TURUN YLIOPISTON JULKAISUJA
ANNALES UNIVERSITATIS TURKUENSIS

SARJA - SER. D OSA - TOM. 1061

MEDICA - ODONTOLOGICA

DETECTION OF PATHOLOGIC CHANGES FOLLOWING TRAUMATIC BRAIN INJURY USING MAGNETIC RESONANCE IMAGING

by

Nina Brandstack

TURUN YLIOPISTO
UNIVERSITY OF TURKU
Turku 2013

From the Department of Radiology, Turku University Central Hospital, University of Turku, Faculty of Medicine, and Turku Graduate School of Clinical Sciences, Turku, Finland

Research school membership: National Graduate School of Clinical Investigation (CLIGS)

Supervised by

Docent Timo Kurki, MD, PhD
Department of Radiology
University of Turku
PULSSI Medical Centre
Turku, Finland

and

Docent Olli Tenovuo, MD, PhD
Department of Neurology
University of Turku
Turku, Finland

Reviewed by

Professor Paul Parizel, MD, PhD
Department of Medical Imaging
University of Antwerp
Antwerp, Belgium

and

Docent Tapani Tikkakoski, MD, PhD
Department of Radiology
Keski- Pohjanmaa Central Hospital
Kokkola, Finland

Official opponent

Professor Ritva Vanninen, MD, PhD
Department of Radiology
University of Kuopio
Kuopio, Finland

ISBN 978-951-29-5339-4 (PRINT)
ISBN 978-951-29-5340-0 (PDF)
ISSN 0355-9483
Painosalama Oy – Turku, Finland 2013

To my Family

ABSTRACT

Nina Brandstack

DETECTION OF PATHOLOGIC CHANGES FOLLOWING TRAUMATIC BRAIN INJURY USING MAGNETIC RESONANCE IMAGING
From the department of Radiology, University of Turku, Turku, Finland 2013

Background: Approximately two percent of Finns have sequels after traumatic brain injury (TBI), and many TBI patients are young or middle-aged. The high rate of unemployment after TBI has major economic consequences for society, and traumatic brain injury often has remarkable personal consequences, as well. Structural imaging is often needed to support the clinical TBI diagnosis. Accurate early diagnosis is essential for successful rehabilition and, thus, may also influence the patient's outcome. Traumatic axonal injury and cortical contusions constitute the majority of traumatic brain lesions. Several studies have shown magnetic resonance imaging (MRI) to be superior to computed tomography (CT) in the detection of these lesions. However, traumatic brain injury often leads to persistent symptoms even in cases with few or no findings in conventional MRI.

Aims and methods: The aim of this prospective study was to clarify the role of conventional MRI in the imaging of traumatic brain injury, and to investigate how to improve the radiologic diagnostics of TBI by using more modern diffusion-weighted imaging (DWI) and diffusion tensor imaging (DTI) techniques. We estimated, in a longitudinal study, the visibility of the contusions and other intraparenchymal lesions in conventional MRI at one week and one year after TBI. We used DWI-based measurements to look for changes in the diffusivity of the normal-appearing brain in a case-control study. DTI-based tractography was used in a case-control study to evaluate changes in the volume, diffusivity, and anisotropy of the long association tracts in symptomatic TBI patients with no visible signs of intracranial or intraparenchymal abnormalities on routine MRI. We further studied the reproducibility of different tools to identify and measure white-matter tracts by using a DTI sequence suitable for clinical protocols.

Results: Both the number and extent of visible traumatic lesions on conventional MRI diminished significantly with time. Slightly increased diffusion in the normal-appearing brain was a common finding at one week after TBI, but it was not significantly associated with the injury severity. Fractional anisotropy values, that represent the integrity of the white-matter tracts, were significantly diminished in several tracts in TBI patients compared to the control subjects. Compared to the cross-sectional ROI method, the tract-based analyses had better reproducibility to identify and measure white-matter tracts of interest by means of DTI tractography.

Conclusions: As conventional MRI is still applied in clinical practice, it should be carried out soon after the injury, at least in symptomatic patients with negative CT scan. DWI-related brain diffusivity measurements may be used to improve the documenting of TBI. DTI tractography can be used to improve radiologic diagnostics in a symptomatic TBI sub-population with no findings on conventional MRI. Reproducibility of different tools to quantify fibre tracts vary considerably, which should be taken into consideration in the clinical DTI applications.

Keywords: traumatic brain injury, magnetic resonance imaging, diffusion-weighted imaging, diffusion tensor imaging, quantitative tractography, reproducibility

TIIVISTELMÄ

Nina Brandstack

MAGNEETTIKUVANTAMINEN AIVOVAMMOJEN DIAGNOSTIIKASSA

Radiologian oppiaine, Turun Yliopisto, Turku 2013

Tausta: Arviolta noin kahdella prosentilla suomalaisista on aivovammojen jälkitiloja, ja merkittävä osa aivovammapotilaista on nuoria tai keski-ikäisiä. Heikko työhönsijoittuminen aivovamman jälkeen aiheuttaa yhteiskunnalle merkittäviä taloudellisia menetyksiä, ja usein aivovamma aiheuttaa myös huomattavia henkilökohtaisia seurauksia. Kliinisen aivovammadiagnoosin tueksi tarvitaan usein kuvantamistutkimuksia. Riittävän varhaisella ja tarkalla diagnostiikalla on tärkeä merkitys aivovammapotilaan kuntoutuksen suunnittelussa ja siten vaikutusta myös potilaan ennusteeseen. Suurin osa aivovammamuutoksista on joko kortikaalisia kontuusioita tai traumaattisia aksonivaurioita. Useissa tutkimuksissa on todettu näiden muutosten näkyvän paremmin magneettikuvauksessa (MRI) kuin tietokonetomografiatutkimuksessa. Kuitenkin myös perinteisessä MRI tutkimuksessa löydökset ovat usein vähäiset huolimatta potilaan traumaperäisistä oireista.

Tavoitteet ja menetelmät: Tämän prospektiivisen tutkimuksen tavoitteena oli selventää perinteisen magneettitutkimuksen roolia aivovammojen kuvantamisessa sekä parantaa aivovammojen diagnostiikkaa hyödyntämällä uusia diffuusiokuvantamis- (DWI) ja diffuusiotensorikuvantamis- (DTI) tekniikoita. Arvioimme seurantatutkimuksessa kontuusiomuutosten ja aksonivaurioiden näkymistä perinteisessä magneettitutkimuksessa viikon ja vuoden kuluttua aivovammasta. Tutkimme tapaus-verrokki asetelmassa normaalilta näyttävän aivokudoksen diffusiivisuutta laajalti eri aivoalueilta DWI-menetelmällä. Selvitimme tapaus-verrokki asetelmassa aivojen pitkien assosiaatioratojen diffusiivisuus-, anisotropia- ja tilavuusmuutoksia DTI-traktografia –menetelmällä oireisilla aivovammapotilailla, joilla ei ollut näkyviä muutoksia perinteisessä magneettikuvauksessa. Selvitimme lisäksi DTI-kuvantamiseen perustuvien kliiniseen työhön sovellettavien erityyppisten traktografiamenetelmien toistettavuutta.

Tulokset: Näkyvien vammamuutosten määrä ja laajuus perinteisissä magneettikuvissa vähenivät merkittävästi vuoden aikana. Hieman lisääntynyt diffuusio normaalin näköisessä aivokudoksessa oli tavallinen löydös viikon kuluttua vammasta, mutta ei ollut vakuuttavasti yhteydessä vamman vakavuuteen. Aivojen valkean aineen ratojen yhtenäisyyttä kuvaavat fraktionaaliset anisotropia -arvot olivat aivovammapotilailla vähentyneet useissa radoissa merkittävästi verrokkiryhmään nähden. DTI traktografiaan perustuvan ns. tract-based –menetelmän toistettavuus oli selvästi 'region of interest' (ROI) –menetelmää parempi.

Johtopäätökset: Perinteinen magneettitutkimus tulisi tehdä riittävän varhain tilanteissa, jolloin uudempia MRI-tekniikoita ei ole käytettävissä ja tietokonetomografialöydös ei selitä potilaan oireita. DWI-kuvantamiseen pohjautuvista aivokudoksen diffusiivisuusmittauksista voi olla hyötyä akuutin aivovamman dokumentoinnissa. DTI-traktografia –menetelmällä on mahdollista parantaa radiologista diagnostiikkaa oireisilla aivovammapotilailla, joilla perinteinen kuvantamislöydös on normaali, mikä tulisi huomioida aivovammapotilaita hoitavissa ja kuvantavissa yksiköissä. Erityyppisten traktografiamenetelmien toistettavuus vaihtelee merkittävästi, mikä tulisi ottaa huomioon kliinisissä sovelluksissa.

Avainsanat: aivovamma, magneettikuvaus, diffuusiokuvaus, diffuusiotensorikuvaus, kvantitatiivinen traktografia, toistettavuus

TABLE OF CONTENTS

ABBREVIATIONS

ADC	Apparent diffusion coefficient
AF	Arcuate fasciculus
AOC	Alteration of consciousness
CR	Coefficient of repeatability
CSF	Cerebrospinal fluid
CT	Computed tomography
CV	Coefficient of variation
DAI	Diffuse axonal injury
DTI	Diffusion tensor imaging
DTT	Diffusion tensor tractography
DWI	Diffusion-weighted imaging
FA	Fractional anisotropy
FLAIR	Fluid-attenuated inversion recovery
GM	Grey matter
HISC	Head injury symptom checklist
ICC	Intraclass correlation
fMRI	Functional Magnetic Resonance Imaging
GCS	Glasgow Coma Scale
GOS	Glasgow Outcome Scale
GOS-E	Glasgow Outcome Scale- Extended
IFOF	Inferior fronto-occipital fasciculus
ILF	Inferior longitudinal fasciculus
LOC	Loss of consciousness
MD	Mean diffusivity
MR	Magnetic resonance
MRI	Magnetic resonance imaging
mTBI	Mild traumatic brain injury
OR	Odds ratio
PCS	Post-concussive syndrome
PPCS	Persistent post-concussive syndrome
PTA	Posttraumatic amnesia

RF	Radiofrequence
ROI	Region of interest
SC	Superior cingulum
SD	Standard deviation
SLF	Superior longitudinal fasciculus
TAI	Traumatic axonal injury
TBI	Traumatic brain injury
TC	Temporal cingulum
TE	Time of echo
TR	Time of repetition
UF	Uncinate fasciculus
WM	White matter

LIST OF ORIGINAL PUBLICATIONS

This thesis is based on the following original publications, which are referred to in the text by their Roman numerals.

I. **MR imaging of head trauma: Visibility of contusions and other intraparenchymal injuries in early and late stage**
Brandstack Nina, Kurki Timo, Tenovuo Olli, and Isoniemi Heli.
Brain Injury 2006;20:409-16.

II. **Diffusivity of normal appearing brain in acute traumatic brain injury**
Brandstack Nina, Kurki Timo, Hiekkanen Heli, and Tenovuo Olli.
Clinical Neuroradiology 2011;21:75-82.

III. **Quantitative Diffusion Tensor Tractography of Long Association Tracts in Patients with Traumatic Brain Injury without Findings in Routine MRI**
Brandstack Nina, Kurki Timo, and Tenovuo Olli.
Radiology 2013, in press.

IV. **Reproducibility of Tract-based and Region of Interest DTI Analysis of Long Association Tracts**
Brandstack Nina, Kurki Timo, Laalo Jussi, Kauko Tommi, and Tenovuo Olli.
Submitted.

The original publications have been reproduced with the permission of the copyright holders.

1. INTRODUCTION

Traumatic brain injury (TBI) is a major cause of death and severe disability under the age of 45 years in Western countries (MacKenzie, 2000). The estimated annual total number of new patients suffering from TBI in Finland is 21 000–31 000 (Koskinen and Alaranta, 2008). Knowledge of the extension and the exact sites of injury in the invidual patient can provide important information for planning long-term rehabilitation, and may have an influence on the long-term outcome. Imaging techniques are in a special position to validate functional deficits related to TBI since lack of motivation and somatoform disorders can influence the accuracy of neuropsychological tests and self-reported symptoms. As many TBIs occur in traffic accidents, work-related accidents, sport-related injuries, or assaults, their medicolegal consequences may also be significant.

Traumatic axonal injuries (TAI) and cortical contusions constitute the majority of traumatic brain lesions. Traumatic axonal injuries are thought to be present in the majority of all severe head injuries, and also to be responsible for the long-lasting or persistent problems after mild injuries. Although TBI may result in physical impairment, the more problematic consequences involve the individual`s cognition, emotional functioning, and behaviour (NIH Consensus Development Panel on Rehabilitation of Persons with Traumatic Brain Injury, 1999). Association pathways of the brain are typically involved in cognitive tasks, and these are among the most commonly damaged tracts after TBI.

In the acute management of a TBI patient, computed tomography (CT) is routinely the first imaging method (Coles, 2007; Toyama et al. 2005) since it can be performed rapidly, and pathology that is critical for the early medical management of cerebral trauma can be detected by CT (Bigler, 2011). However, several studies have shown magnetic resonance imaging (MRI) to be superior to CT in the detection of intraparenchymal traumatic lesions, both in the acute and chronic stages, and regardless of the injury severity. However, TBI often leads to persistent symptoms even in cases without findings in conventional MRI (Fork et al., 2005; Scheid et al., 2006). The clinical use of MRI in the imaging of TBI has not achieved a universally accepted role. MRI studies are often performed in the subacute and chronic stages of recovery from TBI, with the intention of answering specific neuropsychiatric questions regarding patient status and/or to assist in evaluating outcome (Ashwal et al., 2006; Levine et al., 2006). The sensitivity in detecting TBI-related anatomical abnormalities by MRI depends on the image sequence and methods used. Clinical MRI studies are still often performed with old or poorly equipped MRI systems and with ordinary visual evaluation. A seldom raised aspect is the role of early imaging in documenting the intracranial injury, since late conventional MRI is thought to underestimate especially the extent of TAI. Recently, much interest has been directed towards the potential of diffusion-weighted imaging (DWI) and diffusion tensor imaging (DTI) techniques to investigate traumatic axonal injury. At the moment, DTI may be the most promising neuroimaging technique to

examine structural connectivity in the brain in various conditions, but its reproducibility has not been adequately studied.

This thesis presents results regarding the visibility of intraparenchymal injuries with time on conventional MRI. Additionally, the study aims to establish the role of more modern MR imaging techniques including DWI and DTI in the radiological diagnostics of traumatic brain injury. Results regarding the abnormalities in quantitative DTI tractography of long association tracts in subjects with symptoms after TBI without traumatic findings on routine MRI are presented. Also the occurrence and distribution of diffusion changes throughout the normal-appearing brain in acute TBI are depicted. Another focus of this thesis is to introduce reproducible DTI protocols and analysis methods applicable for clinical use in the diagnostics of TBI and other cognitive disorders.

2. REVIEW OF THE LITERATURE

2.1. TRAUMATIC BRAIN INJURY (TBI)

2.1.1. Definition and classification

TBI is a complex process, and there is no single classification encompassing all the clinical, pathological, and molecular features of TBI. Because the clinical definition from the Department of Veteran Affairs and the Department of Defence (VA/DoD 2009) is among the most recently developed and addresses issues specific to TBI among military service members, veterans, and civilians (Orman et al., 2011), it is summarized here:

TBI is a traumatically induced structural injury and/or physiological disruption of brain function as a result of an external force that is indicated by new onset of at least one of the following clinical signs, immediately following the event:

1. Any period of loss of or a decreased level of consciousness;
2. Any loss of memory for events immediately before or after the injury;
3. Any alteration in mental state at the time of the injury (confusion, disorientation, slowed thinking, etc., also known as alteration of consciousness (AOC));
4. Neurological deficits (weakness, loss of balance, change in vision, praxis, paresis/paraplegia, sensory loss, aphasia, etc.) that may or may not be transient;
5. Intracranial lesion.

External forces may include any of the following events: the head being struck by an object, the head striking an object, the brain undergoing an acceleration/deceleration movement without direct external trauma to the head, a foreign body penetrating the brain, forces generated from events such as a blast or explosion, or other force yet to be defined (Orman et al., 2011).

The most common clinical indicators used to assess acute brain injury severity include length of loss of consciousness (LOC) and posttraumatic amnesia (PTA), and the most widely used tool for assessing level of consciousness is the Glasgow Coma Scale (GCS) (Teasdale and Jennett, 1974). The GCS includes three categories: eyes opening (score 1-4), best verbal response (score 1-5), and best motor response (1-6). The total score of the GCS ranges from 3 to 15, low scores indicating a low level of consciousness. PTA is defined as lasting up to the earliest time point after which continuous memory has returned. Based on the length of AOC, LOC or PTA, TBI can be categorized as mild, moderate, or severe. Acute injury severity is best determined at the time of the injury (VA/DoD 2009). Clinical criteria for determining acute severity are summarized in **Table 1**.

The terms concussion and mild traumatic brain injury (mTBI) have been used interchangeably and both are used in the literature cited in this study. Although the criteria especially for the mTBI/ concussion vary across studies, most agree that the common criteria include GCS scores of 13-15, brief LOC, brief PTA, and negative head CT scan (VA/DoD 2009).

Table 1. Severity of brain injury stratification.

Criteria	Mild	Moderate	Severe
Structural imaging	Normal[a]	Normal or abnormal	Normal or abnormal
LOC	0-30 minutes;	>30 minutes and	>24 hours;
	0 = very mild*	<24 hours	>7 days= very severe*
AOC[b]	≤ 24 hours	>24 hours	Severity based on other criteria
PTA	≤1 day;	>1 and <7 days	>7 days;
	<10min= very mild*	>4 weeks= very severe*	
GCS[c]	13-15	9-12	3-8

[a] Minor abnormalities possibly not related to the brain injury may be present on structural imaging in the absence of loss of consciousness (LOC), alteration of consciousness (AOC), and posttraumatic amnesia (PTA).

[b] Alteration of mental state must be immediately related to trauma to the head. Typical symptoms would be looking and feeling dazed and uncertain what is happening, confusion, difficulty thinking clearly or responding appropriately to mental status questions, and being unable to describe events immediately before or after the trauma event.

[c] Some studies report the best available Glasgow Coma Scale (GCS) score within the first 6 hours or some other time period.

Modified from VA/DoD (2009) Clinical Practice Guideline.

* Adapted from Finnish Clinical Practice Guideline for TBI (Käypä hoito, Aivovammat, 2008).

2.1.2. The mechanism and pathophysiology of TBI

Pathological classifications of TBI may be anatomical, describing injuries as focal or diffuse, or pathophysiological, based on primary and secondary injuries. TBI may be produced by blunt force head injury (contact or noncontact), penetrating injuries, or blast injuries. The forces and, correspondingly, the range of pathologies associated with each mechanism are different. Focal pathologies caused by TBI include scalp lacerations, skull fractures, contusions/lacerations, intracranial hemorrhage, and focal lesions secondary to raised intracranial pressure. Diffuse pathologies include global ischemic injury, traumatic axonal injury/ diffuse vascular injury, and brain swelling.

Traditionally, TBI has been classified as an open (typical of gunshot wounds) or closed injury, depending on the presence of disruption of the dura mater. Most of all TBIs are closed injuries, and they can be both focal or diffuse. The term diffuse axonal injury (DAI) has lately been reserved for the clinical syndrome with supporting neuroradiological changes, while the term used for the pathological/neuroradiological demonstration of damaged axons in a pattern supporting a traumatic etiology is traumatic axonal injury (TAI) (Smith, 2011). In the literature cited in this study, however, both the terms, DAI

and TAI, have mainly been used interchangeably for the neuroradiological demonstration of damaged axons.

Pathologies caused by TBI are not static but change with time. These pathologies are modified by other pathophysiological parameters, which enables therapeutic interventions. Even the effects of a mild TBI may lead to long-term problems. More studies are needed to identify patients at risk of long-term neurodegeneration and to understand the underlying pathological processes.

A TBI-associated lesion may occur anywhere in the brain, but lesions often predominantly affect cortical structures (contusions) and white matter (WM) pathways (TAI).

Contusions

Contusions typically involve the crests of gyri and are often superficial, involving the grey matter only, but they may extend into underlying white matter and form a hematoma. By definition the pia mater is intact in overlying contusions but torn in lacerations. Contusions typically involve the frontal poles, the inferior frontal lobe, the temporal poles and lateral and inferior aspects of the temporal lobes, and the cortex below and above the Sylvian fissure (Smith, 2011). Contusions involving the occipital lobes and cerebellum are rare due to the smooth inner surface of the posterior fossa of the skull (compared with the bony ridges of the anterior and middle fossae). Still, contusions may be seen in atypical places directly related to skull fractures. The pattern of contusions may be coup (contusions involving frontal and temporal lobes following a fall forward), contrecoup (contusions involving frontal and temporal lobes following a backward fall), or underlying fractures.

Traumatic axonal injury (TAI)

Traumatic axonal injuries contributes to at least 35% of the mortality and morbidity of TBI cases without space-occupying lesions, and TAI is an important cause of severe disability in head injury survivors (Smith, 2011).

The location and severity of traumatic axonal injury is related to various factors (Adams et al., 1982; Parizel et al., 1998) including the characteristics of acceleration/deceleration forces during the trauma incident, and differences in attenuation and rigidity between adjacent cerebral structures. Originally, trauma-induced axonal injury was thought to be the result of axons being disconnected at the time of the impact (primary axotomy). However, current opinion based on many experimental studies (Farkas and Povlishock, 2007) is that the forces modify focal axonal sections, resulting in mechanoporation with calcium influx and microtubule disruption causing local axonal transport impairment and axonal swelling, followed by detachment over a period of time after injury (Smith, 2011).

Association pathways (i.e. fibre bundles that travel to other cortical areas in the same hemisphere), the commissural fibres (i.e. fibres that pass to the contralateral hemisphere)

of the corpus callosum, and several projection fibres (i.e. fibres that connect the cerebral cortex with the subcortical centres, the brain stem, and the spinal cord) belong to the most commonly damaged tracts after TBI (Kinnunen et al., 2011; Kraus et al., 2007; Lipton et al., 2008; Nakayama et al., 2006; Niogi et al., 2008; Rutgers et al., 2008; Salmond et al., 2006; Xu et al., 2007). The association tracts commonly damaged include the uncinate fasciculus (UF), cingulum, superior lognitudinal fasciculus (SLF), inferior lognitudinal fasciculus (ILF), and the inferior fronto-occipital fasciculus (IFOF) (Kinnunen et al., 2011; Kraus et al., 2007; Niogi et al., 2008; Xu et al., 2007). Association tracts are typically involved in cognitive tasks such as executive functions, attention, memory, and learning (Niogi and Mukherjee, 2010; Schmahmann et al., 2008). The UF (Catani and Mesulam, 2008; Duffau et al., 2009; Schmahmann et al., 2008) and cingulum (Schmahmann et al., 2008) are also involved in emotion processing. Association tracts may also be components in processing visual spatial information (IFOF), may serve as the conduit for the neural systems serving visual awareness and engagement in the environment (SLF), or may be important for the auditory-related processing (AF). Damage in ILF may cause visual agnosia and prosopagnosia (Schmahmann et al., 2008). The corpus callosum transfers motor, sensory, and cognitive information between the brain hemispheres, and is involved in several functions of the body, such as, tactile localization, eye movement, and maintaining the balance of arousal and attention. The projection fibres consist of fibres uniting the cortex with the lower parts of the brain and with the spinal cord, and they are involved in several, for example sensoral and motor, functions of the brain.

2.1.3. Epidemiology of TBI

TBI is a major cause of death and severe disability under the age of 45 years in Western countries (MacKenzie, 2000). Road traffic accidents, falls, and violence are the major causes of TBI (Murray et al., 1999; Tagliaferri et al., 2006).

Data on hospital discharges and on deaths at national or local level do allow head injuries to be identified by the International Classification of Diseases (ICD) codes (Jennett, 1996).

Epidemiology of TBI, however, is difficult to describe accurately because of inconsistencies in the TBI definition and classification, along with discrepancies in data collection. Especially true number of mild TBI cases is difficult to define, since it might be that only about 25% of individuals with TBI are treated in hospitals (Sosin et al., 1996). Moreover, the TBI mortality rate and distribution of etiological factors may change over time, and differ between regions (Dutton et al., 2010; Murray et al., 1999).

There are significant differences in the death rates even between Finland and the other Nordic countries; in the study of Sundstrøm et al. (2007), Finland had about twice as high a TBI mortality rate as the other Nordic countries. It seems unlikely that all the observed variation would be explained by differences in the criteria used to define TBI or

to identify patients, since Sundstrøm et al. also found a similar difference when analyzing data from a completely different source: Finnish neurosurgeons performed approximately twice as many operations for acute TBI as their Nordic colleagues. Alcohol misuse is a well-known modifiable risk factor for TBI and other injuries. In Finland, the drinking pattern is more intoxication-oriented compared to the other Nordic countries (Ramstedt, 2007). This high frequency of heavy drinking may partly explain the higher number of TBI deaths among Finns. Moreover, assaults as a cause of moderate or severe TBI are relatively common in Finland (13%) compared to the other European countries (0.7%-3.6%) studied by Hukkelhoven et al. (2002), which may also reflect the drinking habits.

According to the systematic review article of brain injury epidemiology in Europe, the incidence in adults with all TBI severities, is in the range of 150-300 per 100 000 per year (Tagliaferri et al. 2006). In their study, Koskinen and Alaranta (2008) found that, during 1991-2005, the average incidence of hospitalized TBI as the primary diagnosis in patients who did not have a previous TBI in their medical history, was 101/ 100 000 in Finland, and ca. 59% of the patients were males. Falls were the most common and traffic accidents the second most common external cause of TBI in all age groups. Falls were especially common in the youngest and oldest age groups. In the age group of 20–29 years, falls (36%) and traffic accidents (34%) were almost equally common. The estimated annual total number of new patients suffering from TBI in Finland is 21 000–31 000 (Koskinen and Alaranta, 2008).

2.1.4. Outcome

Definition

Different tools to define outcome after TBI have been developed. The Glasgow Outcome Scale (GOS) (Jennett and Bond, 1975) has been generally accepted and is one of the most widely used methods of analyzing outcome in head-injured patients (Teasdale et al., 1998). It is used to rate a patient's overall outcome, taking into consideration cognitive and physical impairments, as well as disability in everyday activities. The five categories of the original scale are: dead (1), vegetative (2), severely disabled (3), moderately disabled (4), and good recovery (5). The extended version of the scale (GOS-E) (Wilson et al., 1998) divides each of the latter three categories into two, making eight categories, and thus describes the upper range of outcome in more detail.

Severe and moderate TBI

According to a meta-analysis of studies on severe TBI (Stein et al., 2010), case fatality rates are approximately 35% and have not decreased since 1990. Progress in diagnosis and treatment has been balanced by factors that are associated with poorer outcome, including more virulent mechanisms of injury and higher age of TBI patients (see, e.g. Dutton et al., 2010; Sundstrøm et al., 2007). Mortality (at 6 months or at one year) in moderate TBI has ranged between 9% and 21% in multicentre studies (Andriessen et al., 2011, Compagnone et al., 2009; Murray et al., 1999).

Subjects with moderate-to- severe injuries face a spectrum of cognitive, social, emotional, and behavioural changes from their pre-traumatic level. In the recent multicentre study of Andriessen et al. (2011), 52% of severe and 45% of moderate TBI survivors still experienced some level of disability (GOSE ≤6) at one year post-injury, emphasizing the severe and long-term consequences of the disorder. The major cognitive and functional deficits after moderate TBI include memory problems, concentration difficulties, fatigue, and headaches (Vitaz et al., 2003). The psychosocial problems of decreased social contact, depression, and loneliness remain a persistent long-term problem for the majority of individuals with severe TBI (Morton and Wehman, 1995).

Mild TBI (mTBI)

Mild traumatic brain injury represents about 70-90% of all treated TBIs, and the population-based rate is propably two to six times higher than that of hospital-treated mild TBIs (Holm et al., 2005).

Most individuals with mild TBI recover completely without permanent cognitive or psychological sequelae (Iverson, 2005). A sub group of the patients, however, have prolonged post-concussive symptoms, including cognitive (e.g., poor memory and concentration), somatic (e.g., headache, dizziness), and affective (e.g., depression, irritability) symptoms diminishing the quality of life (McCauley et al., 2001) for these patients. In late, persistent, or prolonged post-concussive syndrome (PPCS), these symptoms last for over six (Evans, 2004) or by other standards, three (Bigler, 2008) months. However, it is not clear that there is a postconcussive "syndrome" per se, or rather common symptoms that occur to greater or lesser degrees in a particular individual as a function of a specific injury and relevant premorbid factors (McAllister, 2011). In the literature, besides the definition, also the indicator of poor outcome varies (from self-reported symptoms to more objective measures, including neurocognitive tests). Several reviews, summarized by McAllister (2011), are consistent in concluding that at group level, most spontaneous cognitive recovery after mTBI is complete by 3 months postinjury. According to the estimate of Iverson (2005), 10-20% of mTBI patients experience incomplete recovery, which makes mTBI a major public health concern.

Predicting outcome after TBI

There are several pre-injury, time of injury and post- injury factors that have been shown to have an influence on outcome after TBI. These factors include age at injury (Kuo et al., 2011), gender (Berry et al., 2009), genetic factors (McAllister, 2011), level of education (Sherer et al., 2002), compensation/litigation (Holm et al., 2005), the force and direction of impact (Ommaya et al., 2002), the size, location, and nature of the brain lesion (Jacobs et al., 2011; Lehtonen et al., 2005; Medana and Esiri, 2003; Perlbarg et al., 2009), preoperative midline shift (Kuo et al., 2011), PTA (Walker et al., 2010), GCS (Machamer et al., 2005), and the psychological and emotional state of the patient (Whelan-Goodinson et al., 2008). The recently published 30-year follow-up study of

Himanen et al. (2011) suggests that besides the higher age at injury and presence of other illnesses, the vocational outcome is associated with long-term survival.

Although a large scale of prognostic factors for traumatic brain injury has been evaluated, clinical classification and conventional neuroimaging techniques fail to accurately predict long-term outcome (Lee et al., 2008; Le and Gean, 2009). Recently, two promising multifactoral models has been developed for outcome prediction in moderate-to-severe TBI: The Corticosteroid Randomization after Significant Head Injury (CRASH) trial (http://www.crash.lshtm.ac.uk/Risk%20calculator/index.html) and the International Mission on Prognosis and Analysis of Clinical Trials in TBI (IMPACT) (http://www.tbi-impact.org/) are freely available on the internet. In both models, age and clinical features with or without CT classification are used as predictive factors. Also blood glucose and hemoglobin values (IMPACT) and geographical location (CRASH) have been taken into account. However, even if these models provide robust probabilistic estimates of outcome in groups of patients, they cannot accurately predict outcome in individuals (Menon and Zahed, 2009).

Structural neuroimaging exams are often administered for diagnostic purposes after traumatic brain injury. Ideally, neuroimaging would provide accurate diagnosis of injury severity that would be informative considering acute treatment and also be predictive for indiduals long-term functional recovery. According to several earlier studies, diffusion-weighted imaging (DWI) (Goetz et al., 2004; Shanmuganathan et al., 2004) and diffusion tensor imaging (DTI) (Caeyenberghs et al., 2010; Kraus et al., 2007; Messé et al., 2011; Niogi et al., 2008; Wang et al., 2008; Warner et al., 2010) may be valuable MRI techniques in outcome prediction.

Predicting whether patients will go on to develop persistent postconcussive syndrome after mild TBI is particularly challenging. However, to find predictors for PPCS is of potential interest, because neurobehavioural rehabilitation reduces the risk of persistent symptoms, and treatment failure is common if symptoms persist after 3 to 6 months (Mittenberg et al., 1996). Previous data of Lange et al. (2012) do not support an association between white matter integrity in the corpus callosum and self-reported postconcussion syndrome 6 to 8 weeks after mild TBI.

2.2. STRUCTURAL IMAGING OF TBI

2.2.1. Structural and technical principles

Structural imaging refers to various techniques including computed tomography (CT) and magnetic resonance (MR) imaging (MRI) that generate static views of the brain. The brain can be compartmentalized into white matter (WM), gray matter (GM), and cerebral spinal fluid (CSF) –filled spaces. The brain's vasculature can be separately imaged using structural imaging techniques, but the vasculature makes up only a small percentage of total brain volume and the microvasculature within GM and WM becomes (except in

cases of a present disease state or hemorrhage) classified as the tissue that it is embedded in. Although a focal lesion, such as a contusion, may appear as an impressive localized lesion on structural imaging, the true neurobehavioural effect of that region of damage is how it disrupts the network of WM pathways (Bigler, 2011). Besides TBI, the changes in the integrity of the WM pathways are related to a wide variety of brain disorders at any age, and the ability to measure these pathways is of great interest in many clinical settings.

CT and MRI analyse a three-dimensional volume or slice of patient tissue to produce a two-dimensional image. The resulting image is built of a matrix of picture elements (pixels), each one representing a volume element (voxel) of patient tissue. CT and MRI define a numeric value for each pixel. The matrix of picture elements that make up each image is usually between 128 x 256 (32 768 pixels) and 560 x 560 (313 600 pixels). To produce an anatomic image, shades of gray are assigned to ranges of pixel values. To analyze optimally all the anatomic information of any particular slice, the image is viewed at different window-width and window-level settings optimized for different tissues (Brant, 2007).

2.2.2. Computed Tomography (CT)

CT produces a volume of data in order to demonstrate different bodily structures based on their ability to block the X-ray beam. CT is highly accurate in the detection of acute intra-axial and extra-axial hemorrhage, as well as skull, temporal bone, facial and orbital fractures (Barr et al., 2007).

In the acute management of a TBI patient, CT imaging is routinely the first imaging method (Coles, 2007; Toyama et al. 2005). CT scans can be performed rapidly, and pathology that is critical for the early medical management of cerebral trauma, such as various types of hemorrhages, clinical presentation of cerebral edema, and presence of midline shift, can be detected by CT (Bigler, 2011). Moreover, CT can safely be performed also in monitored patients and in patients with (or without knowledge about having) a pacemaker or other implanted metallic device. From the rehabilitation and neurobehavioural outcome perspective, CT imaging, however, has a very limited role. Plenty of studies have shown magnetic resonance imaging (MRI) to be superior to CT in the detection of intraparenchymal traumatic lesions, both in the acute and chronic stages, and regardless of the injury severity (See, e.g. Fiser et al., 1998; Hadley et al., 1988; Ichise et al., 1994; Jenkins et al., 1986; Newton et al., 1992; Paterakis et al., 2000). Especially traumatic axonal injuries and brainstem injuries are much better seen by MRI than by CT (Barr et al., 2007).

2.2.3. Conventional Magnetic Resonance (MR) Imaging (MRI)

Magnetic resonance imaging is based on the ability of a small number of tissue protons to absorb and emit radio wave energy when the body is placed within a magnetic field.

Whereas CT evaluates only one tissue parameter, X-ray attenuation, MRI analyzes multiple tissue characteristics including proton (hydrogen) density, T_1 and T_2 relaxation times of tissue, and blood flow within tissue (Brant, 2007). A small number of tissue protons align with the main magnetic field and are afterwards displaced from their alignment by application of radiofrequency (RF) gradients. When the RF gradient is terminated, the displaced protons realign with the main magnetic field, releasing a pulse of energy that is detected, localized, and processed by a computer algorithm (Brant, 2007). Different tissues absorb and release radiowave energy at different characteristic rates. Longitudinal or spin-lattice relaxation (T_1) is the mechanism by which an excited magnetization vector returns to equilibrium (conventionally shown along the z axis), and T_1 is basically a measure of how quickly a tissue can become magnetized. Tranverse or spin-spin relaxation (T_2) is the mechanism by which the excited magnetization vector (conventionally shown in the x-y plane) disintegrates, and it indicates how quickly a tissue loses its magnetization. T_2 is always at least slightly faster than longitudinal relaxation. Proton density-weighted images, respectively, emphasise proton density differences in tissues.

The present MRI technology is based on a relatively large scale of MR sequence techniques, with several variations used by different MR manufacturers. In this study, the following MRI sequences are considered conventional:

Spin-echo (SE)

SE pulse sequences produce conventional (standard) T1-weighted images (T1WIs), T2WIs, and proton density-weighted images. For the SE sequences, the operator selects two major components of MR instrument settings: time of repetition (TR) and time of echo (TE). TR is the time between RF pulses, and the MR signal is acquired at time TE after excitation. SE T1WIs are obtained by selecting short TR ($\leq$ 500 ms) and short TE ($\leq$ 20 ms), and SE T2WIs by selecting long TR (2000 ms) and long TE (70ms). Proton density-weighted images are obtained by minimizing both T1 and T2 effects, which emphasize hydrogen-density differences in tissues. In proton density-weighted images, a long TR (2000 to 3000 ms) and a short TE (25 to 30 ms) are used.

Fast spin-echo (FSE)

Fast spin-echo sequences, also known as turbo spin-echo (TSE), multiple spin-echo, echo train, or rapid-acquisition relaxation-enhanced (RARE) sequences, markedly reduce image acquisition time. Blurring occurs because of lower signal intensity compared with SE sequences (Brant, 2007).

Inversion recovery (IR)

Inversion recovery pulse sequences are used mainly to emphasize differences in T1 relaxation times of tissues. A delay time, the time of inversion (TI), is added to the TE, and TR instrument settings selected by the operator (Brant, 2007).

Fast spoiled gradient echo (FSPGR)

Fast spoiled gradient-echo of three-dimensional MRI data improves the anatomical display of the sulcal structure of the hemispheric convexities.

MRI studies are often performed in the subacute and chronic stages of recovery from TBI, with the intention of answering specific neuropsychiatric questions regarding patient status and/or to assist in evaluating outcome (Ashwal et al., 2006; Levine et al., 2006). However, the clinical use of MRI in imaging of TBI has not achieved a universally accepted role. Even if MRI is superior in demonstrating traumatic lesions compared to CT, its role in clinical decision-making or outcome prediction has given conflicting results (Paterakis et al., 2000; Levin et al., 1987; Wilson et al., 1988; Mitchener et al., 1997; Van der Naalt et al., 1999; Hofman et al., 2001; Hughes et al., 2004; Scheid et al., 2003; Gerber et al., 2004; Tong et al., 2004; Schaefer et al., 2004; Huisman et al., 2004; Pierallini et al., 2000). In more recent studies (Niogi et al., 2008; Lee et al., 2008) the number of abnormalities in conventional MRI has not correlated with cognitive performance in TBI. A seldom raised viewpoint is, however, the role of early imaging in documenting the intracranial injury, since late imaging may fail to demonstrate the trauma- induced changes. Previous systematic studies clarifying the visibility of traumatic axonal injury lesions with time have been practically non-existent.

2.2.4. More advanced MR imaging techniques

The sensitivity in detecting TBI-related anatomical abnormalities by MRI depends on the image sequence and methods used. TBI often leads to persistent symptoms even in patients without findings in conventional MRI (Sheid et al., 2006; Fork et al., 2005). Several studies with relatively modern imaging techniques, such as haemorrhage sensitive T2*-weighted gradient-echo imaging or susceptibility- weighted imaging, magnetization transfer imaging, diffusion-weighted imaging, and diffusion tensor imaging have demonstrated that both CT and conventional MRI are rather insensitive tools for detecting traumatic axonal injury and underestimate the extent of injury (see, e.g. Scheid et al., 2003; Tong et al. 2004; Hofman et al., 2002; Schaefer et al., 2004; Huisman et al., 2004, respectively). The frequency of axonal injury is, however, underestimated even with MR imaging techniques including fluid-attenuated inversion recovery (FLAIR), T2-weighted fast spin-echo, and T2*-weighted gradient-echo (GE) sequences (Parizel et al., 2005; Schaefer et al., 2004). Recently, much interest has been directed towards the potential of DWI and DTI techniques to investigate traumatic axonal injury. Since this study consentrates on the DWI and DTI techniques, the literature of these methods is more widely reviewed.

2.2.4.1. *T2*-Weighted Imaging and Susceptibility-Weighted Imaging (SWI)*

The presence of paramagnetic blood breakdown products results in magnetic susceptibility differences and creates local magnetic field inhomogeneties, which manifest as marked hypointensity on T2*-images. T2*-weighted gradient-echo MR imaging and the high-

resolution gradient-echo MRI technique, known as susceptibility-weighted imaging, are known to be useful in detecting hemorrhagic lesions associated with DAI (Scheid et al., 2003; Tong et al., 2004). SWI is 3-6 times more sensitive than conventional T2*-weighted gradient-echo (T2*GE) sequences in detecting the size, number, volume, and distribution of hemorrhagic lesions in DAI (Mittal et al., 2009).

2.2.4.2. *Magnetization transfer imaging (MTI)*

Magnetization transfer imaging contrast is obtained by applying a radio frequency only to the proton magnetization of the macromolecules based on the interaction and exchange of protons unbound in free water with those bound with macromolecules (Bagley et al., 2000). Tissue damage is visualized as a reduced proton exchange, or a decrease in the magnetization transfer ratio (MTR), whereas an elevated proton exchange, or increased MTR, is evidence of possible remyelination or resolution of edema.

2.2.4.3. *Diffusion-weighted imaging (DWI)*

DWI is an imaging technique acquired on a standard MRI scanner that provides information about the random thermal motion of water molecules and allows quantification of tissue architecture through the degree of tissue water diffusion within a voxel (Basser, 1995). This determination of 'apparent diffusion coefficient' (ADC) or 'mean diffusivity' (MD), represents the overall average measure of diffusion and can provide information on pathological changes not detectable on visual evaluation. The terms mean diffusivity and apparent diffusion coefficient have been used interchangeably and both are used in the literature cited in this study. Based on DWI the lesions can be characterized into those with increased diffusivity and those with decreased diffusivity (Arfanakis et al., 2002; Assaf et al., 1997). The value of DWI in the evaluation of TAI and other traumatic lesions in the acute stage have been demonstrated in several previous studies (Alsop et al., 1996; Ezaki et al., 2006; Hergan et al., 2002; Kinoshita et al., 2005; Liu et al., 1999; Schaefer et al., 2004). Besides the improved lesion detection and characterization, DWI may provide clinically relevant quantitative data on traumatic tissue changes outside the visible lesions.

The precise mechanisms underlying the changes in ADC values associated with acute head trauma are still not fully understood. Huisman et al. (2003) have assumed that the increased ADCs may be a marker of less severe injury, most likely representing vasogenic edema. In severe TBI, edema has been reported to be predominantly cellular, with decreased ADC in the acute stage (Marmarou et al., 2006). During the first and second days after the injury, the majority of shearing injuries have shown decreased ADC values (Assaf et al., 1997; Schaefer et al., 2004). However, after the first days, mainly increased ADCs have been reported, and at one week these have been dominant (Assaf et al., 1997). Earlier studies have shown increased ADC values in normal-appearing brain in acute TBI (Goetz et al., 2004; Inglese et al., 2005; Shanmuganathan et al., 2004), but also reduced ADC in the normal-appearing corpus callosum due to acute TAI has been

reported (Huisman et al., 2004). Previous reports have demonstrated ADC changes of normal appearing brain tissue only in some regions (Goetz et al., 2004; Huisman et al., 2004; Inglese et al., 2005), or in whole-brain histograms (Shanmuganathan et al., 2004). Increased ADC values in whole-brain peak ADCs have shown significant correlation with GCS (Shanmuganathan et al., 2004), and according to the earlier study of Goetz et al. (2004), increased ADC values of normal-appearing white matter in TBI patients correlate with injury severity. The extent of pathological changes, however, has not been clarified. In most previous studies of ADC changes in TBI, the measurements have been performed only in structures in which traumatic axonal injury generally occurs, such as frontotemporal white matter, corpus callosum and corona radiata.

2.2.4.4. *Diffusion tensor imaging (DTI)*

DTI is an imaging technique acquired on a standard MRI scanner that can be used to non-invasively estimate white matter tract integrity in vivo. Both 1.5 T and 3 T MRI scanners are common in clinical use, but typical current "best practice" 3 T DTI protocols differ considerably from older 1.5 T versions, with the inclusion of modern array head coils resulting in higher signal to noise ratios (Vollmar et al., 2010).

DTI provides information about the random thermal motion of water molecules, and allows quantification of tissue architecture through both, the degree and directionality of tissue water diffusion (Basser, 1995; Basser and Pierpaoli, 1996). In human tissues, water diffusion is not free in all directions but hindered and restricted by the presence of barriers, including cell cytostructure and membranes. For example, in the white matter (WM) regions of the brain, where the neuronal projections or axons are similarly aligned, water diffusion is generally greater in the direction along axons than perpendicular to them. This characteristic is referred to as anisotropic diffusion. Anisotropy is commonly expressed relative to the magnitude of the diffusion tensor as the fractional anisotropy (FA) (Basser and Pierpaoli, 1996). The gray matter regions are characterized by a less ordered tissue structure, and diffusion, in contrast to white matter, tends to be less anisotropic and more uniform in all directions.

DTI technique describes the diffusion process using three eigenvectors, of which the magnitudes, the eigenvalues (Alexander et al., 2007), quantify the diffusion in three orthogonal directions. The anisotropic diffusion in the WM of the brain is determined by several factors including the thickness of the myelin sheath and of the axons, and the distribution of directions, and the density of WM fibre tracts. FA ranges from 0 to 1, where 0 represents completely isotropic diffusion (free diffusion) and 1 represents the most anisotropic diffusion (diffusion restricted to one direction). In addition, radial diffusivity (RD) denotes the extent of diffusion perpendicular to the direction of maximal diffusivity, which presumably includes diffusion through intracellular and extracellular space perpendicular to the predominant orientation of the axons. Mean diffusivity (MD), or 'apparent diffusion coefficient' (ADC), refers to the average of the three eigenvalues and represents the overall average measure of diffusion. DTI can, at the macroscopic scale

of a voxel, detect the extent of directional bias of diffusion occurring at the microscopic level. Thus, DTI is able to distinguish between regions where fibres are highly aligned in the voxel from those where fibres are less coherent. Orientation of specific tracts can be visualised on two-dimensional colour maps based on eigenvalues and eigenvectors of the diffusion tensor ellipsoid that determine the shape and orientation, respectively, at each image pixel (Mori et al., 1999).

DTI analysis may be performed in several ways including whole-brain histogram analysis, voxel-based analysis (VBA), region-of-interest (ROI) method, and quantitative fibre tracking (Quantitative diffusion tensor tractography, qDTT):

Whole-Brain histogram Analysis

Histogram analysis can be used to produce a summary of changes to the whole brain. It is fast, easy to implement, does not require spatial warping, and can be used for single-subject analysis. However, whole-brain histogram analysis cannot provide regionally specific information, and thus may lack utility for correlation to deficits in particular in cognitive and behavioural domains (Niogi and Mukherjee, 2010).

Voxel-Based Analysis (VBA)

The VBA analysis method is fully automated, data-driven, and easily reproducible. However, inaccurate spatial normalization of data may introduce errors, which makes interpreting the results problematic since positive results may be a result of local misalignment (Niogi and Mukherjee, 2010). VBA analysis examines the entire brain without the need for a priori hypotheses, which is why these studies are best suited to determine common areas of damage by comparing TBI cohorts to normal controls. However, the spatial heterogeneity of injury in TBI reduces the sensitivity of group comparisons using VBA, because VBA only detects those changes that are common across the entire experimental cohort when compared with normal controls. Moreover, it has not been universally accepted that VBA has sufficient statistical power for single-subject analysis, which may limit its clinical utility (Niogi and Mukherjee, 2010).

Region-Of-Interest (ROI) method

One commonly used approach to DTI analysis is based on cross-sectional identification of a tract of interest by drawing regions of interest (ROIs) for the specified volume. Fractional anisotropy and mean diffusivity can be derived from the calculated tensor for each voxel within the ROI as a quantitative measure of the directionality and total amount of water diffusion of the given ROI volume (Basser and Pierpaoli, 1996). ROI measurement has the advantage of being relatively easy to implement, but, on the other hand, there are several limitations related to the manual ROI definition including inconsistency in placing ROIs and contamination from the CSF or grey matter, as well as inclusion of white matter from different projections (Kanaan et al., 2006; Snook et al., 2007). Additionally, the decision to include or exclude subjects who have incidental

findings, such as nonspecific white matter hyperintensities, can influence the results of a study (Iverson et al., 2011). Most importantly, ROIs typically encompass only a small region of the entire 3D course of the tract of interest, thus reducing the generalizability of the results (Niogi and Mukherjee, 2010).

Quantitative Diffusion Tensor Tractography (qDTT)

Quantitative diffusion tensor tractography is a relatively new method to perform DTI analysis that can be used to assess entire pathways according to diffusion tensor anisotropy, derived from DTI (Wang et al., 2008; Wilde et al., 2008). Fibre tracking algorithms, which use the information provided by the diffusion tensor concerning the orientation of fibres within a voxel, can be used to calculate the representation of WM tracts in 3D, which, in turn, allows DTI metrics such as mean diffusivity, fractional anisotropy, and volume of the whole tract to be measured. The major potential limitation with quantitative tractography is that mTBI may possibly manifest with only small foci of damage, even at the microstructural level, which may be difficult to detect when examining whole-tract scalar measures in pathways that are mostly normal along their 3D trajectories (Niogi and Mukherjee, 2010).

Pathologic processes that alter the microstructure such as loss or disorganization of fibres associated with breakdown of myelin and downstream nerve terminals, neuronal swelling or shrinkage, and increased or decreased extracellular space, may affect both diffusion and anisotropy. FA and ADC/MD together are considered proxies for white matter integrity (Ducreaux et al., 2005). In particular, fractional anisotropy (FA) values have been promising to reveal injured white matter, and in general, FA values are considered more sensitive to show abnormalities than ADC values (Huisman et al., 2004; Inglese et al., 2005; Kraus et al., 2007; Lipton et al., 2009; Niogi and Mukherjee, 2010; Warner et al., 2010). A variety of diffusion changes associated with traumatic axonal injury in subjects with TBI have been reported. However, TAI have been mainly characterized by reduced FA (Arfanakis et al., 2002; Bendlin et al., 2008; Huisman et al., 2004; Kraus et al., 2007; Rutgers et al., 2008; Wang et al., 2008) and/or increased ADC (Bendlin et al., 2008; Huisman et al., 2004; Inglese et al., 2005; Nakayama et al., 2006; Rugg-Gunn et al., 2001).

Earlier clinical DTI studies on TBI have found reduced FA in several white matter areas, both within lesions and in tissue appearing normal on conventional MRI (Arfanakis et al., 2002; Chan et al., 2003; Huisman et al., 2004; Inglese et al., 2005; Nakayama et al., 2006; Ptak et al., 2003; Rugg-Gunn et al., 2001; Salmond et al., 2006). At post-acute stages, ADC in areas of decreased FA has been found to be normal or increased (Chan et al., 2003; Inglese et al., 2005; Nakayama et al., 2006; Rugg-Gunn et al., 2001; Salmond et al., 2006). Several studies have shown widely distributed pathological FA values for various tracts in severe TBI, whereas in milder cases, the injuries may be more local (Niogi et al., 2008; Kraus et al., 2007; Bendlin et al., 2008). Most previous DTI studies of TBI using quantitave tractography have been limited to one or two tracts, such as the

corpus callosum (Nakayama et al., 2006; Wilde et al., 2008), or to only moderate/severe TBI (Warner et al., 2010; Wang et al., 2008; Caeyenberghs et al., 2010).

2.2.5. Structural imaging of TBI: Recent development

At the moment, DTI may be the most promising neuroimaging technique in TBI; besides the improved detection and localisation of the traumatic lesions without ionizing radiation, these lesions have in several studies correlated well with cognitive performance (Caeyenberghs et al., 2010; Kraus et al., 2007; Messé et al., 2011; Niogi et al., 2008; Wang et al., 2008; Warner et al., 2010). Moreover, the addition of DTI measures adjusted for age, gender, and admission GCS score may significantly improve prognostic models (Betz et al., 2012).

Although these results are promising, healthy scepticism and caution should be exercised with regard to interpreting their meaning because there is no consensus about which methods of data analysis to use and relatively few investigations have been conducted, of which most have been small in sample size and have examined patients at only one time point after injury (Grossman et al., 2010). Diffusion tensor tractography is increasingly used to examine structural connectivity in the brain in various conditions, but its reproducibility is understudied; there is still a need for reproducible imaging protocols and analysis methods applicable for clinical use in the diagnostics of TBI and other cognitive disorders.

2.3. RELIABILITY OF THE STRUCTURAL IMAGING OF TBI

The concept of reliability is complex, but the decicion on whether a particular measurement or tool is of any value or not, may basically be made by answering the two questions: How reliable the measurement or tool is in itself, and how reliable raters are in performing it.

Pathology that is critical to the early medical management of cerebral trauma can be detected by CT (Bigler, 2011), but radiologists on call miss a significant number of brain contusions on acute brain CT, and there is significant variation in the detection of brain contusions even among the most experienced readers (Laalo et al., 2009). In MRI (unlike in CT), there are, besides the reader-dependent factors, numerous variables that can be adjusted during the acquisition of imaging that may have an influence on the reliability. As described before, a growing body of literature suggests that both CT and conventional MRI techniques are rather insensitive tools for detecting the full extent of TBI, especially traumatic axonal injury, and they fail to accurately predict long-term outcome.

Several factors that influence the reproducibility of DTI measures have been studied in the literature. Scanner parameters including field strength and parallel imaging cf. non-parallel imaging (Alexander et al., 2006), as well as acquisition parameters such as voxel

size (Vollmar et al., 2010), and use of motion correction (Ling et al., 2011), and cardiac gating (Chung et al., 2010), as well as the number of diffusion-weighted directions (Ni et al., 2006; Wang et al., 2012), may all influence the reproducibility of the DTI-based analyses. Besides the technical factors, the role of rater performance in reproducibility is important, since many of the technical factors may be standardized relatively easily.

In clinical DTI studies, tracts of interest are often identified on cross-sectional images, and the measurement is performed by drawing ROIs. Limitations related to the manual ROI definition include inconsistency in placing ROIs and contamination from the CSF or grey matter, as well as inclusion of white matter from different projections (Kanaan et al., 2006; Snook et al., 2007). The degree of reproducibility in cross-sectional ROI approaches is related to the ROI shape and size; a small ROI is more prone to noise and partial volume effects so that increasing ROI size improves reproducibility, as long as contamination from surrounding structures with markedly different voxel values is avoided (Vollmar et al., 2010).

Another tool used to identify specific white matter tracts is DTI tractography. There are various ways to perform quantitative fibre tracking analysis. The most common methods for isolating fibre tracts include the use of multiple inclusion and exclusion ROIs (Mori and van Zijl, 2002; Wakana et al., 2007). Tract-based analyses have several advantages compared to the cross-sectional ROI analyses including the larger number of voxels that reduce the measurement variance (Kanaan et al., 2006), as well as the higher level of automaticity and lower level of subjectivity (Borich et al., 2012). The need for many ROIs to isolate the specific tract can, however, diminish the reliability of the tract-based methods (Malykhin et al., 2008). Another tractography-technique-related limitation is the choosing of the appropriate FA threshold and deflection angle (Niogi and Mukherjee, 2010). By using a low FA threshold, the peripheral parts of the tracts are included, while tractography with a high FA threshold concentrates on the core of the tract. Moreover, the large volume variability of the WM tracts (Hasan et al., 2010) can decrease the accuracy of FA and MD measurements. Additional measurements from the core of the tract may be used to confirm the tract-based results (Kurki et al., 2012; Yasmin et al., 2008). Tractography with a high FA threshold provides a novel alternative possibility to perform the central part analysis in corresponding volumes representing the region with highest FA values in each subject (Kurki et al., 2012).

Reliability results concerning the cross-sectional ROI measurements in a healthy population have been somewhat controversial (Bisdas et al., 2008; Brander et al., 2010; Vollmar et al., 2010), and the reproducibility of the ROI measurements has been shown to vary regionally and depending on the ROI drawing method (Bisdas et al., 2008; Brander et al., 2010; Hakulinen et al., 2012). Only few previous reliability studies on the cross-sectional ROI measurements of the association tracts have been published. Previously, the semi-automated tractography-based MD and FA measurements have been reported to provide good to excellent intra- and inter-rater reliability in a healthy adult population (Wakana et al., 2007), and an excellent intra- and inter-rater reliability

in a combination of healthy and depressed subjects (Malykhin et al., 2008) in several WM tracts. Reliability of the tract volumes has shown more variation, and in the study of Malykhin et al. (2008), the least reliable measurements were for volumes of the uncinate fasciculus and rostral cingulum.

In general, no single reliability estimate should be used for reliability studies (Bruton et al., 2000). As a reliability estimate, it is preferable to use the ICC rather than the CV, as the former relates the size of the error variation to the size of the variation of interest, but even the ICC analysis has some limitations; Since the ICC is calculated as a comparison of variation between cases (e.g., volume, FA or MD values of the tracts in the study population) to variation within cases (e.g., across raters), ICC may be high with a relatively large variation within cases if there is a large variation between cases (Bruton et al., 2000). Bland and altman's 95% limits of agreement test gives additional information on the magnitude of disagreement between measurements that can be interpreted clinically (Bruton et al., 2000; Rankin and Stokes, 1998). However, in previous DTI reliability studies, the use of this coefficient of repeatability (CR) analysis has been rare.

Besides TBI, the changes in the integrity of the white matter tracts are related to a wide variety of brain disorders at any age, which should be kept on mind in the clinical interpretation of the results, as well as in legal proceedings (Wortzel et al., 2011). Reduced FA and an increased MD are nonspecific findings in most pathological processes affecting the brain's parenchyma (Hakulinen et al., 2012; Wortzel et al., 2011). Reduced FA has been detected even in heavy cigarette smokers and in internet addiction disorder subjects (Lin et al., 2012; Lin et al., 2013). It is known that age also affects both FA and ADC values, and that small changes occur across the lifespan and even in different ways in men and women (Sullivan et al., 2001; Hakulinen et al., 2012). At present, there is no gold standard for validating diffusion measures, which are dependent on the scanning protocols, software methods, and observers (Hakulinen et al., 2012). There is a need for reproducible imaging protocols and analysis methods applicable for clinical use in the diagnostics of TBI and other cognitive disorders.

3. AIMS

The general aim was to clarify the role of magnetic resonance imaging in the detection of pathologic changes following traumatic brain injury and investigate how to improve the radiological diagnostics of traumatic brain injury patients with persistent symptoms but with only few or no findings on conventional structural imaging (i.e. CT or routine MRI).

The specific aims were:

1. to investigate the visibility of traumatic brain lesions on conventional MRI in early and late phase, with the hypothesis that there is a significant decrease in both the number and the volume of traumatic lesions with time

2. to study the occurrence and distribution of ADC changes throughout the normal-appearing brain in acute TBI and, by correlating the ADC values with both the acute injury severity and outcome scores, to estimate the clinical significance of the eventual changes

3. to evaluate the number and type of abnormalities in quantitative diffusion tensor tractography of long association tracts in subjects with symptoms after traumatic brain injury without traumatic findings on routine MRI

4. to analyze the reproducibility of tractography-based measurements compared with conventional core ROI measurements by using a DTI sequence suitable for clinical protocols

4. SUBJECTS AND METHODS

4.1. SUBJECTS WITH TBI (I-II) AND CONTROLS (II)

In this prospective study, 36 consecutive unselected patients attending the emergency department of Turku University Hospital due to TBI were studied one week (7 ± 2 days) and again one year (384 ± 29 days) after the trauma. The severity of the brain trauma was assessed by the Glasgow Coma Scale on arrival at the hospital and by the duration of post-traumatic amnesia (PTA), which was defined as the earliest time point for the recovery of continuous memory. The outcome was assessed one year after trauma by the Glasgow Outcome Scale, extended version (GOS-E) (Jennett et al., 1981). Subjective symptoms, both at one week and at one year after the injury, were assessed by the Head Injury Symptom Checklist (HISC) (McLean et al., 1984).

The inclusion criteria for the TBI patients were: 1) acute (<3 days) brain trauma with one or more of the following: a) loss of consciousness for at least one min, b) PTA for at least 30 min, c) focal neurological symptoms or signs of brain injury during the first three days (hemiparesis, confusion etc.) not attributable to other factors, and d) neuroradiological findings demonstrating acute TBI; 2) age 16 - 70 years; and 3) informed consent. Patients with other central nervous system diseases or contraindications to MRI were excluded.

The temporally first 10 of the patients were studied on a Siemens Magnetom 1.5 T MR imaging system, and the following 26 were studied on a General Electric Signa 1.5 T MRI system. Only the patients studied on the General Electric Signa MRI system were included in study **II**. Of these 26 patients, another four were excluded because of the lack of their MR imaging information at the time of analysis.

The TBI patients were divided into two groups; patients without visible lesions on conventional MRI images (n=16 (**I**); n=7 =Group A (**II**)), and patients with visible traumatic lesions (n=20 (**I**); n=15 =Group B (**II**)). In study **II,** fourteen healthy subjects served as controls and formed group C. The age and sex of the patients and controls did not differ significantly from each other.

Detailed dermographics of the patients and control subjects are presented in **Table 2**.

The majority of patients were male (69.4% (**I**); 77.3% (**II**)), and the mean age of the patients was 42.2 ± 16.7 years (**I**) and 41.8 ± 18.1 years (**II**). Most injuries were caused by a fall.

TABLE 2. Demographics of the subjects in studies **I** and **II**. Mean ± SD.

	Group A, **I** Normal MRI (n=16)	Group B, **I** Positive MRI (n=20)	Group A, **II** Normal MRI (n=7)	Group B, **II** Positive MRI (n=15)
Age[1] (range)	43 ± 14 (20-67)	42 ± 19 (16-67)	40 ± 18 (20-67)	42 ± 19 (18-67)
Sex[2] (m/f, n)	9/7	16/4	4/3	13/2
Cause of injury[2] (n)				
- traffic accident	3	7	1	6
- fall	13	13	6	9
Duration of PTA[3] (n)				
- < 24 h	15	10		
- 1 – 7 days	1	6		
- > 7 days	-	4		
Initial GCS score[3]	14.2 ± 0.7	13.1 ± 2.7	14.3 ± 0.8	12.7 ± 3.0
- range	13-15	6-15	13-15	6-15
GOS-E at one year[3]	7.1 ± 0.6	6.5 ± 1.1	7.1 ± 0.7	6.5 ± 1.2
- range	6-8	5-8	6-8	5-8
HISC at one year[1]	4.4 ± 4.4	5.4 ± 6.1		
- range	0-15	0-18		

PTA=Post Traumatic Amnesia; GCS=Glasgow Coma Scale; GOS-E= Glasgow Outcome Scale, extended version; HISC=Head Injury Symptom Checklist
[1]Spearman's Rank Correlation test
[2]One-way ANOVA
[3]Kruskal-Wallis test **Significant at level $p<0.01$

4.2. SUBJECTS WITH TBI AND CONTROLS (III, IV)

Demographics of the patients and control subjects are presented in **Table 3**.

III: The study population consisted of 153 consecutive clinical patients referred to 3 T MRI with advanced imaging sequences between July 2008 and December 2011, because of persisting symptoms following TBI, and with no TBI findings on conventional MRI. Subjects who had nonspecific white matter T2 hyperintensities with a maximum diameter less than 5 mm were included. This study was prospective, but the clinical characterization of the examined patients was studied retrospectively.

The criteria for including subjects with TBI in this study were: 1) TBI leading to loss of consciousness and/or posttraumatic amnesia; 2) no other neurological or vascular disease, chronic hypertension or chronic alcoholism; 3) no signs of other brain diseases in routine MRI; 4) age 16-56 years, 5) time from injury ≥ 3 months, and 6) detailed clinical background information available. Of the 153 examined patients, 30 were excluded for lack of detailed clinical information, 13 for a time between trauma and MRI of less than three months, two for an age of over 56 years, and one for a lack of MRI data analysis, resulting in a total of 106 patients who were finally included (55 men and 51 women; aged 16-56 years, mean 38.4±10.8 years (male 38.4±11.7 and female 38.3±10.0 years)).

Group C, **II** Controls (n=14)	p-values A-B (**I**)	A-B (**II**)	A-C	B-C
43 ± 17 (21-73)	0.696	0.919	0.857	0.966
10/4	0.131	0.136	0.537	0.329
	0.439	0.248		
	0.005**			
	0.592	0.396		
	0.122	0.254		
	0.900			

An experienced neurological evaluation and a detailed neuropsychological examination had been performed in all patients, revealing common TBI-related symptoms and cognitive deficits (such as fatigue, impaired memory, attentional deficits, executional deficits) clinically related to their TBI. Severity of the TBI was defined on the basis of both the GCS and duration of the PTA as follows: mild TBI= GCS 13-15, PTA < 24 h; moderate TBI= GCS 9-12, PTA 1-7 days; severe TBI= GCS 3-8, PTA 1-4 weeks; very severe TBI= PTA more than 4 weeks. If the GCS and PTA criteria were not consistent, the more severe criteria were chosen. Outcome was assessed using the GOS-E.

The injury was caused by a traffic accident in 55 patients, by a fall in 27 patients, by violence in 8 patients, and by other factors in 16 patients. The median time interval between injury and MR investigation was 41 months (minimum 3 months, maximum 40 years).

The results were compared with 62 age- and sex-matched healthy persons with no signs of brain abnormalities in MRI (31 men and 31 women; aged 16–55 years, mean 35.5±11.3 years). A questionnaire was used to exclude earlier TBI and other brain diseases in control subjects.

IV: To evaluate intra-rater and inter-rater reproducibility of the DTI-related measurements, DTI images of 15 participants (n= 10 TBI sequels and 5 healthy subjects: mean age

39.7±12.2 years, range 18-58 years) were randomly selected from a larger population that had been studied with DTI methodology; these participants formed *Group A*. Ten participants (n= 3 TBI sequels and 7 healthy subjects: mean age 36.0±11.4 years, range 18-55 years) were scanned twice 8-210 days apart in the same scanner with the same DTI protocol to find measurements with high between-scan consistency; they formed *Group B*.

TABLE 3. Demographics of the subjects in studies **III** and **IV**. Mean ± SD.

	Controls (**III**) (n = 62)	Patients (**III**) (n = 106)	Group A[3] (**IV**) (n = 15)	Group B[4] (**IV**) (n = 10)
Age (range) y	35.5 ± 11.3 (16-55)	38.4 ± 10.8 (16-56)	39.7±12.2 (18-58)	36.0±11.4 (18-55)
Sex (m/f, n)	31/31	55/51	10/5	4/6
Cause of injury (n)				
- traffic accident		55		
- fall	27			
- violence		8		
- other[1]	16			
Initial severity[2] (range)	1.9 ± 0.79 (1-4)			
GOS-E score (range)	6.2 ± 0.95 (8-4)			
Median time from TBI (range, months)	41 (3-14 827)			

[1] Mostly bike accidents and object strikes the head

[2] Severity of TBI was defined on the basis of both Glasgow Coma Scale (GCS) and duration of posttraumatic amnesia (PTA) as follows: 1= mild TBI= GCS 13-15, PTA < 24 h; 2= moderate TBI= GCS 9-12, PTA 1-7 days; 3= severe TBI= GCS 3-8, PTA 1-4 weeks; 4= very severe TBI= PTA more than 4 weeks. If the GCS and PTA criteria were not consistent, the more severe criteria were chosen.

[3] n= 10 TBI sequels and 5 healthy subjects

[4] n= 3 TBI sequels and 7 healthy subjects

4.3. GATHERING THE DATA

4.3.1. MR image acquisition and image processing (I-II)

I: Ten patients were studied on a 1.5 T scanner (Siemens Magnetom, Erlangen, Germany), and the following sequences were used for analysis:

1) T2-weighted axial turbo spin echo (TSE) 3500/93 ms (repetition time/ echo time) with one acquisition, slice thickness 5.0 mm, data acquisition matrix 192 256 and field of view (FOV) 23.0 cm;

2) T1-weighted coronal SE 600/15 with two acquisitions, slice thickness 7.0 mm, data acquisition matrix 192 256 and FOV 23.0 cm;

3) T2/PD-weighted coronal TSE 3500/93/19 with one acquisition, slice thickness 4.0 mm, data acquisition matrix 192 256 and FOV 23.0 cm.

I, II: Twenty-six patients were studied on an another 1.5 T scanner (General Electric Signa, Milwaukee, Wisconsin, USA) using the following sequences:

1) T2-weighted axial fast spin echo (FSE) 4520/81.6 with 2 acquisitions, slice thickness 5.0 mm, data acquisition matrix 512 x 224 and FOV 24 x 18 cm,

2) Fluid-attenuated T2-weighted fast spin echo inversion recovery (FSEIR) 10002/172.5 with inversion time 2200 ms, one acquisition, slice thickness 7.0 mm; data acquisition matrix 256 x 192 and FOV 24.0 cm, and

3) 3D fast spoiled gradient echo (FSPGR) 11.3/4.2, flip angle 20, one acquisition, data acquisition matrix 256 x 192, slice thickness 1.2 mm, FOV 22 x 17.6 cm, 124 contiguous axial slices with no interslice gap.

Because of missing MRI information for four patients at the time of analysis, only twenty-two of the patients were included in study **II**.

II: Additional T2*-weighted or EPI images were used to detect hemorrhagic lesions. DWI was performed using a spin-echo echo planar imaging sequence (SE-EPI) 3000/98.8, one acquisition, slice thickness 5.0 mm, data acquisition matrix 128 x 128, FOV 24.0 cm. The diffusion was measured in three orthogonal directions using b values of 0 and 1,000 s/mm^2. Three sets of orthogonal images and one set of trace images were generated on line. The images were transferred to a separate workstation, and ADC maps were generated using commercial software (Functool, GE Medical Systems).

4.3.2. Classification of the intraparenchymal lesions (I)

Primary intraparenchymal lesions were classified by their location into cortical contusions and other traumatic lesions. The MRI examinations were analysed by an experienced neuroradiologist. The early and late MRI studies were reviewed on different days.

Contusions

Rating of contusions was done according to their number in different anatomical locations and their size. The extent of contusions was rated on a scale of 0–3, in which 1 indicated contusions that included cortex and white matter (WM), together less than 0.5 cm, while 2 indicated contusions that included WM from 0.5 to 2 cm, and 3 indicated contusions that included WM over 2 cm.

Other intraparenchymal lesions

Other intraparenchymal lesions were characterized as traumatic on the basis of their location and characteristics. These lesions consisted of traumatic axonal injuries and traumatic deep grey matter injuries. In general, haemorrhagic white matter changes without adjacent cortical lesion or lesions at the grey–white matter junction, in the corpus callosum, corona radiata and the dorsolateral upper brain stem, were characterized as

traumatic axonal injury. Visual rating of the traumatic lesions on T2-weighted images was done using a modified Scheltens semi-quantative rating scale (Scheltens et al., 1993). Both T2-hyperintense and T2-hypointense changes were included.

4.3.3. Apparent diffusion coefficient (ADC) measurements and regions of interest (ROI) analysis (II)

The ADC measurements were performed in 46 different anatomical locations including bilateral cortical, subcortical and deep brain structures using the region-of-interest (ROI) method.

The measurements were performed using oval ROIs under the guidance of an experienced neuroradiologist (TK). The white matter atlas (Diffusion Tensor Imaging Atlas of the Brain's White Matter Tracts (www.dtiatlas.org/) was used to ensure correct ROI placements and delineations.

All measurements were performed outside the visible traumatic lesions. In addition to EPI (b=0) and trace diffusion images, orthogonal images were also used to separate various white matter structures. The fasciculi were visualized as structures with relatively low signal intensity on images with anteroposterior (y direction) diffusion gradient and relatively high signal intensity on other orthogonal images. Based on orthogonal DWI, superior longitudinal and subcallosal fasciculi and cingulum could be differentiated from other white matter, but in inferior frontal and deep temporal regions with several adjacent fasciculi, specific tracts were not defined.

4.3.4. Cognitive and neuropsychological methods (I-III)

The Glasgow Outcome Scale (GOS) has been generally accepted and one of the most widely used methods of analyzing outcome in head-injured patients (Teasdale et al., 1998). It is used to rate a patient's overall outcome, taking into consideration cognitive and physical impairments, as well as disability in everyday activities. In this thesis (**I-III**), the extended version of the scale (GOS-E) has been used, because patients with a mild or moderate head injury in general end up with good recovery or only moderate disability, and this scale describes this upper range of outcome in more detail. Additionally, subjective symptoms both at one week and at one year after the injury, were assessed by the Head Injury Symptom Checklist (McLean et al., 1984) (**I**).

4.3.5. MR image acquisition and image processing (III-IV)

MR data acquisition

MRI was performed at 3 T (Achieva, Philips Medical Systems, Best, the Netherlands) using a sensitivity encoding (SENSE) 8-channel transmit-receive head coil. The imaging protocol consisted of the following:

1) transverse T2-weighted turbo spin echo images (TR/TE 3405/80 ms, 40 slices with 3.0 mm thickness, 0.3 mm gap; 296 x 560r matrix, turbo factor 11, FOV 240 mm, RFOV 84%, number of excitations 1, imaging time 2 min 37 s);

2) coronal fluid attenuated inversion recovery (FLAIR) images (TR/TE/TI 11000/125/2800 ms, 32 slices with 4.0 mm thickness, 1.0 mm gap; 231 x 512r matrix, turbo factor 31, FOV 230 mm, RFOV 88%, number of excitations 1, imaging time 2 min 45 s);

3) sagittal 3D FLAIR images (TR/TE/TI 8000/338/2400 ms, 200 slices with 1.6 mm thickness, interval 0.8 mm; 129 x 432r matrix, turbo factor 110, FOV 230 mm, RFOV 100%, number of excitations 1, imaging time 4 min 00 s);

4) sagittal 3DT1 turbo field echo images, (TR/TE/TI 8.3/3.8/1032 ms, 165 slices with 1.0 mm thickness, 0.0 mm gap; 242 x 288r matrix, turbo factor 240, flip angle 8^{o}, FOV 244 mm, RFOV 105%, number of excitations 1, imaging time 5 min 29 s);

5) transverse susceptibility-weighted fast field echo images (venous BOLD) (TR/TE 17/24 ms, 144 slices with 1.8 mm thickness, interval 0.9 mm; 201 x 512r matrix, turbo factor 21, flip angle 15^{o}, FOV 230 mm, RFOV 80%, number of excitations 1, imaging time 2 min 52 s); transverse minimum intensity projection images were created from the original images with a slice thickness of 4.0 mm;

6) transverse DTI images; diffusion-weighted turbo spin echo EPI images (TR/TE 5877/62, 60 slices with 2.0 mm thickness, gap 0.0 mm, 112 x 128r matrix, turbo factor 59, EPI factor 59, FOV 224 mm, RFOV 100%, number of excitations 2, imaging time 3 min 52 s); b values of 0 and 800 sec/mm2 and 15 different gradient encoding directions were used, and isotropic images with 2.0 x 2.0 x 2.0 mm voxel size were obtained.

The images were post-processed with the Philips Diffusion Registration Tool (Philips, Medical Systems, Best, the Netherlands) to remove distortions and misalignments due to shear and eddy current, as well as head motion (Netsch et al., 2004; Rohde et al., 2004). All transverse images were obtained according to the line between the lower border of the genu and splenium of the corpus callosum.

Only DTI images were used for analysis in the reliability study (**IV**). Before the further DTI- based analysis, DTI images were, however, fused with the 3DT1 images to obtain more exact anatomical landmarks.

MR data analysis

The conventional images of the subjects were evaluated, as was the original report, by a neuroradiologist (TK) to exclude the signs of TBI on conventional images (**III**).

The association tracts studied were the uncinate fasciculus (UF) (**III,IV**), superior cingulum (SC) (**III,IV**), temporal cingulum (TC) (**III**), superior longitudinal fasciculus (SLF, only fibres between the frontal and parietal/occipital lobes were included) (**III,IV**), arcuate fasciculus (AF, only fibres between the temporal and frontal lobes were included) (**III,IV**), inferior fronto-occipital fasciculus (IFOF, encompassing only connections between the frontal and occipital lobes) (**III,IV**) and inferior longitudinal fasciculus (ILF, fibres between the occipital lobe and medial temporal, inferior temporal, or fusiform gyri were included) (**III**).

One of the raters (TK) had four years' experience in DTI data acquisition and analysis, and provided DTI-specific training for the other two raters (radiology resident (NB) (**III, IV**) and neuroradiologist (JL) (**IV**)). White matter integrity was quantified by the FA and ADC values. The principal fibre orientation within each pixel was visualized by colour-coded orientation maps (red: right-left, blue: superior-inferior, green: anterior-posterior). Regardless of the analysis approach used (**IV**), the same fused DTI and 3DT1 images with a FA colour map for each subject were used for each subsequent analysis step.

III: The tract-based DTI analyses of the 14 assosiation tracts in TBI patients and healthy controls were assessed by one rater with 14 years' experience in neuroradiology. An increase in diffusivity, or decrease in volume or FA of more than 2 SD from the mean of controls was regarded as abnormal. Separate normal values were used for men and women. To determine the intra-rater reliability of the ROI placements related to the tract-based measurements, repeated placement of ten tracts (both UF, SC, SLF, AF and IFOF) on a subset of 10 TBI images at an interval of over two weeks was studied. To determine the inter-observer reliability, another rater analyzed ten of the tracts from a subset of 10 TBI images.

IV (*Group A*): The tract-based and cross-sectional DTI analyses of the 10 association tracts were assessed by three raters to evaluate the inter-rater reliability. One rater was selected to repeat the measures after a two-week period to evaluate the intra-rater reliability.

IV (*Group B*): To study the between-scan reproducibility, two repeated tract-based DTI-analysis approaches using data from the DTI examinations at two different time point (8-210 days apart) were assessed for each of the 10 participants by one rater.

*Cross-sectional ROI quantification (***IV***)*

The UF, SC, SLF, AF and IFOF were delineated bilaterally with defined standard ROI sizes (5-8 voxels for UF, AF and ILF; 10-15 voxels for SC and SLF) and locations (UF: blue in the axial slice, level of the upper temporal lobe; SC: green in the coronal slice, level of the corpus mamillare; SLF: green in the axial slice, level of the upper back part of the aqueduct; AF: blue in the axial slice, level of the middle splenium of the corpus callosum; IFOF: green in the coronal slice, above the blue UF) **(Figure 1).**

The manually defined ROIs were drawn consulting FA maps, colour maps and a white matter atlas. The mean FA and ADC values from the voxels within the manually defined ROIs were recorded for subsequent statistical analyses. Each ROI measurement was repeated three times, and values from the ROI with the lowest FA were used for further analysis.

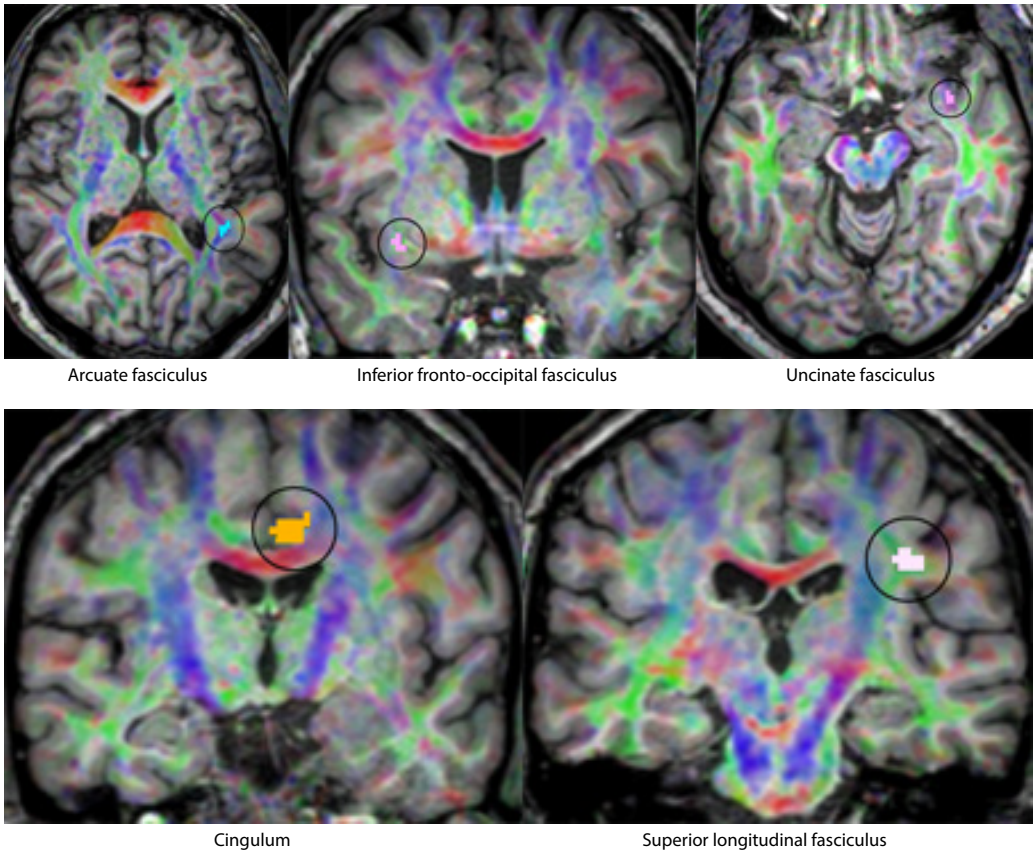


Figure 1. ROI places for the cross-sectional quantification.

Semi-automated tract-based quantification

Deterministic DTI tractography (FiberTrak package, Philips) was performed, and the tracts were defined by means of two or three free-hand inclusion ROIs, placed in standard positions according to anatomical landmarks. Regions of interest were drawn on the baseline image of the DTI scans. All ROIs were placed bilaterally. For uncinate fasciculus, two ROIs were drawn on each side of the same coronal slice, one just anterior to the temporal stem where the frontal cortex begins, and the other in the temporal pole. For the superior cingulum bundle, two ROIs were drawn in the antero-posterior direction on coronal slices around the cingulum on each side, posterior ROIs at the level of the upper back part of the aqveduct, and anterior ROIs between the commissura

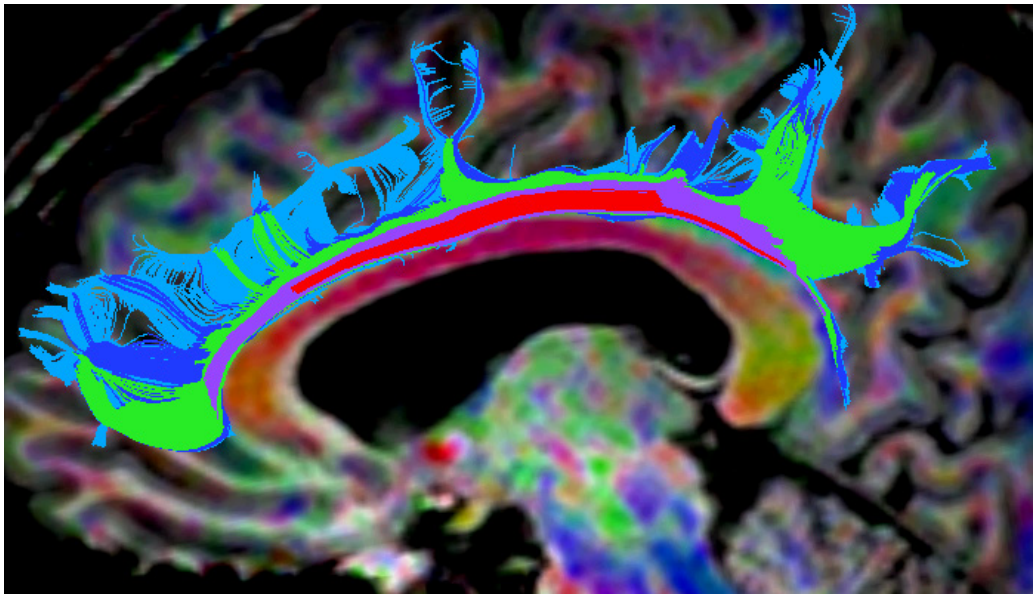
anterior and back part of the rostrum of the corpus callosum. For the temporal cingulum bundle, two ROIs were drawn on coronal slices around the cingulum (parahippocampal gyrrus) on each side, one ROI at the level of the upper back part of the aqueduct, and another ROI at the level of the middle splenium of the corpus callosum. For the superior longitudinal fasciculus, two ROIs were drawn on coronal slices. The anterior ROI was drawn just behind the corpus mamillare, and the posterior ROI at the level of the upper back part of the aqueduct. Connections to the temporal lobe were manually removed with exclusion ROIs at the level of the middle part of the splenium; this removed part of the SLF formed the arcuate fasciculus. For the inferior occipito-frontal fasciculus, two ROIs were drawn on coronal slices, one just anterior to the temporal stem where the frontal cortex begins, and one in the occipital cortex in the most anterior slice where the occipital cortex was visualised. For the inferior longitudinal fasciculus, two ROIs were drawn on coronal slices, one at the level of the upper back part of the aqueduct and one in the occipital cortex in the most anterior slice where the occipital cortex was visualised.

After the ROIs were delineated and the fibre tracts were reconstructed, aberrant fibres were manually removed with exclusion ROIs to include only fibres within the desired tract. Fibres crossing the midline and fibres to the thalami or brainstem were removed from all trajectories. Volume and FA and MD values from the remaining fibres were used for subsequent statistical analysis.

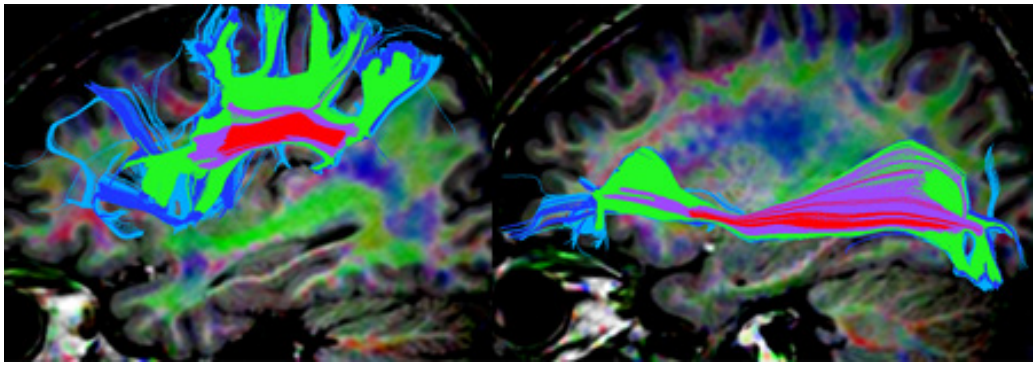
III: The tractography was performed using the following FA and turning angle threshold to terminate the tracking process: FA 0.15/angle 27°.

IV (*Group A*): DTI tractography was performed using the following FA and turning angle threshold to terminate the tracking process: FA 0.15/angle 27° (UF, SC, SLF and ILF) or FA 0.15/angle 60° (AF). Additional volume-based analysis in the central part of the tract was performed, by repeating fiber tracking at increased FA thresholds with the same ROIs until the desired volume was achieved. Tract volumes closest to the predetermined size were used in the study: 3 cm3 for UFs, SCs and AFs; 6 cm3 for SLFs and IFOFs.

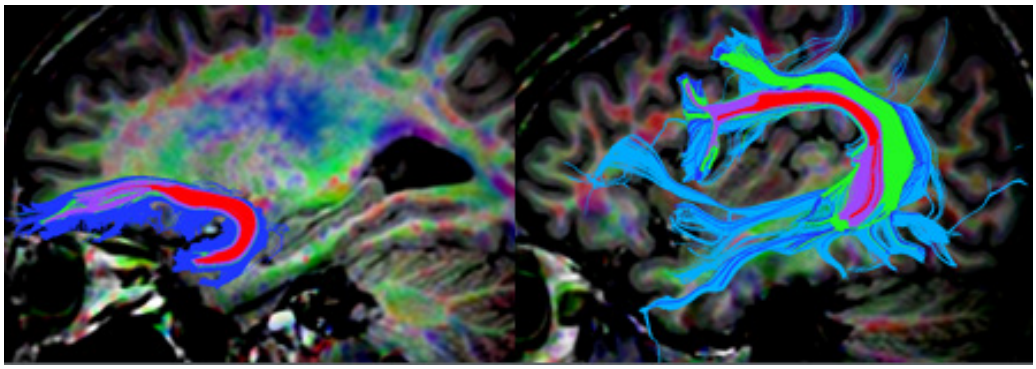
IV (*Group B*): The tractography was performed using two different FAs and turning angle thresholds for each tract to evaluate whether the lower FA and higher turning angle threshold decrease the reliability: FA 0.15/angle 27°; FA 0.15/angle 60°; FA 0.30/angle 27° (**Figure 2**). The additional volume-based analysis in the central part of the tract was performed with two predetermined sizes for each tract: (3cm3 and 1cm3 for both UFs, SCs and AFs; 6cm3 and 2cm3 for both SLFs and IFOFs) (**Figure 2**).



Cingulum



Superior longitudinal fasciculus Inferior fronto-occipital fasciculus



Uncinate fasciculus Arcuate fasciculus

Figure 2. Tractography of the superior cingulum, superior longitudinal fasciculus, inferior fronto-occipital fasciculus, uncinate fasciculus and arcuate fasciculus using the following FA and turning angle threshold to terminate the tracking process: FA 0.15/angle 27° (light blue); FA 0.15/angle 60° (dark blue); FA 0.30/angle 27° (green). The additional volume-based analysis in the central part of the tract was performed with two predetermined sizes: 3cm3 (purple) and 1cm3 (red).

4.4. STATISTICAL DATA ANALYSIS

The Wilcoxon signed rank test was used for the statistical analyses of MRI changes between various time points (**I**). The associations between radiological findings and clinical variables were studied with Spearman's correlation coefficient, one-way ANOVA, and Kruskal-Wallis statistics (**I, II**).

The Mann-Whitney U test was used for the statistical analyses of changes in ADC values between the various groups (**II**). The association between ADC values and GCS or GOS-E scores was analysed with the Kruskal-Wallis test (**II**).

The results of study **III** are based on repeated measures analysis of covariance, which takes into account correlated responses between and within subjects and fasciculi. Estimates are adjusted for multiplicity using the simulation method in the SAS software MIXED procedure. Gender, age, time from injury, trauma mechanism, and severity of the TBI were considered as covariates for the tests. Pearson's correlations were employed to demonstrate the relationships between various quantitative values. Differences in the correlations between patients and controls were evaluated using the same Repeated Measures ANCOVA design. The significant differences were defined by the slope estimates of the higher order interaction terms. The association between tract characteristics and outcome, was assessed using multivariate logistic regression with odds ratios, and 95% confidence intervals were calculated. To determine the intra-observer reliability of the ROI placements related to the tract-based measurements, repeated placement of ten tracts (both UF, SC, SLF, AF and ILF) on a subset of 10 TBI images at an interval of over two weeks was studied. To determine the inter-observer reliability, another rater analyzed ten of the tracts from a subset of 10 TBI images. Two-way mixed effects models (i.e. ICC (3,1), where rater and rating effects were random and measure effects of individual ratings were fixed, were used to calculate ICCs between raters and within rater (Shrout and Fleiss, 1979). The condition "absolute agreement" was chosen. Raters were considered as having a fixed effect because they were not randomly selected. As with other reliability coefficients, there is no standard acceptable level of reliability using the ICC. In our study, similarly to the recent study of Borich et al. (2012), ICC values greater than 0.75 were considered to demonstrate excellent reliability, values between 0.40 and 0.75 fair to good reliability, and values less than 0.40 poor reliability. Chinn (1991) recommends that any measure should have an intra-class correlation coefficient of at least 0.6 to be useful.

IV: To assess the inter-rater and intra-rater reliability of each analysis method, intra-class correlation coefficients (ICCs) for mean tract volumes, for mean FA, and for mean MD of each association tract were calculated. Separate two-way random effects models (i.e. ICC (2,1), where raters were assumed to be a random subset of all possible raters, were used to calculate ICCs among raters and within one rater for each approach (Shrout and Fleiss, 1979). The condition "absolute agreement" was chosen. This model was chosen because it is an appropriate model to assess inter-rater reliability with more than

two raters, as well as intra-rater reliability with multiple values from one rater (Portney and Watkins, 2000; Shrout and Fleiss, 1979). Additional ICC analyses were carried out to assess the between-scan reliability of the tract-based analysis methods. Internal consistency was calculated by using standardized Cronbach's alpha. A previously described interpretation of Coronbach's alpha was used (George and Mallery, 2003). An additional Bland-Altman reliability analysis was done, by calculating the Coefficient of Repeatability (CR), which represents the value below which the absolute difference between two repeated test results may be expected to lie with a probability of 95%. For comparison with literature data, the coefficient of variation (CV) was also computed according the equation, where the coefficient of variation (CV) is defined as the ratio of the standard deviation to the mean.

I-IV: Differences were considered significant at $p < 0.05$. The statistical analyses were performed with the SAS 8.1 system (**I**), SPSS 13.0 system (**II**), and SPSS 19.0 for Mac and SAS 9.3 software for Windows (**III, IV**).

4.5. ETHICAL CONSIDERATIONS

After a complete description of the study had been given to the subjects, written informed consent was obtained. The protocol was approved by the Conjoint Ethics Committee of Turku University and Turku University Hospital.

5. RESULTS

5.1. VISIBILITY OF CONTUSIONS AND OTHER INTRAPARENCHYMAL INJURIES IN CONVENTIONAL MRI IN EARLY AND LATE STAGE AFTER TBI (I)

Traumatic brain lesions were detected in 20 of the 36 patients in acute phase MRI; contusions were detected in 18 and other lesions in 12 cases. Contusions were found in the same 18 patients on control MR images at one year, but other lesions were seen only in 10 cases.

Contusions

The number of contusions detected in different anatomical locations had decreased at the re-examination by 29.3% (from 133 to 94, $p < 0.001$). Thus, almost a third of all contusions seen at one week could not be detected in the chronic phase MR images. The mean sum of the extent of contusions decreased from 14.3 ± 10.4 in the acute MRI to 8.0 ± 6.1 at one year ($p<0.001$).

Other intraparenchymal lesions

The number of anatomical regions with other traumatic lesions was reduced from 65 in the first MRI study to 45 lesions at one year. The mean semi- quantitative scores of lesions decreased from 11.8 ± 9.9 in early MRI to 8.8 ± 8.6 in late MRI ($p<0.01$).

Clinical correlations

The disappearance of lesions was not connected with the age or sex of the patient. Nor was the mechanism of injury associated with the observed MRI change. Those patients with more severe injuries (either GCS $\leq$ 12 or PTA > 24 hours) showed significantly greater change both in the number and extent of non-contusional lesions compared to patients with mild injuries. There was not a similar association with contusions. The one-year outcome measured by GOS was significantly associated with the change in the extent of non- contusional lesions; those showing a greater change had a poorer outcome. In logistic regression analysis taking into account the severity of the injury, the extent of non-contusional changes was not independently associated with GOS. The change in subjective post-concussive symptoms between one week and one year was not significantly associated with the MRI changes.

5.2. DIFFUSIVITY OF NORMAL-APPEARING BRAIN IN ACUTE TBI (II)

Group A
(i.e. TBI patients without visible lesions in conventional MRI, n=7)

Increased ADC values (> mean + 2SD) were detected in 33.0 % of measurements (106 of 322 measurements). Three or more pathological values were seen in 86 % of

subjects. Four patients (57 % of subjects) had increased values in more than 25 % of ROIs.

A decreased (< mean - 2SD) ADC value was observed only in the globus pallidus of two patients, and in the cerebellum of one patient (0.9 % of measurements).

Group B
(i.e. TBI patients with visible traumatic lesions in conventional MRI, n=15)

Nine patients had both contusions and TAI lesions, four patients had contusions without visible TAI, and two patients had only TAI lesions.

Increased (> mean + 2SD) ADC values were detected in 24.8 % of measurements (171 of 690 measurements). Three or more pathological values were seen in 80 % of subjects. Five patients (33 % of subjects) had increased values in more than 25 % of ROIs. One subject showed increased ADC values in all supratentorial white matter and deep grey matter areas, midbrain, and cerebellum.

Decreased (< mean - 2SD) ADC values were detected in 4.2 % of measurements. Decreased ADC values were found in eight patients in the various white matter regions, thalamus, and basal ganglia. In one patient, a decreased ADC was present in 10 different ROIs, but otherwise only some pathological areas were detected.

Group C
(i.e. Control subjects, n=14)

The lowest ADC values were measured in the superior longitudinal fasciculus, subcallosal fasciculus, cingulum, corona radiata, cerebellar white matter, and globus pallidus. The highest ADC values were found in the hippocampus, corpus callosum, inferior frontal white matter, and temporopolar white matter. Increased (> 2SD) ADC values were detected in 3.0 % and decreased (> 2SD) ADC values in 0.9 % of the measurements in control subjects.

Differences between the groups and correlation to the GCS and outcome

The ADC values between the two TBI groups did not differ in any region; pathological values were detected most often in the genu of the corpus callosum (12/22 ROIs), pallidum (19/44 ROIs), corona radiata (18/44 ROIs), and superior longitudinal fasciculus (17/44 ROIs). An intermediate amount of pathological findings was found in lobar white matter regions (12-17/44 ROIs). Pathological values were least common in pons (2/44 ROIs), cingulum (8/44 ROIs), cerebellum (9/44 ROIs), and midbrain (9/44 ROIs).

There were significant differences in the ADC values between the controls and both patient groups in several lobar white matter regions and fasciculi, in the corpus callosum and corona radiata, as well as in some deep grey matter structures. The ADC

values of the various regions, however, did not correlate with the GCS scores or with the GOS-E scores in any region. There was no significant correlation between the number of regions with pathological values and the GCS or GOS-E scores, either. No correlation was found even when the number of regions with increased and decreased ADC values was compared separately with the GCS and the GOS-E. Of the eight patients with the most extensive ADC changes, five had GCS 15 and three had GOS-E 8.

5.3. QUANTITAVE DIFFUSION TENSOR TRACTOGRAPHY OF LONG ASSOCIATION TRACTS IN PATIENTS WITH TBI WITHOUT FINDINGS IN ROUTINE MRI (III)

Healthy controls

The right AF was missing in 13 cases, whereas other tracts were present in all control subjects. There was large variability in the sizes of both AFs. The standard deviations of volumes in the other tracts were 22.3 - 34.6 % from the mean value. The standard deviations of FA values were 3.1 - 6.0 % from the respective mean values, and the variation in FA values was largest in both AFs and TCs. The standard deviations of ADC values were 2.0 - 4.2 % from the respective mean values. The largest variation in ADC values was detected in the left UC, both TCs, both IFOFs, and both ILFs.

There was significant negative correlation between FA values and volumes in both SLFs, AFs, IFOFs, and ILFs. No significant correlations were found between the ADC values and tract volumes. FA and ADC values had a negative correlation in all tracts except for the left SC and left ILF.

In healthy controls, 50 (80.6%) did not have any pathological FA values differing from the mean normal value by more than 2 SD, 9 (14.5%) had one, one (1.6%) had two and two (3.2%) had three tracts with abnormal FA value. No one had more than three tracts with pathological FA values.

Subjects with TBI

The AF was missing on the right side in 19 and on the left side in one of the 106 patients. Other tracts were present in all cases.

Abnormal volumes were detected in 5.3 %, abnormal FA values in 10.7 %, and abnormal ADC values in 12.1 % of the tracts (n=1458). In the tract volumes, no significant difference between patients and controls was found. In the FA values, a significant difference ($p<0.05$) between patients and controls was found in both UFs, both IFOFs, and the right ILF, and in the ADC values of both UFs, the right SC, the right SLF, the right AF, and both IFOFs.

Abnormal volumes (10% right, 15% left), FA (16% right, 25% left) and ADC (22% right, 19% left) values were most common in the UFs, and abnormal FA and ADC values were relatively common (>10%) also in the IFOFs, ILFs, and SLFs. Either FA or volume (or both) abnormality was most common in the left UF (32 %), relatively common (>10 %) in the right UF, both TCs, SLFs, and IFOFs, and in the right ILF. They were relatively rare (<10 %) in the SCs, AFs, and left ILF. Time from injury, trauma mechanism or severity of the TBI did not have a statistically significant influence on the results. When calculated with different normal values for men and women, 3.9 % more abnormal values were detected: 0.6 % fewer abnormal FAs, 4.5 % more abnormal ADCs, and 12 % more abnormal volumes.

In TBI patients, 12% had four or more pathological FA values that differed from the mean normal value by more than 2 SD, 51% did not have any, 17% had one, 10% had two, and 9% had three pathological FAs. Volume, FA and ADC values were not significantly correlated to the outcome.

Correlations for normal control subjects between volume, FA and ADC values in the fourteen association tracts were compared with the respective correlations for TBI patients. The only significant difference between the correlations was that the negative correlation between FA and ADC values in the left ILF was stronger in the patient group.

Intra-rater reliability for the measurements was excellent (ICC $\geq$ 0.897), except for the volume measurements in the left SLF (ICC= 0.537), and for the ADC measurements in the right UF (ICC= 0.644). Inter-rater reliability for the measurements was excellent (ICC$\geq$ 0.766), except for the volume measurements in the left SLF (ICC= 0.722).

5.4. REPRODUCIBILITY OF TRACT-BASED AND REGION OF INTEREST DTI ANALYSIS OF LONG ASSOCIATION TRACTS (IV)

Standard deviations of the measurements

Standard deviations of the tract volumes varied greatly depending on the tract; the largest variations were found in both AFs and in the right SLF (SD in the range of 1000-1400 cm3). Volume variation was smallest for both UFs and SCs (SD 300-400 cm3). No systemic differences in the SDs between the tracts either for the FA (0.015-0.030 whole tract; 0.020-0.045 central part) or MD (0.015-0.035 whole tract; 0.015-0.030 central part) were detected. The tract volume measurements from the middle of the tract had some variation (SD in the range of 24-245cm3, 4.8-25%) from the desired volume depending on the tract.

Inter-rater reliability (Group A)

Results for each CR analysis between each rater are presented in **Table 4**. There were no systematic differences in the results, depending on the experience of the raters. Results for each ICC, CR, and CV analysis are summarized in **Table 5**.

ICC values for the whole tract volume measurements showed good or excellent reproducibility (ICC range: 0.639-0.971), but all the CR values (average between raters) were over 10% from the respective mean tract volumes. For both the tract-based methods, reproducibility of the FA and MD measurements was found to be excellent (ICC range: 0.761-0.997). CR values for the mean FA and MD measurements were less than 5% from the respective mean tract FA and MD, except for the FA of the left AF (7.1%). For the measurements in the middle of the tract, CR values were less than 6% (FA) and 2% or less (MD) from the respective mean FA and MD. CV estimates of the tract-based inter-rater variability in MD were in the range of 0.1%–1.0%, and for FA 0.6%–1.8%.

ICC values for the cross-sectional method showed fair to good reliability for FA measurements (ICC range: 0.502-0.690) in the left UF, right SC, right SLF and right IFOF, and for MD measurements (ICC range: 0.447-0.679) in both SCs, left SLF, right AF, and both IFOFs; otherwise, the FA concordance and the MD concordance were found to be poor (ICC $\leq$ 0.388). CV estimates of variability in MD were in the range of 2.2%–5.5%, and for FA 5.1%–11.0%. CR values for the cross-sectional ROI-based FA and MD measurements were relatively high; only the mean CR values for the MD in both the SCs and SLFs were less than 10% from the respective mean MD.

Intra-rater reliability (Group A)

Results from each ICC, CR, and CV analysis are summarized in **Table 5.**

For both the tract-based methods, the reliability of FA and MD measurements was found to be excellent (ICC range: 0.907-0.999), except for MD measurements of the right UF with good intra-rater reliability (ICC= 0.738). CR values for all the tract-based FA and MD measurements were less than 5% from the respective mean FA and MD values. CV estimates of the tract-based intra-rater variability in MD were in the range of 0.0%–0.8%, and for FA 0.1%–1.1%. ICC values for the whole tract volume measurements showed excellent reliability (ICC range: 0.831-0.998, $p < 0.0001$), but the CR values were over 10% from the respective mean tract volumes in all the tracts except for both SCs.

For the cross-sectional method, intra-rater reliability was found to vary from fair to excellent for FA measurements (ICC range: 0.418-0.813) in all tracts except for the left AF (ICC=0.271) and for MD measurements (ICC range: 0.515-0.810) in all tracts except for the left AF (ICC=0.314) and the left UF (ICC=0.228). CV estimates of variability in MD were in the range of 1.2%–3.9% and for FA 4.1%–7.9%. CR values for the cross-sectional ROI measurements in general were relatively high; the mean CR values for

all FA measurements were more than 10% from the respective mean FA. The mean CR values for the MD measurements in both the SCs, SLFs, AFs and the right IFOF were, however, less than 10% from the respective mean MD.

Between-scan reliability (Group B)

According to the ICC analysis, the between-scan reliability of the FA and MD measurements was found to be mainly excellent for both the tract-based methods. Reproducibility varied between fair to good and excellent, except for poor reproducibility in MD measurements in the left IFOF with 0.15 FA threshold. Measurements for the whole tract volume with turning angle threshold 27° showed excellent reproducibility with both 0.15 and 0.30 FA threshold (ICC range: 0.787-0.918 and 0.766-0.932, respectively). With the the higher turning angle threshold (60°), reproducibility was good or excellent (ICC range: 0.745-0.906), except for the volume measurements in the left AF (ICC 0.459) and in the right SLF (ICC 0.661). However, neither the two used FAs (0.15/ 0.30) or the turning angle (27°/ 60°) thresholds had a systematic influence on the reproducibility of the FA, MD, and whole tract volume measurements.

The reproducibility of the FA and MD measurements in the central part of the tract varied from good to excellent (ICC range: 0.746-0.989 and 0.634-0.964, respectively) in all the tracts with both the pretermined sizes (3cm3 and 1cm3 for both the UFs, SCs and AFs; 6cm3 and 2cm3 for both the SLFs and IFOFs). However, a tendency towards better reproducibility of the central tract measurements with the smaller predetermined sizes could be detected.

CV estimates of the tract-based between-scan variability in MD were in the range of 0.6%–2.5%, and for FA in the range of 0.7%–3.5%.

No systematic bias (i.e., one set of repeated measures is not consistently higher or lower than the other set) was found between the scans.

Internal consistency (Group A)

Internal consistency of the measurements yielded results similar to these of the ICC analysis.

Table 4. Standard deviations (SD) of the measurements, and inter-rater and intra-rater coefficients of repeatability (CR) of each analysis method for the right (dx) and the left (sin) uncinate fasciculus (UF), superior cingulum (SC), superior longitudinal fasciculus (SLF), arcuate fasciculus (AF), and inferior fronto-occipital fasciculus (IFOF). Inter-rater CR values between each pair of raters (R1-R2; R1-R3; R2-R3) are presented.

	Analysis	**Inter-rater CR**				**Intra-rater CR**		**Inter-rater CR**				**Intra-rater CR**
		R1-R2	R1-R3	R2-R3	SD	R2-R2		R1-R2	R1-R3	R2-R3	SD	R2-R2
UF dx	***Tract based***						**sin**					
	Mean V tot	138	190	212	371	176		254	335	393	333	264
	Mean FA tot	8	9	7	20	11		15	14	16	30	9
	Mean MD tot	10	19	19	24	40		38	28	28	32	25
	Mean FA std	8	12	14	35	13		33	21	27	42	4
	Mean MD std	8	6	5	21	19		6	7	12	19	2
	ROI based											
	Mean FA	138	145	183	53	96		205	219	161	81	184
	Mean MD	114	141	165	37	103		97	91	91	33	101
SC dx	***Tract based***						**sin**					
	Mean V tot	142	450	415	366	48		177	470	417	385	139
	Mean FA tot	5	10	9	16	6		15	17	14	18	10
	Mean MD tot	4	7	7	21	5		8	9	7	16	4
	Mean FA std	10	15	15	37	7		6	44	45	42	7
	Mean MD std	4	6	7	19	2		3	3	3	17	4
	ROI based											
	Mean FA	79	123	128	61	90		95	161	168	48	86
	Mean MD	44	75	54	32	37		70	62	59	35	65
SLF dx	***Tract based***						**sin**					
	Mean V tot	1220	1030	572	1105	586		1620	1560	719	682	650
	Mean FA tot	12	12	7	14	6		19	20	11	18	9
	Mean MD tot	6	10	9	19	3		6	10	11	23	7
	Mean FA std	19	21	10	34	13		36	34	15	27	9
	Mean MD std	4	4	3	18	7		14	17	8	19	10
	ROI based											
	Mean FA	99	97	87	52	94		190	138	119	39	106
	Mean MD	86	84	65	21	46		65	74	63	30	49
AF dx	***Tract based***						**sin**					
	Mean V tot	627	800	986	1126	676		1950	1540	1510	1246	1100
	Mean FA tot	11	19	17	20	16		21	32	37	17	19
	Mean MD tot	7	7	14	18	10		16	18	22	21	15
	Mean FA std	12	19	24	38	14		6	34	33	36	6
	Mean MD std	12	6	15	16	10		6	18	23	24	8
	ROI based											
	Mean FA	211	132	237	49	125		123	140	189	40	140
	Mean MD	107	110	71	34	57		72	93	85	17	73
IFOF dx	***Tract based***						**sin**					
	Mean V tot	750	1000	1030	632	271		691	1150	885	607	276
	Mean FA tot	12	14	12	21	5		16	19	12	21	9
	Mean MD tot	32	29	8	23	8		40	45	15	35	30
	Mean FA std	31	22	37	37	10		14	32	30	22	11
	Mean MD std	8	15	14	23	4		8	14	12	27	6
	ROI based											
	Mean FA	186	157	144	73	114		169	194	149	49	149
	Mean MD	98	157	114	49	76		101	87	65	46	106

Table 5. Inter-rater and *intra-rater* reliability of each analysis method for uncinate fasciculus, superior cingulum, superior longitudinal fasciculus, arcuate fasciculus, and inferior fronto-occipital fasciculus. Results with acceptable reproducibility (CR/Average less than 10%; ICC >0.6) are bolded.

Fasciculus	Analysis	CR/Average	ICC (±95% CI)	CR/Average	ICC (±95% CI)
UF dx	***Tract based***				**sin**
	Mean V tot	15.1	**0.971** (0,940-0.986)	25.8	**0.681** (0.428-0.835)
		14.6	***0.972*** *(0,920-0.990)*	*20.8*	***0.914*** *(0.769-0.970)*
	Mean FA tot	**2.2**	**0.980** (0.959-0.991)	**3.8**	**0.967** (0.932-0.984)
		2.9	***0.968*** *(0.910-0.989)*	***2.2***	***0.989*** *(0.967-0.996)*
	Mean MD tot	**1.9**	**0.943** (0.884-0.973)	**3.8**	**0.882** (0.766-0.942)
		4.8	***0.738*** *(0.391-0.902)*	***3.0***	***0.922*** *(0.790-0.973)*
	Mean FA std	**2.5**	**0.987** (0.972-0.994)	**5.9**	**0.948** (0.894-0.975)
		3.0	***0.981*** *(0.947-0.994)*	***0.8***	***0.999*** *(0.996-1.000)*
	Mean MD std	**0.8**	**0.988** (0.975-0.994)	**1.0**	**0.973** (0.943-0.987)
		2.3	***0.922*** *(0.789-0.972)*	***0.2***	***0.999*** *(0.996-1.000)*
	ROI based				
	Mean FA	28.0	0.388 (0.035-0.654)	36.0	0.502 (0.176-0.728)
		16.7	***0.813*** *(0.539-0.932)*	*32.3*	***0.612*** *(0.177-0.847)*
	Mean MD	17.9	0.179 (-0.190-0.504)	12.4	0.388 (0.035-0.654)
		13.2	*0.515 (0.036-0.802)*	*13.5*	*0.228 (-0.293-0.645)*
SC dx	***Tract based***				**sin**
	Mean V tot	24.8	**0.892** (0.784-0.947)	23.8	**0.893** (0.787-0.948)
		3.4	***0.998*** *(0.995-0.999)*	***9.0***	***0.985*** *(0.958-0.995)*
	Mean FA tot	**2.0**	**0.967** (0.931-0.984)	**3.6**	**0.915** (0.829-0.959)
		1.4	***0.986*** *(0.960-0.995)*	***2.3***	***0.969*** *(0.911-0.989)*
	Mean MD tot	**0.8**	**0.989** (0.976-0.995)	**1.0**	**0.971** (0.940-0.986)
		0.6	***0.994*** *(0.982-0.998)*	***0.5***	***0.994*** *(0.984-0.998)*
	Mean FA std	**2.7**	**0.984** (0.966-0.992)	**5.8**	**0.911** (0.820-0.957)
		1.3	***0.996*** *(0.989-0.999)*	***1.2***	***0.997*** *(0.992-0.999)*
	Mean MD std	**0.7**	**0.988** (0.976-0.994)	**0.4**	**0.997** (0.993-0.998)
		0.3	***0.998*** *(0.994-0.999)*	***0.6***	***0.991*** *(0.975-0.997)*
	ROI based				
	Mean FA	20.3	**0.677** (0.422-0.833)	24.8	0.361 (0.004-0.636)
		16.7	***0.765*** *(0.441-0.912)*	*14.8*	***0.766*** *(0.444-0.912)*
	Mean MD	**7.4**	**0.673** (0.415-0.830)	**8.1**	**0.679** (0.425-0.834)
		4.8	***0.810*** *(0.533-0.930)*	***8.3***	***0.621*** *(0.190-0.851)*
SLF dx	***Tract based***				**sin**
	Mean V tot	27.4	**0.911** (0.820-0.957)	51.5	**0.639** (0.364-0.811)
		17.3	***0.961*** *(0.891-0.986)*	*27.6*	***0.831*** *(0.578-0.939)*
	Mean FA tot	**2.6**	**0.933** (0.863-0.968)	**4.0**	**0.899** (0.797-0.951)
		1.4	***0.982*** *(0.948-0.994)*	***2.2***	***0.969*** *(0.912-0.989)*
	Mean MD tot	**1.0**	**0.974** (0.947-0.988)	**1.2**	**0.980** (0.959-0.991)
		0.4	***0.996*** *(0.988-0.999)*	***0.8***	***0.990*** *(0.971-0.997)*
	Mean FA std	**3.4**	**0.969** (0.936-0.985)	**5.8**	**0.864** (0.734-0.933)
		2.6	***0.983*** *(0.952-0.994)*	***1.9***	***0.987*** *(0.962-0.995)*
	Mean MD std	**0.5**	**0.995** (0.989-0.997)	**1.7**	**0.941** (0.880-0.972)
		0.9	***0.983*** *(0.951-0.994)*	***1.3***	***0.972*** *(0.920-0.990)*
	ROI based				
	Mean FA	16.3	**0.690** (0.442-0.840)	26.7	0.255 (-0.113-0.561)
		16.4	***0.641*** *(0.222-0.860)*	*19.7*	***0.624*** *(0.196-0.853)*
	Mean MD	10.0	0.188 (-0.182-0.511)	**8.8**	0.565 (0.261-0.768)
		5.8	***0.742*** *(0.397-0.903)*	***6.3***	***0.762*** *(0.436-0.911)*

Fasciculus	Analysis	CR/Average	ICC (±95% CI)	CR/Average	ICC (±95% CI)
AF dx	***Tract based***				**sin**
	Mean V tot	63.8	**0.889** (0.750-0.953)	63.2	**0.821** (0.656-0.911)
		50.6	***0.929*** *(0.768-0.980)*	*37.6*	***0.931*** *(0.811-0.976)*
	Mean FA tot	**3.8**	**0.886** (0.739-0.953)	**7.1**	**0.761** (0.554-0.879)
		3.9	***0.928*** *(0.765-0.979)*	***4.6***	***0.958*** *(0.883-0.985)*
	Mean MD tot	**1.2**	**0.894** (0.755-0.956)	**2.3**	**0.917** (0.833-0.960)
		1.3	***0.964*** *(0.879-0.990)*	***1.8***	***0.957*** *(0.880-0.985)*
	Mean FA std	**3.8**	**0.968** (0.923-0.987)	**4.5**	**0.932** (0.861-0.967)
		2.9	***0.982*** *(0.936-0.995)*	***1.1***	***0.997*** *(0.991-0.999)*
	Mean MD std	**1.4**	**0.929** (0.832-0.971)	**2.0**	**0.939** (0.875-0.971)
		1.2	***0.937*** *(0.791-0.982)*	***1.0***	***0.988*** *(0.965-0.996)*
	ROI based				
	Mean FA	35.0	0.182 (-0.265-0.565)	24.1	0.174 (-0.195-0.500)
		23.5	*0.418 (-0.189-0.794)*	*22.8*	*0.271 (-0.250-0.671)*
	Mean MD	12.4	0.447 (0.025-0.734)	10.7	0.021 (-0.339-0.375)
		7.3	***0.698*** *(0.223-0.905)*	***9.4***	*0.314 (-0.206-0.695)*
IFOF dx	***Tract based***				**sin**
	Mean V tot	42.0	**0.775** (0.555-0.887)	39.1	**0.762** (0.555-0.880)
		11.8	***0.978*** *(0.936-0.992)*	*11.5*	***0.977*** *(0.936-0.992)*
	Mean FA tot	**3.2**	**0.939** (0.876-0.971)	**3.3**	**0.933** (0.864-0.968)
		1.0	***0.992*** *(0.976-0.997)*	***1.9***	***0.973*** *(0.923-0.991)*
	Mean MD tot	**2.8**	**0.867** (0.738-0.935)	**4.0**	**0.879** (0.761-0.941)
		1.0	***0.987*** *(0.961-0.995)*	***3.5***	***0.907*** *(0.753-0.967)*
	Mean FA std	**5.7**	**0.924** (0.847-0.963)	**4.7**	**0.842** (0.693-0.922)
		2.0	***0.988*** *(0.965-0.996)*	***2.0***	***0.974*** *(0.926-0.991)*
	Mean MD std	**1.5**	**0.963** (0.924-0.983)	**1.3**	**0.978** (0.954-0.990)
		0.5	***0.996*** *(0.989-0.999)*	***0.8***	***0.993*** *(0.979-0.998)*
	ROI based				
	Mean FA	27.1	0.575 (0.274-0.773)	26.2	0.262 (-0.105-0.566)
		18.0	***0.735*** *(0.385-0.900)*	*22.3*	*0.473 (-0.020-0.781)*
	Mean MD	15.5	0.471 (0.136-0.709)	11.1	**0.670** (0.412-0.829)
		9.6	***0.721*** *(0.359-0.894)*	*13.9*	*0.578 (0.125-0.831)*

ICC= intraclass correlation; CR= coefficient of repeatability; UF= uncinate fasciculus; SC= superior cingulum; SLF= superior longitudinal fasciculus; AF= arcuate fasciculus; IFOF= inferior fronto-occipital fasciculus; dx= right; sin= left; std= standard volume in the middle of the tract

6. DISCUSSION

6.1. METHODOLOGICAL CONSIDERATIONS

6.1.1. Subjects

Brain lesions detected by MRI are present in most patients hospitalized after closed head injury (Fiser et al., 1998; Levin et al., 1992; Van der Naalt et al., 1999). Although the criteria for hospitalization may vary according to cultural and other factors, the presence of traumatic MRI changes has also been shown in patients who have not been considered to need hospitalization (Hofman et al., 2002; Hughes et al., 2004). Our study material (**I, II**) included also mild TBI cases without need of hospitalization, and only few severe cases. It should be recognized, in any case, that our study material is selected since it might be that only about 25% of the individuals with TBI are treated in hospitals; a vast number of subjects with TBI never seek medical care (Sosin et al., 1996). In general, selection bias is common in traumatic brain injury research, and limits the clinical usefulness and generalizability of study findings (Luoto et al., 2013). The representativeness of the TBI group in our third sub-study (**III)** may be considered low, since only the subpopulation of TBI sequels with no traumatic findings on routine MRI, was included. Thus, our results can obviously not be generalized widely to subjects with TBI. On the other hand, fully unselected TBI materials may not be substantial in clinical practice since the majority of patients with mild injury recover without long-term cognitive impairment (Belanger et al., 2005). In this respect, our special subpopulation of TBI subjects may actually be considered as a strength.

Even if a relative small study population (n= 15 and n= 10) in our reproducibility study (**IV**) can be considered as a limitation of our study, it is comparable to previous reliability studies: Borich et al., 2012 (n=20), Bisdas et al., 2008 (n=12), Malykhin et al., 2008 (n=8), Vollmar et al., 2010 (n=9), and Wakana et al., 2007 (n=10). Still, the CR values for the ROI measurements in our study could have been smaller, if our study population had been bigger (Rankin and Stokes, 1998). In our study (**III**), we had a relatively large group of 106 TBI patients. However, since we had mainly patients with good or moderate outcome (GOS-E 5-7) with only a small variation in FA values, the size of the study sample proved to be small for showing any connection between FA values and outcome. Another limitation in this study (**III**) was that the detailed clinical characterization of the examined patients was studied retrospectively. The clinical characterization was based on the medical records in several medical centres, and the reliability of the retrospective evaluation may have been affected by inconsistent information in the medical records. It should also be noted, that we did not have pre-injury DTI of the TBI subjects, and the time between injury and MRI scan varied from three months to even 40 years, which may limit the intepretation of the results.

In the other two studies (**I, II**), the study sample is moderate (n= 36 and n= 22, respectively) for a clinical imaging study. We could not show any association between the ADC values and severity of injury, whether measured as acute GCS or one-year outcome (**II**). However, since we had only few severe cases and relatively small ADC changes, it is possible that a larger study sample could change our results.

6.1.2. Limitations of the technique

Although modern imaging techniques have been shown to be beneficial in the demonstration of traumatic changes, clinical MRI studies are commonly performed with old or poorly equipped MRI systems and/or with ordinary visual evaluation. In our first sub-study (**I**), the visibility of changes was, therefore, analysed only in conventional sequences. A FLAIR sequence was used in the majority of cases, because previous studies have shown it to be equal or superior to more conventional SE images in the detection of traumatic lesions (Hofman et al., 2001; Pierallini et al., 2000). However, excluding the hemorrhage sensitive T2* and SWI sequences from the analysis may be considered as a limitation of the study, since these sequences are currently often easily achievable. Moreover, in our sub-studies **I,** and **II**, the slice thickness 5.0-7.0 mm in part of the sequences was relatively high, resulting in sub-optimal resolution in these sequences.

A weakness of our second sub-study (**II**) is the lack of standardization and normalization of the subject brains. However, the ROI placements were based on the specific white matter tract atlas and performed under the guidance of an experienced neuroradiologist. To improve differentiation of white matter regions we used orthogonal diffusion images for the localization of several anisotropic white matter tracts, which are not visualized on echo planar or trace diffusion images. In white matter tracts, the diffusion is high along the main axis of the fibres and restricted perpendicular to the axons, which leads to relative hypointensity of the tracts in the direction of the diffusion gradient, and relative hyperintensity of the tracts perpendicular to the direction of the diffusion gradient. We could separate some fasciculi, corona radiata and the internal capsule more realiably on the basis of the orthogonal images than on other images.

Limitations of the DTI tractography technique (**III, IV**) include concurrent FA and volume decline, and the effect of crossing tracts on the results. The large variability of the tract volumes (in both controls and subjects with TBI) and the dependence of mean FA values on volume can decrease the accuracy of the technique. It is important to notice that a study with different field strength, non-parallel imaging or different image acquisition parameters could have yielded different results. Additional sources of error contribute to data variability: motion during the scans (despite the motion correction) (Ling et al., 2011), signal-to-noise level, gradient stability, and differences in subject positioning between subjects and between examinations (Wang et al., 2012). The program chosen for the semi-automated tract-based quantification might also have an influence on the results. Moreover, it should be noticed that, although DTI provides directional

information at the voxel level, it provides no accurate information about the connection between neighbouring voxels. In our studies, cardiac gating was not applied. It is well known that cardiac pulsation can have a profound effect on DTI results; this could be a major contribution to the reproducibility noted in our studies. However, cardiac gating has not been widely accepted in clinical studies because it lengthens the scanning time, and subject motion conditions and arrhythmia can occasionally cause gating instability (Landman et al., 2007).

Each application of DTI has an optimal or near- optimal experimental design. For TBI patients, for instance, it may be difficult to remain recumbent during the long imaging acquisition time, and it can be desirable to limit the number of diffusion-weighted images to minimize the acquisition time and head movements. Tractography results of large bundles using 12 diffusion encoding directions has been shown to have similar reproducibility compared to those using 60 directions, but 12 direction may be less sensitive to smaller pathways (Heiervang et al., 2006). We considered 15 diffusion encoding directions acceptable in our study, since we included relatively large fibre tracts and wanted to use DTI protocols suitable for clinical use with a reasonable acquision time.

6.2. VISIBILITY OF INTRAPARENCHYMAL INJURIES IN CONVENTIONAL MRI IN EARLY AND LATE STAGE AFTER TBI (I)

Contusions

The size of contusions was markedly reduced in our study at one year, with a total volume reduction of 44%. Earlier studies have shown a volume reductions of 85.2% during the first month (Levin et al., 1987), 14.4% between 2–5 days and 6 months (Hofman et al., 2001), and 65.1% between 2–18 days and 4–12 months (Stamatakis et al., 2002). However, these studies have either not separated contusions from other lesions (Levin et al., 1987; Hofman et al., 2001) or used different field strength during the follow-up (Stamatakis et al., 2002). Nearly a third of all contusions seen at one week could not be recognized in the chronic phase in our study, and in some cases, the residual deficit would probably have escaped detection without the initial scan.

Other intraparenchymal lesions

Diffuse axonal injury is thought to be present in the majority of all severe head injuries. Moreover, it is suggested to be responsible for the long-lasting or persistent problems after mild injuries (Voller et al., 2001). Until recently, no technique has been accurate in diagnosing and assessing the distribution and severity of traumatic axonal injuries. Although modern imaging techniques have been shown to be beneficial in the demonstration of traumatic changes, especially traumatic axonal injury, clinical MRI studies are still often performed with ordinary visual evaluation. In our study patients, lesions suggestive of DAI could be seen in only 12 cases, and both the number and extent

of lesions were markedly reduced during the one-year follow-up. In two patients, the DAI lesions were no longer visible in the later study. To the authors' knowledge, previous systematic studies clarifying the visibility of DAI lesions with time are practically non-existent. The early study of Wilson et al. (1988) demonstrated that 50% of sub-cortical white matter lesions had disappeared during a follow-up of 11 months. Stamatakis et al. (2002) reported a volume reduction of 86.6% in diffuse lesions between 2–18 days and 4–12 months after the injury, but their initial imaging was obtained with a 0.15 T and the follow-up with a 1.5 T field scan. In our material, 30.7% of visible non-contusional intra-parenchymal lesions disappeared between one week and one year after the injury, with a semi-quantitative volume reduction of 25.4%. Although many studies have shown that T2*-weighted, susceptibility-weighted, diffusion-weighted, or diffusion-tensor imaging techniques improve the detection of both acute and chronic traumatic axonal injuries compared with conventional sequences, they too may show a reduction in trauma-induced changes with time (Arfanakis et al., 2002; Danielsen et al., 2003; Le et al., 2005; Schaefer et al., 2004; Takayama et al., 2000; Wardlaw and Statham, 2000). Indeed, only those imaging methods which give quantitative measures of brain morphology (Bigler et al., 2002; Blatter et al., 1997; Gale et al., 2005; MacKenzie et al., 2002; Thatcher et al., 1997; Tomaiuolo et al., 2004) or neuronal integrity (Garnett et al., 2000) have shown an increasing amount of pathology with time after TBI.

Possible mechanisms behind the reduction of lesions

The mechanisms behind lesion reduction or total disappearance are not fully understood, but probably some of these lesions are related to edema in the acute phase after trauma (Barzo et al., 1997; Takayama et al., 2000). Also the disappearance or resolution of DAI lesions may be related to the early phase vasogenic or intracellular oedema. It could be speculated that a marked change in the visibility of lesions could be associated with better outcome. In this study, the patients with more severe traumas showed significantly more changes in the visibility of MRI lesions. It seems unlikely that this is a causative association, but probably merely reflects a mathematical effect, i.e. those having more and greater lesions also have a greater probability of visible changes. Similarly, the significant negative association between the GOS score and the extent of the DAI change is, according to the logistic regression analysis, only a reflection of more severe injury.

Clinical correlations

This study is too small to determine whether a greater change in the visibility of MRI lesions is associated with better recovery, but this rather limited material did not suggest such a connection. This is in accordance with earlier studies which have questioned the clinical value of early conventional MRI in outcome prediction (Fiser et al. 1998; Hughes et al., 2004; Paterakis et al., 2000). As late imaging may fail to demonstrate the trauma- induced changes, as shown in our study, an early MRI is essential in this respect, especially if only conventional MRI is available. Moreover, as the long-term outcome seems to be intimately connected with the development of atrophic changes (Bigler,

1995; Himanen, 2005), their evaluation and connection to the actual trauma may be impossible without the possibility of comparison with early phase imaging.

6.3. DIFFUSIVITY OF NORMAL APPEARING BRAIN IN ACUTE TBI (II)

Earlier reports have demonstrated ADC changes of normal-appearing brain tissue in some regions (Goetz et al., 2004; Huisman et al., 2004; Inglese et al., 2005) or in whole-brain histograms (Shanmuganathan et al., 2004) after TBI, but in this study the distribution of pathological diffusivity in various brain regions was clarified more extensively than previously.

In the control brains, we also measured the diffusivity of several fascicles that had not been included in previous reports (Ahlhelm et al., 2004; Engelter et al., 2000; Helenius et al., 2002). The cingulum, superior longitudinal fasciculus, and subcallosal fasciculus had relatively low diffusivity, which can be related to the compact structure and heavy myelination in these structures. ADCs in the other measured fasciculi were slightly higher and in the same range as values in the lobar white matter. The values in the lobar white matter and subcortical grey matter were mostly intermediate, which is consistent with previous results (Ahlhelm et al., 2004; Engelter et al., 2000; Helenius et al., 2002). The diffusivity in the corpus callosum was higher than in other white matter regions, which is also in accordance with previous studies (Arfanakis et al., 2002; Tench et al., 2002).

Both increased and decreased diffusivity has been shown in normal-appearing brain after TBI (Goetz et al., 2004; Huisman et al., 2004; Inglese et al., 2005; Shanmuganathan et al., 2004). After the first few days, mainly increased ADCs have been reported, and at one week these have been dominant (Assaf et al., 1997). The precise mechanisms underlying the changes in ADCs associated with acute head trauma are still not fully understood. Axonal damage can range from stretching, with associated poration that if not severe may seal over, to axotomy (Farkas and Povlischock, 2007). The possible mechanisms of ADC increase in normal-appearing tissue are trauma-induced vasogenic edema, changes of cytoarchitecture with neurofilament alignment, and early neurodegeneration (Ebisu et al., 1993; Goetz et al., 2004; Hanstock et al., 1994; Hergan et al., 2002; Schaefer et al., 1997; Schaefer et al., 2000). Several of our patients had cortical contusions, which can cause edema in the surrounding normal-appearing tissue. However, in our material, increased ADC values were almost as common in patients without visible lesions as in patients with visible TBI. Therefore, it seems that cortical contusions are not an important cause of the ADC increases in normal-appearing brain tissue.

We could demonstrate that ADC increase is a common phenomenon after a TBI and can occur in most white matter and deep grey matter regions. Increased ADC values were common in our material, both in patients with and without visible traumatic lesions. On the contrary, reduced diffusivity was relatively uncommon in our material.

It can be supposed that at one week after injury most early phase ADC decreases have already disappeared and possibly changed to ADC increases. Diffusivity changes were most often detected in brain areas generally associated with the common occurrence of traumatic axonal injury, such as the corpus callosum and corona radiata; they were also common in the superior longitudinal fasciculus and subcallosal fasciculus and relatively common in lobar white matter. However, the changes were generally also detected in other supratentorial grey and white matter structures, indicating a widespread diffusion change.

Our material included a number of mild cases without a need for hospitalisation. Approximately one third of the patients were without visible traumatic lesions, all of them having mild TBI according to the GCS (= 13-15). The lack of an association between the severity of injury and ADC values in our study may be due to the relatively small number of patients and the few severe cases. More sensitive clinical techniques (neuropsychological assessments including follow-up) could change our results. However, increased diffusivity seems to be a sensitive marker of intracranial injury and present even in the mildest cases without visible lesions.

6.4. QUANTITAVE DIFFUSION TENSOR TRACTOGRAPHY OF LONG ASSOCIATION TRACTS IN PATIENTS WITH TBI WITHOUT FINDINGS IN ROUTINE MRI (III)

We were able to demonstrate that variability in mean FA and ADC values in normal controls is generally small, which renders 3T DTI tractography a useful method in clinical diagnostics. With the tractography method, a wide distribution of FA and ADC changes in various association tracts could be detected in TBI subjects who showed no signs of contusions, microbleeds, local atrophies, or remnants of superficial hemorrhages on routine MRI. The left UF was the most common site of abnormality, but abnormalities were also common in several other tracts.

In control brains, the mean FA values of the SLF, AF, IFOF, and ILF were dependent on the tract size, which can be related to the variability in the relative amount of peripheral white matter in the tractograms, but can also reflect the structural variability of the whole tract. Postmortem studies, concluded by Hasan et al. (2010), have shown that white matter fibre tracts can be heterogeneous. In our study, the intra- and inter-rater reliability was high. Thus, the large variability in volumes more likely results from the structural variability of the tracts than from inconsistent ROI placement. However, the large standard deviations in the size and FA values in the AF, and the large standard deviations in the FA and ADC values in the TC make these tracts less suitable for quantitative diagnostics. Different normal values for genders may not be needed for the studied association tracts since only marginally fewer FA changes and slightly more ADC changes were detected with different normal values for men and women. However, using gender-dependent normal values, many more abnormal volumes in the TBI group

could be detected, suggesting that closer matching of normal controls may increase the sensitivity of the method.

A previous tractography study has reported decreased volumes in the corpus callosum, fornix and peduncle projections in subjects with TBI (Wang et al., 2008). We did not find any significant trauma-induced volume decline in the group comparison, which is probably related to the large normal variation in size of the association tracts and to the small number of severe cases in our study.

As described above, diffusion changes in traumatic axonal injury have mainly been characterized by reduced FA and/ or increased mean diffusivity, and FA values are generally considered more sensitive to show abnormalities than ADC values. In our study, ADC values showed more abnormalities than FA values, probably due to the effect of crossing tracts on ADC values (Mukherjee et al., 2008a; Mukherjee et al., 2008b) and, also, in the tracts traversing close to the lateral ventricles, contamination from the cerebrospinal fluid (Concha et al., 2005).

The high frequency of axonal injury in the UF has also been detected in previous studies (Bendlin et al., 2008; Niogi et al., 2008). In our material with only one patient with poor outcome (GOS-E ≤ 4), injury to the cingulum seems to be relatively rare compared with other reports (Bendlin et al., 2008; Kraus et al., 2007; Niogi et al., 2008; Rutgers et al., 2008).

DTI is more sensitive than conventional MRI in detecting subtle, but clinically meaningful, changes following TBI. As was expected, we could not show a connection between FA values and outcome in our study patients with mainly a relatively similar outcome. However, several recent studies (see e.g. Caeyenberghs et al., 2010; Messé et al., 2011; Warner et al., 2010) have shown DTI findings to have good correlation with clinical status and outcome. Recovery, in general, is much more accelerated during the first five months than the last seven months of the first year post-injury, suggesting the need for early restorative treatments. Only some of the brain areas, including manual motor and visuospatial domains, are likely to recover more slowly, and they might have a wider therapeutic window (Christensen et al. 2008). This makes the role of accurate early diagnosis highly important. Modern imaging techniques are in a special position to validate functional deficits related to TBI since lack of motivation and somatoform disorders can influence the accuracy of neuropsychological tests and self-reported symptoms. Anosognosia (i.e. lack of awareness of deficits) is common in more severe injuries (Hart et al., 2004) and influences an individual's subjective evaluations of his or her cognitive and behavioural changes, which complicates the identification of TBI symptoms and their consequences in these patients. Thus, at least in cases with trauma-related symptoms without declarative findings on routine imaging techniques (i.e. CT or conventional MRI), DTI may be crucial in refining TBI diagnosis, prognosis, and management.

As many TBIs occur in traffic accidents, work-related accidents, sports-related injuries or assaults, their medicolegal consequences may also be significant. Accurate diagnosis of brain injury is also critical to discriminate true injury from other disorders, as well as malingering symptoms.

6.5. REPRODUCIBILITY OF TRACT-BASED AND REGION OF INTEREST DTI ANALYSIS OF LONG ASSOCIATION TRACTS (IV)

To provide an acceptable measure of change in diffusion measurements with advancing age, disease progression, or intervention, it is essential to establish measurement reliability. The reliability of a method consists of several aspects including the validity, sensitivity, and reproducibility of the method. As described above, many factors that influence the reliability of DTI measures have been studied in the literature. The role of rater performance in reliability is important, since many of the technical factors are more easily standardized. Our study provides an explicit evaluation of the reproducibility of tractography measurements of ten association tracts in the brain essential for cognitive brain functions, using a clinical DTI protocol at 3.0 T. The examined fibre variables were FA and MD, together with the volumetric measurements, including central part analysis in corresponding volumes representing the region with highest FA values in each subject. Different types of intra-rater, inter-rater, and between-scan reliabilities were compared to find important sources of variability, and to evaluate whether increasing the FA threshold from 0.15 to 0.30 and decreasing the turning angle threshold from 60° to 27°, as well as decreasing the predetermined central part volume, improved the reproducibility.

In our study, the only operator-dependent step in the analyses was the subjective placement of the ROIs in the cross-sectional images for the tract delineation and for the ROI-based measurements. We used colour-coded diffusion maps that show fibre orientation to improve the definition of the ROIs from the two-dimensional images (Hermoye et al., 2006), and placed the ROIs to the shape of the structure to diminish the partial volume effects (Snook et al., 2007). Standard ROI sizes and positions were used. The most common methods to perform quantitative fibre tracking analysis, such as the method used in our study, include the use of multiple inclusion and exclusion ROIs. As for accuracy of tracking results, we reconstructed tracts that are well documented in previous anatomical studies using ROIs based on a priori knowledge. The macroscopic configurations of these reconstructed tracts are thus likely to reflect true fibre bundles, but it is possible that some parts of the trajectory may contain inaccuracies due to partial volume effects, noise, and crossing fibres. We tried to minimize the impact of the skills of the raters to follow the set rules for anatomic delineations by adequately training and supervising the raters; there were no systematic differences in our results depending on the experience of the raters.

In the reproducibility of the cross-sectional ROI measurements, the highest agreement has been found in the corpus callosum and the lowest for the corona radiata, internal

capsule, centrum semiovale, CST, and cerebral peduncle (Bisdas et al., 2008; Bonekamp et al., 2007; Brander et al., 2010; Ozturk et al. 2008; Vollmar et al., 2010). In our study, the lowest agreement in the cross-sectional ROI measurements was found in the UF and AF, which may be related to the tract shape and relatively high amount of crossing fibres in these areas. Only few previous reliability studies on the cross-sectional ROI measurements of the association tracts have been published. Vollmar et al. (2010) included the left uncinate fasciculus in their cross-sectional ROI analysis, and Bonekamp et al. (2007) included both cingulum bundles in their reproducibility analysis in children and adolescents. Vollmar et al. (2010) had better reproducibility for the FA measurements (scan-rescan ICC 0.91) compared to our study (intra-rater ICC 0.502 and inter-rater ICC 0.612), which may be related to their use of cardiac gating and bigger ROI size. In our study, both intra-rater and inter-rater comparisons revealed approximately twofold higher variability for FA values (mean CV 5.5% and 8.2%, respectively) compared to MD measurements (mean CV 2.5%, and 3.5%, respectively), which is in agreement with data reported by Bonekamp et al. (2007). The reproducibility of the ROI-based FA and MD measurements in the cingulum was markedly higher in their study compared to ours according to the ICC analysis, but it should be noticed that they also had a higher standard deviation of the FA measurements and relatively poor reproducibility of the FA measurements in the cingulum using the Bland-Altman reliability analysis (CR 12.7% from the mean FA). Our results are in accordance with the previous studies that have suggested that tract-based analysis may be more specific (Hong et al., 2008; Kanaan et al., 2006; Partridge et al., 2005) and reliable (Borich et al., 2012; Partridge et al., 2005; Wang and Melhelm, 2005) than ROI-based analysis to evaluate the integrity of white matter fibres. In our study, the tract-based method showed better reproducibility of FA and MD measurements in all the studied tracts compared to the cross-sectional ROI method.

For the tract-based method, the intra- and inter-rater CVs and ICCs for FA and MD were comparable with other data (Danielian et al., 2010 (UF); Malykhin et al., 2008 (UF, cingulum); Wang et al., 2012 (UF, cingulum, AF, IFOF)). In our study, the inter-session reproducibility for FA and MD measurements was, however, markedly higher compared to the recent results of Wang et al. (2012) when 15 gradient directions, such as in our study, were used. This may be related to the careful subject positioning and minor motion during the scan in our study. In accordance with previous studies (Danielian et al., 2010; Wang et al., 2012), intra- and inter-rater reproducibilities were higher compared to the between-scan reproducibility. In our study, between-scan reproducibility varied from good to excellent, except for the fair reproducibility in FA measurements in the left SC with FA 0.30/angle 27° and in volume measurements in the left AF with FA 0.15/angle 60°, and the poor reproducibility in MD measurements in the left IFOF with FA 0.15/angle 27°. We found no systematic difference in the reproducibility of the FA, MD and volume measurements depending on the FA or turning angle threshold. Measures of mean fractional anisotropy (FA) and mean diffusivity (MD) along the tracts were more reproducible than measures of tract volume, in accordance with previous studies

(Heiervang et al., 2006; Malykhin et al., 2008). Since the large variability in WM tract volumes and structure determined by tractography can decrease the accuracy of FA and MD measurements, additional measurements from the core of the tract have been recommended to confirm the tract-based results (Yasmin et al., 2008). According to our results, tractography provides a highly reproducible method to perform this central part analysis.

As a reliability estimate, it is preferable to use the ICC rather than the CV, as the former relates the size of the error variation to the size of the variation of interest. Even the ICC analysis has, however, some limitations; Since the ICC is calculated as a comparison of variation between cases (e.g., volume, FA or MD values of the tracts in the study population) to variation within cases (e.g., across raters), ICC may be high with a relatively large variation within cases if there is a large variation between cases (Bruton et al., 2000). Since, in general, no single reliability estimate should be used for reliability studies, and since also the ICC analysis has limitations, we considered it important to complement the ICC analyses by calculating the Bland and Altman 95% limits of agreement. The Bland and Altman 95% limits of agreement test gives a Coefficient of repeatability (CR) value that gives information on the magnitude of disagreement between measurements, for example, if CR for a FA measurement is 20, the disagreement between rater (intra-rater reliability) or raters (inter-rater reliability) is 20 or less with 95% likelihood (**Table 4**). Even though the Bland and Altman reliability analysis gives important additional information that can be interpreted clinically, its use has been rare in previous DTI studies. We found relatively high CR values for the ROI-based FA and MD measurements in several tracts, even if the ICC values were good or excellent (**Table 5**). On the other hand, besides the excellent intra- and inter-rater reliability of the tract-based methods according to the ICC analyses, our study showed low CR values for the FA (CR/ Average = 0.8-7.1%) and MD (CR/ Average = 0.2-4.8%) measurements in all the studied association tracts, indicating reasonable reliability of the tract-based methods in clinical use. However, the large variation in volumes seems to partly explain the good ICCs for the whole tract volumes in our study, since there was also relatively large variation in the measurements between raters, and the CRs of the volumes were large.

Our results make the use of tract-based volume analyses and ROI analyses in the study of the association tracts questionable, a finding, which should be taken into consideration in clinical applications. Additional investigations with a larger study population (n>50) and other clinical DTI study protocols are recommended to confirm our CR results. Because concurrent FA and volume changes complicate the grading of injury severity based on quantitative imaging data, further studies on whether the use of volume-related normal values could improve the accuracy of the method would also be valuable.

Inter-rater, intra-rater and between-scan reliability studies allow for an estimation of reproducibility, but another important aspect of reliability is whether a measurement is sensitive to the parameters of interest or not. Large variation in measurement of interest

in a normal population makes it difficult to detect changes in a diseased population, particularly at an individual level. We evaluated the difference between the TBI and healthy subjects by tractography-based analyses in our third sub-study (**III**), and found that the DTI tractography- based FA and MD analyses are suitable for clinical use also in this respect.

7. SUMMARY OF FINDINGS AND CONCLUSIONS

7.1. SUMMARY OF FINDINGS AND CLINICAL IMPLICATIONS

Study I: Both the number and extent of visible traumatic lesions on conventional MRI diminish significantly with time. As conventional MR imaging is still applied in clinical practice, it should be carried out soon after the injury at least in CT-negative cases. The absence of visible traumatic lesions especially in late conventional MRI does not exclude TBI, which has to be taken into account in medicolegal cases.

Study II: Slightly increased diffusion in normal-appearing brain is a common finding at one week after TBI, both in patients with and without visible lesions on conventional MRI. Diffusivity changes can occur in all supratentorial white matter and deep grey matter structures. Decreased ADCs at one week are uncommon in patients with mild TBI. This information may be used to improve documentation of the intracranial injury, but we could find no association between the ADC values and injury severity, whether measured as acute GCS or one-year outcome.

Study III: DTI tractography at 3 Tesla is potentially capable of improving the radiological diagnostics in the subpopulation of symptomatic TBI patients with no visible signs of intracranial or intraparenchymal abnormalities on routine MRI. Our results further stress the use of more sensitive imaging techniques if a chronic post-traumatic aetiology is suspected.

Study IV: According to this DTI tractography study of ten association tracts, the tract based FA and MD measurements are highly reproducible. Tract-based methods for volume measurements have, however, relatively low reproducibility. Compared to the commonly used cross-sectional ROI method, the tract-based analysis seem to be a more robust way to identify and measure white matter tracts of interest, which should be taken into consideration in clinical DTI applications. Neither the turning angle threshold (27°/60°) nor FA threshold (0.15/0.30) had a systematic influence on reproducibility.

7.2. FUTURE PROSPECTS FOR RESEARCH

The role of neuroimaging in the diagnosis of TBI is evolving. In general, the structural imaging techniques play a role in diagnosis, while the pathophysiology, symptom genesis, and mechanisms of recovery have recently been clarified by functional imaging techniques. However, DTI may also have the potential to add to our understanding of the natural course of traumatic axonal injuries and of the mechanisms of neuroplasticity and repair operating during recovery from TBI. DTI may also be useful to assess the effectiveness of treatment.

The diffusivity changes in normal-appearing brain in our study did not correlate either with GCS or outcome, and further investigations are needed to evaluate the possibilities of clinical applications of these findings.

We were able to demonstrate that DTI tractography at 3 Tesla is potentially capable of improving the radiological diagnostics in TBI patients with no findings on routine MRI. The relationship between FA values and outcome in this subpopulation remained unclear, since we had mainly (89.6%) patients with good or moderate outcome (GOS-E 5-7) with only a small variation in FA values. In our study, the detailed clinical characterization of the examined patients was studied retrospectively, which did not allow extensive uniform neuropsychological assessments. Further studies with more sensitive clinical techniques (neuropsychological assessments including follow-up) would be needed to evaluate whether the FA values are associated with the severity of injury in this TBI subpopulation. To find predictors for persistent postconcussive syndrome after mild TBI, is of potential interest, because neurobehavioural rehabilitation reduces the risk of persistent symptoms, and treatment failure is common if symptoms persist after 3 to 6 months. We found that the uncinate fasciculus and several other association tracts are common sites of injury in TBI patients with persistent symptoms with no findings on conventional MRI. Future prospective evaluation of whether DTI abnormalities in these tracts are connected with the risk of persistent symptoms would be valuable.

A few preliminary DTI studies at 7 T have been published recently (see e.g. Polders et al., 2011; Zhan et al., 2013), but further studies are needed to evaluate the potential advantages of the method in TBI research and diagnostics.

This thesis has concentrated mainly on the structural magnetic resonance imaging of traumatic brain injury. However, DTI or other structural methods cannot assess the "function" or underlying cerebral metabolic rate and cerebral blood flow in the brain. Besides the radioactive isotope based single-photon emission computed tomography (SPECT) and positron emission tomography (PET) techniques, the non-ionising functional magnetic resonance imaging (fMRI) and magnetic resonance spectroscopy (MRS), are relatively new functional imaging techniques increasingly used in TBI research (Anderson et al., 2011). Further studies are needed to establish the clinical applicability of these methods in TBI. In the future, comparison of modalities in a single study is also important, since it will help to establish how the modalities can be complementary to one another.

8. ACKNOWLEDGEMENTS

This study was carried out at the Department of Radiology, University of Turku. It was part of a more extensive study on traumatic brain injury coordinated by the Department of Neurology, University of Turku. I am grateful to the former Head of the Department of Radiology, Professor Martti Kormano, who first introduced me to this project and gave me the opportunity to be a part of this challenging and engrossing brain research area. I would also like to thank the present Head of the Department of Radiology, Professor Hannu Aronen, for his quite recent assistance.

I am most grateful to my supervisors of this thesis, Docent Timo Kurki and Docent Olli Tenovuo, for the privilege of working with them. Without their knowledge, professionalism, and enthusiasm, as well as support and encouragement in my research and writing throughout the years, this thesis would have been impossible. Thank you Timo and Olli!

I want to thank my co-authors Doctor Heli Hiekkanen, Doctor Jussi Laalo, and Biostatistician Tommi Kauko for their friendly collaboration. I also wish to thank Biostatistician Mari Koivisto for her assistance in statistical matters. I sincerely thank Jaqueline Välimäki, MA, for reviewing the language of this thesis.

Docent Tapio Tikkakoski and Professor Paul Parizel, the official reviewers of this thesis, are warmly acknowledged. I thank Tapio especially for his constructive criticism and detailed advice that helped me to improve my work. I express my sincere gratitude to Paul who dedicated himself to reviewing this work on a tight schedule. I am also grateful for his skilled support and advice.

I want to thank Docent Seppo Koskinen for his support, and assistance with my research visit at the Karolinska Institutet in autumn 2011. Radiologists at the Karolinska Institutet are acknowledged for their warm welcome. My “brain trauma sister” Harriet Nyström is especially acknowledged for the pleasant scientific conversations at the Karolinska Institutet’s Neuro Center during my three months stay in Stockholm.

I am also grateful to Docents Oili Salonen and Jukka Turkka, the members of my thesis committee, for their skilled support and encouragement.

I am especially grateful to the patients who participitated in this study, thus making it possible. I hope the findings of this work will be used in improving the diagnosis of patients with traumatic brain injury.

In my life outside the research world, I am grateful to

My best medical school friends, Heidi Immonen, Marja Sarkola, Anna Koskinen and Rita Arjonen, for sharing the unforgottable and eventful evolution from young medical students into medical doctors and still being there.

My current radiologist colleagues at Helsinki University Central Hospital, for intelligent advice in clinical work, and also for their pleasant company, which makes the everyday working life enjoyable.

All my dear friends, especially Päivi Kovanen, Veera Pikkarainen and Tintti Eteläpää, for providing me with other perspectives outside scientific work and sharing the ups and downs of life. Kristo, Jaakko, Tuomo, Jukka, Mia, Janne, Anna, Tytti, Laura, Arttu, Niina, and Karkki are also especially acknowledged.

My loving parents Erkki Brandstack and Hannele Sundfors, Mamma Anne Brandstack, Guy Sundfors, grandmother Irja Kaipainen, as well as sisters Nora Brandstack and Kirsi Juurinen and their husbands Teemu and Pekka, for their support and help in life.

My parents-in-law Anna-Liisa and Erkki, and brothers-in law for being there, and my sister-in-law Satu, and Matti, who have helped us tremendously and been our children's babysitter when we have been busy with our theses or needed a break from research world.

I save my warmest and most loving thoughts for my dear husband Janne Härkönen and our children, Aava and Eemil. Janne, no words exist to express my gratitude for all the love, patience, and support you have given me during these years. You have always taken good care of our children when I have needed to work, and you have always believed in me. I am fortunate to have you as my husband. Aava and Eemil, thank you for showing me what is most precious in life. With the three of you, life is a beautiful experience.

This study was financially supported by Turku University Hospital (EVO grants), Helsinki University Central Hospital (EVO grants), and The National Graduate School of Clinical Investigation, all of which are warmly acknowledged.

Nina Brandstack

Turku, March 2013

REFERENCES

Adams JH, Graham DI, Murray LS, Scott G. Diffuse axonal injury due to non missile head injury in humans: an analysis of 45 cases. Ann Neurol 1982;12:557-563.

Ahlhelm F, Hagen T, Schneider G, Dorenbeck U, Nabhan A, Reith W. ADC mapping of normal human brain. Med Sci Monit 2004;10:121-125.

Alexander AL, Lee JE, Lazar M, Field AS. Diffusion Tensor Imaging of the Brain. Neurotherapeutics 2007;4:316-329.

Alexander AL, Lee JE, Wu YC, Field AS. Comparison of diffusion tensor imaging measurements at 3.0 T versus 1.5 T with and without parallel imaging. Neuroimaging Clin N Am 2006;16:299-309.

Alsop DC, Murai H, Detre JA, McIntosh TK, Smith DH. Detection of acute pathologic changes following experimental traumatic brain injury using diffusion-weighted magnetic resonance imaging. J Neurotrauma 1996;13:515-21.

Anderson KE, Hurley RA, Taber KH. Functional Imaging. In: Silver JM, McAllister TW and Yudofsky SC, eds. Textbook of traumatic brain injury. 2nd ed. Washington, DC: American Psychiatric Publishing; 2011. p. 91-113.

Andriessen TMJC, Horn J, Franschman G, Van der Naalt J, Haitsma I, Jacobs B, Steyerberg EW, Vos PE. Epidemiology, Severity Classification, and Outcome of Moderate and Severe Traumatic Brain Injury: A Prospective Multicenter Study. J Neurotrauma 2011;28:2019-2031.

Arfanakis K., Haughton VM, Carew JD, Rogers BP, Dempsey RJ, Meyerand ME. Diffusion tensor MR imaging in diffuse axonal injury. AJNR Am J Neuroradiol 2002;23:794-802.

Ashwal S, Babikian T, Gardner-Nichols J, Freier MC, Tong KA, Holshouser BA. Susceptibility-weighted imaging and proton magnetic resonance spectroscopy in assessment of outcome after pediatric traumatic brain injury. Arch Phys Med Rehabil 2006;87(12 suppl 2):50-58.

Assaf Y, Beit-Yannai E, Shohami E, Berman E, Cohen Y. Diffusion- and T2-weighted MRI of close- head injury in rats: a time course study and correlation with histology. J Magn Reson Imaging 1997;15:77-85.

Bagley LJ, McGowan JC, Grossman RI, Sinson G, Kotapka M, Lexa FJ, Berlin JA, McIntosh TK. Magnetization Transfer Imaging of Traumatic Brain Injury. J Magn Reson Imaging 2000;11:1-8.

Barr RM, Gean AD, Le TH. Craniofacial Trauma. In: Brant WE and Helms CA, eds. Fundamentals of Diagnostic Radiology. 3rd ed. Philadelphia by Lippincott Williams & Wilkins; 2007. p. 55-85.

Barzo P, Marmarou A, Fatouros P, Ito J, Corwin F. MRI diffusion-weighted spectroscopy of reversible and irreversible ischemic injury following closed head injury. Acta Neurochir Suppl 1997;70:115-118.

Basser PJ. Inferring microstructural features and the physiological state of tissues from diffusion-weighted images. NMR Biomed 1995;8:333-344.

Basser PJ, Pierpaoli C. Microstructural and physiological features of tissues elucidated by quantitative-diffusion-tensor MRI. J Magn Reson 1996 B;111:209-219.

Belanger HG, Curtiss G, Demery JA, Lebowitz BK, Vanderploeg RD. Factors moderating neuropsychological outcomes following mild traumatic brain injury: a meta-analysis. J Int Neuropsychol Soc 2005;11:215-227.

Bendlin BB, Ries ML, Lazar M, Alexander AL, Dempsey RJ, Rowley HA, Sherman JE, Johnson SC. Longitudinal changes in patients with traumatic brain injury assessed with diffusion-tensor and volumetric imaging. Neuroimage 2008;42:503-514.

Berry C, Ley EJ, Tillou A, Cryer G, Margulies DR, Salim A. The effect of gender on patients with moderate to severe head injuries. J Trauma 2009;67:950-953.

Betz J, Zhuo J, Roy A, Shanmuganathan K, Gullapalli RP. Prognostic value of diffusion tensor imaging parameters in severe traumatic brain injury. J Neurotrauma 2012;29:1292-1305.

Bigler ED. Neuropsychology and clinical neuroscience of persistent post-concussive syndrome. JINS Journal of the International Neuropsychological Society 2008;14:1-22.

Bigler ED. Structural Imaging. In: Silver JM, McAllister TW and Yudofsky SC, eds. Textbook of traumatic brain injury. 2nd ed. Washington, DC: American Psychiatric Publishing; 2011. p. 73-90.

Bigler ED, Anderson CV, Blatter DD. Temporal lobe morphology in normal aging and traumatic brain injury. AJNR Am J Neuroradiol 2002;23:255-66.

Bigler ED, Snyder JL. Neuropsychological outcome and quantitative neuroimaging in mild head injury. Arch Clin Neuropsychol 1995;10:159-174.

Bisdas S, Bohning DE, *Bešenski* N, Nicholas JS, Rumboldt Z. Reproducibility, interrater agreement, and age-related changes of fractional anisotropy measures at 3T in healthy subjects: Effect of the applied b-value. AJNR Am J Neuroradiol 2008;29:1128-1133.

Blatter DD, Bigler ED, Gale SD, Johnson SC, Anderson CV, Burnett BM, Ryser D, Macnamara SE, Bailey BJ. MR-based brain and cerebrospinal fluid measurement after traumatic brain injury: correlation with neuropsychological outcome. AJNR Am J Neuroradiol 1997;18:1-10.

Bonekamp D, Nagae LM, Degaonkar M, Matson M, Abdalla WMA, Barker PB, Mori S, Horskáa A. Diffusion tensor imaging in children and adolescents: reproducibility, hemispheric, and age-related differences. Neuroimage 2007;34:733-742.

Borich MR, Wadden KP, Boyd LA. Establishing the reproducibility of two approaches to quantify white matter tract integrity in stroke. Neuroimage 2012;59:2393-2400.

Brander A, Kataja A, Saastamoinen A, Ryymin P, Huhtala H, Öhman J, Soimakallio S, Dastidar P. Diffusion tensor imaging of the brain in a healthy adult population: Normative values and measurement reproducibility at 3 T and 1.5 T. Acta Radiol 2010;7:800-807.

Brant WE. Diagnostic Imaging Methods. In: Brant WE, Helms CA, eds. Fundamentals of diagnostic radiology. 3rd ed. Philadelphia*:* Lippincott Williams & Wilkins; 2007. p. 3-28.

Bruton A, Conway JH, Holgate ST. Reliability: What is it and how is it measured? Physiotherapy 2000;86:94–99.

Caeyenberghs K, Leemans A, Geurts M, Taymans T, Linden CV, Smits-Engelsman BCM, Sunaert S, Swinnen SP. Brain-Behavior Relationships in Young Traumatic Brain Injury Patients: DTI Metrics are Highly Correlated with Postural Control. Hum Brain Mapp 2010;31:992-1002.

Catani M, Mesulam M. The arcuate fasciculus and the disconnection theme in language and aphasia: history and current state. Cortex 2008;44:953-961.

Chan JHM, Tsui EYK, Peh WC, Fong D, Fok KF, Leung KM, Yuen MK, Fung KK. Diffuse axonal injury: detection of changes in anisotropy of water diffusion by diffusion-weighted imaging. Neuroradiology 2003;45:34-38.

Chinn S. Repeatability and method comparison. Thorax 1991;46:454-456.

Christensen BK, Colella B, Inness E, Hebert D, Monette G, Bayley M, Green RE. Recovery of cognitive function after traumatic brain injury: A multilevel modeling analysis of Canadian outcomes. Arch Phys Med Rehabil 2008;89:3-15.

Chung SW, Courcot B, Sdika M, Moffat K, Rae C, Henry RG. Bootstrap quantification of cardiac pulsation artifact in DTI. Neuroimage 2010;49:631-640.

Coles JP. Imaging after brain injury. Br J Anaesth 2007;99:49-60.

Compagnone C, d'Avella D, Servadei F, Angileri FF, Brambilla G, Conti C, Cristofori L, Delfini R, Denaro L, Ducati A, Gaini SM, Stefini R, Tomei G, Tagliaferri F, Trincia G, Tomasello F. Patients with moderate head injury: a prospective multicenter study of 315 patients. Neurosurgery 2009;64:690-696.

Concha L, Gross DW, Beaulieu C. Diffusion Tensor Tractography of the Limbic System. AJNR Am J Neuroradiol 2005;26:2267-2274.

Danielsen ER, Christensen PB, Arlien-Søborg P, Thomsen C. Axonal recovery after severe traumatic brain injury demonstrated in vivo by 1H MR spectroscopy. Neuroradiology 2003;45:722-724.

Ducreux D, Huynh I, Fillard P, et al. Brain MR diffusion tensor imaging and fibre tracking to differentiate between two diffuse axonal injuries. Neuroradiology 2005;47:604-608.

Duffau H, Gatignol P, Moritz-Gasser S, Mandonnet E. Is the left uncinate fasciculus essential for language? A cerebral stimulation study. J Neurol 2009;256:382-389.

Dutton RP, Stansbury LG, Leone S, Kramer E, Hess JR, Thomas M, Scalea TM. Trauma Mortality in Mature Trauma Systems: Are We Doing Better? An Analysis of Trauma Mortality Patterns, 1997–2008. J Trauma. 2010;69:620-626.

Ebisu T, Naruse S, Horikawa Y, Ueda S, Tanaka C, Uto M, Umeda M, Higuchi T. Discrimination between different types of white matter edema with diffusion-weighted MR imaging. J Magn Reson Imaging 1993;3:863-868.

Engelter ST, Provenzale JM, Petrella JR, DeLong DM, MacFall JR. The effect of aging on the apparent diffusion coefficient of normal-appearing white matter. AJR Am J Roentgenol 2000;175:425-430.

Evans RW. Post-traumatic headaches. Neurological Clinics 2004;22:237-249.

Ezaki Y, Tsutsumi K, Morikawa M, Nagata I. Role of diffusion-weighted magnetic resonance imaging in diffuse axonal injury. Acta Radiol 2006;47:733-740.

Farkas O, Povlischock TJ. Cellular and subcellular change evoked by diffuse traumatic brain injury: a

complex web of change extending far beyond focal damage. Progress Brain Res 2007;161:43-59.

Fiser SM, Johnson SB, Fortune JB. Resource utilization in traumatic brain injury: the role of magnetic resonance imaging. Am Surg 1998;64:1088-1093.

Fork M, Bartels C, Ebert AD, Grubich C, Synowitz H, Wallesch CW. Neuropsychological sequelae of diffuse traumatic brain injury. Brain Inj 2005;19:101-108.

Gale SD, Baxter L, Roundy N, Johnson SC. Traumatic brain injury and grey matter concentration: a preliminary voxel based morphometry study. J Neurol Neurosurg Psychiatry 2005;76:984-988.

Garnett MR, Blamire AM, Corkill RG, Cadoux-Hudson TA, Rajagopalan B, Styles P. Early proton magnetic resonance spectroscopy in normal-appearing brain correlates with outcome in patients following traumatic brain injury. Brain 2000;123:2046-2054.

George D, Mallery P. SPSS for Windows step by step: A simple guide and reference, 2003. 11.0 update, 4th ed. Boston: Allyn & Bacon.

Gerber DJ, Weintraub AH, Cusick CP, Ricci PE, Whiteneck GG. Magnetic resonance imaging of traumatic brain injury: relationship of T2*SE and T2GE to clinical severity and outcome. Brain Inj 2004;18:1083-1097.

Goetz P, Blamire A, Rajagopalan B, Cadoux-Hudson T, Young D, Styles P. Increase in apparent diffusion coefficient in normal appearing white matter following human traumatic brain injury correlates with injury severity. J Neurotrauma 2004;21:645-654.

Grossman EJ, Inglese M, Bammer R. Mild traumatic brain injury: is diffusion imaging ready for primetime in forensic medicine? Top Magn Reson Imaging 2010;21:379-386.

Hadley DM, Teasdale GM, Jenkins A, Condon B, Macpherson P, Patterson J, Rowan JO. Magnetic resonance imaging in acute head injury. Clin Radiol 1988;39:131-139.

Hakulinen U, Brander A, Ryymin P, Öhman J, Soimakallio S, Helminen M, Dastidar P, Eskola H. Repeatability and variation of region-of-interest methods using quantitative diffusion tensor MR imaging of the brain. BMC Medical Imaging 2012;12:30.

Hanstock CC, Faden AI, Bendall MR, Vink R. Diffusion-weighted imaging differentiates ischemic tissue from traumatized tissue. Stroke 1994;25:843-848.

Hart T, Sherer M, Whyte J, Polansky M, Novack TA. Awareness of behavioral, cognitive, and physical deficits in acute traumatic brain injury. Arch Phys Med Rehabil 2004;85:1450-1456.

Hasan KM, Kamali A, Abid H, Kramer LA, Fletcher JM, Ewing-Cobbs L. Quantification of the spatiotemporal microstructural organization of the human brain association, projection and commissural pathways across the lifespan using diffusion tensor tractography. Brain Struct Funct 2010;214:361-373.

Heiervang E, Behrens TE, Mackay CE, Robson MD, Johansen-Berg H. Between session reproducibility and between subject variability of diffusion MR and tractography measures. Neuroimage 2006;33:867-877.

Helenius J, Soinne L, Perkiö J, Salonen O, Kangasmäki A, Kaste M, Carano RAD, Aronen HJ, Tatlisumak T. Diffusion-weighted MR imaging in normal human brains in various age groups. AJNR Am J Neuroradiol 2002;23:194-199.

Hergan K, Schaefer PW, Sorensen AG, Gonzalez RG, Huisman TA. Diffusion-weighted MRI in diffuse axonal injury of the brain. Eur J Radiol 2002;12:2536-2541.

Hermoye L, Saint-Martin C, Cosnard G, Lee S-K, Kim J, Nassogne M-C, Menten R, Clapuyt P, Donohue PK, Hua K, Wakana S, Jiang H, van Zijl PCM, Mori S. Pediatric diffusion tensor imaging: Normal database and observation of the white matter maturation in early childhood. Neuroimage 2006;29:493-504.

Himanen L, Portin R, Hämäläinen P, Hurme S, Hiekkanen H, Tenovuo O: Risk factors for reduced survival after traumatic brain injury: a 30-year follow-up study. Brain Inj 2011;25:443-452.

Himanen L, Portin R, Isoniemi H, Helenius H, Kurki T, Tenovuo O. Cognitive functions in relation to MRI findings 30 years after traumatic brain injury. Brain Inj 2005;19:123-133.

Hofman PA, Stapert SZ, van Kroonenburgh MJ, Jolles J, de Kruijk J, Wilmink JT. MR imaging, single-photon emission CT, and neurocognitive performance after mild traumatic brain injury. AJNR Am J Neuroradiol 2001;22:441-449.

Hofman PAM, Verhey FRJ, Wilmink JT, Rozendaal N, Jolles J. Brain lesions in patients visiting memory clinic with postconcussional sequelae after mild to moderate brain injury. J Neuropsychiatry Clin Neurosci 2002;14:176-184.

Holm L, Cassidy JD, Carroll LJ, Borg J. Summary of the WHO collaborating centre for neurotrauma task force on mild traumatic brain injury. J Rehabil Med 2005;37:137-141.

Hong YH, Sung JJ, Kim SM, Park KS, Lee KW, Chang KH, Song IC. Diffusion tensor tractography-

based analysis of the pyramidal tract in patients with amyotrophic lateral sclerosis. J Neuroimaging 2008;18:282–287.

Hughes DG, Jackson A, Mason DL, Berry E, Hollis S, Yates DW. Abnormalities on magnetic resonance imaging seen acutely following mild traumatic brain injury: correlation with neuropsychological tests and delayed recovery. Neuroradiology 2004;46:550-558.

Huisman TAGM, Schwamm LH, Schaefer PW, Koroshetz WJ, Shetty-Alva N, Ozsunar Y, Wu O, Sorensen AG. Diffusion tensor imaging as potential biomarker of white matter injury in diffuse axonal injury. AJNR Am J Neuroradiol 2004;25:370-376.

Huisman TA, Sorensen AG, Hergan K, Schaefer PW. Diffusion-weighted imaging for the evaluation of diffuse axonal injury in closed head injury. J Comput Assist Tomo 2003;27:5-11.

Hukkelhoven C, Steyerberg E, Farace E, Habbema D, Marshall L, Maas A. Regional differences in patient characteristics, case management and outcomes in traumatic brain injury: experience from the tirilazad trials. Neurosurgery 2002;97:549-557.

Ichise M, Chung D-C, Wang P, Wortzman G, Gray BG, Franks W. Technetium-99m-HMPAO SPECT, CT, and MRI in the evaluation of patients with chronic traumatic brain injury: a correlation with neuropsychological performance. J Nucl Med 1994;35:217-226.

Inglese M, Makani S, Johnson G, Cohen BA, Silver JA, Gonen O, Grossman RI. Diffuse axonal injury in mild traumatic brain injury: a diffusion tensor imaging study. J Neurosurgery 2005;103:298-303.

Iverson GL. Outcome from mild traumatic brain injury. Curr Opin Psychiatry 2005;18:301-317.

Iverson GL, Hakulinen U, Wäljas M, Dastidar P, Lange RT, Soimakallio S, Öhman J. To exclude or not to exclude: white matter hyperintensities in diffusion tensor imaging research. Brain Inj 2011;25:1325-1332.

Iverson GL, Jacobs B, Beems T, van der Vliet TM, Diaz-Arrastia RR, Borm GF, Vos PE. Computed tomography and outcome in moderate and severe traumatic brain injury: Hematoma, volume and midline shift revisited. J Neurotrauma 2011;28:203-215.

Jenkins A, Teasdale G, Hadley MDM, Macpherson P, Rowan JO. Brain lesions detected by magnetic resonance imaging in mild and severe head injuries. The Lancet 1986;ii:445-446.

Jennett B. Epidemiology of head injury. J Neurol Neurosurg Psychiatry 1996;60:362-369.

Jennett B, Bond M. Assessment of outcome after severe brain damage: a practical scale. Lancet 1975;i:480-484.

Jennett B, Snoek J, Bond MR, Brooks N. Disability after severe head injury: observations on the use of the Glasgow Outcome Scale. J Neurol Neurosurg Psychiatry 1981;44:285-293.

Kanaan RA, Shergill SS, Barker GJ, Catani M, Ng VW, Howard R, McGuire PK, Jones DK. Tract-specific anisotropy measurements in diffusion tensor imaging. Psychiatry Res Neuroimaging. 2006;146:73-82.

Kinnunen KM, Greenwood R, Powell JH, Leech R, Hawkins PC, Bonnelle V, Patel MC, Counsell SJ, Sharp DJ. White matter damage and cognitive impairment after traumatic brain injury. Brain 2011;134:449-463.

Kinoshita T, Moritani T, Hiwatashi A, Wang HZ, Shrier DA, Numaguchi Y, Westesson P-LA. Conspicuity of diffuse axonal injury lesions on diffusion-weighted MR imaging. Eur J Radiol 2005;56:5-11.

Koskinen S, Alaranta H. Traumatic brain injury in Finland 1991-2005: a nationwide register study of hospitalized and fatal TBI. Brain Inj 2008;22:205-214.

Kraus MF, Susmaras T, Caughlin BP, Walker CJ, Sweeney JA, Little DM. White matter integrity and cognition in chronic traumatic brain injury: a diffusion tensor imaging study. Brain 2007; 130:2508-2519.

Kuo J-R, Lo C-J, Lu C-L, Chio C-C, Wang C-C, Lin K-C. Prognostic predictors of outcome in an operative series in traumatic brain injury patients. J Formos Med Assoc 2011;110:258-264.

Kurki T, Laalo J, Oksaranta O. Diffusion tensor tractography of the uncinate fasciculus: pitfalls in quantitative analysis due to traumatic volume changes. J Magn Reson Imaging 2012, in press.

Käypä hoito; Aivovammat. Current Care guideline; Adult traumatic brain injury (online). Working group set by the Finnish Medical Society Duodecim and the Finnish Neurological Society, the Societas Medicinae Physicalis et Rehabilitationis Fenniae, the Finnish Neurosurgical Society, the Finnish Neuropsychological Society, and the Finnish Insurance Medicine Society. Helsinki: Finnish Medical Society Duodecim, 2008 (referred 14.2.2011). Available online at: www.kaypahoito.fi

Laalo JP, Kurki TJ, Sonninen PH, Tenovuo OS. Reliability of diagnosis of traumatic brain injury by computed tomography in the acute phase. J Neurotrauma. 2009;26:2169-2178.

Landman BA, Farrell JAD, Jones CK, Smith SA, Prince JL, Moria S. Effects of diffusion weighting schemes on the reproducibility of DTI-derived fractional anisotropy, mean diffusivity, and principal eigenvector measurements at 1.5T. Neuroimage 2007;36:1123-1138.

Lange RT, Iverson GL, Brubacher JR, Mädler B, Heran MK. Diffusion tensor imaging findings are not strongly associated with postconcussional disorder 2 months following mild traumatic brain injury. J Head Trauma Rehabil 2012;27:188-198.

Le TH, Gean AD. Neuroimaging of traumatic brain injury. Mt Sinai J Med. 2009;76:145-162.

Le TH, Mukherjee P, Henry RG, Berman JI, Ware M, Manley GT. Diffusion tensor imaging with three-dimensional fiber tractography of traumatic axonal shearing injury: an imaging correlate for the posterior callosal "disconnection" syndrome: case report. Neurosurgery 2005;56:189.

Lee H, Wintermark M, Gean AD, Ghajar J, Manley GT, Mukherjee P. Focal lesions in acute mild traumatic brain injury and neurocognitive outcome: CT versus 3T MRI. J Neurotrauma 2008;25:1049-1056.

Lehtonen S, Stringer AY, Millis S, Boake C, Englander J, Hart T, High W, Macciocchi S, Meythaler J, Novack T, Whyte J. Neuropsychological outcome and community re-integration following traumatic brain injury: The impact of frontal and non-frontal lesions. Brain Inj 2005;19:239-256.

Levin HS, Amparo E, Eisenberg HM, Williams DH, High WM Jr, McArdle CB, Weiner RL. Magnetic resonance imaging and computerized tomography in relation to the neurobehavioral sequelae of mild and moderate head injuries. J Neurosurg 1987;66:706-713.

Levin HS, Williams DH, Eisenberg HM, High Jr WM, Guinto Jr FC. Serial MRI and neurobehavioural findings after mild to moderate closed head injury. J Neurol Neurosurg Psychiatry 1992;55:255-262.

Levine B, Fujiwara E, O'Connor C, Richard N, Kovacevic N, Mandic M, Restagno A, Easdon C, Robertson IH, Graham SJ, Cheung G, Gao F, Schwartz ML, Black SE. In vivo characterization of traumatic brain injury neuropathology with structural and functional neuroimaging. J Neurotrauma 2006;23:1396-1411.

Lin F, Wu G, Zhu L, Lei H. Heavy smokers show abnormal microstructural integrity in the anterior corpus callosum: A diffusion tensor imaging study with tract-based spatial statistics. Drug Alcohol Depend 2013;129:82-87.

Lin F, Zhou Y, Du Y, Qin L, Zhao Z, Xu J, Lei H. Abnormal White Matter Integrity in Adolescents with Internet Addiction Disorder: A Tract-Based Spatial Statistics Study. PLoS ONE 2012;7:e30253.

Ling J, Merideth F, Caprihan A, Pena A, Teshiba T, Mayer AR. Head Injury or Head Motion? Assessment and Quantification of Motion Artifacts in Diffusion Tensor Imaging Studies. Hum Brain Mapp 2011;000:000–000 (online only).

Lipton ML, Gellella E, Lo C, Gold T, Ardekani BA, Shifteh K, Bello JA, Branch CA. Multifocal white matter ultrastructural abnormalities in mild traumatic brain injury with cognitive disability: a voxel-wise analysis of diffusion tensor imaging. J Neurotrauma 2008;25:1335-1342.

Lipton ML, Gulko E, Zimmerman ME, Friedman BW, Kim M, Gellella E, Gold T, Shifteh K, Ardekani BA, Branch CA. Diffusion-tensor imaging implicates prefrontal axonal injury in executive function impairment following very mild traumatic brain injury. Radiology 2009;252:816-824.

Little DM, Kraus MF, Joseph J, Geary EK, Susmaras T, Zhou XJ, Pliskin N, Gorelick PB. Thalamic integrity underlies executive dysfunction in traumatic brain injury. Neurology 2010;74:558-564.

Liu AY, Maldjian JA, Bagley LJ, Sinson GP, Grossman RI. Traumatic brain injury: diffusion-weighted MR imaging findings. AJNR Am J Neuroradiol 1999;20:1636-1641.

Luoto T, Tenovuo O, Kataja A, Brander A, Ohman J, Iverson GL. Who gets recruited in mild traumatic brain injury research? J Neurotrauma 2013;30:11-16.

Machamer J, Temkin N, Fraser R, Doctor JN, Dikmen S. Stability of employment after traumatic brain injury. J Int Neuropsychol Soc 2005;11:807-816.

MacKenzie EJ. Epidemiology of injuries: current trends and future challenges. Epidemiol Rev 2000;22:112-119.

MacKenzie JD, Siddiqi F, Babb JS, Bagley LJ, Mannon LJ, Sinson GP, Grossman RI. Brain atrophy in mild or moderate traumatic brain injury: a longitudinal quantitative analysis. AJNR Am J Neuroradiol 2002;23:1509-1515.

Malykhin N, Concha L, Seres P, Beaulieu C, Coupland NJ. Diffusion tensor imaging tractography and reliability analysis for limbic and paralimbic white matter tracts. Psychiatry Res Neuroimaging 2008;164:132-142.

Marmarou A, Signoretti S, Fatouros PP, Portella G, Aygok GA, Bullock MR. Predominance of cellular edema in traumatic brain swelling in patients with severe head injuries. J Neurosurgery 2006;104:720-730.

Mayer AR, Ling J, Mannell MV, Gasparovic C, Phillips JP, Doezema D, Reichard R, Yeo RA. A prospective diffusion tensor imaging study in mild traumatic brain injury. Neurology 2010;74:643-650.

McAllister TW. Genetic Factors. In: Silver JM, McAllister TW and Yudofsky SC, eds. Textbook of traumatic brain injury. 2nd ed. Washington, DC: American Psychiatric Publishing; 2011. p. 37-48.

McAllister TW. Mild Brain Injury. In: Silver JM, McAllister TW and Yudofsky SC, eds. Textbook of traumatic brain injury. 2nd ed. Washington, DC: American Psychiatric Publishing; 2011. p. 239-264.

McCauley SR, Boake C, Levin HS, Contant CF, Song JX. Postconcussional disorder following mild to moderate traumatic brain injury: anxiety, depression, and social support as risk factors and comorbidities. J Clin Exp Neuropsychol 2001;23:792-808.

McLean A, Dikmen S, Temkin N, Wyler AR, Gale JL. Psychosocial functioning at 1 month after injury. Neurosurgery 1984;14:393-399.

Medana I, and Esiri M. Axonal damage: a key predictor of outcome in human CNS diseases. Brain 2003;126:515-530.

Menon DK, Zahed C. Prediction of outcome in severe traumatic brain injury. Current Opinion in Critical Care 2009;15:437-441.

Messé A, Caplain S, Paradot G, Garrigue D, Mineo JF, Soto Ares G, Ducreux D, Vignaud F, Rozec G, Desal H, Pélégrini-Issac M, Montreuil M, Benali H, Lehéricy S. Diffusion tensor imaging and white matter lesions at the subacute stage in mild traumatic brain injury with persistent neurobehavioral impairment. Hum Brain Mapp 2011;32:999-1011.

Mitchener A, Wyper DJ, Patterson J, Hadley DM, Wilson JT, Scott LC, Jones M, Teasdale GM. SPECT, CT, and MRI in head injury: acute abnormalities followed up at six months. J Neurol Neurosurg Psychiatry 1997;62:633-636.

Mittal S, Wu Z, Neelavalli J, Haacke EM. Susceptibility-Weighted Imaging: Technical Aspects and Clinical Applications, Part 2. AJNR Am J Neuroradiol 2009;30:232-252.

Mittenberg W, Tremont G, Zielinski RE, Fichera S, Rayls KR. Cognitive-behavioral prevention of postconcussion syndrome. Arch Clin Neuropsychol 1996;11:139-145.

Mori S, Crain BJ, Chacko VP, van Zijl PC. Three-dimensional tracking of axonal projections in the brain by magnetic resonance imaging. Ann Neurol 1999;45:265-269.

Mori S, van Zijl P. Fiber tracking: principles and strategies -a technical review. NMR Biomed 2002;15:468-480.

Morton MV, Wehman P. Psychosocial and emotional sequelae of individuals with traumatic brain injury: a literature review and recommendations. Brain Inj 1995;9:81-92.

Mukherjee P, Berman JI, Chung SW, Hess CP, Henry RG. Diffusion tensor MR imaging and fiber tractography: theoretic underpinnings. AJNR Am J Neuroradiol 2008a;29:632-641.

Mukherjee P, Chung SW, Berman JI, Hess CP, Henry RG. Diffusion tensor MR imaging and fiber tractography: technical considerations. AJNR Am J Neuroradiol 2008b;29:843-852.

Murray GD, Teasdale GM, Braakman R, Cohadon F, Dearden M, Iannotti F, Karimi A, Lapierre F, Maas A, Ohman J, Persson L, Servadei F, Stocchetti N, Trojanowski T, Unterberg A. The European Brain Injury Consortium survey of head injuries. Acta Neurochir 1999;14:223-236.

Nakayama N, Okumura A, Shinoda J, Yasokawa Y-T, Miwa K, Yoshimura S-I, Iwama T. Evidence for white matter disruption in traumatic brain injury without macroscopic lesions. J Neurol Neurosurg Psychiatry 2006;77:850-855.

Netsch T, van Muiswinkel A. Quantitative evaluation of image-based distortion correction in diffusion tensor imaging. IEEE Trans Med Imaging 2004;23:789-798.

Newton MR, Greenwood RJ, Britton KE, Charlesworth M, Nimmon CC, Carroll MJ, Dolke G. A study comparing SPECT with CT and MRI after closed head injury. J Neurol Neurosurg Psychiatry 1992;55:92-94.

Ni H, Kavcic V, Zhu T, Ekholm S, Zhong J. Effects of number of diffusion gradient directions on derived diffusion tensor imaging indices in human brain. AJNR Am J Neuroradiol 2006;27:1776-1781.

NIH Consensus Development Panel on Rehabilitation of Persons With Traumatic Brain Injury. Rehabilitation of Persons With Traumatic Brain Injury. JAMA 1999;282:974-983.

Niogi SN, Mukherjee P. Diffusion Tensor Imaging of Mild TBI. J Head Trauma Rehabil 2010;25:241-255.

Niogi SN, Mukherjee P, Ghadar J, Johnson C, Kolster RA, Sarkar R, Lee H, Meeker M, Zimmerman RD, Manley GT, McCandliss BD. Extent of microstructural white matter injury in postconcussive syndrome correlates with impaired cognitive reaction time: a 3T diffusion tensor imaging study of mild traumatic brain injury. AJNR Am J Neuroradiol 2008;29:967-973.

Ommaya AK, Goldsmith W, Thibault L. Biomechanics and neuropathology of adult and paediatric head injury. Br J Neurosurg 2002;16:220-242.

Orman JAL, Kraus JF, Zaloshnja E, Miller T. Epidemiology. In: Silver JM, McAllister TW and Yudofsky SC, eds. Textbook of traumatic brain injury. 2nd ed. Washington, DC: American Psychiatric Publishing; 2011. p.3-22.

Ozturk A, Sasson AD, Farrell JAD, Landman BA, da Motta ACBS, Aralasmak A, Yousem DM. Regional differences in diffusion tensor imaging measurements: Assesment of intrarater and interrater variability. AJNR Am J Neuroradiol 2008;29:1124-1127.

Parizel PM, Van Goethem JW, Ozsarlak O, Maes M, Phillips CD. New developments in the neuroradiological diagnosis of craniocerebral trauma. Eur Radiol 2005;15:569-581.

Parizel PM, Ozsarlak, Van Goethem JW, van den Hauwe L, Dillen C, Verlooy J, Cocyns P, De Schepper AM**.** Imaging findings in diffuse axonal injury after closed head trauma. Eur Radiol 1998;8:960-965.

Paterakis K, Karantanas AH, Komnos A, Volikas Z. Outcome of patients with diffuse axonal injury: the significance and prognostic value of MRI in the acute phase. J Trauma 2000;49:1071-1075.

Partridge SC, Mukherjee P, Berman JI, Henry RG, Miller SP, Lu Y, Glenn OA, Ferriero DM, Barkovich AJ, Yigneron DB. Tractography-based quantitation of diffusion tensor imaging parameters in white matter tracts of preterm new-borns. J Magn Reson Imaging 2005;22:467-474.

Perlbarg V, Puybasset L, Tollard E, Lehericy S, Benali H, Galanaud D. Relation between brain lesion location and clinical outcome in patients with severe traumatic brain injury: A diffusion tensor imaging study using voxel-based approaches. Hum Brain Mapp 2009;30:3924-3933.

Pierallini A, Pantano P, Fanzotti LM, Bonamini M, Vichi R, Zylberman R, Pisarri F, Colonnese C, Bozzao L. Correlation between MRI findings and long-term outcome in patients with severe trauma. Neuroradiology 2000;42:860-867.

Polders DL, Leemans A, Hendrikse J, Donahue MJ, Luijten PR, Hoogduin JM. Signal to Noise Ratio and Uncertainty in Diffusion Tensor Imaging at 1.5, 3.0, and 7.0 Tesla. J Magn Reson Imaging 2011;33:1456-1463.

Portney L, Watkins M. Foundations of Clinical Research: Applications to Practice, Second Edition. Appleton & Lange, Norwalk, CN; 2000.

Ptak T, Sheridan RL, Rhea JT, Gervasini AA, Yun JH, Curran MA, Borszuk P, Petrovick L, Novelline RA. Cerebral fractional anisotropy score in trauma patients: A new indicator of white matter injury after trauma. Am J Roentgenol 2003;181:1401-1407.

Ramstedt, M. Variations in alcohol-related mortality in the Nordic countries after 1995 –continuity or change? Nordic Studies on Alcohol and Drugs 2007;24:5-15.

Rankin G, Stokes M. Reliability of assessment tools in rehabilitation: An illustration of appropriate statistical analyses. Clinical Rehabilitation 1998;12.

Rohde GK, Barnett AS, Basser PJ, Marenco S, Pierpaoli C. Comprehensive approach for correction of motion and distortion in diffusion-weighted MRI. Magn Reson Med 2004;51:103-114.

Rugg-Gunn FJ, Symms MR, Barker GJ, Greenwood R, Duncan JS. Diffusion imaging shows abnormalities after blunt head trauma when conventional magnetic resonance imaging is normal. J Neurol Neurosurg Psychiatry 2001;70:530-533.

Rutgers DR, Toulgoat F, Cazejust J, Fillard P, Lasjaunias P, Ducreux D. White matter abnormalities in mild traumatic brain injury: A diffusion tensor imaging study. AJNR Am J Neuroradiol 2008;29:514-519.

Salmond CH, Menon DK, Chatfield DA, Williams GB, Pena A, Sahakian BJ, Pickard JD. Diffusion tensor imaging in chronic head injury survivors: Correlations with learning and memory indices. Neuroimage 2006;29:117-124.

Schaefer PW, Buonanno FS, Gonzalez RG, and Schwamm LH. Diffusion-weighted imaging discriminates between cytotoxic and vasogenic edema in a patient with eclampsia. Stroke 1997;28:1082-1085.

Schaefer PW, Grant PE, Gonzalez RG. Diffusion-weighted MR imaging of the brain. Radiology 2000;217:331-345.

Schaefer PW, Huisman TAGM, Sorensen G, Gonzalez RG, Schwamm LH. Diffusion-weighted MR imaging in closed head injury: high correlation with initial Glasgow Coma Scale score and score on Modified Rankin Scale at discharge. Radiology 2004;233:58-66.

Scheid R, Preul C, Gruber O, Wiggins C, von Cramon DY. Diffuse axonal injury associated with chronic traumatic brain injury: evidence from T2*-weighted gradient-echo imaging at 3 T. AJNR Am J Neuroradiol 2003;24:1049-1056.

Scheid R, Walther K, Guthke T, Preul C, von Cramon DY. Cognitive sequelae of diffuse axonal injury. Arch Neurol 2006;63:418-424.

Scheltens P, Barkhof F, Leys D, Pruvo JP, Nauta JJ, Vermersch P, Steinling M, Valk J. A semiquantative rating scale for the assesment of signal hyperintensities on magnetic resonance imaging. J Neurol Sci 1993;114:7-12.

Schmahmann JD, Smith EE, Eichler FS, Filley CM. Cerebral white matter: neuroanatomy, clinical neurology, and neurobehavioral correlates. Ann N Y Acad Sci 2008;1142:266-309.

Shanmuganathan K, Gullapalli RP, Mirvis SE, Roys S, Murthy P. Whole-brain apparent diffusion coefficient in traumatic brain injury: correlation with Glasgow Coma Scale Score. AJNR Am J Neuroradiol 2004;25:539-544.

Sherer M, Novack TA, Sander AM, Struchen MA, Alderson A, Thompson, RN. Neuropsychological assessment and employment outcome after traumatic brain injury: a review. Clin Neuropsychol 2002;16:157-178.

Shrout P, Fleiss J. Intraclass correlations: uses in assessing rater reliability. Psychol Bull 1979;2:420-428.

Smith C. Neuropathology. In: Silver JM, McAllister TW and Yudofsky SC, eds. Textbook of traumatic brain injury. 2nd ed. Washington, DC: American Psychiatric Publishing; 2011. p. 23-36.

Snook L, Plewes C, Beaulieu C. Voxel based versus region of interest analysis in diffusion tensor imaging of neurodevelopment. Neuroimage 2007;34:243-252.

Sosin DM, Sniezek JE, Thurman DJ. Incidence of mild and moderate brain injury in the United States, 1991. Brain Inj 1996;10:47-54.

Stamatakis EA, Wilson JTL, Hadley DM, Wyper DJ. SPECT imaging in head injury interpreted with statistical parametric mapping. J Nucl Med 2002;43:476-483.

Stein SC, Georgoff P, Meghan S, Mizra K, Sonnad SS. 150 years of treating severe traumatic brain injury: a systematic review of progress in mortality. J. Neurotrauma 2010;27:1343-1353.

Sullivan EV, Adalsteinsson E, Hedehus M, Ju C, Moseley M, Lim KO, Pfefferbaum A. Equivalent disruption of regional white matter microstructure in ageing healthy men and women. Neuroreport 2001;12:99-104.

Sundstrøm T, Sollid S, Wentzel-Larsen T, Wester K. Head Injury Mortality in the Nordic Countries. J Neurotrauma 2007;24:147-153.

Tagliaferri F, Compagnone C, Korsic M, Servadei F, Kraus J. A systematic review of brain injury epidemiology in Europe. Acta Neurochir 2006;148:255-268.

Takayama H, Kobayashi M, Sugishita M, Mihara B. Diffusion-weighted imaging demonstrates transient cytotoxic edema involving the corpus callosum in a patient with diffuse brain injury. Clin Neurol Neurosurg 2000;102:135-139.

Teasdale G, Jennett B. Assessment of coma and impaired consciousness. A practical scale. Lancet 1974;304:81-84.

Teasdale GM, Pettigrew LE, Wilson JT, Murray G, Jennett B. Analyzing outcome of treatment of severe head injury: a review and update on advancing the use of the Glasgow Outcome Scale. J Neurotrauma 1998;15:587-597.

Tench CR, Morgan PS, Wilson M, Blumhardt LD. White matter mapping using diffusion tensor MRI. MRM Magnetic Resonance in Medicine 2002;47:967-972.

Thatcher RW, Camacho M, Salazar A, Linden C, Biver C, Clarke L. Quantitative MRI of the gray-white matter distribution in traumatic brain injury. J Neurotrauma 1997;14:1-14.

Tomaiuolo F, Carlesimo GA, Di Paola M, Petrides M, Fera F, Bonanni R, Formisano R, Pasqualetti P, Caltagirone C. Gross morphology and morphometric sequelae in the hippocampus, fornix, and corpus callosum of patients with severe non-missile traumatic brain injury without macroscopically detectable lesions: a T1 weighted MRI study. J Neurol Neurosurg Psychiatry 2004;75:1314-1322.

Tong KA, Ashwal S, Holshouser BA, Nickerson JP, Wall CJ, Shutter LA, Osterdock RJ, Haacke EM, Kido D. Diffuse axonal injury in children: clinical correlation with hemorrhagic lesions. Ann Neurol 2004;56:36-50.

Toyama Y, Kobayashi T, Nishiyama Y, Satoh K, Ohkawa M, Seki K. CT for acute stage of closed head injury. Radiat Med 2005;23:309-316.

Van der Naalt J, Hew JM, van Zomeren AH, Sluiter WJ, Minderhoud JM. Computed tomography and magnetic resonance imaging in mild to moderate head injury: Early and late imaging related to outcome. Ann Neurol 1999;46:70-78.

Vitaz TW, Jenks J, Raque GH, Shields CB. Outcome following moderate traumatic brain injury. Surg Neurol 2003;60:285-291.

Voller B, Auff E, Schnider P, Aichner F. To do or not to do? Magnetic resonance imaging in mild traumatic brain injury. Brain Inj 2001;15:107-115.

Vollmar C, O'Muircheartaigh J, Barker GJ, Symms MR, Thompson P, Kumari V, Duncan JS, Richardson MP, Koepp MJ. Identical, but not the same: Intra-site and inter-site reproducibility of fractional anisotropy measures on two 3.0 T scanners. Neuroimage 2010;51:1384-1394.

Wakana S, Caprihan A, Panzenboeck MM, Fallon JH, Perry M, Gollub RL, Hua K, Zhang J, Jiang H, Dubey P, Blitz A, van Zijl P, Mori S. Reproducibility of quantitative tractography methods applied to

cerebral white matter. Neuroimage 2007;36:630-644.

Walker WC, Ketchum JM, Marwitz JH, Chen T, Hammond F, Sherer M, Meythaler J. A multicentre study on the clinical utility of post-traumatic amnesia duration in predicting global outcome after moderate-severe traumatic brain injury. J Neurol Neurosurg Psychiatry 2010;81:87-89.

Wang JY, Bakhadirov K, Devous MDS, Abdi H, McColl R, Moore C, de la Plata CDM, Ding K, Whittemore A, Babcock E, Rickbeil T, Dobervich J, Kroll D, Dao B, Mohindra N, Madden CJ, Diaz-Arrastia R. Diffusion tensor tractography of traumatic diffuse axonal injury. Arch Neurol 2008;65:619-626.

Wang JY, Abdi H, Bakhadirov K, Diaz-Arrastia R, Devous MD. A comprehensive reliability assessment of quantitative diffusion tensor tractography. Neuroimage 2012;60:1127-1138.

Wang S, Melhem ER. Amyotrophic lateral sclerosis and primary lateral sclerosis: the role of diffusion tensor imaging and other advanced MR-based techniques as objective upper motor neuron markers. Ann N Y Acad Sci 2005;1064:61-77.

Wardlaw JM, Statham PFX. How often is haemosiderin not visible on routine MRI following traumatic intracerebral haemorrhage? Neuroradiology 2000;42:81-84.

Warner MA, de la Plata CM, Spence J, Wang JY, Harper C, Moore C, Devous M, Diaz-Arrastia R Assessing Spatial Relationships between Axonal Integrity, Regional Brain Volumes, and Neuropsychological Outcomes after Traumatic Axonal Injury. J Neurotrauma 2010;27:2121-2130.

Whelan-Goodinson R, Ponsford J, Schönberger M. The association between psychiatric state and outcome following traumatic brain injury. J Rehabil Med 2008;40:850-857.

Wilde EA, McCauley SR, Hunter JV, Bigler ED, Chu Z, Wang ZJ, Hanten GR, Troyanskaya M, Yallampalli R, Li X, Chia J, Levin HS. Diffusion tensor imaging of acute mild traumatic brain injury in adolescents. Neurology 2008;70:948-955.

Wilson JT, Pettigrew LE, Teasdale GM. Structured interviews for the Glasgow Outcome Scale and the Extended Glasgow Outcome Scale: Guidelines for their use. J Neurotrauma 1998;15:573-585.

Wilson JTL, Wiedmann KD, Hadley DM, Condon B, Teasdale G, Brooks DN. Early and late magnetic resonance imaging and neuropsychological outcome after head injury. J Neurol Neurosurg Psychiatry 1988;51:391-396.

Wortzel HS, Kraus MF, Filley CM, Anderson A, Arciniegas DB. Diffusion Tensor Imaging in Mild Traumatic Brain Injury Litigation. J Am Acad Psychiatry Law 2011;39:511-523.

Xu J, Rasmussen Jr I, Lagopoulos J, Håberg A. Diffuse axonal injury in severe traumatic brain injury visualized using high-resolution diffusion tensor imaging. J Neurotrauma 2007;24:753-765.

Yasmin H, Nakata Y, Aoki S, Abe O, Sato N, Nemoto K, Arima K, Furuta N, Uno M, Hirai S, Masutani Y, Ohtomo K. Diffusion abnormalities of the uncinate fasciculus in Alzheimer's disease: diffusion tensor tract-specific analysis using a new method to measure the core of the tract. Neuroradiology 2008;50:293-299.

Zhan L, Mueller BA, Jahanshad N, Jin Y, Lenglet C, Yacoup E, Sapiro G, Ugurbil K, Harel N, Toga A, Lim K, Thompson PM. Magnetic Resonance Field Strength Effects on Diffusion Measures and Brain Connectivity Networks. Brain Connect 2013;30.