



Contents lists available at ScienceDirect

Behavioural Brain Research

journal homepage: www.elsevier.com/locate/bbr



Research report

Both hemorrhagic and non-hemorrhagic traumatic MRI lesions are associated with the microstructural damage of the normal appearing white matter

Arnold Toth^{a,b,*}, Balint Kornyei^a, Noemi Kovacs^a, Tamas Rostas^b, Andras Buki^{a,d},
Tamas Doczi^{a,c,d}, Peter Bogner^{a,b}, Attila Schwarcz^{a,d}

^a Department of Neurosurgery, Pécs Medical School, Rét. u. 2, H-7623 Pécs, Hungary

^b Department of Radiology, Pécs Medical School, Ifjuság str. 13, H-7624 Pécs, Hungary

^c Diagnostic Center of Pécs, Rét. u. 2, H-7623 Pécs, Hungary

^d MTA-PTE Clinical Neuroscience MR Research Group, Hungary

HIGHLIGHTS

- Non-hemorrhagic lesion presence is related to decreased microstructural integrity.
- Basal ganglia area microbleeds are related to decreased microstructural integrity.
- Focal MRI pathology evaluation may substitute quantitative diffusion tensor imaging.

ARTICLE INFO

Article history:

Received 29 June 2016

Received in revised form 11 October 2016

Accepted 22 February 2017

Available online xxx

Keywords:

MRI

Traumatic brain injury

Susceptibility weighted imaging

Diffusion tensor imaging

Traumatic microbleeds

Diffuse axonal injury

ABSTRACT

Traumatic microbleeds (TMBs) and non-hemorrhagic lesions (NHLs) on MRI are regarded as surrogate markers of diffuse axonal injury. However, the actual relation between lesional and diffuse pathology remained unclear, since lesions were related to clinical parameters, largely influenced by extracranial factors.

The aim of this study is to directly compare TMBs, NHLs and their regional features with the co-existing diffuse injury of the normal appearing white matter (NAWM) as measured by diffusion tensor imaging (DTI).

Thirty-eight adults with a closed traumatic brain injury (12 mild, 4 moderate and 22 severe) who underwent susceptibility weighted imaging (SWI), T1-, T2 weighted and FLAIR MRI and routine CT were included in the study. TMB (on SWI) and NHL (on T1-, T2 weighted and FLAIR images) features and Rotterdam scores were evaluated. DTI metrics such as fractional anisotropy (FA) and mean diffusivity (MD) were measured over different NAWM regions. Clinical parameters including age; Glasgow Coma Scale; Rotterdam score; TMB and NHL features were correlated to regional NAWM diffusivity using multiple regression.

Overall NHL presence and basal ganglia area TMB load were significantly, negatively correlated with the subcortical NAWM FA values (partial $r = -0.37$ and -0.36 ; $p = 0.006$ and 0.025 , respectively).

The presence of any NHL, or TMBs located in the basal ganglia area indicates diffuse NAWM damage even after adjusting for clinical and CT parameters. To estimate DAI, a conventional lesional MRI pathology evaluation might at least in part substitute the use of quantitative DTI, which is yet not widely feasible in a clinical setting.

© 2017 Elsevier B.V. All rights reserved.

Abbreviations: AD, axial diffusivity; DAI, diffuse axonal injury; FA, fractional anisotropy; FLAIR, fluid attenuated inversion recovery; GCS, Glasgow Coma Scale; GRE, gradient recalled echo; MD, mean diffusivity; NAWM, normal appearing white matter; NHL, non-hemorrhagic lesion; RD, radial diffusivity; SWI, susceptibility weighted imaging; TBI, traumatic brain injury; TE, echo time; TI, inversion time; TMB, traumatic microbleed; TR, repetition time.

* Corresponding author at: Department of Neurosurgery, Rét. u. 2, H-7623 Pécs, Hungary.

E-mail address: prsarn@gmail.com (A. Toth).

<http://dx.doi.org/10.1016/j.bbr.2017.02.039>

0166-4328/© 2017 Elsevier B.V. All rights reserved.

Please cite this article in press as: A. Toth, et al., Both hemorrhagic and non-hemorrhagic traumatic MRI lesions are associated with the microstructural damage of the normal appearing white matter, Behav Brain Res (2017), <http://dx.doi.org/10.1016/j.bbr.2017.02.039>

1. Introduction

Traumatic brain injury (TBI) constitutes a public health problem worldwide [1]. Diffuse axonal injury (DAI) is a substantial pathological component of brain injury, and is highly related to outcome [2]. DAI (or traumatic axonal injury) can occur both in severe and mild TBI [2] at a certain extent. However, in consequence of its microscopic range, it is basically “invisible” to standard TBI imaging protocols.

Interest in the application of susceptibility weighted imaging (SWI) [3] in TBI has recently been increasing rapidly, since it has been shown to be a very sensitive imaging method for the detection of the possible DAI-associated hemorrhagic foci, also known as traumatic microbleeds (TMBs) [4]. SWI is a fully velocity-compensated, high-resolution 3D gradient echo sequence that uses magnitude and filtered-phase information, both separately and in combination, and is therefore able to create a strong contrast for the susceptibility effects of microbleeds [3]. Other imaging methods, such as CT, T2- and T2*-weighted MRI or FLAIR are also able to depict punctual DAI-related lesions, though less reliably than SWI [5–8]. SWI allows patients with a DAI to be dichotomized as hemorrhagic or non-hemorrhagic, and this has been proposed to be of clinical relevance [9,10]. Moreover, the number, localization, type and volume of TMBs have been linked to the severity of clinical state as well as the outcome [7,8,11–19].

Another advanced MRI method, diffusion tensor imaging (DTI), has been proposed to sensitively detect white matter damage itself by a very extensive body of literature [20]. DTI does not only detect Brownian diffusion extent, but also diffusion directionality (anisotropy) and direction [21,22]. Data suggest that the different DTI parameters, such as fractional anisotropy (FA), mean diffusivity (MD), axial diffusivity (AD) and radial diffusivity (RD) may reflect the damage of the white matter [20]. Though the detected actual traumatic DTI parameter alterations vary largely across studies, in the relatively late phases of TBI, studies mostly agree that FA is decreased, while MD is increased due to axonal pathology. However, to date DTI assessment of axonal injury has been based almost exclusively on group analyses, with the use of post-processing computation and statistics [20]. The main reasons limiting the interpretation of the DTI data at a subject level are the high inter-subject variations (even within the healthy population), and the relatively low specificity of DTI to axonal injury.

In contrast to DTI, SWI might be easily applied at an individual level. A conventional, qualitative morphological assessment might define TMB load, extent and anatomical distribution per patient. Therefore, SWI appears to be the most feasible advanced MRI method for the indirect evaluation of axonal damage in a clinical environment, because group analysis and statistics are not necessary for its interpretation.

However, thus far no consensus has been reached concerning the optimal clinical use of SWI, T2*GRE or other MRI modalities including the ones depicting non-hemorrhagic lesional pathology (NHL), in TBI. Studies largely agree that TMBs are related to the clinical severity or outcome, but they are heterogeneous in the aspects of the investigated population size, injury severity, image acquisition method, outcome assessment method, as well as the detection and definition of TMB [7,8,11–19]. Though TMBs are accepted as surrogate markers of DAI, it is of note that they are rather due to microvascular injury, than axonal injury, and these two are not necessarily strictly related. The clinical state of the patient, however is probably rather dependent on the diffuse axonal-, and not microvascular injury.

The main problem in understanding the role of SWI in TBI can be summarized as: SWI data were related to clinical parameters that are largely influenced by extracranial factors and not directly to the actual axonal pathology. For example, Glasgow Coma Scale (GCS)

may be influenced by alcohol or drug intoxication and accompanying neurological and psychiatric diseases, outcome scores as the Glasgow Outcome Score [23] may be influenced by the presence of extra-cranial injuries, the therapy and rehabilitation process, and the adherence of the patient, etc. As a consequence, it is still unclear if TMBs, or NHLs, and which lesion parameters (number, volume or localization) are indicating the higher rate of axonal damage.

Theoretically, these issues might be resolved when TMB and NHL parameters are correlated with the simultaneously acquired DTI data, since DTI has been widely accepted to sensitively indicate microstructural damage [20], however, because of its technical features and necessary group analyses, up to now it has been not widely used in a clinical setting.

The purpose of this study is to directly compare the diffuse microstructural injury of the normal appearing white matter as measured by DTI with the clinically readable hemorrhagic and non-hemorrhagic traumatic lesion features in TBI patients, to better understand the association between diffuse axonal injury and lesional pathologies.

2. Methods

2.1. Subjects

This retrospective study was conducted on a database consisting of 66 adults with a closed TBI who underwent the MR imaging protocol (see Section 2.2) from the trauma center at the University of Pécs. Exclusion criteria included a previous documented TBI, any known neurological (including migraine) or psychiatric disease, uncontrolled hypertension, a history of anticoagulant therapy or coagulopathy, or a severe mass lesion or brain distortion precluding reliable DTI analysis. These criteria allowed the inclusion of 38 patients, for clinical and demographic details see Table 1.

20 age- and sex-matched healthy volunteers following the same exclusion criteria were recruited as a control group in the study (see Table 1).

Ethical approval was received from the Institutional Review Board (IRB). Written informed consent was obtained from all the participants (or their legally authorized representatives) in the study.

2.2. Imaging protocol

An acute (<24 h) CT scan was performed for all TBI patients according to the institutional TBI CT protocol. CT scan was not performed for the healthy control group.

MRI data were acquired in the chronic phase after the injury (min. 1 month, average 2 years $\pm$ 15 months after the injury). MRI was performed on a Magnetom[®] TIM Trio[®] 3 Tesla scanner (Siemens, Erlangen, Germany) with a 12 channel standard head coil. The control group underwent the same MRI protocol as the TBI patients:

The sequences consisted of a high-resolution T1-weighted scan (MP-RAGE), T2-weighted scan, FLAIR, DTI and SWI. After the localizer scanning for proper orientation, shimming was carried out to maximize field homogeneity.

T1-weighted high-resolution images were obtained using a three-dimensional (3D) MP-RAGE sequence (TR/TI/TE = 1900/900/3.41 ms; flip angle = 9°; 160 axial slices; slice thickness = 0.94 mm; no inter-slice gap; field of view [FOV] = 210 mm $\times$ 240 mm; matrix size = 224 $\times$ 256; receiver bandwidth = 180 Hz/pixel).

T2-weighted images were acquired using a turbo spin echo sequence (TR/TE = 6000/93 ms; flip angle = 120°; 30 axial slices; slice thickness = 4 mm; no inter-slice gap;

Table 1
Demographic data of patients and controls.

Data	Patients (n = 38)	Controls (n = 20)	p
Age (y, SD, min–max)			
Men	35 (13.6) 18–68	38.4 (16.2) 19–72	0.6 ^a
Women	48 (22.1) 20–83	45.4 (14.2) 24–60	0.77 ^a
Total	39.7 (17.4) 18–83	40.9 (15.58) 19–72	0.79 ^a
No. of women	12	7	0.78 ^b
Severity (mild/moderate/severe)	13/4/21		

^a Two-tailed *t*-test.^b Chi-square test.

FOV = 193 mm × 220 mm; matrix size = 280 × 320; receiver bandwidth = 220 Hz/pixel.

FLAIR images were acquired using: TR/TI/TE = 15,700/2750/105 ms; flip angle = 120; 100 axial slices; slice thickness = 1.5 mm; no inter-slice gap; field of view [FOV] = 220 mm × 220 mm; matrix size = 192 × 192; receiver bandwidth = 400 Hz/pixel.

DTI was performed using a two dimensional single-shot diffusion-weighted, spin-echo, echo-planar imaging (EPI) sequence. DTI was achieved using 20 optimum non-collinear encoding directions with a diffusion weighting of $b = 700 \text{ s/mm}^2$, and a single volume was collected with no diffusion gradients applied (TR/TE = 8500/90 ms; 60 axial slices, slice thickness = 2 mm; FOV = 208 mm × 256 mm; matrix size = 208 × 256; receiver bandwidth = 1563 Hz/pixel; 4 averages. The total acquisition time was 12 min). Advanced shimming was performed before the DTI acquisition to optimize the homogeneity of the magnetic field across the brain and to minimize EPI distortions.

The 3D gradient echo SWI sequence was run with the following parameters: (TR/TE = 27/20 ms; 72 axial slices; slice thickness = 1.5 mm; flip angle = 15; matrix size = 182 × 256; FOV = 173 mm × 230 mm; receiver bandwidth = 120 Hz/pixel).

Subjects were positioned supine in the scanner. Foam cushioning was used to immobilize the head within the coil to minimize motion degradation.

2.3. Image analyses – focal pathology

TMBs were identified on the SWI scans by two independent raters (A.T. and a board certified neuroradiologist T.R.), blinded to the clinical data. TMBs were defined as punctuate, ovoid or curvilinear hypointensities located in the white matter or white-gray matter border that are clearly not consistent with blood vessels or artifacts. To aid such exclusion of mimicking hypointensities and a more precise anatomical localization, both the high resolution T1 weighted- and the T2 images were registered to the SWI images using the FMRIB's Linear Image Registration Tool (FLIRT) [24], which allowed a multi-modal assessment of TMB sites. TMBs associated with T1 or T2 signal alteration were not excluded. TMBs adjacent to contusions, intraventricular hemorrhage or bone-air interface artifacts (e.g. near mastoid process) and external ventricular drain were excluded. The following TMB features were recorded: TMB presence; number of TMBs in the brainstem; "basal ganglia area" (including the caudate nucleus, thalamus, putamen, pallidum, subthalamic nucleus, and all white matter located in this region); corpus callosum; and "subcortical white matter" (including the corona radiata and centrum semiovale); and overall TMB number. Afterwards, TMBs identified by both raters were automatically included in the final registry of TMBs per scan. As a second step, for TMBs detected only by one rater, raters took a second look and decided in mutual consent to include or not in the final TMB registry. FSL "mask" tool (FSL [25], FMRIB's Software Library, www.fmrib.ox.ac.uk/fsl, Oxford) and manual tracing was used to create a

TMB map for each patient, this was used to exclude TMB sites from DTI analyses (see Section 2.4).

The detection, recording and masking of the non-hemorrhagic DAL associated lesions (NHL) on T1-, T2-weighted and FLAIR images were performed in the same way as of the TMBs except the lesion definitions. NHLs were defined as lesions of mixed intensity on T1 weighted images, and hyperintense lesions on T2 and FLAIR images. The raters made a cautious effort using the multi-modal approach (as mentioned above) to exclude non-specific scars, lacunes, chronic hypertensive or ischemic microvascular lesions, enlarged Virchow-Robin spaces, etc.

Inter-rater reliability of the first-step TMB and NHL measurement was determined using Intraclass Correlation Coefficient [26]. Two-way model, absolute agreement type was applied, single measurement reliability was calculated.

The standard acute CT scans were read by a board certified neuroradiologist (T.R.) to assess the Rotterdam score [27] for each patient.

2.4. Image analysis – DTI

Initial diffusion image processing was performed to generate fractional anisotropy (FA), mean diffusivity (MD), axial diffusivity (AD = $L1$) and radial diffusivity (RD = $(L2 + L3)/2$) maps using the FDT (FMRIB's Diffusion Toolbox) tool [25]. The steps included eddy current correction and motion correction using a 12-parameter affine registration to a reference volume (i.e. non-diffusion weighted data, $b = 0 \text{ s/mm}^2$), averaging of the four sets of 20 diffusion directions, and automated brain extraction with brain extraction tool [28], which was manually supervised to avoid incorrect brain extraction. Diffusion data were then fed into DTIFit [25] to calculate the diffusion tensor model for each brain voxel and subsequently compute FA, MD, AD and RD values from the tensor's three eigenvalues [29].

To assess regional mean FA values, FA maps were registered to the high-resolution T1 weighted MPRAGE images (using FLIRT [24]) and then, blinded to clinical data, masks were created in all possible slices by manual tracing (B.K., FSL tools [25]) based on anatomical borders ending up in a 3D mask for the brainstem, corpus callosum, and the subcortical white matter (including the corona radiata and the centrum semiovale) (see Fig. 1). These masks were overlaid on the co-registered T1 images presenting a clearer gray matter–white matter interface and anatomical substrates, and were corrected in the axial, sagittal and coronal image planes. To exclude lesion sites, the TMB and NHL maps (see focal pathology image analyses) were then subtracted from this mask in the MPRAGE space using co-registration if necessary (i.e. for SWI, FLAIR and T2 based lesion maps). The so created mask was then overlaid on the native space FA, MD, AD and RD maps, to calculate the corresponding DTI parameter averages (also see Fig. 1). All co-registration steps were manually controlled.

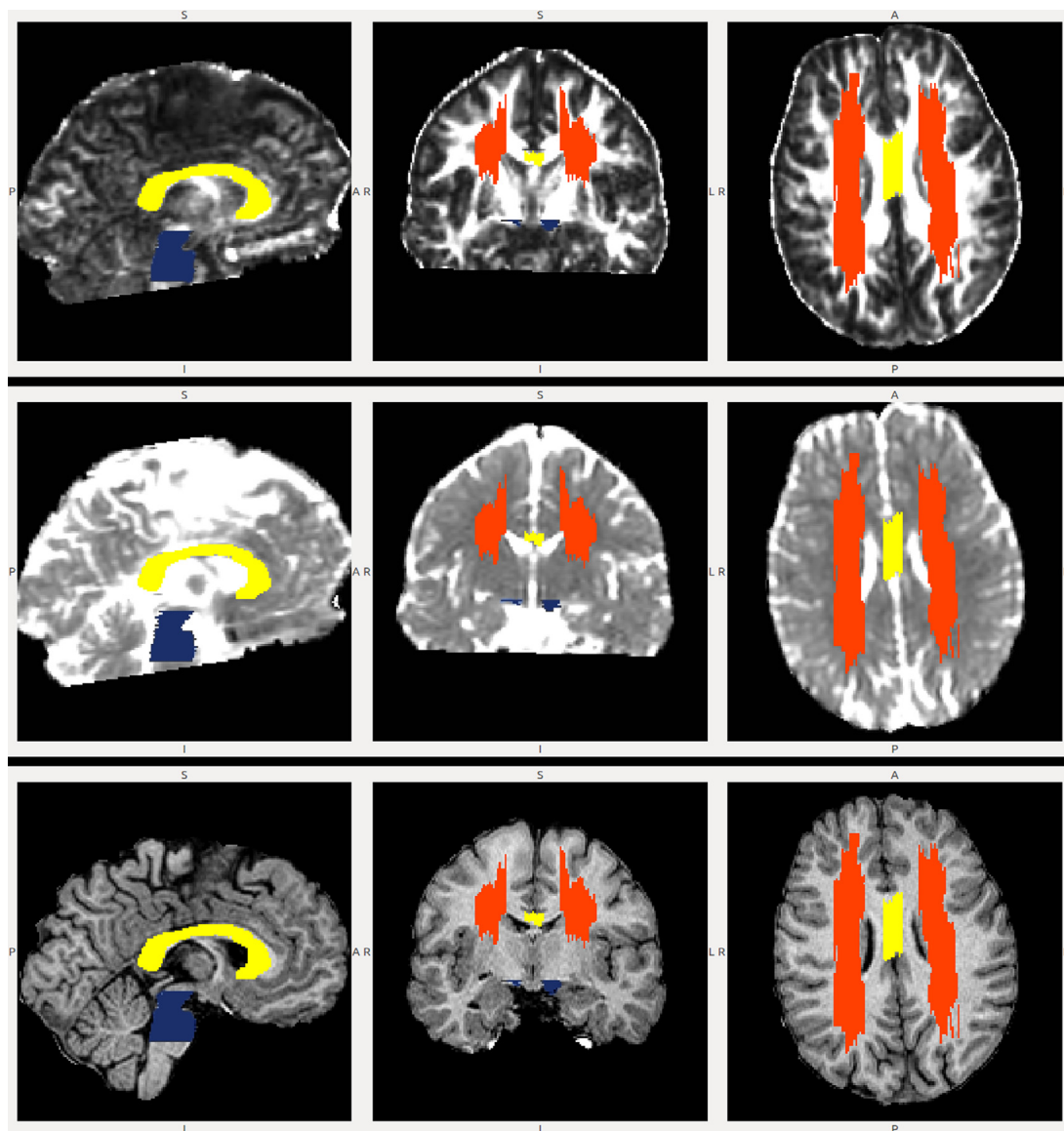


Fig. 1. Illustration of the region of interests for the measurement of regional DTI parameters. Masks: red = subcortical white matter; yellow = corpus callosum; blue = brainstem. Rows: 1. = masks overlaid on FA maps; 2. = masks overlaid on MD maps; 3. = masks overlaid on MPRAGE. Columns: 1. = sagittal plane; 2. = coronal plane; 3. = axial plane. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of the article.)

2.5. Statistical analyses

MedCalc 15.4 (MedCalc Software bvba, Ostend, Belgium) [30] was used for descriptive and analytical statistics. Multiple regression model (stepwise method, variable entered if $p < 0.05$, variable removed if $p > 0.1$) was applied as follows: independent factors (potentially associated with white matter damage and DTI parameters): acute Glasgow Coma Scale – GCS, age, presence or absence of TMBs and NHLs, number of TMBs and NHLs in the brainstem, area of basal ganglia, corpus callosum, subcortical white matter, and Rotterdam scores. Dependent variables: regional (subcortical white matter, corpus callosum and brain stem) DTI parameter (FA, MD, AD, RD) averages.

To check if TBI induced the suspected DTI alterations, the healthy control DTI data were compared to the TBI DTI data.

In order to reveal relations between the CT findings, MRI and clinical variables, Rotterdam scores were also related to age, GCS, TMB and NHL data.

Beyond checking the effect of age on DTI parameters (see above), relationship between age and GCS, and lesion parameters were checked, to exclude possible interactions and to reveal if lesion occurrence is related to age. NHL and TMB data were compared and correlated. GCS was also related to NHL and TMB data, to assess the relationship between acute clinical severity and lesion occurrence.

Student's *t*-test was used for normal distributed data comparisons. Spearman's rank correlation was used to test association when ordinal variables were included. Chi-square test was used for categorical variables. A *p*-value of < 0.05 was considered statistically significant.

3. Results

3.1. Lesion load

Out of the 38 patients, TMBs on SWI were found in 14 cases (in 1 out of 13 (7.7%) mild (GCS = 14–15), in 1 out of 4 (25%) moderate (GCS = 9–13), and in 12 out of 21 (57.1%) severe (GCS 3–8)

Table 2
TMB^a and NHL^b distribution over regions.

	Subcortical WM	Corpus callosum	Basal ganglia area	Brainstem	Overall
TMB number (SWI)					
Mild (1/13) ^c	1	0	0	0	1
Moderate (1/4)	3	1	0	0	4
Severe (12/21)	80	21	24	8	133
Overall (14/38)	84	22	24	8	138
NHL number (T1, T2 and FLAIR)					
Mild (2/13)	1	0	0	0	1
Moderate (1/4)	7	0	0	0	7
Severe (11/21)	20	2	3	1	26
Overall (14/38)	28	2	3	1	34

^a Traumatic microbleed.

^b Non-hemorrhagic lesion.

^c (Affected/all).

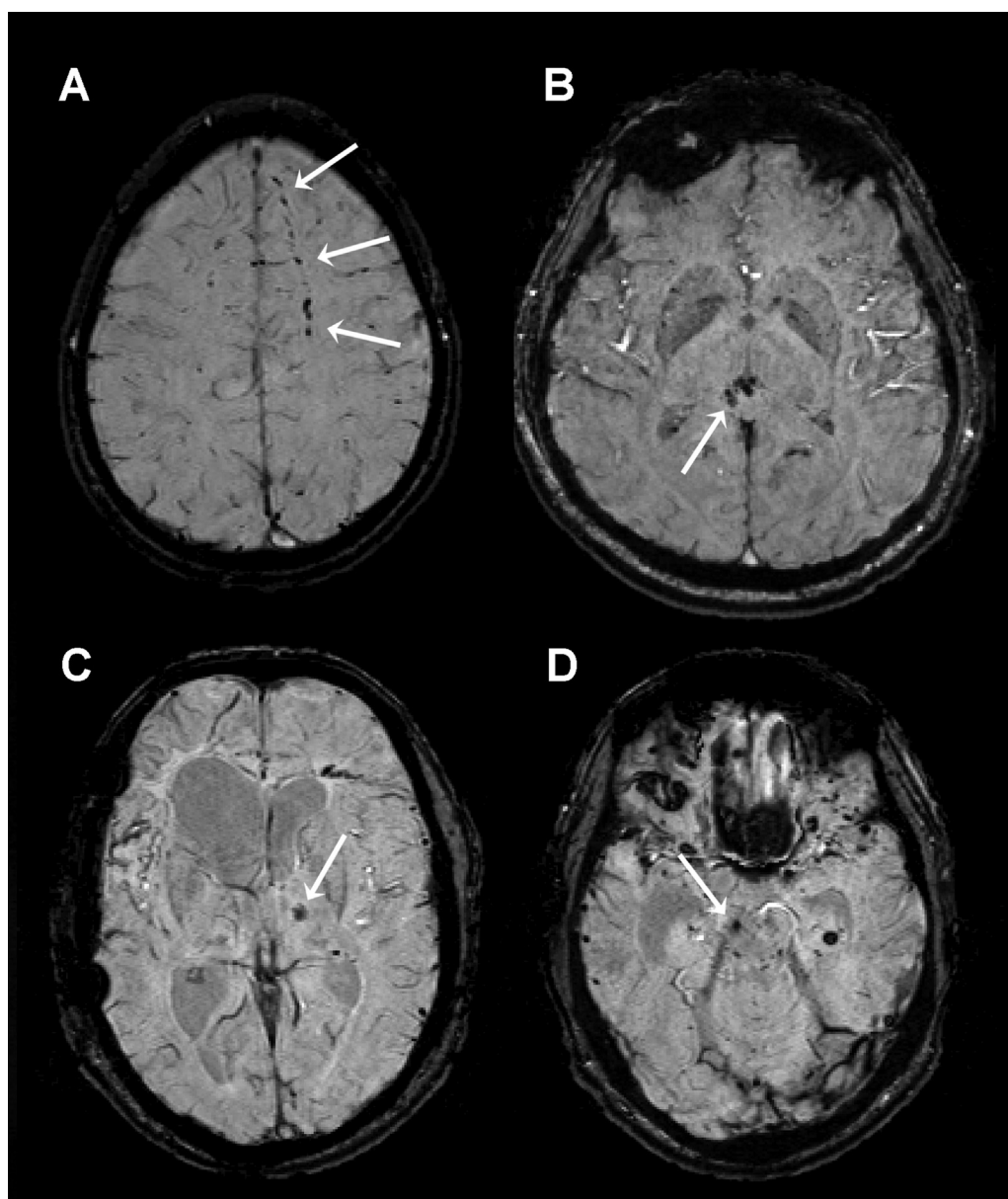


Fig. 2. Illustration of TMBs revealed by SWI in the investigated regions. TMBs (indicated by white arrows) can be seen in (A) subcortical white matter, (B) corpus callosum, (C) basal ganglia area, (D) brainstem.

TBI patients), see Table 2. TMB count range per patient was 1–43 (mean = 13, SD = 14.58). Out of all patients, overall 138 TMBs were

found, in 10 cases in the subcortical white matter (sum 84 TMBs), in 8 cases (sum 22 TMBs) in the corpus callosum, in 8 cases in the area

of basal ganglia (sum 24 TMBs) and in 4 cases in the brainstem (sum 8 TMBs), often combined, see Table 2. Fig. 2 shows TMB examples in the different regions. No TMBs were found in the healthy controls.

Out of the 38 patients, NHLs on T1, T2 weighted and FLAIR images together were found in 14 cases as well (in 2 out of 13 (15.4%) mild, in 1 out of 4 (25%) moderate, and in 11 out of 21 (52.4%) severe TBI patients), see Table 2. NHL count range per patient was 1–7 (mean = 2.78, SD = 2.12). Overall 34 NHLs were found, in 12 cases in the subcortical white matter (sum 28 NHLs), in 1 case (sum 2 NHLs) in the corpus callosum, in 3 cases in the area of basal ganglia (sum 3 NHLs) and in 1 case in the brainstem (sum 1 NHL), often combined, see Table 2. No NHLs were found in the healthy controls.

Overall TMB and NHL number per patient showed a significant positive correlation ($\rho = 0.54$, $p = 0.004$). Fig. 3 shows NHL examples in the different regions.

Neither TMB nor NHL parameters were significantly related to age.

The rate of lesions detected by only one rater in the first step, and debated in a second step by both raters whether including them or not in the final lesion registry was 14% of all detections for rater 1, and 8% of all detections for rater 2. The intraclass correlation coefficient between the two raters for the first step of detections was 0.983. Discrepancies mainly arise in case of adjacent lesions, as regarding them one large or two separate smaller lesions.

3.2. DTI parameters

In the TBI patients, the following regional DTI parameter averages were measured (see Table 3): FA: subcortical white matter = 0.41203 (SD = 0.0367), corpus callosum = 0.5666 (SD = 0.1027), brainstem = 0.3768 (SD = 0.0499); MD: subcortical white matter = 0.847×10^{-5} mm²/s (SD = 0.06597), corpus callosum = 1.116×10^{-5} mm²/s (SD = 0.251), brainstem = 1.164×10^{-5} mm²/s (SD = 0.206). AD: subcortical white matter = 1.2357×10^{-3} mm²/s (SD = 0.086), corpus callosum = 1.8654×10^{-3} mm²/s (SD = 0.2384), brainstem = 1.6764×10^{-3} mm²/s (SD = 0.2311); RD: subcortical white matter = 0.6578×10^{-3} mm²/s (SD = 0.0705), corpus callosum = 0.8339×10^{-3} mm²/s (SD = 0.2111), brainstem = 1.012×10^{-3} mm²/s (SD = 0.2133).

In the age and sex matched healthy controls, the following regional DTI parameter averages were measured (see Table 3): FA: subcortical white matter = 0.4676 (SD = 0.0353), corpus callosum = 0.7049 (SD = 0.0357), brainstem = 0.4189 (SD = 0.0458); MD: subcortical white matter = 0.7638×10^{-5} mm²/s (SD = 0.0306), corpus callosum = 0.8986×10^{-5} mm²/s (SD = 0.1478), brainstem = 1.0366×10^{-5} mm²/s (SD = 0.2389). AD: subcortical white matter = 1.1899×10^{-3} mm²/s (SD = 0.2223), corpus callosum = 1.7379×10^{-3} mm²/s (SD = 0.044), brainstem = 1.5857×10^{-3} mm²/s (SD = 0.1295); RD: subcortical white matter = 0.6113×10^{-3} mm²/s (SD = 0.05601), corpus callosum = 0.6915×10^{-3} mm²/s (SD = 0.0558), brainstem = 0.8985×10^{-3} mm²/s (SD = 0.1312).

Two-tailed Student's *t*-test yielded significant differences between TBI patients and controls both in FA and MD values in all regions, see Table 3 (*p* values were the following for FA: subcortical white matter < 0.0001, corpus callosum < 0.0001, brainstem = 0.001; for MD: subcortical white matter = 0.001, corpus callosum = 0.0004, brainstem = 0.02). Regarding AD and RD, significant difference between TBI patients and controls was only found in the corpus callosum RD averages (*p* values were the following for AD: subcortical white matter = 0.3, corpus callosum = 0.10, brainstem = 0.24; for RD: subcortical white matter = 0.059, corpus callosum = 0.04, brainstem = 0.12).

3.3. Multiple regression analysis of clinical, imaging parameters and DTI data

Age, NHL presence, and basal ganglia area TMB load was significantly, negatively related to the subcortical white matter FA values (partial $r = -0.45$, -0.38 , -0.36 ; $p = 0.007$, 0.03, 0.03, respectively, coefficient of determination $R^2 = 0.4$).

GCS was significantly, positively correlated to corpus callosum FA values (partial $r = 0.57$, $p = 0.0002$, coefficient of determination $R^2 = 0.33$), and negatively correlated to corpus callosum RD values (partial $r = -0.42$, $p = 0.0092$, coefficient of determination $R^2 = 0.3$).

Age was significantly, positively correlated to both subcortical white matter and corpus callosum MD values (partial $r = 0.64$, 0.58, respectively; $p < 0.0001$ for both, coefficient of determination $R^2 = 0.45$ and 0.34, respectively). Age was also significantly, positively correlated to both subcortical white matter and corpus callosum AD values (partial $r = 0.58$ and 0.73, respectively, $p = 0.0002$ and $p < 0.0001$, respectively, coefficient of determination $R^2 = 0.33$ and 0.53, respectively), and to both subcortical white matter and corpus callosum RD values (partial $r = 0.49$ and 0.35; p values = 0.002 and 0.036; coefficient of determination $R^2 = 0.23$ and 0.3, respectively).

No other significant correlations were found between clinical, imaging variables and DTI values.

For details, see Table 4.

3.4. Rotterdam score

Rotterdam score was not independently related to any DTI parameters (see Section 3.3). Rotterdam score was significantly, negatively related to GCS ($\rho = -0.615$, $p < 0.0001$) and positively to TMB presence ($\rho = 0.361$, $p = 0.026$), but not NHLs, nor age.

3.5. GCS

GCS was an independent factor significantly associated with corpus callosum FA (see Section 3.3, and Table 4). GCS was also significantly, negatively related to the presence of TMBs and NHLs (TMBs: $\rho = -0.43$, $p = 0.006$; NHLs: $\rho = -0.423$, $p = 0.0081$). No significant relation was found between GCS and age.

4. Discussion

This study aimed to reveal the significance of TMBs seen on SWI and NHLs seen on conventional MRI in TBI by directly comparing them to the normal appearing white matter damage rate, i.e. the DTI alterations. Such direct comparison might help understanding the relationship between the microvascular injury linked TMBs and the co-existing diffuse axonal damage. An attempt was made to assess regional features, and to adjust for potential clinical influencing factors as GCS and age, and CT factor defined by the Rotterdam score.

DTI is regarded to be one of the most sensitive MRI methods in detecting axonal damage [20]. The DTI derived metric FA has been shown by the vast majority of DTI studies to be decreased, while MD to be increased after various severities and timespans after brain injury, that has been explained as an effect of axonal/myelin disintegration [10,20,31–40]. This is in line with our present finding of the significantly decreased FA and increased MD in TBI patients over all regions when compared to age and sex matched healthy controls. By a somewhat lower number of studies investigating RD as well, RD was found to be increased after injury [41–44], that was also supported by our finding of the significantly increased RD values of the corpus callosum, and near-significantly increased RD values of the subcortical white matter of the TBI patients compared to healthy controls. Relying on these data, in the present study,

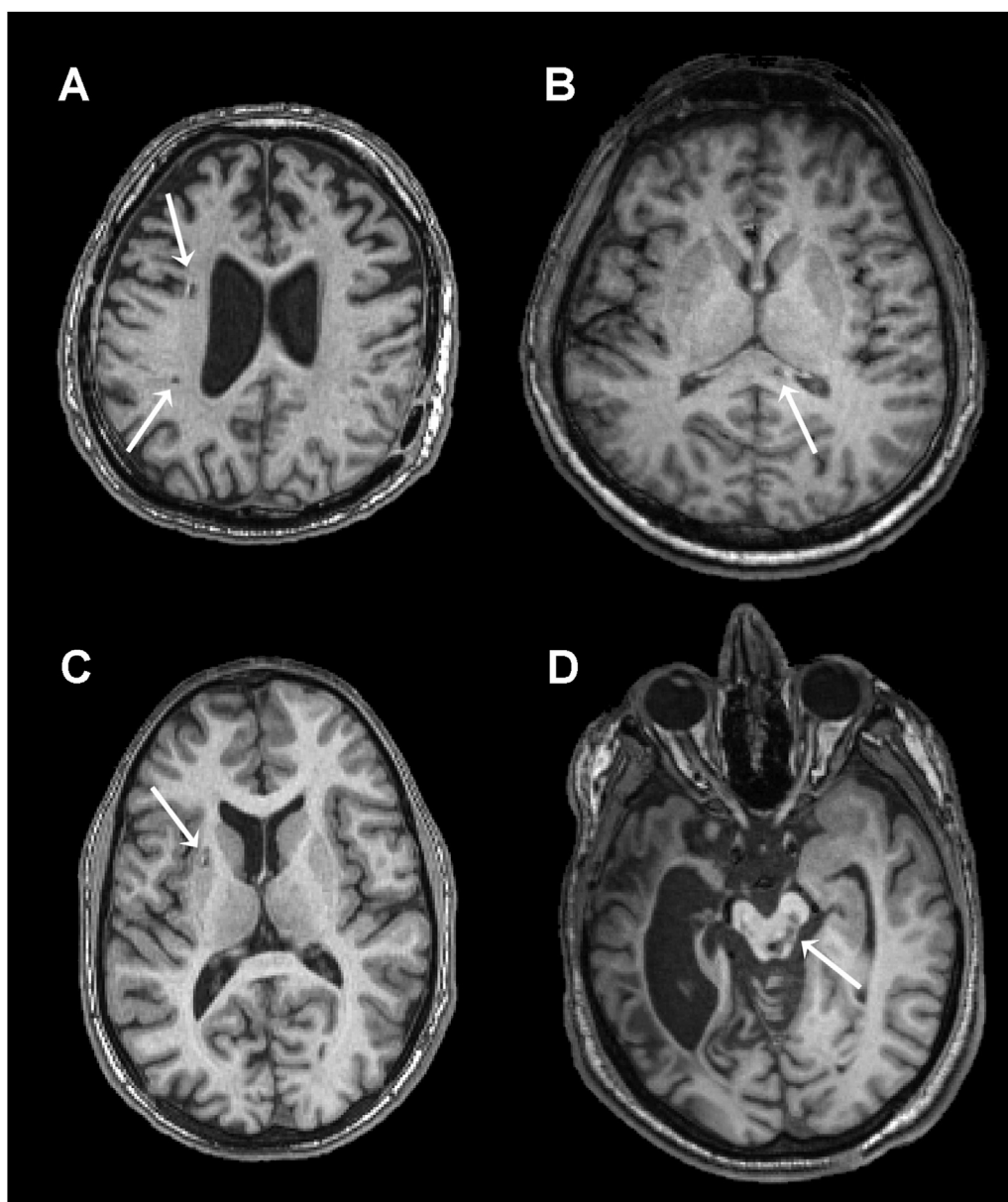


Fig. 3. Illustration of NHLs revealed by MPRAGE in the investigated regions. NHLs (indicated by white arrows) can be seen in (A) subcortical white matter, (B) corpus callosum, (C) basal ganglia area, (D) brainstem.

Table 3

DTI^a parameter averages over regions in TBI^b patients and controls.

	Subcortical WM ^c			Corpus callosum			Brainstem		
	TBI	Control	<i>p</i> ⁱ	TBI	Control	<i>p</i>	TBI	Control	<i>p</i>
FA ^d	0.41203 (0.0367) ^h	0.4676 (0.0353)	<0.0001	0.5666 (0.1027)	0.7049 (0.0357)	<0.0001	0.3768 (0.0499)	0.4189 (0.0458)	0.001
MD ^e	0.847 (0.06597)	0.7638 (0.0306)	0.001	1.116 (0.251)	0.8986 (0.1478)	0.0004	1.164 (0.206)	1.0366 (0.2389)	0.02
AD ^f	1.2357 (0.086)	1.1899 (0.2223)	0.3	1.8654 (0.2384)	1.7379 (0.044)	0.1	1.6764 (0.2311)	1.5857 (0.1295)	0.24
RD ^g	0.6578 (0.0705)	0.6113 (0.05601)	0.059	0.8339 (0.2111)	0.6915 (0.0558)	0.04	1.012 (0.2133)	0.8985 (0.1312)	0.12

^a Diffusion tensor imaging.

^b Traumatic brain injury.

^c White matter.

^d Fractional anisotropy.

^e Mean diffusivity.

^f Axial diffusivity.

^g Radial diffusivity.

^h Numbers in parentheses indicate standard deviations.

ⁱ Two-tailed Student's *t*-test, MD values: 10⁻⁵ mm²/s, AD and RD values: 10⁻³ mm²/s.

Table 4
Factors related to regional DTI parameters in multiple regression model.

		Independent variables	Coefficient of determination R ²	Std. error	r _{partial}	t	p
FA	Corona radiata/centrum semiovale	Age	0.405	0.0002834	−0.4509	−2.902	0.0066
		NHL presence		0.01149	−0.3762	−2.332	0.0259
		TMB load in basal ganglia area		0.004058	−0.3607	−2.221	0.0333
		Corpus callosum	0.327	0.002988	0.5715	4.178	0.0002
MD	Corona radiata/centrum semiovale	Age	0.451	4.763×10^{-7}	0.6382	4.904	<0.0001
		Corpus callosum	0.343	1.312×10^{-6}	0.5854	4.332	<0.0001
AD	Corona radiata/centrum semiovale	Age	0.33	5.128×10^{-7}	0.57	4.237	0.0002
		Corpus callosum	0.53	1.562×10^{-6}	0.727	6.36	<0.0001
RD	Corona radiata/centrum semiovale	Age	0.24	1.191×10^{-6}	0.49	3.35	0.0019
		Corpus callosum	0.3	1.24×10^{-5}	−0.42	−2.755	0.0092
		Age		3.35×10^{-6}	0.35	2.18	0.036

FA, fractional anisotropy; MD, mean diffusivity; AD, axial diffusivity; RD, radial diffusivity; NHL, non-hemorrhagic lesion; TMB, traumatic microbleed; GCS, Glasgow Coma Scale.

decreased FA, increased MD and RD are accepted as indicators of axonal damage rate. The fact that AD showed no relations either to TBI severity measures or imaging markers, moreover showed no significant difference when compared to healthy controls in our study, is congruent with several previous studies that found AD to be unaffected by TBI, or get normalized in the chronic phase of TBI [41,42,45].

Both the presence of NHLs at any region (in T1-, T2-weighted and FLAIR images), and the TMB lesion load at the basal ganglia area (on SWI) were independently, significantly related to low subcortical white matter FA values, i.e. to a diffuse white matter damage of this region. Overall TMB presence, or overall TMB count itself was not independently related. This implies that TMB formation at other sites (e.g. subcortical white matter) might be rather dependent on the patients' individual microvascular susceptibility to shears, and not to the actual axonal damage, while TMB development in the basal ganglia area might be due to a certain type and rate of mechanic stress that is associated with a diffuse injury of the white matter of higher regions. Or, TMBs at other sites, though might be prominently present at a high number and overall volume, are highly inter-related with other factors as e.g. GCS, and alone do not further discriminate the FA values. These findings are somewhat contradictory to studies in which SWI (or T2* GRE) lesion number or volume, irrespective from localization, has been shown to be associated with outcomes, and therefore probably with axonal injury [7,8,11,13,15,46]. However, most of these studies were conducted on children and did not focus on brain regions. A study investigating multi-modal MRI TBI outcome prediction has shown that besides T2 weighted imaging and FLAIR, SWI alone did not discriminate by outcome [47]. Studies focusing on regional aspects of TMBs have underscored that TMBs in deeper brain regions are predicting poorer outcomes, or are associated with a poorer actual clinical state/injury severity [12,16,48]. Keeping the Adams' localization based DAI grading in mind, this is not surprising [49]. When adjusting for age, GCS, Rotterdam scores, as performed in this study, subcortical white matter and corpus callosum TMBs, though they occur at a much higher rate than NHLs do (as presented in this study as well), do not seem to be able to further discriminate the underlying microstructural damage of the normal appearing white matter. Age was not correlated with TMBs in our study, which supports that the recorded TMBs were not significantly biased by cerebral microbleeds of other pathological origin (aging/small vessel disease). This was important since small vessel disease related cerebral microbleeds themselves are known to be associated with a diffuse poorer microstructural integrity [50].

The linkage between the presence of NHL (even if a single NHL is present on T1-, T2-weighted or FLAIR images) and the lower FA of the subcortical white matter suggests that the appearance of NHLs might be more directly related to white matter dam-

age than the TMBs', and though they occur less frequently, their presence is prominently indicative of an underlying diffuse axonal pathology beyond the actual focal injury. This might explain the previous studies finding a relationship between focal parenchymal MRI abnormalities and injury severity and outcome [11,47,51–53]. It has to be kept in mind though, that in the present study the NHLs were assessed by combining three modalities (T1-, T2-weighted and FLAIR images), so it is not a single modality that catches up with similar information provided by SWI. No attempt was made to assess these modalities separately. One reason for this was to keep the study robust, other that the evaluation of the NHLs seemed to be most reliable when adding up the information from the co-registered images of the different modalities. Such co-registration can not only be performed in a research environment, it is available in most current MRI workstations, therefore can be achieved in a clinical setting as well. The fact that age did not correlate with NHLs supports that NHL raters were reliably able to discriminate non-specific aging scars from true NHLs. Still, though observing NHLs was presented to be just as important as the TMBs in the basal ganglia area, the reading of the single modality SWI to evaluate TMBs is apparently simpler than a multi-modal assessment of NHLs.

It might be raised that because of the relatively late (chronic) imaging performed in this study, some TMBs or NHLs could have been missed. Concerning TMBs, this is very unlikely. Though TMBs were shown to possibly change (expand) in the very acute phase after the injury [54], in the chronic phase, apart from a possible slight attenuation, they were mostly shown to be constant in their appearance even through years [55–57]. Regarding NHLs, we are not aware of long-term studies investigating lesion progression or cessation, we cannot exclude that some lesions disappeared till the imaging time point. A similar study performed in the acute phase might help further refinement of the relationship between NHLs and diffuse axonal damage.

Of the investigated parameters, GCS was independently positively correlated with FA and negatively with RD values of the corpus callosum. GCS was recorded in the acute phase, and the FA and RD values were measured in the chronic, thus the damage of the commissural white matter associated to the altered levels of consciousness appears to be a long-term consequence. The association between the damage of the corpus callosum, consciousness and neurocognitive impairment, and the long-term FA reduction and RD increase of the corpus callosum in TBI patients is supported by several papers [32,39–41,43,58–63]. The most interesting additional finding of this study is that neither corpus callosum TMBs or NHLs were independently, specifically associated with the clinically important microstructural disruption at this region. Thus, one might expect a clinically significant microstructural injury of the corpus callosum even despite of apparent macroscopic focal pathol-

ogy including any MRI modality, a finding also reported previously [64].

This study found a significant correlation between age and low subcortical white matter FA, increased subcortical white matter MD, AD, RD and increased corpus callosum MD, AD and RD. Except for subcortical white matter FA and corpus callosum RD, age was the only related independent factor. The strong association between normal aging and diffusivity alteration (FA decline, MD, AD and RD increase) as seen in the present work, has been widely characterized by previous studies [65–69]. Unfortunately, TBI is regarded to cause mostly the same directions in diffusivity changes, which makes the differentiation between the two effects challenging. In case of the subcortical white matter FA, both trauma related TMBs and NHLs, as well as age were yielded as independent factors. It is highly possible, that white matter aging is triggered by TBI. A set of previous studies shown that FA is not only acutely decreased after TBI over certain brain regions, but FA further decreases over years in the affected regions [40]. However, MD and AD increase was specifically linked to aging in our study. One explanation might be that MD alteration in the chronic phase is less specific for TBI, i.e. MD change due to the injury melts into the more pronounced effect of aging. A more detailed regional analysis might help in distinguishing these effects, since both normal aging and TBI are characterized by typical spatial patterns of diffusivity alteration [70–72].

We aimed to check if one of the most powerful CT prognostic scores, the Rotterdam score is able to predict white matter microstructure damage, compared to the MRI techniques. Rotterdam scores were very strongly correlated with GCS, and after adjusting for GCS, Rotterdam scores were not able to further explain any diffusivity differences in the TBI population. By these means, MRI detection of TMBs and NHLs outperformed CT.

The most limiting circumstance in this study was the relatively late (chronic) image acquisition. Because of this, raters might have missed NHLs, and diffusivity alteration might have been affected by non-specific factors (aging, neuro-rehabilitation, etc.). At the same time, the long term follow-up data are valuable since outcome prediction is a major aim of TBI imaging. Other limiting factor was the use of manual tracing and region-of-interest technique for the calculation of regional DTI metrics. Unfortunately, because of the prominent structural differences between the patients, inter-subject co-registration was unreliable and often impossible, that made voxel-wise techniques useless. By overcoming these issues, probably a more precise understanding of focal lesion-diffuse injury association would become achievable.

4.1. Conclusion

Both hemorrhagic (TMBs on SWI), and non-hemorrhagic (NHLs on T1-, T2 weighted and FLAIR images) features of DAI are associated with widespread chronic diffusivity alterations in the normal appearing white matter even after adjusting for age, GCS and Rotterdam scores. The presence of NHLs, even when only a single NHL is present, indicates a diffuse microstructural disintegration. Surprisingly, though SWI typically detects a higher number of parenchymal lesions in DAI then other modalities due to its high sensitivity, not overall lesion presence, -count or -volume, but only TMB load of the basal ganglia area was independently related with decreased normal appearing white matter FA levels. One explanation might be that TMBs of higher level white matter regions (corpus callosum, subcortical white matter) are rather dependent of an individual microvascular vulnerability than of the linked axonal damage.

This finding also indicates that regional heterogeneity of injury could significantly affect the natural history as well as the effect of therapeutic interventions emphasizing the need for individualized diagnostic scores and treatment approaches.

Based on the better understanding of the relation between macroscopic focal- and microscopic diffuse pathology, in order to estimate DAI in a clinical environment, a conventional lesional MRI pathology evaluation might at least in part substitute the use of quantitative DTI.

Conflict of interest

The authors report no conflicts of interest.

Acknowledgements

This study was funded by Hungarian Brain Research Program – Grant No. KTIA_13_NAP-A-II/8, Grant No. KTIA_13_NAP-A-II/9, SROP-4.2.2.A-11/1/KONV-2012-0017 and Hungarian Scientific Research Fund Grant No. OTKA/K-120356.

References

- [1] J. Bruns Jr., W.A. Hauser, The epidemiology of traumatic brain injury: a review, *Epilepsia* 44 (Suppl. 10) (2003) 2–10.
- [2] V.E. Johnson, W. Stewart, D.H. Smith, Axonal pathology in traumatic brain injury, *Exp. Neurol.* 246 (2013) 35–43.
- [3] S. Mittal, Z. Wu, J. Neelavalli, E.M. Haacke, Susceptibility-weighted imaging: technical aspects and clinical applications, part 2, *Am. J. Neuroradiol.* 30 (2) (2009) 232–252.
- [4] D.J. Sharp, T.E. Ham, Investigating white matter injury after mild traumatic brain injury, *Curr. Opin. Neurol.* 24 (6) (2011) 558–563.
- [5] M.H. Beauchamp, M. Ditchfield, F.E. Babl, M. Kean, C. Catroppa, K.O. Yeates, V. Anderson, Detecting traumatic brain lesions in children: CT versus MRI versus susceptibility weighted imaging (SWI), *J. Neurotrauma* 28 (6) (2011) 915–927.
- [6] A.L. Cheng, S. Batool, C.R. McCreary, M.L. Lauzon, R. Frayne, M. Goyal, E.E. Smith, Susceptibility-weighted imaging is more reliable than T2*-weighted gradient-recalled echo MRI for detecting microbleeds, *Stroke* 44 (10) (2013) 2782–2786.
- [7] B.H. Geurts, T.M. Andriessen, B.M. Goraj, P.E. Vos, The reliability of magnetic resonance imaging in traumatic brain injury lesion detection, *Brain Inj.* 26 (12) (2012) 1439–1450.
- [8] G. Spitz, J.J. Maller, A. Ng, R. O'Sullivan, N.J. Ferris, J.L. Ponsford, Detecting lesions after traumatic brain injury using susceptibility weighted imaging: a comparison with fluid-attenuated inversion recovery and correlation with clinical outcome, *J. Neurotrauma* 30 (24) (2013) 2038–2050.
- [9] A. Di Ieva, T. Lam, P. Alcaide-Leon, A. Bharatha, W. Montanera, M.D. Cusimano, Magnetic resonance susceptibility weighted imaging in neurosurgery: current applications and future perspectives, *J. Neurosurg.* (2015) 1–13.
- [10] A. Toth, N. Kovacs, G. Perlaki, G. Orsi, M. Aradi, H. Komaromy, E. Ezer, P. Bukovics, O. Farkas, J. Janszky, T. Doczi, A. Buki, A. Schwarcz, Multi-modal magnetic resonance imaging in the acute and sub-acute phase of mild traumatic brain injury: can we see the difference? *J. Neurotrauma* 30 (1) (2013) 2–10.
- [11] E.L. Yuh, P. Mukherjee, H.F. Lingsma, J.K. Yue, A.R. Ferguson, W.A. Gordon, A.B. Valadka, D.M. Schnyer, D.O. Okonkwo, A.I. Maas, G.T. Manley, T.-T. Investigators, Magnetic resonance imaging improves 3-month outcome prediction in mild traumatic brain injury, *Ann. Neurol.* 73 (2) (2013) 224–235.
- [12] A. Iwamura, T. Taoka, A. Fukusumi, M. Sakamoto, T. Miyasaka, T. Ochi, T. Akashi, K. Okuchi, K. Kichikawa, Diffuse vascular injury: convergent-type hemorrhage in the supratentorial white matter on susceptibility-weighted imaging in cases of severe traumatic brain damage, *Neuroradiology* 54 (4) (2012) 335–343.
- [13] M.H. Beauchamp, R. Beare, M. Ditchfield, L. Coleman, F.E. Babl, M. Kean, L. Crossley, C. Catroppa, K.O. Yeates, V. Anderson, Susceptibility weighted imaging and its relationship to outcome after pediatric traumatic brain injury, *Cortex* 49 (2) (2013) 591–598.
- [14] J. Liu, Z. Kou, Y. Tian, Diffuse axonal injury after traumatic cerebral microbleeds: an evaluation of imaging techniques, *Neural Regen. Res.* 9 (12) (2014) 1222–1230.
- [15] C.A. Colbert, B.A. Holshouser, G.S. Aaen, C. Sheridan, U. Oyoyo, D. Kido, S. Ashwal, Value of cerebral microhemorrhages detected with susceptibility-weighted MR imaging for prediction of long-term outcome in children with nonaccidental trauma, *Radiology* 256 (3) (2010) 898–905.
- [16] T. Babikian, M.C. Freier, K.A. Tong, J.P. Nickerson, C.J. Wall, B.A. Holshouser, T. Burley, M.L. Riggs, S. Ashwal, Susceptibility weighted imaging: neuropsychologic outcome and pediatric head injury, *Pediatr. Neurol.* 33 (3) (2005) 184–194.
- [17] K.A. Tong, S. Ashwal, B.A. Holshouser, J.P. Nickerson, C.J. Wall, L.A. Shutter, R.J. Osterdock, E.M. Haacke, D. Kido, Diffuse axonal injury in children: clinical correlation with hemorrhagic lesions, *Ann. Neurol.* 56 (1) (2004) 36–50.
- [18] R.J. Mannion, J. Cross, P. Bradley, J.P. Coles, D. Chatfield, A. Carpenter, J.D. Pickard, D.K. Menon, P.J. Hutchinson, Mechanism-based MRI classification of

- traumatic brainstem injury and its relationship to outcome, *J. Neurotrauma* 24 (1) (2007) 128–135.
- [19] K.G. Moen, V. Brezova, T. Skandsen, A.K. Haberg, M. Folvik, A. Vik, Traumatic axonal injury: the prognostic value of lesion load in corpus callosum, brain stem, and thalamus in different magnetic resonance imaging sequences, *J. Neurotrauma* 31 (17) (2014) 1486–1496.
- [20] M.B. Hukower, D.B. Poliak, S.B. Rosenbaum, M.E. Zimmerman, M.L. Lipton, A decade of DTI in traumatic brain injury: 10 years and 100 articles later, *Am. J. Neuroradiol.* 34 (11) (2013) 2064–2074.
- [21] C. Pierpaoli, P. Jezzard, P.J. Basser, A. Barnett, G. Di Chiro, Diffusion tensor MR imaging of the human brain, *Radiology* 201 (3) (1996) 637–648.
- [22] C. Pierpaoli, P.J. Basser, Toward a quantitative assessment of diffusion anisotropy, *Magn. Reson. Med.* 36 (6) (1996) 893–906.
- [23] J.T. Wilson, L.E. Pettigrew, G.M. Teasdale, Structured interviews for the Glasgow Outcome Scale and the extended Glasgow Outcome Scale: guidelines for their use, *J. Neurotrauma* 15 (8) (1998) 573–585.
- [24] M. Jenkinson, P. Bannister, M. Brady, S. Smith, Improved optimization for the robust and accurate linear registration and motion correction of brain images, *Neuroimage* 17 (2) (2002) 825–841.
- [25] S.M. Smith, M. Jenkinson, M.W. Woolrich, C.F. Beckmann, T.E. Behrens, H. Johansen-Berg, P.R. Bannister, M. De Luca, I. Drobnjak, D.E. Flitney, R.K. Niazy, J. Saunders, J. Vickers, Y. Zhang, N. De Stefano, J.M. Brady, P.M. Matthews, Advances in functional and structural MR image analysis and implementation as FSL, *Neuroimage* 23 (Suppl. 1) (2004) S208–S219.
- [26] P.E. Shrout, J.L. Fleiss, Intraclass correlations: uses in assessing rater reliability, *Psychol. Bull.* 86 (2) (1979) 420–428.
- [27] A.I. Maas, C.W. Hukkelhoven, L.F. Marshall, E.W. Steyerberg, Prediction of outcome in traumatic brain injury with computed tomographic characteristics: a comparison between the computed tomographic classification and combinations of computed tomographic predictors, *Neurosurgery* 57 (6) (2005) 1173–1182 (discussion 1173–82).
- [28] S.M. Smith, Fast robust automated brain extraction, *Hum. Brain Mapp.* 17 (3) (2002) 143–155.
- [29] P.J. Basser, J. Mattiello, D. LeBihan, Estimation of the effective self-diffusion tensor from the NMR spin echo, *J. Magn. Reson. B* 103 (3) (1994) 247–254.
- [30] F. Schoonjans, A. Zalata, C.E. Depuydt, F.H. Comhaire, MedCalc: a new computer program for medical statistics, *Comput. Methods Programs Biomed.* 48 (3) (1995) 257–262.
- [31] K. Arfanakis, V.M. Haughton, J.D. Carew, B.P. Rogers, R.J. Dempsey, M.E. Meyerand, Diffusion tensor MR imaging in diffuse axonal injury, *Am. J. Neuroradiol.* 23 (5) (2002) 794–802.
- [32] M. Inglese, S. Makani, G. Johnson, B.A. Cohen, J.A. Silver, O. Gonen, R.I. Grossman, Diffuse axonal injury in mild traumatic brain injury: a diffusion tensor imaging study, *J. Neurosurg.* 103 (2) (2005) 298–303.
- [33] J.Y. Wang, K. Bakhadirov, H. Abdi, M.D. Devous Sr., C.D. Marquez de la Plata, C. Moore, C.J. Madden, R. Diaz-Arrastia, Longitudinal changes of structural connectivity in traumatic axonal injury, *Neurology* 77 (9) (2011) 818–826.
- [34] J.J. Bazarjian, J. Zhong, B. Blyth, T. Zhu, V. Kavcic, D. Peterson, Diffusion tensor imaging detects clinically important axonal damage after mild traumatic brain injury: a pilot study, *J. Neurotrauma* 24 (9) (2007) 1447–1459.
- [35] R. Kumar, R.K. Gupta, M. Husain, C. Chaudhry, A. Srivastava, S. Saksena, R.K. Rathore, Comparative evaluation of corpus callosum DTI metrics in acute mild and moderate traumatic brain injury: its correlation with neuropsychometric tests, *Brain Inj.* 23 (7) (2009) 675–685.
- [36] R. Yallampalli, E.A. Wilde, E.D. Bigler, S.R. McCauley, G. Hanten, M. Troyanskaya, J.V. Hunter, Z. Chu, X. Li, H.S. Levin, Acute white matter differences in the fornix following mild traumatic brain injury using diffusion tensor imaging, *J. Neuroimaging* 23 (2) (2013) 224–227.
- [37] K. Caeyenberghs, A. Leemans, M. Geurts, T. Taymans, C.V. Linden, B.C. Smits-Engelsman, S. Sunaert, S.P. Swinnen, Brain-behavior relationships in young traumatic brain injury patients: DTI metrics are highly correlated with postural control, *Hum. Brain Mapp.* 31 (7) (2010) 992–1002.
- [38] L. Ljungqvist, D. Nilsson, M. Ljungberg, A. Sorbo, E. Esbjornsson, C. Eriksson-Ritzen, T. Skoglund, Longitudinal study of the diffusion tensor imaging properties of the corpus callosum in acute and chronic diffuse axonal injury, *Brain Inj.* 25 (4) (2011) 370–378.
- [39] T.C. Wu, E.A. Wilde, E.D. Bigler, X. Li, T.L. Merkley, R. Yallampalli, S.R. McCauley, K.P. Schnelle, A.C. Vasquez, Z. Chu, G. Hanten, J.V. Hunter, H.S. Levin, Longitudinal changes in the corpus callosum following pediatric traumatic brain injury, *Dev. Neurosci.* 32 (5–6) (2010) 361–373.
- [40] J. Dinkel, A. Drier, O. Khalilzadeh, V. Perlberg, V. Czernecki, R. Gupta, F. Gomas, P. Sanchez, D. Dormont, D. Galanaud, R.D. Stevens, L. Puybasset, for NICR, Long-term white matter changes after severe traumatic brain injury: a 5-year prospective cohort, *Am. J. Neuroradiol.* 35 (1) (2014) 23–29.
- [41] A. Sidaros, A.W. Engberg, K. Sidaros, M.G. Liptrot, M. Herning, P. Petersen, O.B. Paulson, T.L. Jernigan, E. Rostrup, Diffusion tensor imaging during recovery from severe traumatic brain injury and relation to clinical outcome: a longitudinal study, *Brain* 131 (Pt 2) (2008) 559–572.
- [42] V.F. Newcombe, G.B. Williams, D. Coffings, J. Cross, T.A. Carpenter, J.D. Pickard, D.K. Menon, Aetiological differences in neuroanatomy of the vegetative state: insights from diffusion tensor imaging and functional implications, *J. Neurol. Neurosurg. Psychiatry* 81 (5) (2010) 552–561.
- [43] R.C. Tasker, A.G. Westland, D.K. White, G.B. Williams, Corpus callosum and inferior forebrain white matter microstructure are related to functional outcome from raised intracranial pressure in child traumatic brain injury, *Dev. Neurosci.* 32 (5–6) (2010) 374–384.
- [44] M.B. Oni, E.A. Wilde, E.D. Bigler, S.R. McCauley, T.C. Wu, R. Yallampalli, Z. Chu, X. Li, J.V. Hunter, A.C. Vasquez, H.S. Levin, Diffusion tensor imaging analysis of frontal lobes in pediatric traumatic brain injury, *J. Child Neurol.* 25 (8) (2010) 976–984.
- [45] A. Messe, S. Caplain, G. Paradot, D. Garrigue, J.F. Mineo, G. Soto Ares, D. Ducreux, F. Vignaud, G. Rozec, H. Desal, M. Pelegriani-Issac, M. Montreuil, H. Benali, S. Lehericy, Diffusion tensor imaging and white matter lesions at the subacute stage in mild traumatic brain injury with persistent neurobehavioral impairment, *Hum. Brain Mapp.* 32 (6) (2011) 999–1011.
- [46] S. Ashwal, T. Babikian, J. Gardner-Nichols, M.C. Freier, K.A. Tong, B.A. Holshouser, Susceptibility-weighted imaging and proton magnetic resonance spectroscopy in assessment of outcome after pediatric traumatic brain injury, *Arch. Phys. Med. Rehabil.* 87 (12 Suppl. 2) (2006) S50–S58.
- [47] C.A. Chastain, U.E. Oyoyo, M. Zipperman, E. Joo, S. Ashwal, L.A. Shutter, K.A. Tong, Predicting outcomes of traumatic brain injury by imaging modality and injury distribution, *J. Neurotrauma* 26 (8) (2009) 1183–1196.
- [48] J.H. Park, S.W. Park, S.H. Kang, T.K. Nam, B.K. Min, S.N. Hwang, Detection of traumatic cerebral microbleeds by susceptibility-weighted image of MRI, *J. Korean Neurosurg. Soc.* 46 (4) (2009) 365–369.
- [49] J.H. Adams, D. Doyle, I. Ford, T.A. Gennarelli, D.I. Graham, D.R. McLellan, Diffuse axonal injury in head injury: definition, diagnosis and grading, *Histopathology* 15 (1) (1989) 49–59.
- [50] S. Akoudad, M. de Groot, P.J. Koudstaal, A. van der Lugt, W.J. Niessen, A. Hofman, M.A. Ikram, M.W. Vernooij, Cerebral microbleeds are related to loss of white matter structural integrity, *Neurology* 81 (22) (2013) 1930–1937.
- [51] J. van der Naalt, J.M. Hew, A.H. van Zomeren, W.J. Sluiter, J.M. Minderhoud, Computed tomography and magnetic resonance imaging in mild to moderate head injury: early and late imaging related to outcome, *Ann. Neurol.* 46 (1) (1999) 70–78.
- [52] A. Jenkins, G. Teasdale, M.D. Hadley, P. Macpherson, J.O. Rowan, Brain lesions detected by magnetic resonance imaging in mild and severe head injuries, *Lancet* 2 (8504) (1986) 445–446.
- [53] H.S. Levin, E. Amparo, H.M. Eisenberg, D.H. Williams, W.M. High Jr., C.B. McArdle, R.L. Weiner, Magnetic resonance imaging and computerized tomography in relation to the neurobehavioral sequelae of mild and moderate head injuries, *J. Neurosurg.* 66 (5) (1987) 706–713.
- [54] A. Toth, N. Kovacs, V. Tamas, B. Kornyei, M. Nagy, A. Horvath, T. Rostas, P. Bogner, J. Janszky, T. Doczi, A. Buki, A. Schwarcz, Microbleeds may expand acutely after traumatic brain injury, *Neurosci. Lett.* 617 (2016) 207–212.
- [55] A. Messori, G. Polonara, C. Mabilgia, U. Salvolini, Is haemosiderin visible indefinitely on gradient-echo MRI following traumatic intracerebral haemorrhage? *Neuroradiology* 45 (12) (2003) 881–886.
- [56] K.G. Moen, T. Skandsen, M. Folvik, V. Brezova, K.A. Kvistad, J. Rydland, G.T. Manley, A. Vik, A longitudinal MRI study of traumatic axonal injury in patients with moderate and severe traumatic brain injury, *J. Neurol. Neurosurg. Psychiatry* 83 (12) (2012) 1193–1200.
- [57] B.H. Geurts, T.M. Andriessen, B.M. Goraj, P.E. Vos, The reliability of magnetic resonance imaging in traumatic brain injury lesion detection, *Brain Inj.* (2012).
- [58] M.C. Chang, S.H. Jang, Corpus callosum injury in patients with diffuse axonal injury: a diffusion tensor imaging study, *NeuroRehabilitation* 26 (4) (2010) 339–345.
- [59] L. Ewing-Cobbs, M.R. Prasad, P. Swank, L. Kramer, C.S. Cox Jr., J.M. Fletcher, M. Barnes, X. Zhang, K.M. Hasan, Arrested development and disrupted callosal microstructure following pediatric traumatic brain injury: relation to neurobehavioral outcomes, *Neuroimage* 42 (4) (2008) 1305–1315.
- [60] M.R. Kennedy, J.R. Wozniak, R.L. Muetzel, B.A. Mueller, H.H. Chiou, K. Pantekoek, K.O. Lim, White matter and neurocognitive changes in adults with chronic traumatic brain injury, *J. Int. Neuropsychol. Soc.* 15 (1) (2009) 130–136.
- [61] W. Yuan, S.K. Holland, V.J. Schmithorst, N.C. Walz, K.M. Cecil, B.V. Jones, P. Karunanayaka, L. Michaud, S.L. Wade, Diffusion tensor MR imaging reveals persistent white matter alteration after traumatic brain injury experienced during early childhood, *Am. J. Neuroradiol.* 28 (10) (2007) 1919–1925.
- [62] J. Xu, I.A. Rasmussen, J. Lagopoulos, A. Haberg, Diffuse axonal injury in severe traumatic brain injury visualized using high-resolution diffusion tensor imaging, *J. Neurotrauma* 24 (5) (2007) 753–765.
- [63] R. Kumar, M. Husain, R.K. Gupta, K.M. Hasan, M. Haris, A.K. Agarwal, C.M. Pandey, P.A. Narayana, Serial changes in the white matter diffusion tensor imaging metrics in moderate traumatic brain injury and correlation with neuro-cognitive function, *J. Neurotrauma* 26 (4) (2009) 481–495.
- [64] N. Nakayama, A. Okumura, J. Shinoda, Y.T. Yasokawa, K. Miwa, S.I. Yoshimura, T. Iwama, Evidence for white matter disruption in traumatic brain injury without macroscopic lesions, *J. Neurol. Neurosurg. Psychiatry* 77 (7) (2006) 850–855.
- [65] E.V. Sullivan, A. Pfefferbaum, Neuroradiological characterization of normal adult ageing, *Br. J. Radiol.* 80 Spec No 2 (2007) S99–S108.
- [66] S.T. Engelter, J.M. Provenzale, J.R. Petrella, D.M. DeLong, J.R. MacFall, The effect of aging on the apparent diffusion coefficient of normal-appearing white matter, *Am. J. Roentgenol.* 175 (2) (2000) 425–430.
- [67] E.V. Sullivan, E. Adalsteinsson, M. Hedehus, C. Ju, M. Moseley, K.O. Lim, A. Pfefferbaum, Equivalent disruption of regional white matter microstructure in ageing healthy men and women, *Neuroreport* 12 (1) (2001) 99–104.
- [68] I.J. Bennett, D.J. Madden, C.J. Vaidya, D.V. Howard, J.H. Howard Jr., Age-related differences in multiple measures of white matter integrity: a diffusion tensor imaging study of healthy aging, *Hum. Brain Mapp.* 31 (3) (2010) 378–390.

- [69] R. Kumar, A.S. Chavez, P.M. Macey, M.A. Woo, R.M. Harper, Brain axial and radial diffusivity changes with age and gender in healthy adults, *Brain Res.* 1512 (2013) 22–36.
- [70] A. Pfefferbaum, E. Adalsteinsson, E.V. Sullivan, Frontal circuitry degradation marks healthy adult aging: evidence from diffusion tensor imaging, *Neuroimage* 26 (3) (2005) 891–899.
- [71] A. Stadlbauer, E. Salomonowitz, G. Strunk, T. Hammen, O. Ganslandt, Age-related degradation in the central nervous system: assessment with diffusion-tensor imaging and quantitative fiber tracking, *Radiology* 247 (1) (2008) 179–188.
- [72] A. Pfefferbaum, E.V. Sullivan, Increased brain white matter diffusivity in normal adult aging: relationship to anisotropy and partial voluming, *Magn. Reson. Med.* 49 (5) (2003) 953–961.