first year after injury. Our results will extend the findings of Venkatesan et al. (2015) in which the progression of FC changes from the acute to chronic phase of moderate-to-severe TBI was investigated. The chronology of data is critical for understanding within-subject temporal dynamics. These results will provide implications for neuro-recovery. This study aims to compare DMN FC between TBI patients and HC and attempt to corroborate hyperconnectivity findings in patients who sustained diffuse TBI.

Our first specific aim is to conduct cross-sectional comparisons of FC between HC and patients with TBI at 3-, 6-, and 12-month post-injury. Based on the literature, we predict hyperconnectivity responses in the DMN of the TBI patient group at 3, 6, and 12 months post-injury compared to HC. Our second aim is to determine the change in magnitude of FC in the DMN of the TBI group over time. We predict the magnitude of hyperconnectivity to reduce over time due to neuro-recovery mechanisms.

This study will provide a strong foundation for future studies involving TBI and FC evolution over time. A long-term goal of this study is to translate the understanding of FC changes in the DMN during the critical recovery phase into better treatment options and courses of neurocognitive therapy for those affected by brain injury.

2. Methods

2.1. Participants

This study is part of a larger longitudinal multi-modal neuroimaging study that enrolled 42 moderate-to-severe TBI patients between 18-64 years of age, and 35 demographically-matched HC (Table 2). Patients were classified as moderate-to-severe non-penetrating TBI, as determined by (1) Glasgow Coma Scale (GCS) <13, (2) a loss of consciousness (LOC) ≥ 12 hours, or (3) post traumatic amnesia (PTA) ≥ 24 hours. The GCS score is determined from the Emergency Department and is not due to sedation, paralysis, or intoxication. Patients were recruited from the inpatient rehabilitation unit on a medical campus. Controls were recruited as significant others, friends, or relatives of the patients. Data was collected from patients at 3 months, 6 months, and 12 months post-injury, with relatively little variation in this time point (Table 1). If patients did not participate in all three session they were excluded from this study,

removing 13 patients in total. Other exclusion criteria include psychiatric illnesses like schizophrenia or bipolar disorder, history of substance abuse, history of previous TBI, central nervous system disease, or seizure disorder. Exclusion criteria also include pregnancy, non-fluency in English, magnetic implants, claustrophobia, or other reasoning which renders the patient incapable to complete all three fMRI sessions. The controls were enrolled based on the same exclusion criteria. In addition to exclusion criteria, we also removed subjects due to excessive motion in the scanner. Excessive motion was determined by outlier cleaning method, Artifact Detection Technique (ART), which identifies scans past a specific threshold of motion and global signal change. We chose to set ART scrubbing threshold to 0.9 mm beyond the mean for motion parameters and 5 standard deviations beyond the mean for global BOLD signal (GBS) change parameters. This flagged 2 controls and 2 patients with less than 80% valid scans (defined as scans that were within the criteria for motion and GBS), who were then removed from the study. Figure 1 exemplifies ART outlier detection results. All subjects provided written informed consent and were compensated for their time.

Table 1. Average time in days post-injury in TBI patients (N=28).

	First scan (days)	Second scan (days)	Third scan (days)
Mean	101	183.86	367.25
Standard Deviation	19.46	16.19	21.81

Table 2. Patient demographics after removing outliers. M= mean, SD= standard deviation. Age and education reported in years. ED = Emergency Department.

	Control (N=33)	TBI (N=28)
Age (M/SD)	37.72 / 9.95	32.75 / 13.84
Gender (Male/Female)	25 / 8	19 / 9
Education (M/SD)	13.25 / 2.17	13.57 / 2.51
GCS on admission to ED (M/SD)	-	5.798 / 1.589

2.2. Data Collection

All participants underwent the same testing procedure at every session. The testing session involved an anatomical scan and a five-minute functional MRI recording, in which the participants were instructed to keep their eyes open and remain awake for the entire procedure, as well as minimize any movement while in the scanner.

A 3T scanner was used to collect three-dimensional (3D), T1 weighted, anatomical images of an MPRAGE (magnetic prepared rapid acquisition gradient echo) sequence at a spatial resolution of 1x1x1mm voxels. Echo planar imaging (EPI) was used for resting state functional 4D image acquisition, collected with a spatial resolution of 3x3x3 mm voxels, flip angle of 90 degrees, and a repetition time (TR) of 3 seconds over the course of 5 minutes for a total of 100 collected functional images. Scans were acquired for HC at one time, while scans for TBI patients were acquired at three different time points (3, 6, and 12 months post-injury). In sum, for each session each subject provided a T1-weighted structural image and a functional time series

using BOLD signal change. The images were preprocessed using SPM12 and analyzed using CONN Toolbox (19b) (Whitfield-Gabrieli & Nieto-Castanon, 2012).

2.3. Data Analysis

After removing subjects due to exclusion criteria and outliers, a total of 33 controls and 28 moderate-to-severe TBI patients remain. This group provides 117 sessions in total. All images were loaded into Matlab (19b) using CONN Toolbox (19c) batch script. All sessions were preprocessed using CONN Toolbox's volume-based analyses for direct normalization to MNI (Montreal Neurological Institute template) space.

We utilized CONN's default MNI-space pipeline for our preprocessing steps. The volume-based pipeline has five major processing steps. First, images are realigned and un-warped to correct for subject motion. This step uses SPM12 and each subject's own reference image. The reference image is the first scan of each subjects' first session, and the original scans are co-registered and resampled to that image. This step creates realigned functional files as well as estimated motion parameters for each subject, which are labeled as a first level covariate.

The second step of the pipeline translates these newly realigned files to a functional center coordinate (0,0,0 mm). After translation, the images undergo slice timing correction (STC). STC corrects for the temporal misalignment, or the inter-slice differences in acquisition time (TA). This step takes the images from the center timepoint of each slice, thereby time-shifting the data and resampling the data using SPM12 STC procedure. We use interleaved (Siemens) slice order and keep resolution and smoothing prompts at defaults.

The next step is outlier identification using Artifact Detection Tools (ART). This procedure flags BOLD signal beyond 5 standard deviation from the mean, as well as framewise

displacement above 0.9 mm from the mean. The framewise displacement is computed at each timepoint using six control points around each side of the brain and estimates the largest displacement among them. The GBS change is also computed at each timepoint, and this transformation is the change in average BOLD signal as applied by SPMs global-mean mask scaled to standard deviation units. This entire process of identifying outliers in head motion parameters as well as GBS can be reduced to scrubbing, or cleaning, the data. After scrubbing, a new reference image is created as an average across all scans excluding their outlier scans. The functional data, however, keeps all scans in the timeseries. The outlier scans in the functional data can then be removed later on, depending on the chosen outlier parameters. We chose to remove subjects with less than 80% valid scans after cleaning, which is comparable to other FC studies. Using 80% required validity parameters, we removed two patients from the data for a total of six sessions total removed due to outliers.

The next step in the pipeline is to segment tissue class and normalize the MNI space. These tasks are done simultaneously through SPM12. The tissue is classified into white matter, gray matter, and cerebrospinal fluid (CSF) using tissue probability maps (TPMs). TPMs classify the image based on the intensity values of the reference image, and the intensity estimation registers the images into these separate segments. The anatomical images are normalized using the raw T1 weighted image as a reference and the functional images are normalized using the mean BOLD signal as a reference. Both sets of images are resampled using 4^{th} order spline interpolation into 1 mm voxels for the anatomical data and 2 mm voxels for the functional data. The newly segmented and normalized functional data is centered to structural coordinates (0,0,0 mm), and this segmentation and normalization step is repeated for structural volumes. Lastly, the data are skull stripped to remove the image of the skull in all sets of images.

The final step in the preprocessing pipeline spatially smooths the images. To reduce the influence of varying anatomies across subjects, a Gaussian kernel of 8 mm is implemented to increase the BOLD signal-to-noise ratio.

Using the CONN graphical user interface (GUI), the denoising step minimizes physiological and other sources of noise. CONN's denoising pipeline includes linear regression of potential confounding factors such as respiration, heartrate, and electrical interferences from the environment, and also includes a temporal band-pass filtering. The linear regression step identifies confounding effects to the estimated BOLD signal which are removed at each voxel for each subject and session. This step reduces inter-subject confounding effects by implementing an anatomical component-based noise correction procedure (aCompCor), as well as reduces additional noise from magnetization of the scanner. The temporal band-pass filtering stage minimizes the influence of physiological noise by filtering out data above 0.09Hz and below 0.008Hz.

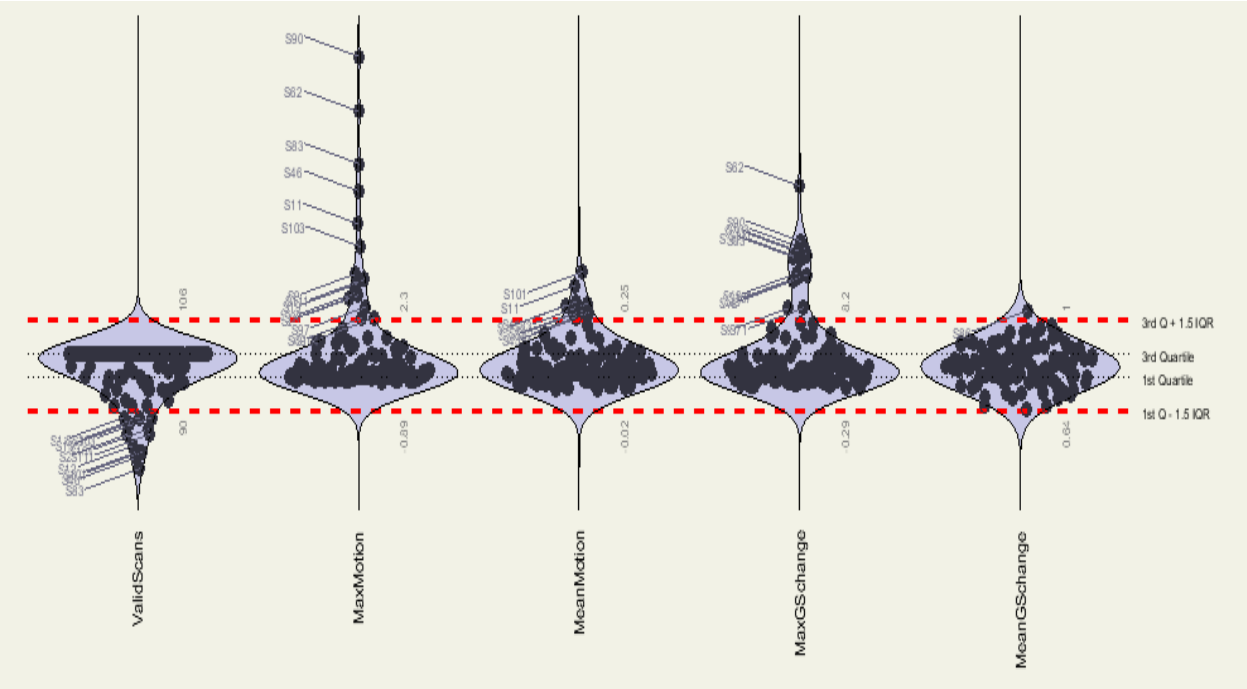

Figure 1. Outlier detection summary, including motion and global BOLD signal change for every subject at every session. Valid scans include the total scans for each subject that fall within the predetermined outlier detection parameters. Voxel movement beyond 0.9 mm of the mean or BOLD signal change within 5 standard deviation of the mean flag the scan as invalid and is removed from subsequent data analysis. Subjects with more than 80% of their scans removed are eliminated from the study. This image reflects only the patients included in the study (after outliers greater than 80% have already been removed). ValidScans = number of valid scans, MaxMotion = magnitude of maximum motion, MeanMotion = mean of motion parameters, MaxGSchange = magnitude of maximum BOLD global signal change, MeanGSchange = mean of BOLD global signal change.

2.3.1. First-Level Within-Group Analysis

We performed seed-to-voxel analysis for the DMN. The seed-to-voxel analysis computes the mean time series for our chosen seed (PCC) and its correlation voxels within the DMN. We chose the PCC as the seed because it is nearly universally accepted as the central node in the DMN (Hillary et al., 2011) and is consistently the reference node in previous resting state fMRI literature (Venkatesan et al., 2015). We implemented a general linear model (GLM) in FC, with no weighting. We did not weight the model because we were investigating resting state. We

used bivariate correlation (Pearson's *r*) to correlate the blood-oxygen-level-dependent (BOLD) relationship between the seed to voxels. The BOLD signal changes measure FC. Correlation values are Fisher (*z*)-transformed to normalize their distribution. The first-level analysis reveals the FC for each subject and identifies the DMN in all groups. Figure 2 confirms the DMN is the network under investigation for both control and TBI groups. These maps were created using SPM software in the CONN results explorer (REX).

2.3.2. Second-Level Between-Group and Longitudinal Analysis

We use seed-based connectivity measures for our second-level analysis of group level differences. This analysis utilizes Fischer-*z* transformations to produce FC maps. A seed-based connectivity map is generated using Fisher-transformed bivariate correlation, which visualizes the correlation from the PCC as to all other brain voxels. This computation correlates the BOLD signal at the reference seed with every individual voxels' BOLD time series. Generating seed-based connectivity maps quantifies the homogeneity of BOLD signal- and therefore the FC- between locations. We conducted paired *t*-tests for longitudinal and independent *t*-tests for cross-sectional analyses. Our longitudinal analysis utilized paired *t*-tests between different time points in order to compare the BOLD signal change in the DMN within the patient group. Our cross-sectional analysis contrasted the patient groups to the HC using voxel-wise independent *t*-tests. The independent *t*-tests compared the DMN FC maps of patients at each time post-injury to that of the control group. Regions with statistically significant differences were identified using a family-wise error (FWE) rate at $p < 0.05$, achieved by an individual voxel threshold $T < 0.005$, with arbitrary cluster size $k = 20$ extend threshold. The maps were made created in CONN Toolbox and displayed using the CONN REX GUI. Lastly, significant regions were labeled using the CONN toolbox significant cluster output data.

3. Results

3.1. Identification of Default Mode Network in Healthy Controls and Patients with Traumatic Brain Injury

Our within group FC maps (as computed by Fisher *z*-transformation) indicate voxel correlations between the PCC (as the seed region) and the DMN. This relationship reveals PCC-DMN connectivity networks for both the HC and TBI groups. See maps in Figure 2. This validates our approach to investigate DMN-related connectivity changes.

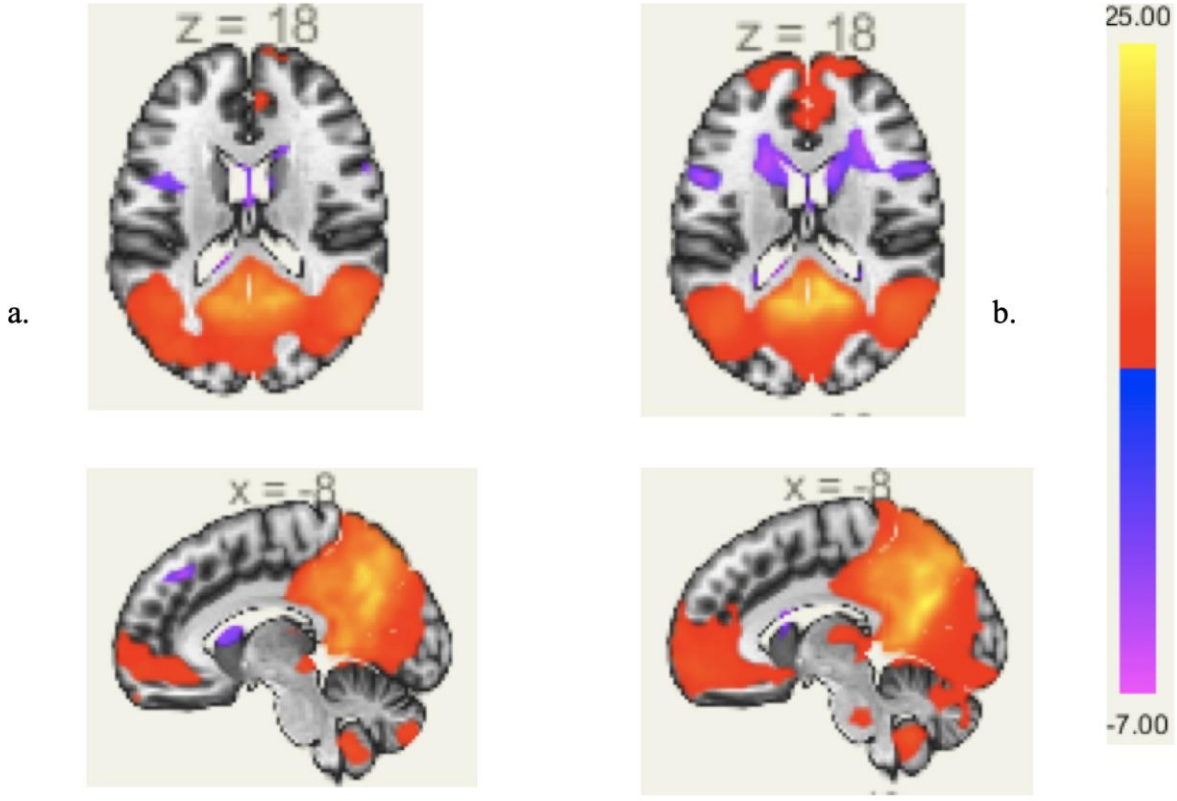

Figure. 2: Axial and sagittal views of whole brain FC with PCC seed in HC (a) and TBI (average across 3, 6, and 12 months post-injury) (b), shown at uncorrected $p < 0.005$ for display purpose. The color bar represents Z-scores of BOLD signal change within the controls (a) and patients (b) Warm colors indicate increased connectivity and cool colors indicate decreased connectivity.

3.2. Cross-Sectional Group Comparison of Default Mode Network Connectivity

Our cross-sectional analysis contrasted the TBI group to the HC group using voxel-wise independent *t*-tests. Cross-sectional data analysis revealed a hyperconnectivity response in the 3 months post-TBI subject group compared to HC (Table 3). The 3 months post-injury TBI group showed evidence for increased positive connectivity, or hyperconnectivity, to the right middle temporal lobe (MTL) (p=0.017) (Figure 3 & 4).

No clusters survived when comparing TBI patients at 6 months post-injury to HC. When comparing TBI patients at 12 months post-injury to HC, we found decreased connectivity in left lateral occipital cortex (left-LOC) (Table 3). The 12 months post-injury TBI group showed evidence for decreased positive connectivity to the left-LOC, or hypoconnectivity (p=0.0006) (Figure 5 & 6)

Table 3. Cross-sectional differences in PCC connectivity. TBI= traumatic brain injury, HC= healthy control

	Location	*p*- FDR corrected	Cluster size (vox)	Peak (MNI coordinate)
Positive connectivity 3months post TBI -HC	Right middle temporal cortex posterior division anterior division	0.017	548	60, -12, -24 46, -14, -30 52, -8, -28
Positive connectivity HC- 12month post TBI	Left lateral occipital cortex Inferior division Superior division	0.000	1051	-28, -88, 24 -30, -78, 10 -48, -70, 10

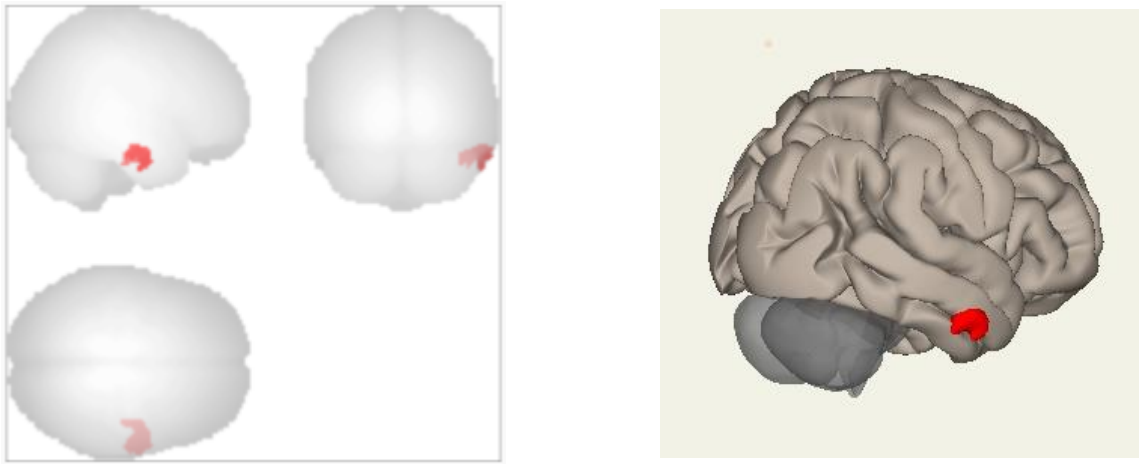

Figure. 3: The right middle temporal gyrus identified by comparing FC with the PCC in TBI patients 3 months post-injury minus HC (p<0.05, k=20). Left: glass brain showing increased activity in the right MTL at 3 months post-injury minus controls. Right: brain surface showing increased activity in the right MTL at 3 months post-injury minus controls.

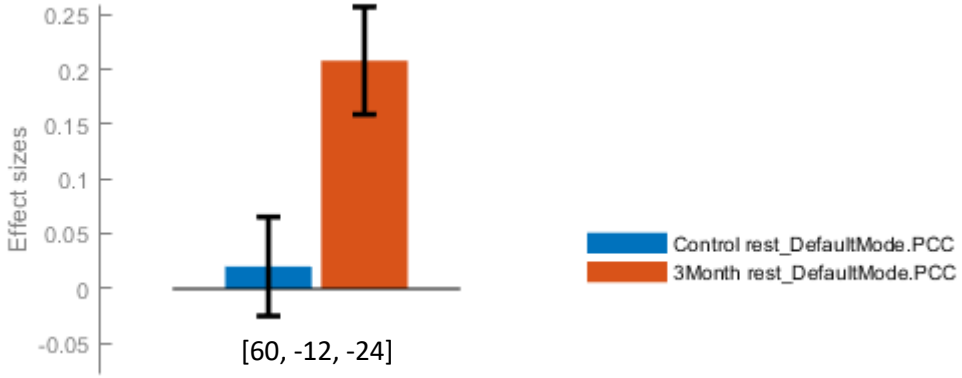

Figure. 4 Fisher z-transformed correlation coefficient (effect size) in peak voxel of the right medial temporal lobe [60, -12, -24] in the TBI group at 3 months post-injury (red) and HC (blue).

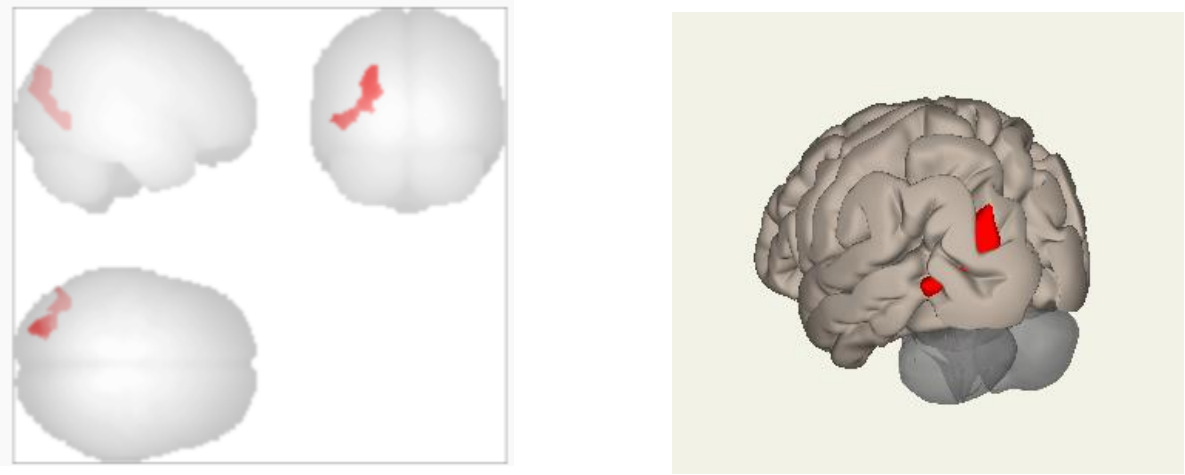

Figure. 5: The left lateral occipital lobe identified by comparing FC with the PCC in TBI patients at 12 months post-injury and HC (p<0.05, k=20). Left: glass brain image showing increased activity in the left-LOC in HC minus TBI at 12 months post-injury. Right: brain surface image showing increased activity in the left-LOC in HC minus TBI at 12 months post-injury.

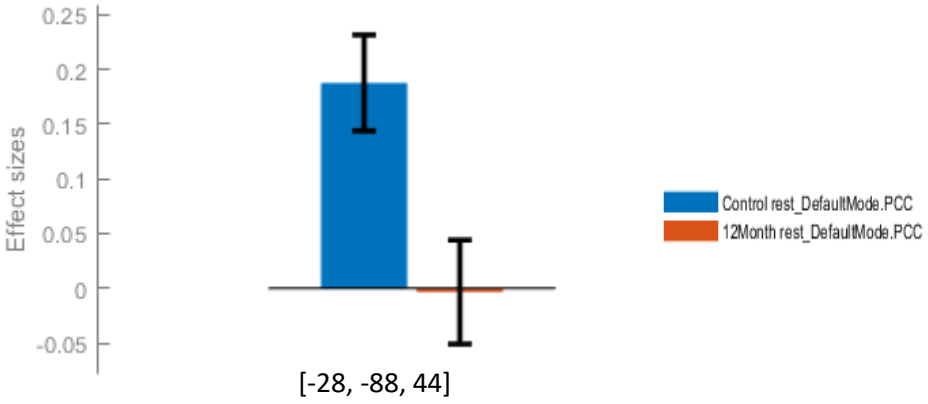

Figure. 6 Fisher *z*-transformed correlation coefficient (effect size) in peak voxel of the left lateral occipital lobe [-28, -88, 44] in the TBI group at 12 months post-injury (red) and HC (blue).

3.3. Longitudinal Changes of Default Mode Network in Patients with Traumatic Brain Injury

No clusters survived from paired *t*-tests comparing different time point within the TBI groups, that is, between 3 and 6 months post-injury, 6 and 12 months post-injury, and 3 and 12 months post-injury. In an exploratory analysis, more lenient parameters uncovered hyperconnectivity in the right MTL at 3 months post-injury as compared to the 6- and 12 months post-injury. Results for the exploratory analysis using relaxed parameters (*p*-value=0.05 and arbitrary clusters size k=5, using paired *t*-tests) can be found in Supplementary Materials.

4. Discussion

We chose to use the PCC because of its prominence as an integral node of the DMN, and also because network hubs have been implicated to be common sites vulnerable to pathology (e.g., Nicolo et al., 2015). The maps created during first-level analysis indeed demonstrated the integral role of PCC through the high correlation of activity at the PCC to mPFC and other regions in the DMN including the precuneus, bilateral angular gyri, lateral occipital lobes, frontal poles, middle temporal gyrus, and cuneate cortex. This pattern of FC between the PCC and DMN nodes is present in both the HC and TBI groups, which confirms the DMN is in fact the network under investigation in this study.

4.1. Group Differences in Default Mode Network Connectivity

The current findings reveal hyperconnectivity within the DMN of TBI patients at 3 months post-injury, and hypoconnectivity within the DMN of TBI patients at 12 months post-injury. Consistent with our hypothesis, cross-sectional group comparison between HC and TBI patients at 3 months post-injury reveals hyperconnectivity between the PCC and other regions of

the DMN in TBI patients. Hyperconnectivity was seen between the PCC and the right MTL in patients with TBI at 3 months post-injury. Other time points did not indicate a significant hyperconnectivity response. Our hypothesis first states that hyperconnectivity will be present in the TBI population, and secondly that hyperconnectivity will decrease as time post-injury increases. This prediction is supported by our results because hyperconnectivity is most robust at 3 months post-injury and is not present (therefore decreases) at later time points post-injury. Finally, hypoconnectivity was not part of our initial predictions, however is in line with our hypothesis indicating decreased FC over time.

4.1.1. Hyperconnectivity at 3 Months Post-Injury

It has been proposed that TBI patients compensate for deficits after injury with increased connectivity in network regions (Hillary & Grafman, 2017). This is evident in our finding that hyperconnectivity is seen in the DMN of patients with TBI at 3 months post-injury compared to controls. The MTL is classified as a node within the DMN network, to which patients' PCC in the subacute phase (3 months post-injury) exhibited increased FC. These results are consistent with findings in Hillary et al., (2011), wherein the TBI group had increased FC at resting state to the right MTL in a seed-to-voxel analysis during resting state.

Hyperconnectivity to the MTL could be evidence for a compensation mechanism in the injured DMN. HC may not need to synchronize the PCC to MTL at such high intensity, but the TBI patients may require the increased connectivity to this region of the DMN in order to maintain healthy levels of functioning within the network. At this stage, our results suggest that the right MTL takes on a larger role in processing within the DMN by increasing its synchronicity to the PCC, a network-guiding node.

Another consideration for hyperconnectivity in the right MTL is as an extension of DMN centrality. The PCC may be dispersing its centrality by increasing its functional connections with the right temporal lobe, perhaps to take some strain off the PCC. Hawellek et al. (2011) described the central nodes as a region to which collateral connections are directed during recovery. This could become exhausting for the PCC to direct and control the network during a state of hyperconnectivity; as a consequence, a shift of the metabolic load to alternative regions within the network is yet another compensation the brain may make to maintain its functional integrity.

Also indicating hyperconnectivity in right hemisphere regions is Venkatesan and Hillary's (2019) research. They found both the left and right PCC showed increased connectivity to regions in the right hemisphere in TBI patients, although this finding was in the frontoparietal control network (FCN) at resting state. Our results of the DMN at resting state also indicated hyperconnectivity responses in the right hemisphere. Taken together, this could suggest an overall increased FC load in the right hemisphere in the TBI population. It should be noted though, that the patients in the Venkatesan and Hillary (2019) study were further into the chronic stage of injury than our cohort of patients, averaging 5.75 years post-injury. Evidence of hyperconnectivity in the right hemisphere of TBI patients at a more chronic stage post-injury (e.g., Venkatesan & Hillary, 2019) may be an indication of the trajectory of recovery, perhaps eventually relieving the right MTL of its hyperconnectivity and making interhemispheric shifts as patients progress in the chronic phase. These findings suggest that the changes in FC after TBI are likely to be both dynamic and transient, dependent upon time post-injury. Finally, Venkatesan and Hillary (2019) also found the increase in FC in the right hemisphere predicted differences in cognition, where the Cognitive Composite score of many attention and higher-

order cognition tasks were positively associated with the right PCC modularity. The Cognitive Composite score assesses neuropsychological testing, which includes visual and attentional tasks, trail making tasks and reasoning tasks. This relationship between FC and cognitive performance further highlights the cognitive relevance of network dynamics. Based on these findings, we speculate that the increase in FC to the right hemisphere that we found may be predictive of cognitive recovery mechanisms.

4.1.2. Hypoconnectivity at 12 Months Post-Injury

Hypoconnectivity in the left-LOC of the TBI group at 12 months post-injury may also reflect a transfer of centrality. The left-LOC is an extension of the PCC seed, indicating decreased FC to its adjacent regions. This response may relate to the metabolic pressures of the PCC as the central node. One interpretation of this indicates a major change in FC patterns as patients enter the chronic stage, wherein the PCC extension becomes less active, perhaps due to exhaustive metabolic pressure throughout the first year post-injury as the central node directing activity within the network. Hypoconnectivity at the chronic stage is in line with Frank Hillary's research (2011) and a literature review surmising hypoconnectivity in the PCC and precuneus (Hillary et al., 2014).

Hillary (2011) states that the brain's fundamental and initial response to injury is hyperconnectivity, which weakens over time (supported by Roy et al., 2018). The posterior hypoconnectivity found in our study is also widely observed in other connection studies (Sheline & Raichle, 2013; Tijims et al., 2013), and is likely to progress to more anterior regions as neural atrophy and degeneration progresses (as illustrated in Hillary & Grafman, 2017). Advanced structural atrophy describes the resource loss consequential to TBI at the chronic phase (Hillary & Grafman, 2017). Hypoconnectivity in the posterior regions at this stage could be predictive of

progressive increases in hypoconnectivity in the anterior regions. Consistent with their findings, our data reveals hypoconnectivity in the posterior region of the cortex as patients enter their first year post-injury. The prediction that posterior hypoconnectivity progresses in the anterior direction as time progresses post-TBI inspires follow-up studies with our patient cohort.

4.1.3. Absence of Group Difference at 6 Months Post-Injury

We found a lack of difference in FC between HC and patients with TBI at 6 months post-injury. The reason for this may be small sample size, lenient outlier cleaning criteria, or heterogeneity of patient FC trajectory. However, it is also possible that DMN FC in patients as a group actually normalizes at the 6-month post-injury time point. In line with this idea, previous literature has provided evidence for the greatest clinical improvement in the first year of recovery (Novak et al., 2001), and this return to near-normal FC levels by 6 months post-injury could reflect such improvements. Furthermore, behavioral recovery is known to occur within the first 6 months following TBI (Millis et al., 2001), and the return to baseline connectivity levels that we observed at 6 months after injury could be indicative of this behavioral healing process.

The neural mechanism for clinical gains in cognitive performance remains unknown, but perhaps FC changes could reflect the neural mechanism necessary for compensatory changes. FC changes contribute to the overall health of the network, so it is not far-fetched to believe these changes could be the necessary mechanism for cognitive recovery. Alternatively, the pattern of FC changes could be dependent on individual anatomies and injuries- in which case-group level statistics would not be sufficient to calculate a model for recovery. This interpretation coincides with the diverse nature of the disease, whereby the heterogeneity of TBI may indicate heterogenous patterns of network recovery and calls for patient-specific analysis rather than traditional group-level analysis.

4.2. Lack of Longitudinal Functional Connectivity Changes during the First Year after Traumatic Brain Injury?

The intention of longitudinal data analysis was to compare each time points directly in order to examine the trajectory of FC changes. The contrasts between time points within the patient group do not reveal significant FC changes. Although our longitudinal data analysis, which directly compared different time points, lacked significant results, further exploratory analysis provided indirect evidence for dynamic temporal changes in TBI patients. Upon further investigation of the longitudinal data with more lenient map thresholds (p=0.01), we were able to visualize increased FC tendencies in the right MTL in TBI at 3 months post-injury, 6 months post-injury, and 12 months post-injury; this cluster, however, did not survive statistical significance beyond 3 months post-injury (refer to Supplementary Materials). This pattern could be an indication of persistent hyperconnectivity throughout the first year after TBI. Perhaps hyperconnectivity in the MTL at 3 months post-injury escalates this region's mechanism for recovery. This escalation may allow for sufficient recovery to the extent that the metabolic load at 6 months post-injury returns to a normal level. This interpretation conceptualizes functional connectivity as a dynamic neural compensatory mechanism after TBI.

The finding that the right MTL cluster showed increased FC throughout all time points in the TBI group but failed to reach significance through group-level analysis is in line with previous work wherein TBI patients exhibit the most hyperconnectivity at 3 months after injury and this hyperconnectivity gradually declines in strength over time. It is possible that hyperconnectivity still exists across the 6- and 12 months post-injury time points, but at a lesser degree, which did not reach significance. Another possibility is that the FC changes do decrease over time, but our analysis was unable to detect the change, possibly due to noise in the

measurement or because of heterogeneity in the sample. Increasing the sample size could help to clarify why only a small effect was found in this sample. One prediction from our results could indicate hyperconnectivity as an initial response to TBI, and as recovery time goes by the patterns of recovery may change for each individual subject (see Supplementary Material). The maps found in Supplementary Material revealed a cluster at the right middle temporal cortex across the 6-month time periods of recovery which faded to a smaller and weaker cluster in the same region by 12 months of recovery.

The cross-sectional approach taken together with the longitudinal approach indicates that FC levels are indeed altered throughout the first year of recovery, but the trajectory of these changes across time points were not as robust as we predicted. Further manipulation of threshold parameters allowed for some clusters to survive at a p=0.05 but were not statistically significant. These values do, however, open up a discussion about the trends of FC changes throughout the first year of TBI recovery. Specifically, these findings can be used to generate planned comparisons for future studies.

4.3. Limitations & future directions

Our study was not without limitations. As with all longitudinal studies, it can be a challenge to control for retention rate. To replicate this study in the future, a higher motivation or compensation might keep the retention rate higher to allow for a sufficient sample size. Although comparable to other studies, sample size could be increased to strengthen the significance of our results. Another limitation is the poor functional resolution relative to anatomical resolution in fMRI studies. Additionally, movement in the scanner could become an

issue in fMRI studies, although the preprocessing steps and outlier detection aimed to control for motion parameters as much as possible.

TBI is heterogeneous by nature, which could also limit the strength of our findings. Although we made sure that all of our patients sustained high-impact, high-velocity injuries, and we excluded patients with large focal lesions to make the sample more homogeneous, the pathology of each subject is in no way uniform. Perhaps future studies could control for this by investigating more homogeneous TBI samples with exactly the same mechanism of injury.

Future studies could also include the investigations of more time points, including TBI patients beyond 12 months of recovery. Although previous research has shown that the most significant clinical recovery occurs within the first year following injury (Novak et al., 2001), it is not unreasonable to hypothesize that connectivity changes may exist even beyond the first year of TBI recovery and extend into the more chronic stage (1+year). Additional time points could provide more information on the pattern of FC changes, and whether FC profiles eventually stabilize to a healthy level or if they continue to degrade to lower than healthy levels. In line with incorporating more time points, another suggestion for future studies includes a 9-month and 15-month post injury group to better trace the temporal evolution of the observed FC changes. Since we observed cross-sectional differences as patients progressed from 3 to 6 months, it is fair to believe that changes will have also occurred from 3 to 9 months, as well as 12 to 15 months. The dynamic pattern of rsFC changes that we observed could benefit from additional connectivity information at these time points.

As reviewed by Sharp et al. (2011), brain structure may play a role in FC. The degree of structural damage and resource availability has been characterized by FC profiles, however the point at which FC profiles stabilize is not yet known. Considering the DMN, Sharp et al. (2011)

found that patients with the highest FC values displayed the least cognitive impairment. This demonstration of a direct relationship between cognitive function and FC changes has a profound implication for connectivity profiles predicting cognitive outcome following TBI. Venkatesan et al. (2015) also found the characterization of the PCC connectivity profile to perhaps have clinically predictive values. The clinical implications of compensatory FC responses alluded to in previous work cannot be supported or rejected by our study. However, the current study could be expanded upon by exploring the relationship between FC and cognitive performance outcome at 3-, 6-, and 12-month post injury. Corroborating functional findings with cognitive and behavioral data would expand upon the many inferences made about the consequences of FC changes.

5. Conclusion

Overall, our findings are compatible with previous studies of moderate-to-severe TBI at various stages of recovery. Hyperconnectivity between the PCC and the MTL may reflect the central role of the PCC in the DMN as well as its susceptibility to pathology. We assume that increased FC measured through BOLD signal change reflects an increase in network communication, but the implications of this communication are still being investigated. Our results also provide evidence for hypoconnectivity in the posterior regions of the brain as patients enter the chronic stage, which is also supported by previous literature.

The changes in FC seen in our study provoke further questions about the stability of FC after trauma. Hyperconnectivity may be an acute consequence of TBI, one which the network attempts to balance out across the first year of recovery. We suspect the network is over-

compensating during the acute phase, and as the disease progresses during the first year post-injury, neural degeneration may impact and influence functional changes. The increased connectivity of the MTL to the PCC may reflect the strive to reach a healthy equilibrium in a compromised state, due to tissue loss, inflammation, or other consequences. Further, the lack of significance of our longitudinal analysis may reflect a dynamic system where small adjustments of neural networks are constantly occurring after TBI. Taken together, these data could be interpreted as dynamic short-term changes of rsFC in the DMN that occurs on the route to long-term functional stability. These stability goals, however, are likely hindered by the consequential neurodegeneration found in a subset of TBI patients.

Identifying patterns of connectivity changes in specific networks or network regions could serve as a biomarker for the development of neurocognitive therapies and ultimately help to inform better diagnosis, prognosis, and treatment of TBI related injury.

Supplementary Materials

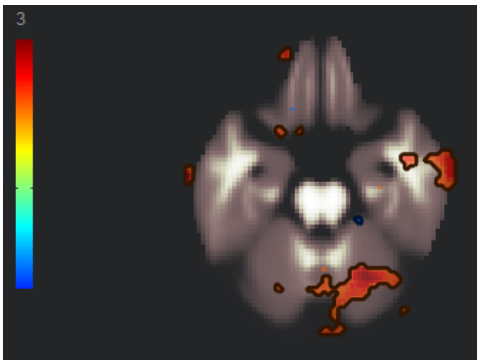

Figure S1. shows the t-maps (p uncorrected <0.05, p value =0.054) of the 3 months after injury patient group minus 6 months after injury patient group. It reveals increased connectivity in the right MTL, which fails to survive statistical testing. This could allude to patterns of hyperconnectivity in the right MTL throughout the first six months of recovery. (This map also shows increased FC in the right (6) cerebellum, which also did not survive statistical testing)

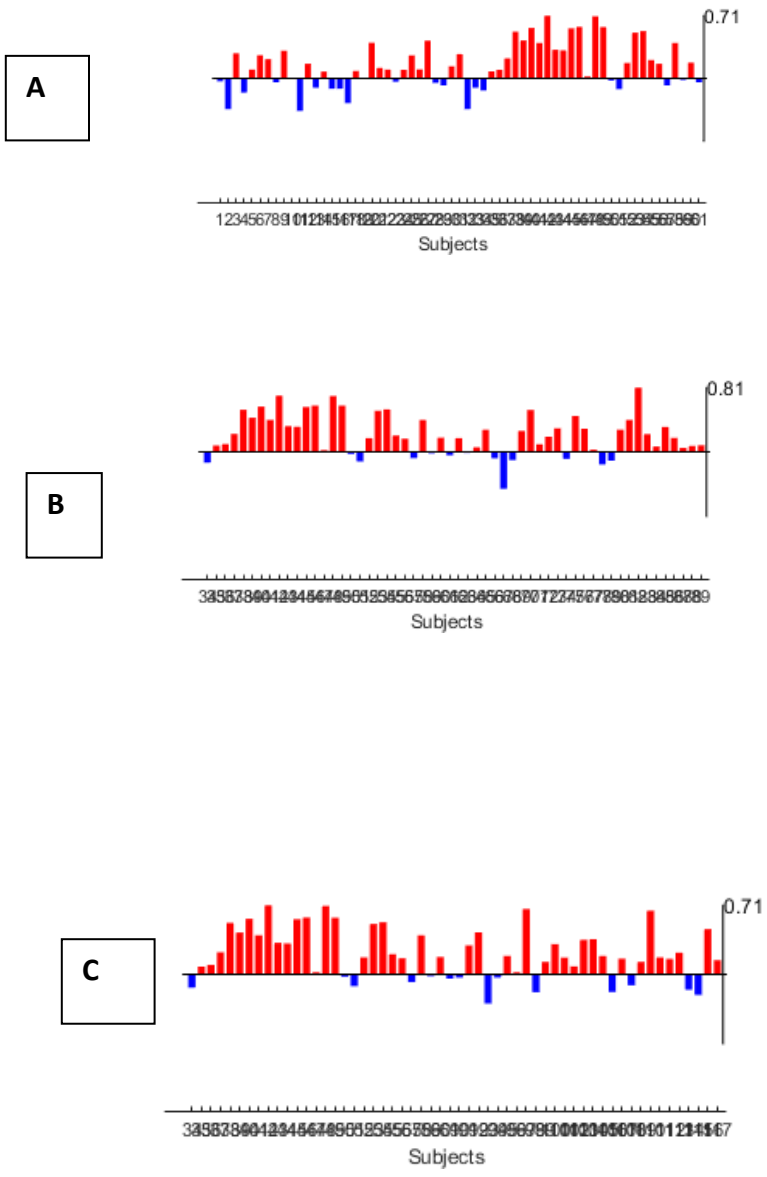

Figure S2. This figure shows the r values for each subject at [60, -12, -24] right MTL voxel. Panel A shows the controls' (the first 28 subjects from the left) r values at this voxel compared to the patients at 3 months post-injury r values at the same voxels (28 rightmost subjects). This provides evidence for increased FC for the patient group at this voxel. Panel B shows the same calculations for patients 3 months after injury on the leftmost side, and the patients 6 months after injury on the rightmost side. This shows evidence for persistent increased FC at the r MTL voxel throughout 6 months. Panel C shows the patients 3 months after injury on the leftmost side compared to the r values at the same voxel for patients at 12 months after injury on the rightmost side of the chart. These values also seem to have increased FC at 12 months. These set of charts provide qualitative evidence for persistent hyperconnectivity in the right MTL of the patient group across all three time points, although any quantitative evidence at 6 and 12 months failed to reach significance.

References

Biswal, Bharat, F. Zerrin Yetkin, Victor M. Haughton, and James S. Hyde. "Functional Connectivity in the Motor Cortex of Resting Human Brain Using Echo-Planar Mri." *Magnetic Resonance In Medicine* 34, no. 4 (October 1995): 537–41. https://doi.org/10.1002/mrm.1910340409.

Bonnelle, V., T. E. Ham, R. Leech, K. M. Kinnunen, M. A. Mehta, R. J. Greenwood, and D. J. Sharp. "Salience Network Integrity Predicts Default Mode Network Function after Traumatic Brain Injury." *Proceedings of the National Academy of Sciences* 109, no. 12 (March 20, 2012): 4690–95. https://doi.org/10.1073/pnas.1113455109.

Buckner, R.L., J.R. Andrews-Hanna, and D.L. Schacter. "The Brain's Default Network: Anatomy, Function, and Relevance to Disease." *Annals of the New York Academy of Sciences* 1124, no. 1 (April 3, 2008): 1–38. https://doi.org/10.1196/annals.1440.011.

Bullmore, Ed, and Olaf Sporns. "Complex Brain Networks: Graph Theoretical Analysis of Structural and Functional Systems." *Nature Reviews Neuroscience* 10, no. 3 (March 2009): 186–98. https://doi.org/10.1038/nrn2575.

Caeyenberghs, Karen, Helena Verhelst, Adam Clemente, and Peter H. Wilson. "Mapping the Functional Connectome in Traumatic Brain Injury: What Can Graph Metrics Tell Us?" *NeuroImage* 160 (October 2017): 113–23. https://doi.org/10.1016/j.neuroimage.2016.12.003.

Castellanos, Nazareth P., Inmaculada Leyva, Javier M. Buldú, Ricardo Bajo, Nuria Paúl, Pablo Cuesta, Victoria E. Ordóñez, et al. "Principles of Recovery from Traumatic Brain Injury: Reorganization of Functional Networks." *NeuroImage* 55, no. 3 (April 2011): 1189–99. https://doi.org/10.1016/j.neuroimage.2010.12.046.

Centers for Disease Control and Prevention. "Traumatic Brain Injury and Concussion." *National Center for Injury Prevention and Control.* (August 28, 2020): https://www.cdc.gov/traumaticbraininjury/index.html.

Eickhoff, S.B., and V.I. Müller. "Functional Connectivity." In *Brain Mapping*, 187–201. Elsevier, 2015. https://doi.org/10.1016/B978-0-12-397025-1.00212-8.

Fox, Michael D., Abraham Z. Snyder, Justin L. Vincent, and Marcus E. Raichle. "Intrinsic Fluctuations within Cortical Systems Account for Intertrial Variability in Human Behavior." *Neuron* 56, no. 1 (October 2007): 171–84. https://doi.org/10.1016/j.neuron.2007.08.023.

Fujiwara, E., Schwartz, M. L., Gao, F., Black, S. E., & Levine, B. (2008). Ventral frontal cortex functions and quantified MRI in traumatic brain injury. *Neuropsychologia*, *46*(2), 461–474. https://doi.org/10.1016/j.neuropsychologia.2007.08.027

Gratton, Caterina, Emi M. Nomura, Fernando Pérez, and Mark D'Esposito. "Focal Brain Lesions to Critical Locations Cause Widespread Disruption of the Modular Organization of the Brain." *Journal of Cognitive Neuroscience* 24, no. 6 (June 2012): 1275–85. https://doi.org/10.1162/jocn_a_00222.

Greer, J. E., M. J. McGinn, and J. T. Povlishock. "Diffuse Traumatic Axonal Injury in the Mouse Induces Atrophy, c-Jun Activation, and Axonal Outgrowth in the Axotomized Neuronal Population." *Journal of Neuroscience* 31, no. 13 (March 30, 2011): 5089–5105. https://doi.org/10.1523/JNEUROSCI.5103-10.2011.

Hagmann, Patric, Leila Cammoun, Xavier Gigandet, Reto Meuli, Christopher J Honey, Van J Wedeen, and Olaf Sporns. "Mapping the Structural Core of Human Cerebral Cortex." Edited by Karl J Friston. *PLoS Biology* 6, no. 7 (July 1, 2008): e159. https://doi.org/10.1371/journal.pbio.0060159.

Hampshire, Adam, Alex MacDonald, and Adrian M. Owen. "Hypoconnectivity and Hyperfrontality in Retired American Football Players." *Scientific Reports* 3, no. 1 (December 2013): 2972. https://doi.org/10.1038/srep02972.

Hawellek, D. J., J. F. Hipp, C. M. Lewis, M. Corbetta, and A. K. Engel. "Increased Functional Connectivity Indicates the Severity of Cognitive Impairment in Multiple Sclerosis." *Proceedings of the National Academy of Sciences* 108, no. 47 (November 22, 2011): 19066–71. https://doi.org/10.1073/pnas.1110024108.

Hayes, Jasmeet P., Erin D. Bigler, and Mieke Verfaellie. "Traumatic Brain Injury as a Disorder of Brain Connectivity." *Journal of the International Neuropsychological Society* 22, no. 2 (February 2016): 120–37. https://doi.org/10.1017/S1355617715000740.

Hillary, F.G., J. Slocomb, E.C. Hills, N.M. Fitzpatrick, J.D. Medaglia, J. Wang, D.C. Good, and G.R. Wylie. "Changes in Resting Connectivity during Recovery from Severe Traumatic Brain Injury." *International Journal of Psychophysiology* 82, no. 1 (October 2011): 115–23. https://doi.org/10.1016/j.ijpsycho.2011.03.011.

Hillary, Frank G., and Jordan H. Grafman. "Injured Brains and Adaptive Networks: The Benefits and Costs of Hyperconnectivity." *Trends in Cognitive Sciences* 21, no. 5 (May 2017): 385–401. https://doi.org/10.1016/j.tics.2017.03.003.

Hillary, Frank G., Cristina A. Roman, Umesh Venkatesan, Sarah M. Rajtmajer, Ricardo Bajo, and Nazareth D. Castellanos. "Hyperconnectivity Is a Fundamental Response to Neurological Disruption." *Neuropsychology* 29, no. 1 (January 2015): 59–75. https://doi.org/10.1037/neu0000110.

Johnson, Victoria E., William Stewart, and Douglas H. Smith. "Axonal Pathology in Traumatic Brain Injury." *Experimental Neurology* 246 (August 2013): 35–43. https://doi.org/10.1016/j.expneurol.2012.01.013.

Kelly, A.M. Clare, Lucina Q. Uddin, Bharat B. Biswal, F. Xavier Castellanos, and Michael P. Milham. "Competition between Functional Brain Networks Mediates Behavioral Variability." *NeuroImage* 39, no. 1 (January 2008): 527–37. https://doi.org/10.1016/j.neuroimage.2007.08.008.

Mathias, Jane L., and Patricia Wheaton. "Changes in Attention and Information-Processing Speed Following Severe Traumatic Brain Injury: A Meta-Analytic Review." *Neuropsychology* 21, no. 2 (2007): 212–23. https://doi.org/10.1037/0894-4105.21.2.212.

Mayo Clinic. "Traumatic Brain Injury*." Mayo Foundation for Medical Education and Research (MFMER).* (February 4, 2021): https://www.mayoclinic.org/diseases-conditions/traumatic-brain-injury/symptoms-causes/syc-20378557.

Millis, S. R., Rosenthal, M., Novack, T. A., Sherer, M., Nick, T. G., Kreutzer, J. S., High, W. M., & Ricker, J. H. (2001). Long-Term Neuropsychological Outcome After Traumatic Brain Injury: *Journal of Head Trauma Rehabilitation, 16*(4), 343–355. https://doi.org/10.1097/00001199-200108000-00005

Nicolo, Pierre, Sviatlana Rizk, Cécile Magnin, Marie Di Pietro, Armin Schnider, and Adrian G. Guggisberg. "Coherent Neural Oscillations Predict Future Motor and Language Improvement after Stroke." *Brain* 138, no. 10 (October 2015): 3048–60. https://doi.org/10.1093/brain/awv200.

Novack, Thomas A., Beverly A. Bush, Jay M. Meythaler, and Kay Canupp. "Outcome after Traumatic Brain Injury: Pathway Analysis of Contributions from Premorbid, Injury Severity, and Recovery Variables." *Archives of Physical Medicine and Rehabilitation* 82, no. 3 (March 2001): 300–305. https://doi.org/10.1053/apmr.2001.18222.

Palacios, E. M., Sala-Llonch, R., Junque, C., Roig, T., Tormos, J. M., Bargallo, N., & Vendrell, P. (2013). Resting-state functional magnetic resonance imaging activity and connectivity and cognitive outcome in traumatic brain injury. *JAMA neurology*, *70*(7), 845-851. doi:10.1001/jamaneurol.2013.38

Payne, William N., Orlando De Jesus, and Andrew N. Payne. "Contrecoup Brain Injury." In *StatPearls*. Treasure Island (FL): StatPearls Publishing, 2021. http://www.ncbi.nlm.nih.gov/books/NBK536965/.

Roy, A., Bernier, R. A., Wang, J., Benson, M., French, J. J., Good, D. C., & Hillary, F. G. (2018). Correction: The evolution of cost-efficiency in neural networks during recovery from traumatic brain injury. *PLOS ONE*, *13*(10), e0206005. https://doi.org/10.1371/journal.pone.0206005

Sharp, David J., Christian F. Beckmann, Richard Greenwood, Kirsi M. Kinnunen, Valerie Bonnelle, Xavier De Boissezon, Jane H. Powell, Serena J. Counsell, Maneesh C. Patel, and Robert Leech. "Default Mode Network Functional and Structural Connectivity after Traumatic Brain Injury." *Brain* 134, no. 8 (August 2011): 2233–47. https://doi.org/10.1093/brain/awr175.

Sharp, David J., Gregory Scott, and Robert Leech. "Network Dysfunction after Traumatic Brain Injury." *Nature Reviews Neurology* 10, no. 3 (March 2014): 156–66. https://doi.org/10.1038/nrneurol.2014.15.

Sheline, Y. I., & Raichle, M. E. "Resting State Functional Connectivity in Preclinical Alzheimer's Disease." *Biological Psychiatry*, *74*(5), (January 7, 2013): 340–347. https://doi.org/10.1016/j.biopsych.2012.11.028

Tijms, B. M., Wink, A. M., de Haan, W., van der Flier, W. M., Stam, C. J., Scheltens, P., & Barkhof, F. "Alzheimer's disease: Connecting findings from graph theoretical studies of brain networks." *Neurobiology of Aging*, *34*(8), (August 2013): 2023–2036. https://doi.org/10.1016/j.neurobiolaging.2013.02.020

Venkatesan, Umesh M., Nancy A. Dennis, and Frank G. Hillary. "Chronology and Chronicity of Altered Resting-State Functional Connectivity after Traumatic Brain Injury." *Journal of Neurotrauma* 32, no. 4 (February 15, 2015): 252–64. https://doi.org/10.1089/neu.2013.3318.

Venkatesan, Umesh M., and Frank G. Hillary. "Functional Connectivity within Lateral Posterior Parietal Cortex in Moderate to Severe Traumatic Brain Injury." *Neuropsychology* 33, no. 6 (September 2019): 893–910. https://doi.org/10.1037/neu0000553.

Whitfield-Gabrieli, S., & Nieto-Castanon, A. "*Conn*: A Functional Connectivity Toolbox for Correlated and Anticorrelated Brain Networks." *Brain Connectivity*, *2*(3), (June 2012): 125–141. https://doi.org/10.1089/brain.2012.0073

Wu, Hsiao-Ming, Sung-Cheng Huang, Naoya Hattori, Thomas C. Glenn, Paul M. Vespa, David A. Hovda, and Marvin Bergsneider. "Subcortical White Matter Metabolic Changes Remote from Focal Hemorrhagic Lesions Suggest Diffuse Injury after Human Traumatic Brain Injury." *Neurosurgery* 55, no. 6 (December 1, 2004): 1306–17. https://doi.org/10.1227/01.NEU.0000143028.08719.42.

Zamora-López. "Cortical Hubs Form a Module for Multisensory Integration on Top of the Hierarchy of Cortical Networks." *Frontiers in Neuroinformatics*, 2010. https://doi.org/10.3389/neuro.11.001.2010.