Diffusion Tensor Imaging Reveals White Matter Differences in Military Personnel Exposed to Trauma with and without Post-traumatic Stress Disorder

Patrick McCunn 1,*, J. Don Richardson 2,3,4,5, Rakesh Jetly 6, Benjamin Dunkley 1,7,8

1 Neurosciences & Mental Health, The Hospital for Sick Children (SickKids) Research Institute, Toronto, Ontario
2 The MacDonald Franklin OSI Research Centre, Lawson Health Research Institute, London, Ontario
3 Department of Psychiatry, Western University, London, Ontario
4 Department of Psychiatry and Behavioral Neurosciences, McMaster University, Hamilton, Ontario
5 Operational Stress Injury Clinic, St. Joseph’s Health Care, London, Ontario, Canada
6 Canadian Forces Health Services Group HQ, Department of National Defence, Ottawa, Ontario
7 Department of Diagnostic Imaging, The Hospital for Sick Children (SickKids), Toronto, Ontario
8 Department of Medical Imaging, University of Toronto, Toronto, Ontario

ARTICLE INFO

Keywords:
Diffusion Tensor Imaging
Magnetic Resonance Imaging
Post-traumatic Stress Disorder
Trauma Exposure
Military Personnel

ABSTRACT

Background: Post-traumatic stress disorder (PTSD) is a debilitating mental health condition that develops in response to exposure to a traumatic event. The purpose of this study was to investigate white matter differences using diffusion tensor imaging (DTI) in trauma exposed military personnel with and without PTSD.

Methods: Data were acquired in compliance with the Hospital for Sick Children and Canadian Armed Forces Research Ethics Boards for the following groups: military personnel with PTSD (PTSD, n = 23), trauma exposed military personnel with no PTSD diagnosis (TE, n = 25) and civilian controls (CC, n = 13). All participants were male. DTI was acquired on a Siemens Trio 3T MRI. Maps of Fractional Anisotropy (FA), Mean Diffusivity (MD), Axial Diffusivity (AD), and Radial Diffusivity (RD) were analyzed using Tract-Based Spatial Statistics (TBSS).

Results: In the PTSD and TE groups, FA was significantly greater within the hippocampus, corpus callosum, cingulum, and several associated white matter tracts. Elevated FA was shown to be largely due to reduced RD suggesting a possible structural substrate that underscores neurophysiological connectivity.

Conclusions: This study reinforces previous findings showing differences in DTI metrics within the limbic system in military personnel exposed to trauma with and without PTSD.

Introduction

Posttraumatic stress disorder (PTSD) is a debilitating mental health condition that develops in response to exposure to a traumatic event (Breslau, 2009; Wynn and Ursano, 2016). PTSD has been characterized by a range of symptoms commonly falling into four main categories: re-experiencing the trauma through intrusive recollections, emotional avoidance, negative alterations to cognition and mood, and increased arousal (American Psychiatric Association, 2013; Friedman et al., 1994). Due to the high levels of environmental stress and traumatic exposure during service, military personnel are at particular risk of developing PTSD (Creamer et al., 2011; Gates et al., 2012). Current lifetime prevalence estimates of PTSD in the military are reported to be greater than 15% (Hines et al., 2014; Richardson et al., 2010).

PTSD is a complex and heterogeneous illness, and the neuropathological processes that underpin the disorder result from a complicated interplay between the type of trauma, history of trauma, and pre-existing cognitive risk factors (Kelmendi et al., 2016; Sherin and Nemeroff, 2011; Yehuda et al., 2015). Currently, no single pathognomonic test exists that can concretely assess these neuropathological processes limiting understanding of the effect of trauma exposure and PTSD and similarly the advancement of clinically relevant diagnostic and prognostic capabilities. As such, much research focus has been on the development of neuroimaging methods capable of determining these mechanistic processes. In particular, diffusion tensor imaging (DTI) has shown great promise as a neuroimaging modality capable of elucidating...
the neuropathological processes, particularly within white matter, underly-
ing PTSD and trauma exposure.

DTI is a powerful magnetic resonance modality that indexes anatomical microstructure and pathological dysregulation of white matter (Basser et al., 1994; Le Bihan et al., 2001; Mori and Zhang, 2006). Many developmental and pathological processes alter tissue microstructure and composition leading to transformed diffusion patterns within the brain (Perpaoli et al., 1996); in particular, these processes are known to be dysregulated by traumatic exposure, and in those that develop it, PTSD (Fani et al., 2012; Gong et al., 2014). DTI measures the geometric diffusion patterns of water molecules within an imaging voxel and their altered scalar diffusion measures, namely fractional anisotropy (FA), mean diffusivity (MD), axial diffusivity (AD) and radial diffusivity (RD). MD represents the overall magnitude of diffusion within a given voxel, FA represents the degree of directionality of diffusion, AD indexes the magnitude of diffusion along the principal axis of diffusion, and RD provides an average value of the magnitude of diffusion along the secondary and tertiary diffusion axes.

Previous studies have reported altered diffusivity profiles in PTSD, but these differences are inconsistent due to a myriad of factors (Daniels et al., 2013), but this is most likely because participants are drawn from heterogeneous samples within and between studies. Further, many studies lack either a non-traumatized or trauma-exposed control group to isolate white matter change related directly to PTSD, rather than stress and traumatic exposure per se. Olson et al. reports decreased FA in the left frontal lobe in PTSD when compared with trauma exposed controls sampled from the general community, but lacked a non-trauma exposed control group (Olson et al., 2017). O’Doherty et. al. reports decreased FA across bilateral anterior thalamic radiation, cingulum cingulate gyrus, superior longitudinal fasciculus, uncinate fasciculus, and in the corpus callosum in both PTSD and trauma exposed partici-
pants when compared with healthy controls (O’Doherty et al., 2018). In individuals suffering PTSD following a sarin attack, Abe et. al. reported increased FA in the cingulum bundle compared to individuals who were also present but did not develop PTSD (Abe et al., 2006). Zhang et. al. similarly found increased FA in subjects associated with a coal mine accident, but within the left superior frontal gyrus, when compared to healthy controls (Zhang et al., 2011). A meta-analysis of adult-onset PTSD showed that FA changes in the anterior and posterior parts of the cingulum were consistent across studies, as well as dysregulated white matter in the superior longitudinal fasciculus and diffuse frontal cortex changes (Siehl et al., 2018). Another recent meta-analysis addition-
ally found that inferior temporal corticospinal tract changes in FA are present in PTSD (Ju et al., 2020). While these studies by no means cover the extent of literature focusing on DTI in PTSD, they provide an overview of the magnitude of diffusion along the secondary and tertiary diffusion axes.

Previous studies have reported altered diffusivity profiles in PTSD, but these differences are inconsistent due to a myriad of factors (Daniels et al., 2013), but this is most likely because participants are drawn from heterogeneous samples within and between studies. Further, many studies lack either a non-traumatized or trauma-exposed control group to isolate white matter change related directly to PTSD, rather than stress and traumatic exposure per se. Olson et al. reports decreased FA in the left frontal lobe in PTSD when compared with trauma exposed controls sampled from the general community, but lacked a non-trauma exposed control group (Olson et al., 2017). O’Doherty et. al. reports decreased FA across bilateral anterior thalamic radiation, cingulum cingulate gyrus, superior longitudinal fasciculus, uncinate fasciculus, and in the corpus callosum in both PTSD and trauma exposed partici-
pants when compared with healthy controls (O’Doherty et al., 2018). In individuals suffering PTSD following a sarin attack, Abe et. al. reported increased FA in the cingulum bundle compared to individuals who were also present but did not develop PTSD (Abe et al., 2006). Zhang et. al. similarly found increased FA in subjects associated with a coal mine accident, but within the left superior frontal gyrus, when compared to healthy controls (Zhang et al., 2011). A meta-analysis of adult-onset PTSD showed that FA changes in the anterior and posterior parts of the cingulum were consistent across studies, as well as dysregulated white matter in the superior longitudinal fasciculus and diffuse frontal cortex changes (Siehl et al., 2018). Another recent meta-analysis addition-
ally found that inferior temporal corticospinal tract changes in FA are present in PTSD (Ju et al., 2020). While these studies by no means cover the extent of literature focusing on DTI in PTSD, they provide an overview of the magnitude of diffusion along the secondary and tertiary diffusion axes.

Similarly, studies specifically of military-related PTSD report het-
erogeneous findings. Complicating this picture is the high prevalence of comorbidities such as depression (Owens et al., 2009; Stander et al., 2014), anxiety (Knowles et al., 2019), and substance abuse (Blanco et al., 2013; Brady et al., 2000; Brenner et al., 1996; Jacobsen et al., 2001). Decreased FA in the white matter tracts of frontal and limbic brain regions in combat related PTSD compared to trauma exposed group without PTSD (Schuff et al., 2011), whereas higher FA has been observed in the thalamocortical and occipitofrontal fasciculus in military personnel with a history of operational stress injuries that result in PTSD, against a comparator group with similar experiences and deployment stress, as well as typically-developed civilian control group without a history of opera-
tional stress injuries. This would allow us to isolate and identify dysre-
gulated white matter due to PTSD, rather than traumatic exposure alone. Using DTI, we examined group differences in whole brain FA, MD, AD, and RD, and furthermore, we determined if self-reported symptom severity (severity scores obtained from the Posttraumatic Stress Disorder Check List (PCL)) correlated with microstructural differences observed in the DTI images of the PTSD group. Based on previous findings in adult-onset PTSD, we hypothesised changes in white matter micro-
structure in frontal regions and the cingulum in PTSD compared to the two control groups.

Methods

Subjects

Data were acquired in compliance with the Hospital for Sick Children and Canadian Armed Forces Research Ethics Boards for the following groups: civilian controls (CC, all male, n = 13, age = 28.7 ± 5.0 years), military personnel with a history of operational stress and traumatic exposure but no PTSD diagnosis (TE, all male, n = 25, age = 32.8 ± 4.5 years) and military personnel with a confirmed PTSD diagnosis (PTSD, all male, n = 23, age = 37.3 ± 6.8 years). All military personnel participants were recruited through military psychiatrists, and control participants through the Hospital for Sick Children.

The PTSD group consisted of military personnel with a clinical diagnosis of PTSD (Diagnostic Stat. Man. Ment. Disord. Fourth Ed. Text Revs., 2000) as determined by a psychiatrist or psychologist specialising in trauma-related mental health injuries at a Canadian Operational Trauma Stress Support Centre (OTSSC). PTSD participants were recruited from active military personnel currently receiving mental health treatment, who were diagnosed between 1 and 4 years prior to taking part in the study and presenting with moderate to severe PTSD (PTSD checklist-military version score of >50) (American Psychiatric Association, 2013). Participants in the TE group were combat-exposed, frontline troops in similar military roles and ranks, with physician confirmed exposure to operational stress and traumatic exposure whilst on deployment but did not develop or meet the clinical criteria for PTSD. Participants in the CC group were recruited through study flyers posted at the Hospital for Sick Children and were screened to confirm they had no history of traumatic exposure.

Participants in all groups were excluded if they had a history of a traumatic brain injury (TBI), as screened by a psychiatrist through a review of their electronic health record, telephone interview, and administration of the Defence and Veteran’s Brain Injury Centre (DVIBC) screening tool (Schwab et al., 2007). Further exclusions included ferrous metal inside the body or implanted medical devices that might be MRI contraindications, neurological disorders, certain ongoing medications (anticonvulsants, and/or benzodiazepines, or other GABA antagonists). PTSD participants undergoing treatment including evidenced-based psychotropic medication(s), such as selective serotonin reuptake inhibitors (SSRIs), serotonin-norephedrine reuptake inhibitors (SNRIs), and Prazosin were allowed to partake in this study.
Cognitive and Behavioral Assessments

All subjects in this study completed demographic questionnaires as well as several cognitive-behavioural assessments including the Wechsler Abbreviated Scale of Intelligence (WASI) (Wechsler, 1999), the Alcohol Use Disorders Identification Test (AUDIT) (Saunders et al., 1993), the Generalized Anxiety Disorder 7-item (GAD-7) scale (Spitzer et al., 2006), and the Patient Health Questionnaire-9 (PHQ-9) (Kroenke et al., 2001). The GAD-7 allows for assessment of symptoms completed the Posttraumatic Stress Disorder Check List (PCL-C) (Blanchard et al., 1996; Weathers et al., 1993), which describes the number of PTSD symptoms and their severity.

Image Acquisition and Analysis

DTI was acquired on a Siemens Trio 3T scanner with a 12-channel head coil using a spin echo EPI acquisition sequence (60 directions, b = 1000 s/mm², TE/TR = 88/8800 ms, FOV = 244 × 244 × 140 mm, resolution: 2 mm isotropic).

Images were processed using the fMRI Software Library (FSL, v.5.0.10) and MRtrix (v. 3.0). Gibbs Ringing Removal (Kellner et al., 2016) followed by PCA denoising (Veraart et al., 2016) was performed first in MRtrix. EDDY (Andersson and Sotiropoulos, 2016) with outlier detection (Andersson et al., 2016), and slice-to-volume motion correction (Andersson et al., 2017) was used to correct for eddy current induced distortions as well as susceptibility-induced distortions. A group QC report was generated using eddy squad v 1.0.2 (Bastiani et al., 2019) and is included as Supplementary Data. Data were brain extracted using BET (Smith, 2002) and the FDT toolbox (v. 5.0) was used to produce maps of FA, MD, AD, and RD. Voxel wise statistical analysis of the data was carried out using Tract-Based Spatial Statistics (TBSS) which projects all participants’ FA onto a mean FA tract skeleton, before applying voxel wise cross-subject statistics. The FMRIB58_FA atlas was used as a target for registration in TBSS. Non-parametric permutation-based testing was performed using Randomise in FSL (Winkler et al., 2014). A one-way ANOVA (1-factor 3-levels) was used with one F-test for the overall effect group effect on diffusion parameters (FA, AD, RD, MD), and 6 contrasts for the individual comparisons of voxel-wise diffusion parameters between groups. Results are reported at the p < .05 level after 10000 permutations, with threshold-free cluster enhancement (TFCE) (Smith and Nichols, 2009) and family-wise error rate correction for multiple comparisons.

To elucidate the nature of any FA differences, FSL’s Cluster was used to identify statistically significant (p < .05) clusters followed by alasso-query to localize against these clusters using the JHU White-Matter Tractography Atlas template. ROI analysis was performed within these clusters independently analyzing mean RD and AD values. Unpaired t-tests were performed in PRISM 8 to compare each of these metrics in each independent cluster between i) PTSD and CC groups and ii) TE and CC groups. Finally, Spearman’s rank-order correlation analysis was performed in PRISM 8, with a Bonferroni correction (corrected statistical significance set at p = .0005) for multiple comparisons to determine if:

I PCL score or each symptom cluster sub-score (Reexperiencing, Avoidance, or Hyperarousal) correlated with DTI metrics (FA, MD, AD and RD) within each of the identified statistically significant clusters in the PTSD group.

II GAD-7 score correlated with DTI metrics within each of the identified statistically significant clusters in the PTSD or TE groups.

III if PHQ-9 score correlated with DTI metrics within each of the identified statistically significant clusters in the PTSD or TE groups.

Results

Patient Demographics

Imaging data were successfully collected from all participants in each group. Participant demographics are listed in Table 1. Age is statistically significant difference between groups as determined by one-way ANOVA (F(2,58) = 10.3401, p < .01), as was alcohol use disorder indexed by the AUDIT (F(2,58) = 3.3317, p = .04). There was no statistically significant difference in WASI between groups (F(2,58) = 2.7708, p = .07).

TBSS results show greater FA in military personnel diagnosed with PTSD versus Civilian Controls

In the PTSD group (Figure 1), three independent clusters were identified with statistically significant (p < .05) differences in FA. All of these clusters showed greater FA in the PTSD group when compared to the CC group. Please refer to Table 2 for cluster locations and values. No statistically significant differences between groups were observed in whole brain MD, AD, or RD.

Within each of the three clusters identified in the PTSD group, RD was the dominant contributor to the observed increased FA values (Figure 2). In each cluster, mean RD was significantly lower in the PTSD group when compared with the CC group. No statistically significant differences in mean AD were observed in any of the clusters. Table 3

TBSS Results show greater FA in trauma exposed military personnel without PTSD versus Civilian Controls

In the TE group (Figure 3), five clusters were identified with statistically significant differences (p < .05) in FA, with all clusters showing higher FA in the TE group when compared to the CC group. Please refer to Table 4 for cluster locations and values. No statistically significant differences were observed in whole brain MD, AD, or RD.

In both cluster five and four of the TE group, significantly decreased RD (p < .05) was the dominant contributor to the observed increased FA values. Figure 4 No other statistically significant differences in mean AD or RD were observed. Table 5

TBSS reveals no observed difference in FA in PTSD versus trauma exposure without PTSD

No statistically significant differences were observed in whole brain FA, MD, AD, or RD when comparing the PTSD and TE groups.

DTI metrics do not correlate with PCL, GAD-7, and PHQ-9 Scores

PTSD severity was indexed using PCL questionnaire scores; these measures were unavailable for two subjects in the PTSD group, and therefore these subjects were removed and not included in those

Table 1

| Group              | n  | Age (years) | WASI   | AUDIT |
|--------------------|----|-------------|--------|-------|
| Civilian Controls  | 13 | 28.7 ± 5.0  | 115.8 ± 7.8 | 5.6 ± 4.2 |
| Trauma Exposed     | 25 | 32.8 ± 4.5  | 117.6 ± 13.9 | 5.4 ± 3.6 |
| PTSD               | 23 | 37.3 ± 6.8  | 109.2 ± 13.5 | 9.4 ± 8.0 |
correlations. Correlational analysis for anxiety (GAD-7) and depression (PHQ-9) versus DTI measures included 21 subjects in the PTSD group, and all 25 subjects in the TE group. Please refer to Table 6 for mean PCL, GAD-7 and PHQ-9 questionnaire data.

No significant correlations were observed between DTI metrics and PTSD severity (PCL) in the PTSD group, or anxiety (GAD-7) or depression (PHQ-9) severity within the PTSD and TE groups. Full statistical results can be found in Supplementary Table 1.

Discussion

In this study, DTI revealed white matter microstructure change indexed by elevated FA in multiple regions of the brain in trauma- and
stress-exposed active military personnel, both with and without PTSD, when compared to typical civilian controls. No whole brain voxel-wise differences were observed between groups in MD, AD, or RD. In the PTSD group, radial diffusivity was found to be the dominant driver of diffusivity changes in all three statistically significant clusters. Similarly, in the TE group, radial diffusivity was the dominant driver in the largest cluster. Finally, within the PTSD group no statistically significant correlations were found between DTI metrics and measures of PTSD, or in the combined cohort of military personnel when correlating against anxiety or depression severity. These results support a body of literature showing dysregulated white matter microstructure and limbic network dysfunction associated with chronic operational stress and further highlights the ability of DTI to detect this dysregulation in-vivo.

White matter microstructure alterations in military personnel with operational stress compared to typical civilian controls

Elevated FA was observed in multiple regions of the brains of military personnel who underwent operational stress and trauma exposure. Further comparisons between subgroups in the military cohort (PTSD versus no PTSD) did not reveal significant differences, suggesting those that exist when compared against controls is likely due to chronic environmental stressors and/or exposure to traumatic events. In the PTSD group, while three independent clusters were identified with elevated FA, over 98% of those voxels were within Cluster 3, indicating a large-scale, interconnected alteration to white matter. This cluster included voxels within the anterior thalamic radiation, corticospinal tract, cingulate gyrus, hippocampus, forceps major, forceps minor, inferior fronto-occipital fasciculus, inferior longitudinal fasciculus, superior longitudinal fasciculus, and uncinate fasciculus. In the TE group, five independent clusters were identified with significantly elevated FA when compared to the CC group. Similar to the PTSD group, a single connected cluster (Cluster 5) contained over 88% of the voxels with elevated FA. This cluster covered the anterior thalamic radiation, corticospinal tract, cingulate gyrus, forceps minor, inferior fronto-occipital fasciculus, inferior longitudinal fasciculus, superior longitudinal fasciculus, uncinate fasciculus, and superior longitudinal fasciculus.

Previous studies using DTI in participants with combat-related PTSD and traumatic exposure have shown changes to FA, predominantly within the frontal and limbic regions, but previously most studies have shown decreased FA rather than an increase, as observed here (Sanjuan et al., 2013; Schuff et al., 2011). Greater FA has been shown in the fronto-occipital fasciculus in veterans with combat-related PTSD, but in that study, the same increases were not seen in combat exposed controls (Aschbacher et al., 2018). Here, we observed these changes in both the PTSD and TE groups, pointing to effects that are driven by operational stress and trauma, rather than a specific psychiatric diagnosis. While these results vary in region and direction, they reinforce the findings that white matter alterations in the limbic system are of central importance to PTSD and operational stress more generally. This is not surprising, as the limbic system and its associated white matter connections are largely responsible for the processing of emotions, memories, and pain (Champney, n.d.; Mega et al., 1997; Willis and Haines, 2018). Extensive human studies have identified alterations and dysfunctions in the limbic system associated with PTSD and trauma exposure, such as reduced hippocampal volume and disrupted functional connectivity in the amygdala (Brunetti et al., 2010; Chao et al., 2013; Gawrysiak et al., 2014; Sripada et al., 2012). Further, in both human and animal studies, it has been shown that stress exposure leads to altered white matter within limbic regions detectable through DTI (Liu et al., 2018; Magalhães et al., 2017; Meng et al., 2018). Thus, this may explain the similarities in the PTSD group and the TE group; the exposure to persistent operational stress and fatigue, psychological difficulties, and traumatic exposure itself may be the driver of dysregulated white matter, while the development of psychiatric illness is a varied functional response to stress and structural remodelling in the brain.

In both PTSD and TE groups, decreased mean RD predominantly contributed to the increased FA observed in this study. RD has been suggested as a potential in-vivo marker of myelin integrity, with decreased RD being associated with increased myelination (Winklewski et al., 2018). There is recent research demonstrating increased myelination within limbic regions such as the hippocampus in military personnel with PTSD (Chao et al., 2015; Jak et al., 2020), but to our knowledge, this has not been previously shown in trauma exposed military personnel without PTSD. This increase in RD could suggest adaptive oligodendrogenesis and increased myelination and provide a structural basis for observations of altered neural dynamics in military-related PTSD, including alterations to excitation and inhibition, and synaptic efficacy that manifests as “hypersynchrony” in neural oscillations, imaged in various studies using magnetoencephalography (MEG) and functional magnetic resonance imaging (fMRI) (Dunkley et al., 2018; Sripada et al., 2012). These changes in RD should be interpreted with caution though and only in the context of describing the observed FA changes, not as a broad, global measure.

DTI measures do not correlate with PTSD, anxiety or depression severity

No statistically significant correlations were observed between any DTI metrics and severity of PTSD, anxiety or depression within the PTSD group. This reiterates previous findings which report little association between PTSD severity and altered diffusivity profiles (Koch et al., 2017; Sweeney et al., 2016). Similar to the PTSD group, no statistically
significant correlations were observed between any DTI metrics and anxiety or depression within the TE group. Previous work has shown some relations between DTI metrics and anxiety in various populations (Wang et al., 2017, 2016), but these correlations are limited and to our knowledge the associations of these metrics have not been previously explored in military personnel. This suggests, unsurprisingly, that amongst these subjects, white matter measures explain little of the covariance in symptoms of depression or anxiety, and that the presence of symptoms is likely driven by a complex multifaceted response to operational stress beyond observable structural circuit changes in the brain.

Table 4
Cluster sizes and location for voxels with statistically significantly (p < .05) greater FA in TE vs CC groups. Cluster locations were determined using the FSL atlastools tool and the JHU White-Matter Tractography Atlas. Each cluster was used as an independent binary mask to determine diffusivity profiles within each ROI. Values are mean ± standard deviation.

| Cluster Number | Region                                              | Average p-value | Number of Voxels | FA (CC) ± | FA (TE) ± |
|----------------|-----------------------------------------------------|-----------------|------------------|-----------|-----------|
| 5              | (L/R) Anterior thalamic radiation, (R) Corticospinal tract, (L/R) cingulate gyrus, Forceps minor, (L/R) Inferior fronto-occipital fasciculus, (R) Inferior longitudinal fasciculus, (L/R) Superior longitudinal fasciculus, (L/R) Uncinate fasciculus, (L/R) Superior longitudinal fasciculus (temporal part) | .027 ± 0.025 | 7980 ± 0.603 | 0.578 ± 0.029 |
| 4              | (L) Inferior fronto-occipital fasciculus            | .013 ± 0.026    | 859 ± 0.494      | 0.463 ± 0.025 |
| 3              | (L) Corticospinal tract, (L) Superior longitudinal fasciculus, (L) Superior longitudinal fasciculus (temporal part) | .026 ± 0.048 | 153 ± 0.434 | 0.408 ± 0.035 |
| 2              | (L) Superior longitudinal fasciculus                | .029 ± 0.062    | 60 ± 0.284       | 0.241 ± 0.068 |
| 1              | (L) Superior longitudinal fasciculus (temporal part) | .044 ± 0.066    | 4 ± 0.413        | 0.408 ± 0.088 |

Figure 4. Mean RD values of cluster 4 and 5 within each cluster in the CC and TE groups. Unpaired t-tests were performed in PRISM 8 to determine the changes in the diffusivity profile which contributed to the observed change in FA in each cluster. In Cluster 4 and 5 RD was lower in the TE group. Values shown are mean ± SEM.

Table 5
Mean DTI values within each cluster in the CC and PTSD groups. Unpaired t-tests were performed in PRISM 8 to determine the differences in the mean diffusivity profile which contributed to the observed change in FA in each cluster. Statistically significantly lower mean RD was found in the TE group in cluster 5 and 4 (Cluster 5: t(36) = 3.017, p = .0047, Cluster 4: t(36) = 3.422, p = .0016). No statistically significant differences were observed in AD. Statistically significant differences are bolded. Values are mean ± standard deviation.

| Cluster Number | AD (x 10^-3 mm^2/s) | RD (x 10^-4 mm^2/s) |
|----------------|---------------------|---------------------|
|                | CC                  | TE                  |
| 4              | 1.217 ± 0.334       | 1.233 ± 0.394       |
| 3              | 1.118 ± 0.317       | 1.131 ± 0.397       |
| 2              | 1.056 ± 0.436       | 1.084 ± 0.505       |
| 1              | 9.560 ± 0.991       | 9.441 ± 0.571       |
|                | 1.071 ± 0.123       | 1.052 ± 0.127       |

Limitations and Future Directions

Many DTI studies of PTSD in military personnel interrogate the interplay between mTBI and PTSD (Bazarian et al., 2013; Isaac et al., 2015). While the importance of this association need not be stated due to the high comorbidity of the two, it is equally important to determine the effect of PTSD in the absence of mTBI as it can be incredibly difficult to distinguish between PTSD and TBI due to overlapping symptom profiles, high-comorbidity, inaccurate symptom reporting, cultural stigma, and the avoidance of disclosing trauma and its effects (Rosen and Ayers, 2020). Thus, this study chose to exclude those who have a history of brain injury in order to isolate the microstructural mechanisms of PTSD and traumatic exposure. Future studies will build on these findings by including participants with a history of mTBI without PTSD, and participants comorbid mTBI and PTSD.

These findings should be interpreted with caveats. Within the PTSD group there were various comorbidities which existed that could not be directly controlled for such as hypothyroidism, chronic pain, and sleep apnea. It is unknown if these conditions were present prior to the development of PTSD, and represent a pre-existing risk factor, or whether exposure to trauma, and/or if these conditions developed as part of a response to stress. These conditions could affect both the DTI results and PTSD symptom burden in unpredictable ways. However, it is well known these conditions are common comorbidities in PTSD and excluding them would be difficult in a naturalistic sample such as ours.
Similarly, the PTSD group were being treated with a variety of medications, leading to potential confounds – however, treatment withdrawal would be unethical given common occurrences of suicidal ideation and suicidality in this population. Further, while our inclusion criteria stated PTSD symptoms have to be present for between 1 and 4 years prior to taking part in the study, the varying time since trauma, length of deployment and exposure to operational stress within the sample meant there was variation in the duration of illness, a confound that is present within the study.

PTSD symptom severity was also not acquired for those within the TE group, unfortunately removing the possibility of contrasting these scores between the TE and PTSD group and precluding us from conducting a whole-brain regression analysis for symptom severity of PTSD. Finally, it should be noted that early life traumatic events were not expressly screened or controlled for but could have been present, and are known to be a predictor of emergent PTSD in later life (Koenen et al., 2007).

Finally, during the DTI processing and cluster extraction, registration was performed between each subject and the JHU White-Matter Tractography Atlas template. This template has an isotropic resolution of 1mm and cluster 1 in both the PTSD vs CC comparison and in the TE vs CC comparison are in fact smaller than the native resolution of the acquired DTI images. While we report these clusters for the sake of completion, we note that the results from these clusters should be interpreted with caution.

Conclusions

This study demonstrated white matter alterations within the limbic system and various associated white matter tracts using DTI in military personnel exposed to persistent operational stress and trauma during deployment compared to healthy controls. There were no observed effects specifically related to PTSD. Elevated FA combined with reduced RD suggests an increase in adaptive oligodendrogenesis and myelination in these regions, and a putative structural basis for alterations in neural dynamics and hyperconnectivity seen in PTSD. Further, PTSD severity was found to not correlate with altered diffusivity profiles, indicating a complex pathophysiological connection between anatomy and symptom presentation that likely includes a complex interplay with functional circuitry not investigated here (Dunkley et al., 2014). Our research presents a link between the microstructural and biophysical brain circuits related to operational stress and traumatic exposure, particularly within a military context, but not necessarily any association with a clinical PTSD diagnosis per se.

Disclosure

The Authors declare that there is no conflict of interest or competing financial interests.

Author Statement

Patrick McCunn – Conceptualization, Methodology, Validation, Formal analysis, Data Curation, Writing - Original Draft, Visualization
J. Don Richardson – Conceptualization, Writing - Review & Editing, Visualization
Rakesh Jetly – Conceptualization, Writing - Review & Editing, Visualization
Benjamin Dunkley – Conceptualization, Data Curation, Writing - Review & Editing, Supervision, Project administration, Funding acquisition

Acknowledgements

The authors would like to thank Drs. Margot Taylor and Elizabeth Pang for their help with this study, Amanda Robertson and Marc Lalancette for help in the data collection, and Matthew Ventresca for help with neurocognitive scoring. This work was supported by funding from Defence Research and Development Canada (DRDC) (contract #W7719-135182/001/TOR) and Innovation for Defence Excellence and Security (IDEaS) Grant.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.jpsychres.2021.113797.

References

Abe, O., Yamazoe, H., Kasai, K., Yamada, H., Aoki, S., Iwami, A., Ohtani, T., Masutani, Y., Kato, N., Ohtomo, K., 2006. Voxel-based diffusion tensor analysis reveals aberrant anterior cingulum integrity in posttraumatic stress disorder due to terrorism. Psychiatry Res. - Neuroimaging. https://doi.org/10.1016/j.pscychresns.2006.01.004.

American Psychiatric Association, 2013. American Psychiatric Association, 2013. Diagnostic and statistical manual of mental disorders (5th ed.). American Journal of Psychiatry. https://doi.org/10.1176/appi.books.9780890425596.743053.

Andersson, J.L.R., Graham, M.S., Drobnjak, I., Zhang, H., Filippini, N., Bastiani, M., 2017. Towards a comprehensive framework for movement and distortion correction of diffusion MR images: Within volume movement. Neuroimage. https://doi.org/10.1016/j.neuroimage.2017.02.085.

Andersson, J.L.R., Graham, M.S., Zolודות, E., Sotirovopoulos, S.N., 2016. Incorporating outlier detection and replacement into a non-parametric framework for movement and distortion correction of diffusion MR images. Neuroimage. https://doi.org/10.1016/j.neuroimage.2016.06.058.

Andersson, J.L.R., Sotirovopoulos, S.N., 2016. An integrated approach to correction for off-resonance effects and subject movement in diffusion MR imaging. Neuroimage. https://doi.org/10.1016/j.neuroimage.2015.10.019.

Aschbacher, K., Mellon, S.H., Wolkowizki, O.M., Henn-Haase, C., Yehuda, R., Flory, J.D., Bierer, L.M., Abu-Amara, D., Marmar, C.R., Mueller, S.G., 2018. Posttraumatic stress disorder, symptoms, and white matter abnormalities among combat-exposed veterans. Brain Imaging Behav. https://doi.org/10.1007/s11682-017-9759-y.

Basser, P.J., Mattiello, J., Lebihan, D., 1994. MR Diffusion Tensor Spectroscopy and Imaging. Biophys. J. 66, 259–267.

Bastiani, M., Coratta, M., Fitzgilbert, S.P., Suri, S., Alfaro-Almagro, F., Sotirovopoulos, S.N., Jbabdi, S., Andersson, J.L.R., 2019. Automated quality control for within and between studies diffusion MRI data using a non-parametric framework for movement and distortion correction. Neuroimage. https://doi.org/10.1016/j.neuroimage.2018.09.073.

Bazarian, J.J., Donnelly, K., Peterson, D.R., Warner, G.C., Zhu, T., Zhong, J., 2013. The Bazarian, J.J., Donnelly, K., Peterson, D.R., Warner, G.C., Zhu, T., Zhong, J., 2013. The relation between posttraumatic stress disorder and mild traumatic brain injury acquired during operations enduring freedom and iraqi freedom. J. Head Trauma Rehabil. https://doi.org/10.1097/HTR.0b013e318256d343.

Bierer, L.M., Ivanov, I., Carpenter, D.M., Wong, E.W., Golier, J.A., Tang, C.Y., Yehuda, R., 2015. White matter abnormalities in Gulf War veterans with posttraumatic stress disorder: A pilot study. Psychoneuroendocriology. https://doi.org/10.1016/j.psyneuen.2014.11.007.

Blanchard, E.B., Jones-Alexander, J., Buckley, T.C., Forneris, C.A., 1996. Psychometric properties of the PTSD checklist (PCL). Behav. Res. Ther. https://doi.org/10.1016/0005-7967(96)00033-2.

Branco, C., Xu, Y., Brady, K., Pérez-Fuentes, G., Okuda, M., Wang, S., 2013. Comorbidity of posttraumatic stress disorder with alcohol dependence among US adults: Results from national epidemiological survey on alcohol and related conditions. Drug Alcohol Depend. https://doi.org/10.1016/j.drugalcdep.2013.04.016.

Breeze, K.T., Kuban, N., Carpenter, D.M., Wong, E.W., Golier, J.A., Tang, C.Y., Yehuda, R., 2015. White matter abnormalities in Gulf War veterans with posttraumatic stress disorder: A pilot study. Psychoneuroendocriology. https://doi.org/10.1016/j.psyneuen.2014.11.007.

Brewerton, T.A., Lucerini, S., 2000. Comorbidity of psychiatric disorders and posttraumatic stress disorder. J. Clin. Psychiatry.
Sweeney, J.A., Kuang, W., Li, J., Bi, F., Bi, F., Sweeney, J.A., Gong, Q., 2016. White Matter Abnormalities in Post-traumatic Stress Disorder Following a Specific Traumatic Event. ElBioMedicine. https://doi.org/10.1016/j.ebiom.2016.01.012.

Veraart, J., Novikov, D.S., Christiaens, D., Ades-aron, B., Sijbers, J., Fieremans, E., 2016. Denoising of diffusion MRI using random matrix theory. Neuroimage. https://doi.org/10.1016/j.neuroimage.2016.08.016.

Wang, C., Costanzo, M.E., Rapp, P.E., Darmon, D., Nathan, D.E., Bashirelahi, K., Pham, D.L., Roy, M.J., Keyser, D.O., 2017. Disrupted gamma synchrony after mild traumatic brain injury and its correlation with white matter abnormality. Front. Neurol. https://doi.org/10.3389/fneur.2017.00571.

Wang, W., Qian, S., Liu, K., Li, B., Li, M., Xin, K., Sun, G., 2016. Reduced white matter integrity and its correlation with clinical symptom in first-episode, treatment-naive generalized anxiety disorder. Behav. Brain Res. https://doi.org/10.1016/j.bbr.2016.08.017.

Wyllie, M.A., Haines, D.E., 2018. The Limbic System, in: Fundamental Neuroscience for Basic and Clinical Applications: Fifth Edition. https://doi.org/10.1016/B978-0-323-39632-5.00031-1.

Winkler, A.M., Ridgway, G.R., Webster, M.A., Smith, S.M., Nichols, T.E., 2014. Permutation inference for the general linear model. Neuroimage. https://doi.org/10.1016/j.neuroimage.2014.01.060.

Winklevski, P.J., Sabisz, A., Naumczyk, P., Jodzio, K., Szurowska, E., Szarmach, A., 2018. Understanding the physiopathology behind axial and radial diffusivity changes—what do we know? Front. Neurol. https://doi.org/10.3389/fneur.2018.00092.

Wynn, G.H., Ursano, R.J., 2016. Posttraumatic stress disorder. The Curated Reference Collection in Neuroscience and Biobehavioral Psychology. https://doi.org/10.1016/B978-0-12-809324-5.05378-5.

Yehuda, R., Hoge, C.W., McFarlane, A.C., Vermetten, E., Lanius, R.A., Nieveldt, C.M., Hobfoll, S.E., Koenen, K.C., Neylan, T.C., Hyman, S.E., 2015. Post-traumatic stress disorder. Nat. Rev. Dis. Prim. https://doi.org/10.1038/nrdp.2015.57.

Zhang, L., Zhang, Y., Li, L., Li, Z., Li, W., Ma, N., Hou, C., Zhang, Zhijun, Zhang, Zhiquang, Wang, L., Duan, L., Lu, G., 2011. Different white matter abnormalities between the first-episode, treatment-naive patients with posttraumatic stress disorder and generalized anxiety disorder without comorbid conditions. J. Affect. Disord. https://doi.org/10.1016/j.jad.2011.03.040.