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Prédisposition génétique à la chronicité des symptômes post-commotionnels à la suite d'un traumatisme crânio-cérébral léger

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Prédisposition génétique à la chronicité des symptômes post-commotionnels à la suite d'un traumatisme crânio-cérébral léger

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Résumé

La prévalence des troubles du sommeil et de douleur chronique est élevée chez le patient ayant subi un traumatisme crânien cérébral léger (TCCL). L'interaction entre ces plaintes est suggérée chez les patients avec un TCCL mais son étiologie reste encore peu connue. Les résultats de recherche présentés dans le premier article de cette thèse suggèrent que les patients avec un TCCL qui souffrent de douleur ont une modification des ondes cérébrales durant leur sommeil, ce qui pourrait expliquer en partie comment les deux symptômes interagissent. De plus, la douleur, surtout si associée à des troubles de l'humeur, semble jouer un rôle majeur dans la persistance des symptômes post-commotionnels.

Le deuxième article de cette thèse décrit une exacerbation des symptômes post-commotionnels chez le patient ayant eu un TCCL et souffrant de douleur. La persistance ou l'apparition de la douleur chronique à long terme serait prédite par le polymorphisme val66met du gène brain-derived neurotrophic factor (BDNF).

Une étude subséquente, présentée dans le troisième article, nous a permis d'approfondir les bases génétiques et cellulaires du rôle du BDNF dans la persistance des symptômes post-commotionnels. Des polymorphismes fréquents dans le gène BDNF ont révélé des variantes liées au mauvais pronostic suite à un TCCL. De plus, l'analyse de cellules extraites de patients ayant subi un TCCL démontrent que l'expression de la protéine BDNF peut être modifiée chez le patient de génotype met66 et ayant subi un TCCL, lui conférant ainsi un rôle neuroprotecteur potentiel.

En résumé, nous avons tenté de démontrer dans cette thèse que la douleur suite à un TCCL joue un rôle important dans les perturbations du sommeil et dans la persistance des symptômes post-commotionnels. Une prédisposition génétique pourrait contribuer à expliquer le mauvais pronostic et la chronicité des symptômes post-commotionnels suite à un TCCL.

Mots-clés : Traumatisme crânien cérébral léger, sommeil, douleur, BDNF

Abstract

Mild traumatic brain injury (MTBI) is a major public health concern as patients are left, amongst other symptoms, with sleep complaints and chronic pain. An interaction between these symptoms is suggested. For instance, a night of poor sleep is usually followed by hypersensitivity to pain and chronic pain always leads to sleep complaints. This interaction is suggested following an MTBI, however, data sustaining that hypothesis are still lacking. Data from the first article suggest that pain and other post-concussion symptoms are correlated with sleep-wake disturbances post-MTBI. MTBI patients with pain have more rapid electroencephalographic (EEG) waves during sleep than those without pain. This may suggest that there is an intrinsic physiological relationship between the two complaints.

Moreover, pain seems to play an important role in the persistence of post-concussive symptoms. The second article of this thesis describes and details the exacerbation of post-concussive symptoms in the presence of pain following MTBI. The val66met polymorphism in the Brain-derived neurotrophic factor (BDNF) gene is an important predisposing factor for chronic pain.

Lastly, a subsequent study, presented in the third article details the genetic and cellular basis of the role of BDNF in the persistence of post-concussive symptoms. Common polymorphisms in the BDNF genes were genotyped and revealed variants related to post-concussive symptoms following MTBI. Moreover, protein expression studies in

lymphoblast cells of MTBI patients showed a modified expression of BDNF with the met genotype that might be neuroprotective.

In summary, this thesis first shows that pain contributes to sleep-wake disturbances following MTBI and that the chronicity of post-concussive symptoms, including chronic pain, may be dependent on polymorphisms in the BDNF gene.

Key words : Mild traumatic brain injury, sleep, pain, brain-derived neurotrophic factor (BDNF)

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Liste des abréviations

AASM : American Academy of Sleep Medicine

ACE : Angiotensin-converting enzyme

ADN :Acide désoxyribonucléique

ANOVA : Analysis of variance

ApoE: Apolipoprotéine E

ARN : Acide ribonucléique

BAI: Beck Anxiety Inventory

BDI: Beck Depression Inventory

BDNF : Brain-derived neurotrophic factor

CDC: Centers for disease control and prevention

COMT: Catechol O-methyl transferase

DRD2: Dopamine receptor D2

ECG : Électrocardiographie

EEG : Électroencéphalographie

EMG: Électromyographie

GCS: Glasgow Coma Scale

IASP : International Association for the Study of Pain

IES-R: Impact of Event Scale - Revised

Il: Interleukines

MAF : Minor allelic frequency

MIDAS: Migraine disability assessment

MTBI: Mild traumatic brain injury

NEFH: Neurofilament, heavy polypeptide

NGB: Neuroglobin

NMDA: N-methyl-D-aspartate

PARP-1: Poly[ADP ribose] polymerase 1

PBS: Phosphate buffer saline

PCS: Pain catastrophizing scale

PSQI: Pittsburgh sleep quality index

PVT: Psychomotor vigilance task

QTL : Quantitative trait loci

QST : Quantitative sensory testing

REM: Rapid eye movement

RT: Reaction time

SD: Standard deviation

SNP : Single nucleotide polymorphism

TCCL : Traumatisme crânien cérébral léger

TNFα : Tumor necrosis factor α

VAS : Visual analog scale

À Sabrina, oui, maman te dédie son livre qui parle de la tête!

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À mes sœurs, je vais bientôt enlever le (c) après PhD et vous pourrez arrêter de m'appeler « could be PhD »!

Introduction

Dans le présent chapitre, un bref survol des caractéristiques épidémiologiques, cliniques et neurophysiologiques du traumatisme crânien cérébral léger sera présenté. La section suivante présentera les troubles du sommeil qui peuvent survenir suite à un traumatisme crânien, ainsi que la relation douleur-sommeil. Une attention spéciale sera dirigée vers les substrats psychophysiques et cellulaires de la douleur en général et suite à un traumatisme crânien en particulier. Ensuite, de nouveaux résultats de recherche sur les prédispositions génétiques qui confèrent la chronicité des symptômes post-commotionnels et de la douleur en particulier seront présentées. Finalement, ce chapitre sera clôturé par une exposition des hypothèses clefs de ce projet de recherche ainsi que des objectifs de la présente thèse.

Le traumatisme crânien léger

Incidence, prévalence, causes, définition

Dans les pays développés, le traumatisme crânien cérébral est la cause principale de décès et d'handicap chez les jeunes adultes [1-2]. À l'exception des infections et des accidents vasculaires cérébraux, le traumatisme crânien cérébral est la forme la plus commune d'atteinte acquise à la tête chez les enfants et les adultes aux États-Unis [2]. L'incidence annuelle des traumatismes crâniens cérébraux est estimée à 400 pour 100 000 de population [3]. Le fardeau économique est donc considérable et est estimé à 60 milliards USD$ par année aux États-Unis seulement [4]. La gestion en soins aigus du patient ayant

subi un traumatisme crânien est si importante pour le clinicien qu'un sondage effectué au Canada et aux États-Unis a montré que la recherche dans ce domaine se doit d'être une priorité [5].

Le Center for Disease Control and Prevention (CDC) a été mandaté par le gouvernement Américain pour étudier la prévalence des traumatismes crâniens, voyant l'importance des effets à long terme de ce fléau. Le rapport 2002-2006 a déterminé que 1.7 millions souffrent d'un traumatisme crânien chaque année, dont 75% sont des traumatismes crâniens cérébraux légers (TCCL) [6]. Même si le TCCL n'est pas mortel, il incombe au patient de nombreuses séquelles physiques et psychologiques pouvant s'étendre sur plusieurs années; d'où sa nomination d'épidémie silencieuse [7]. Les principales causes d'un TCC sont les chutes (35%), les accidents de la route (17%), les altercations et coups lors d'activités sportives (16%) et les bagarres (10%). La majorité des individus atteints sont les hommes entre 25 et 34 ans, ou les enfants de moins de 4 ans [6]. La signature de la guerre actuelle en Irak et en Afghanistan est le TCCL dû à des explosions. Près de 22% des vétérans de cette guerre ont un diagnostic de TCC, en comparaison aux guerres précédentes. Même si la mortalité a baissé grâce à des casques de guerre plus robustes, une majorité des soldats subissent un TCC à tête fermée ou l'équivalent d'un TCC léger [8].

Le concept des traumatismes crâniens a beaucoup évolué dans les dernières décennies. Cependant, la communauté scientifique peine à s'entendre sur le diagnostic et il reste encore beaucoup de travail pour démêler les différentes définitions. Le spectre de sévérité d'un coup à la tête causé par une force mécanique externe varie d'un type

« whiplash » à un coma, ou état végétatif. Le « whiplash » ne fait généralement pas partie des TCC car il n'y a pas évidence d'atteinte cérébrale. Le gradient de sévérité des TCC commence par les commotions cérébrales qui ont souvent une origine sportive et qui sont des TCC légers.

Il existe plusieurs façons de classifier la sévérité des traumatismes crâniens cérébraux.

La première classification est basée sur la sévérité de la blessure et est déterminée par un Score de Coma de Glasgow (GCS). Les traumatismes crâniens sont légers (GCS=13-15), modérés (GCS=12-9) ou sévères (GCS<8). L'échelle de Glasgow se base sur une observation de trois critères : l'ouverture des yeux, la réponse motrice et celle verbale. Le score produit par chaque catégorie informe sur l'état de conscience du patient. Ce score est le plus largement utilisé dans le milieu hospitalier et s'avère très utile dans les cas de traumatismes crâniens modérés à sévères. Il est vrai que dans le cas d'un TCC léger, ce score n'est pas précis, car même s'il y a atteinte cérébrale, la conscience n'est altérée que quelques minutes en général. Il existe d'autres indices de sévérité de traumatismes, dont le Full Outline of UnResponsiveness score (FOUR), le score du CT scan, la mortalité hospitalière, et le Injury Severity Index (ISI). Cependant, même ces scores ne sont pas adaptés pour la sévérité d'un traumatisme crânien léger. Le WHO a émis des directives se basant sur le score de Coma de Glasgow pour diagnostiquer un traumatisme crânien léger, pour un score entre 13 et 15. Tout au long de cette thèse, la classification retenue sera celle

utilisant le Score de Glasgow et portera plus spécifiquement sur les traumatismes crâniens cérébraux légers (GCS 13-15).

La seconde classification est basée sur l'intégrité de la dure-mère, en référence à la nature du traumatisme, par exemple, à crâne ouvert (blessure pénétrante), à crâne fermé ou bien blessure due à une explosion. On peut aussi classifier les traumatismes crâniens selon l'ampleur de la blessure, soit focale ou diffuse [9-11].

Les définitions d'un TCCL dans la littérature sont diverses et variées. En 2004, l'Organisation Mondiale de la Santé a mandaté un groupe de travail en neurotraumatologie pour arriver à un consensus de définition du TCCL : Le TCCL est une lésion cérébrale aigüe résultant en un transfert mécanique de l'énergie d'une force physique externe vers la tête [12]. Récemment, cette définition a été réaffirmée lors d'un consensus pour être lu : « Le traumatisme crânien léger, aussi appelé commotion cérébrale, est défini comme une altération des fonctions cérébrales, ou bien par la présence d'une pathologie du cerveau causée par une force externe » [13].

Une définition opérationnelle a été proposée pour faciliter la comparaison entre les études ainsi que la pose d'un diagnostic clinique précis . D'abord, au moins un symptôme entre confusion et désorientation, une perte de conscience pendant 30 minutes ou moins, une amnésie post-traumatique de moins de 24 heures, et/ou d'autres anomalies transitoires neurologiques. Ensuite, un score de Glasgow entre 13-15 mesuré trente minutes après l'accident [12]. Ces critères sont utilisés pour le recrutement de la cohorte de patients participant dans les études présentés dans le cadre de cette thèse.

Neuropathologie suite à un TCCL

L'électroencéphalographie (EEG) fut le premier outil diagnostic à fournir des évidences de fonctions cérébrales atteintes lors d'un TCCL [14-15]. Cependant, ces évidences sont difficiles à identifier par imagerie ou bien par examen clinique [16]. Lorsqu'une force externe atteint et blesse le cerveau, une altération du flux sanguin ainsi que de la pression intracrânienne causent des dommages aux tissus, appelés dommages primaires. Ces manifestations sont primaires car elles surviennent dans les minutes et les heures suivant le trauma [10]. Les dommages peuvent êtres classés en deux catégories : focaux (au lieu de l'impact) et diffus. Les dommages focaux incluent la présence d'hématomes, de contusions ainsi que de l'enflure. Les blessures diffuses sont causées par la force d'accélération/décélération qui déchire et étire les axones causant des lésions de la substance blanche qui est d'intérêt dans la neurophysiologie d'un TCCL. Une lésion étendue des fibres de matières blanches suivant un traumatisme crânien cérébral est communément appelée une lésion axonale diffuse.

Dans une phase subséquente, des dommages secondaires surviennent. Ces dommages enclenchent des cascades neurochimiques complexes qui sont principalement des blessures axonales et gliales. La figure 1 est tirée d'un article qui décrit la cascade neurochimique[17]. Les cellules agissent principalement par six phénomènes : 1) libération excessive d'acides aminés excitateurs 2) genèse de radicaux libres 3) libération de cytokines inflammatoires 4) protéolyse de la calpaïne 5) extension axonale et, 6) par apoptose [18].

Hypothèse glutamatergique : Il a longtemps été suggéré que le glutamate serait la cause principale de neurotoxicité aigue et chronique suite à un TCCL [19]. Le mécanisme par lequel la mort cellulaire survient est une augmentation de la dépolarisation induite par le glutamate via l'activation des récepteurs (N-methyl-D-aspartate) NMDA, augmentant ainsi la présence de calcium intracellulaire [18]. De plus, un déferlement de sodium et d'eau suit causant ainsi l'engorgement des neurones. Comme la libération du glutamate est excessive, les cellules environnantes subissent aussi des dommages importants [20].

Hypothèse des radicaux libres: Le stress oxydatif active des mécanismes d'inflammation par l'entremise des neutrophiles, la peroxydation des lipides ainsi que la diminution de glutathione [21]. La peroxydation des lipides survient suite au TCC et engendre la production de radicaux libres [22-24].

Les cytokines: L'inflammation est omni-présente lors de blessures cérébrales. Les interleukines et le Tumor Necrosis Factor alpha (TNFα), qui sont des cytokines inflammatoires connues, augmentent drastiquement suite à un TCCL [25]. Cette augmentation exacerbe la libération de radicaux libres et augmente la perméabilité de la barrière hémato-encéphalique, causant ainsi une inflammation [18].

Protéolyse de Calpaïne: La Calpaïne est une cystéine protéase lysosomale calcium-dépendante. L'augmentation intracellulaire de calcium active les protéases, en particulier, la Calpaïne. La Calpaïne joue un rôle important dans le clivage du cytosquelette causant ainsi la mort neuronale [26].

L'étirement axonal: cet étirement représente une déformation mécanique post-TCC avec un élargissement des astrocytes, une perméabilité membranaire, une activation de la Calpaïne ainsi qu'un influx de calcium intracellulaire [18].

L'apoptose: Plusieurs évidences montrent que le TCC cause la mort neuronale, surtout dans l'hippocampe [27-28]. Le mécanisme par lequel cette mort neuronale survient est l'apoptose [18].

La réponse cellulaire suite à un TCCL est aussi caractérisée par une réactivité des astrocytes, une activation de la microglie ainsi qu'un arrêt de la prolifération cellulaire au site de la blessure. Ces mécanismes cellulaires sont mis en branle pour faciliter la récupération des cellules endommagées et assurer la protection des neurones environnants [29].
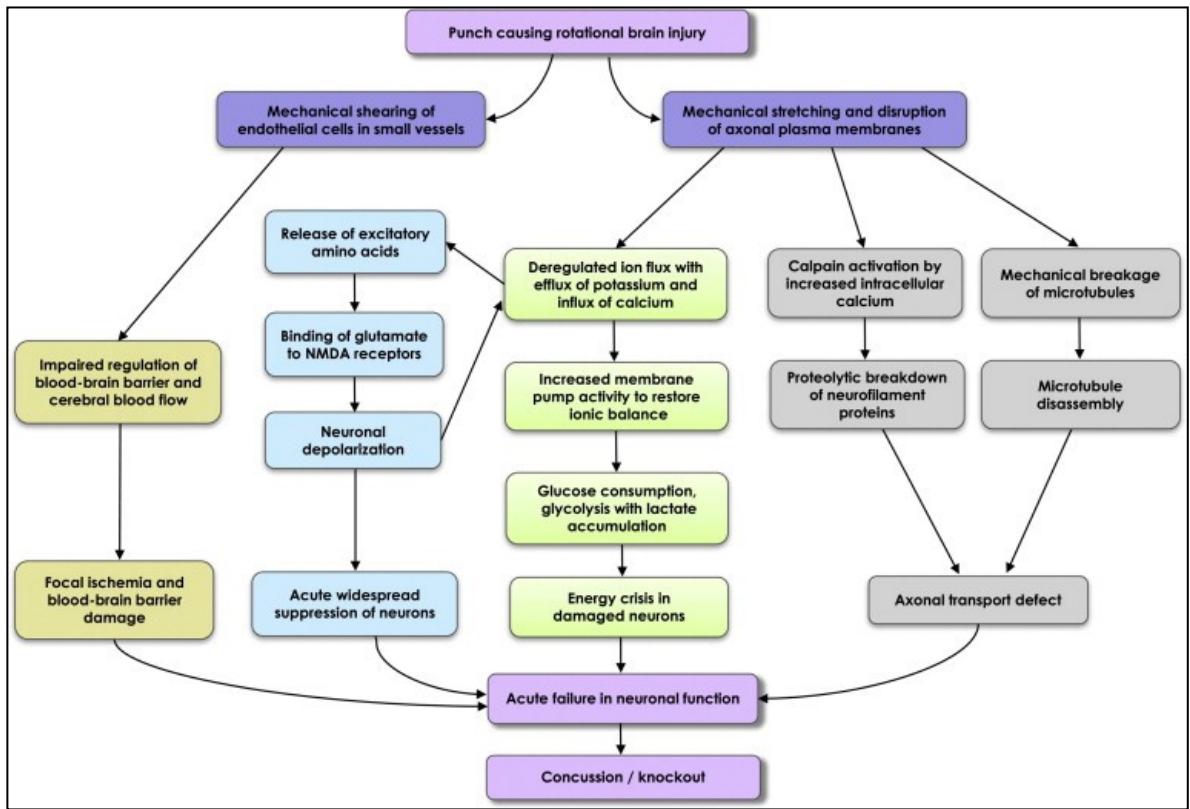
Figure 1: Illustration de quelques exemples de cascades neurochimiques qui surviennent suite à un TCCL [17]

Avec la connaissance des mécanismes énumérés ci-haut, la recherche se dirige vers des hypothèses de neuroprotection, entre autre comment les antagonistes aux récepteurs NMDA pourraient interrompre la neurotoxicité et limiter ainsi les dommages post-TCC. Malheureusement, les essais cliniques pour contrer la neurotoxicité et la présence de radicaux libres ne sont pas concluants [18,30-31].

Le mécanisme de récupération des cellules suite à un TCCL, lui aussi reste encore nébuleux. Dans cette thèse, nous émettrons une nouvelle hypothèse citant que la récupération est dépendante des facteurs de croissance.

Les manifestations primaires provoquent des symptômes précoces (minutes et heures suivant le TCCL) dont les plaintes liées aux céphalées, étourdissements, nausées, vomissements et perte de conscience. Les dommages secondaires quant à eux, sont responsables de l'apparition des symptômes post-commotionnels (jours à semaines suivant le TCCL) qui incluent céphalées, troubles de l'attention et de la mémoire, irritabilité, douleur et troubles de sommeil [32].

Les symptômes post-commotionnels :

Les symptômes post-commotionnels englobent des plaintes cognitives, émotionnelles et somatiques qui surviennent suite à un TCCL. Le DSM-IV propose le diagnostic de symptômes post-commotionnels si au moins trois symptômes sont rapportés durant les trois premiers mois [33]. Le questionnaire du Rivermead Post Concussion est le plus largement utilisé pour le diagnostic [34]. Suite à un TCCL, la majorité des patients retourneront à un état de santé similaire à leur état pré-trauma. Cependant, de 10-20% se plaindront de symptômes post-commotionnels qui persisteront durant la phase chronique, soit plus de trois mois. Plusieurs études se sont attardées sur les raisons qui peuvent expliquer la chronicité des symptômes avec différentes conclusions. Par exemple, une collision par un véhicule automobile ainsi qu'une compensation monétaire seraient des facteurs de mauvais pronostic. L'âge ainsi que la sévérité initiale du traumatisme seraient aussi des facteurs importants [35-38]. Avec l'augmentation de la prévalence du TCCL au

sein de la population militaire, le syndrome de stress post-traumatique vient s'ajouter à la liste des symptômes post-commotionnels [39]. Ces symptômes peuvent affecter la qualité de vie ainsi que l'état de santé générale même dix ans après le traumatisme [40]. D'autres symptômes post-commotionnels incluent les troubles de l'humeur. Les patients ayant subi un TCC sont à risque de développer de la dépression ainsi que de l'anxiété [41]. Plusieurs facteurs pré-trauma peuvent expliquer la prédisposition à développer des troubles de l'humeur, par exemple, une histoire de dépression, la nature de l'accident ainsi qu'une prédisposition génétique [42-43].

Le sommeil suite à un TCCL

Le sommeil est un état physiologique réversible d'altération de la vigilance. Le sommeil est hétérogène de nature et est constitué de périodes caractérisées par des patrons d'activité cérébrale, de mouvements oculaires et de tonus musculaires. Ces périodes alternent entre le sommeil lent (constitué de plusieurs phases) et le sommeil paradoxal [44]. Le sommeil lent est divisé en trois stades : le stade 1, le stade 2 et le stade 3/4 de sommeil lent profond. L'activité cérébrale, telle que détectée par électroencéphalographie (EEG), caractérise le sommeil grâce à l'apparition graduelle d'ondes lentes et de haute amplitude durant le sommeil lent non-paradoxal. Le sommeil paradoxal est quant à lui caractérisé par

des ondes rapides similaires à l'état d'éveil mais accompagné d'atonie à l'électromyogramme (EMG).

De nombreuses études ont montré une prévalence élevée de troubles du sommeil suite à un TCC [45-47]. Les troubles du sommeil suite à un traumatisme crânien peuvent varier, consistant en majorité de somnolence diurne excessive [48]. Les troubles du sommeil les plus communs post-TCCL sont l'hypersomnie, l'insomnie, le délai de phase et la narcolepsie. Ces troubles sont surtout présents en phase aigue, soit les trois premiers mois. En phase chronique, soit de 3 mois à 2 ans post-trauma, la somnolence diurne reste présente dans 50% des cas, l'insomnie et les parasomnies chez 25% respectivement [49]. L'étude de ces troubles de sommeil post-TCC est d'une importance primordiale car ils peuvent engendrer des déficits cognitifs.

L'insomnie est définie par une difficulté répétée à initier, maintenir ou consolider le sommeil et qui résulte en une inhabilité à fonctionner durant la journée [50]. Une revue de 21 études portant sur l'insomnie suite à un TCC a montré que l'insomnie était présente dans plus de 40% des cas pour toutes sévérités de TCC et durant les phases aigues et chroniques [51].

L'étiologie de l'hypersomnie post-traumatique n'est pas encore claire. Tous s'entendent pour affirmer que c'est le traumatisme qui en est le déclencheur, mais le rôle joué par la présence de parasomnies qui peuvent aussi perturber le sommeil reste à déterminer. De plus, il ne faut pas confondre l'hypersomnie et la fatigue. Cette dernière étant aussi très fréquente suite à un TCCL. Pour pouvoir faire cette distinction, des tests

objectifs existent pour mesurer l'ampleur de la somnolence diurne, tel le Multiple Sleep Latency Test et le Multiple Wakefulness Test [52]. Le rapport subjectif d'un patient peut aussi être évalué grâce au questionnaire d'Epworth Sleepiness Scale [53].

Les résultats des études concernant les ondes cérébrales durant le sommeil (analyse de l'EEG) n'ont pas été concluants [54-56]. Même si la sévérité du traumatisme n'interfère pas dans les données d'architecture du sommeil, à l'exception d'une diminution du stade 1 et d'éveils durant le sommeil ainsi qu'une augmentation de l'efficacité du sommeil chez les TCC légers en comparaison aux TCC modérés-sévères en phase chronique, le reste des études n'arrivent pas à une conclusion unanime concernant les changements de l'architecture du sommeil ou bien de l'EEG [49]. Le tableau 1 présente un résumé des résultats d'études sur le sommeil.

Tableau 1 - Les principaux troubles du sommeil et caractéristiques de l'architecture et de l'EEG

a) Les principaux troubles du sommeil

TCC légers		TCC modérés/sévères	
Phase Aigue	**Phase Chronique**	**Phase Aigue**	**Phase Chronique**
Insomnie[49,51]*			
		Somnolence excessive diurne[49,51,57]	
	Parasomnies** [49,58]		Parasomnies** [49,58]
Trouble du rythme circadien***[59]			

*La référence 51 représente une analyse de 21 études sur l'insomnie.

**Les parasomnies incluent : Paralysie du sommeil, somnambulisme, cataplexie, terreurs nocturnes, trouble comportemental en sommeil paradoxal, l'énurésie nocturne, trouble du comportement alimentaire lié au sommeil, narcolepsie sans cataplexie.
***Les troubles du rythme circadien incluent : délai de phase et cycle irrégulier d'éveil-sommeil.

b) Les caractéristiques de l'architecture du sommeil et de l'EEG

Études	Nombre de sujets	Période post-trauma	Nombre de nuits	Résultats
Parsons et al.[54]	8 adolescents	72h 6 semaines 12 semaines	6	-Pas de différence en macrostructure -↓delta, thêta et alpha1 avec le temps.
Kaufman et al.[60]	19 adolescents	3 ans	1	-↓efficacité de sommeil -↑éveils durant le sommeil
Ouellet et al. [61]	14 adultes	21 mois	2	-↑stade 1
Verma et al.[49]	54 adultes	3 mois à 2 ans	1	-↑ efficacité du sommeil -↓ stade 1 -↓éveils durant le sommeil
Schreiber et al.[62]	26 adultes	12 mois à 21 ans	2	-↑stade 2 -↓ sommeil paradoxal
Williams et al.[63]	9 adultes	28 mois	3	-↓efficacité de sommeil -↓latence au sommeil paradoxal
Gosselin et al.[55]	10 athlètes	1 an	2	- pas de différence en macrostructure - pas de différence spectrale
Rao et al.[56]	7 adultes	1 semaine	2	- pas de différence en macrostructure

Tableau tiré et modifié de l'article en annexe: Perturbation du sommeil par la douleur chez les traumatisés crâniens légers.

La douleur suite à un TCCL

La douleur est définie comme étant une expérience sensorielle et émotionnelle désagréable associée à une lésion tissulaire réelle ou potentielle [64]. Cette définition générale, établie par un consensus de l'Association Internationale pour l'Étude de la Douleur (IASP), englobe différentes composantes de l'affection douloureuse, soit émotionnelle, sensorielle et physiologique. Une nouvelle mise à jour de la classification de la douleur a été adoptée en 2012 par l'IASP. Cette classification place les céphalées post-traumatiques dans la catégorie «relatively localized syndromes of the head and neck », soit une douleur continue et diffuse de la tête suite à un TCC accompagnée de changements de personnalité, d'irritabilité, de baisse de concentration, d'étourdissement, de troubles visuels, de baisse de la tolérance à l'alcool, dépression, avec ou sans syndrome de stress post-traumatique [65].

Selon un article de revue récent, 75 % des patients ayant subi un TCCL souffrent de douleur chronique et, de façon surprenante, les patients avec TCC léger en phase chronique, rapportent plus de douleur que les sujets ayant subi un TCC sévère [66]. Les céphalées post-traumatiques seraient la douleur la plus fréquente et représenteraient près de 60% des symptômes post-commotionnels rapportés [67]. Le facteur principal associé à un plus haut risque de présenter des douleurs chroniques suivant un TCCL est la présence de céphalées avant le traumatisme [68-69]. Il a été démontré que les patients ayant subi un TCCL et présentant un syndrome de stress post-traumatique ainsi que des symptômes associés à la dépression sont plus à risque de rapporter de la douleur [70-71].

La chronicité de la douleur ne semble pas être spécifique au TCCL, elle est aussi un fardeau chez les blessés orthopédiques sans atteintes cérébrales [72]. Lors de protocoles expérimentaux sur la détection de modalités sensorielles, aussi connu sous le nom « quantitative sensory testing (QST) », aucune différence significative dans la détection du froid et de la douleur (au froid ou au chaud) n'a été observée chez les patients TCCL, toutefois, le seuil de détection de la chaleur est sensiblement plus élevé chez les TCCL qui souffrent de céphalées et de syndromes de stress post-traumatique par rapport aux sujets sains [73-75]. Ces résultats ont aussi été répliqués par notre groupe [74]. Le QST étant spécifique au système nerveux périphérique, on pourrait se questionner sur l'étiologie centrale de la douleur chez les TCCL [76-77].

Relation douleur-sommeil

Le sommeil, tel que décrit plus haut, est un état où le système nerveux central se caractérise par une diminution de la sensibilité aux stimuli externes, à l'opposé de la douleur qui se définie par un état d'hypervigilance. L'interaction entre ces deux états peut engendrer, chez certains sujets, une relation bidirectionnelle : Un mauvais sommeil crée une hypersensibilité à la douleur et la douleur perturbe le sommeil. Des maladies du sommeil ont aussi été associées à de la douleur dans la population générale. Par exemple, une association entre le ronflement et les céphalées matinales a été rapportée [78]. Dans une étude expérimentale sur la douleur durant le sommeil de sujets sains, il a été montré qu'une douleur musculaire et articulaire, une diminution des ondes lentes et une

augmentation des ondes rapides surviennent [79]. Une autre étude a montré qu'un éveil cortical peut être induit par une douleur expérimentale [80]. Toutefois, ces résultats ne sont pas consistants dans la littérature car ils dépendent de la population étudiée, et de la présence de fibromyalgie ou autre type de douleur [81]. De plus, il y a une controverse quant à la présence d'intrusion d'ondes alpha dans le sommeil lent, qui ne semble pas être spécifique à la douleur [82].

Chez les patients ayant subi un TCCL, l'effet de la douleur sur le sommeil a été suggéré dans des études précédentes. D'abord, une première étude a montré à l'aide de questionnaires, que les patients ayant subi un TCCL et souffrant de douleur rapportent deux fois plus d'insomnie que ceux qui n'en souffrent pas [83]. Notre laboratoire a répliqué les résultats de cette étude en montrant que les céphalées, les plaintes de sommeil ainsi que la dépression sont associés [84]. Ces deux études étant basées sur des questionnaires et sur une étude rétrospective (dans le deuxième cas), elles ne se sont pas attardées sur la présence de modifications physiologiques à l'EEG. Cette affirmation a été la base de l'hypothèse de la première étude présentée dans cette thèse.

La prédisposition génétique

La génétique a longtemps été l'étude des associations d'un gène à la susceptibilité de développer une maladie ou en exprimer certains symptômes. Récemment, l'utilité de la génétique a été aussi de comprendre la guérison ou le pronostic suite à une condition acquise, par exemple, le TCCL. Suite à un TCCL, la majorité des patients retournent à l'état pré-trauma sans séquelles; cependant, un faible pourcentage restera avec des plaintes de symptômes post-commotionnels. La suggestion qu'il existe donc une susceptibilité génétique à chroniciser chez cette population a été soulevée.

Plusieurs gènes candidats ont été recensés (Tableau 2). D'abord, l'apolipoprotéine E (ApoE) a été largement étudiée dans la population TCC (incluant léger, modéré et sévère) et les porteurs de l'allèle Ɛ4 sont les plus à risque d'avoir un mauvais pronostic [85-87]. Le rôle de l'ApoE dans le système nerveux central est un facilitateur de transport de lipides [88]. Dans un modèle de traumatisme cérébral, le rôle suggéré de l'ApoE est l'aide à la réparation cellulaire [89]. À partir de cette découverte, plusieurs gènes candidats qui jouent un rôle dans la réparation cellulaire, l'inflammation, la neurotoxicité ou bien la neuroprotection ont été étudiés [90]. Pour en nommer quelques-uns, les gènes de la famille des neurotrophines et des interleukines s'avèrent être importants.

Tel que décrit dans le tableau 2, la famille des interleukines a été étudiée en relation avec le TCC, cependant, les polymorphismes des gènes n'ont pas pu être associés, hors de tout doute, à un mauvais pronostic [91-92]

La famille des neurotrophines inclue quatre catégories : 1) les « nerve growth factors », 2) les « glial cell-derived growth factors », 3) les « neurokines » et 4) les « non-neuronal growth factors ». Même si la source de ces facteurs de croissance diffère, tous jouent un rôle dans la prolifération des cellules ainsi que dans la plasticité synaptique [93-94]. L'hypothèse de base pour justifier l'étude des neurotrophines suite à un TCC est que les individus ayant une meilleure capacité de rétablissement suite à un TCC portent aussi les allèles convenables à une réparation cellulaire optimale [95]. La neurotrophine la plus étudiée est le « brain derived neurotrophic factor » (BDNF) car elle est la seule à être sécrétée suite à une activation neuronale [96]. Le BDNF est une protéine dont le précurseur est le pro-BDNF. Une fois formé, le BDNF est entreposé dans des vésicules prêtes à être libérées suite à une activation neuronale [93]. Un polymorphisme du gène BDNF du codon 66 situé dans le promoteur du pro-BDNF a été largement étudié. Ce polymorphisme résulte en un changement d'acide aminé d'une valine à une méthionine [97]. Ce polymorphisme semble important dans la cognition ainsi que les fonctions exécutives suite à un TCCL [98-99]. En contrepartie, il existe des études chez le rat qui confèrent un rôle antinociceptif au BDNF [100-101]

Les arguments cités ci-haut placent le BDNF comme étant un gène d'intérêt pour des études plus poussées de facteurs génétiques prédisposant à la chronicité des symptômes post-commotionnels, notamment la douleur, suite à un TCCL. Le BDNF sera donc l'objet d'étude de l'article 3 présenté dans cette thèse

Tableau 2 – Recensement des gènes importants chez les TCC

<u>Gène</u>	<u>Références</u>	<u>Population*</u>	<u>Allèle/SNP</u>	<u>Résultat</u>
ApoE	[87]	77 TCCM/S	Ɛ4	↓performances neuropsychologiques
	[85]	89 TCCL/M/S	Ɛ4	Pronostic clinique défavorable
	[102]	118 TCCL/M	Ɛ4	↑anormale des ondes lentes à l'EEG
	[103]	90 TCCL/M	Ɛ4	Pas de différence
ApoE promoteur	[104]	195 TCCL	G219T	Génotype TT : ↑ risque de rapporter histoire de TCCL
	[105]	196 TCCL	G219T	Génotype TT et Ɛ4 : ↑ risque de rapporter histoire de TCCL
PARP-1	[106]	191 TCCS	rs3219119	Génotype AA : pronostic clinique favorable
Il-6	[92]	62 TCCS	G174C	Pas de différence
Il-1a	[91]	71 TCCL/M/S	IL1A*2	Pas de différence
Il-1b	[107]	69 TCCL/M/S	B2	Allèle 2 : Mauvais pronostic
ACE	[108]	73 TCCM/S	I/D	Allèle D : ↓Performance neuropsychologiques
COMT	[109]	113 TCCL/M/S	Val158Met	Homozygotes performent mieux aux tests cognitifs.
p53	[110]	90 TCCS	Arg72Pro	Arg/Arg : Mauvais pronostic
BDNF	[111]	53 TCCS	Val66Met	Pas d'effet sur récupération suite à état végétatif
	[112]	109 TCC **	rs7124442 rs1519480	↑Récupération de l'intelligence
	[98]	75 TCCL	rs6265	↓ traitement cognitifs
DRD2	[113]	93 TCCL	TAQ1 A	Allèle T : ↓performance cognitive
NEFH	[114]	84 TCCL	rs165602	Pas de différence

*TCCL/M/S : Traumatisme crânien cérébral Léger / Modéré / Sévère

** Sévérité non-spécifiée

Hypothèses et objectifs de la thèse

Les trois articles présentés dans cette thèse tentent d'apporter une réponse sur les facteurs pouvant expliquer, en partie, la chronicité des symptômes, surtout la douleur, chez les victimes de traumatisme crânien cérébral léger (TCCL). Il est essentiel de conserver en mémoire que cette thèse ne vise pas les liens de causalité. La conception d'un projet prospectif a été choisi afin d'identifier des bio-marqueurs et des facteurs en lien avec les plaintes. Une étude utilisant un protocole expérimental avec des approches thérapeutiques pour renverser la douleur et les conséquences sur le sommeil reste à faire afin de démontrer la robustesse de nos hypothèses.

Les objectifs

L'objectif principal de la présente thèse est d'identifier l'effet de la douleur sur le sommeil ainsi que les facteurs qui prédisposeraient les patients suite à un TCCL à souffrir de douleur chronique et de symptômes post-commotionnels.

De façon plus spécifique, **les objectifs par articles** sont :

Dans l'article premier, une description, par une étude polysomnographique, de l'influence et de l'importance de la douleur sur les plaintes subjectives de sommeil suite à un TCCL. Dans la seconde étude, on vise à s'attarder sur les caractéristiques de la douleur en phase aigue ainsi que les facteurs qui prédisposeraient à la chronicité de la douleur en phase chronique tout en investiguant le rôle de facteurs neurotrophiques potentiels.

Enfin, la troisième étude se veut une caractérisation génétique et cellulaire du BDNF, le facteur neurotrophique qui a été identifié dans l'article 2 parmi une population TCCL.

Les hypothèses

- Il existe une signature électroencéphalographique qui expliquerait les plaintes de mauvais sommeil.

- Les plaintes de sommeil de moindre qualité sont dues à des changements fins de l'activité cérébrale exacerbés par la douleur.

- La douleur chronique chez les TCCL est expliquée par la présence de troubles de l'humeur concomitants.

- Un ou des marqueurs génétiques seraient en lien avec les troubles de douleur et de sommeil chez les TCCL.

Articles de thèse

Article 1: Rapid EEG activity during sleep dominates in mild traumatic brain injury patients with acute pain

En révision dans le journal : Journal of Neurotrauma

Article 2: BDNF polymorphism predicts the transition to chronic pain following mild traumatic brain injury

Manuscrit tenu confidentiel due au contenu en cours de brevetage

Article 3: Mutations in BDNF gene affect recovery following mild traumatic brain injury
Article en préparation

Article 1: Rapid EEG activity during sleep dominates in mild traumatic brain injury patients with acute pain

Rapid EEG activity during sleep dominates in mild traumatic brain injury patients with acute pain

Running title: Sleep and pain in mild brain injury

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Abstract

Chronic pain is a highly prevalent post-concussion symptom occurring in a majority of mild traumatic brain injury (mTBI) patients. About half of mTBI patients report sleep–wake disturbances. It is known that pain can alter sleep quality in this population, but the interaction between pain and sleep is not fully understood. This study aimed to identify how pain affects subjective sleep (Pittsburgh Sleep Quality Index PSQI), sleep architecture, and quantitative electroencephalographic (qEEG) brain activity following mTBI.

Twenty-four mTBI patients complaining of sleep–wake disturbances, with and without pain (8 and 16, respectively), were prospectively recruited 45 ($\pm$22.7) days post-trauma on average. Data were compared with those of 18 healthy controls (no sleep or pain complaints). The PSQI, sleep architecture, and qEEG activity were analyzed. Pain was assessed using questionnaires and a 100-mm Visual Analogue Scale (VAS).

mTBI patients reported three times poorer sleep quality than controls on the PSQI. Sleep architecture significantly differed between mTBI patients and controls, but was within normal range. Global qEEG showed lower delta (deep sleep) and higher beta and gamma power (arousal) at certain EEG derivations in mTBI patients compared to controls ($p<0.04$). However, mTBI patients with pain showed greater increase in rapid EEG frequency bands, mostly during REM sleep, and beta bands in non-REM sleep compared to mTBI patients without pain and controls ($p<0.001$).

Pain in mTBI patients was associated with more rapid qEEG activity, mostly during REM sleep, suggesting that pain is associated with poor sleep and is a critical factor in managing post-concussion symptoms.

<u>Introduction</u>

According to the Center for Disease Control (CDC), 1.7 million people sustain a traumatic brain injury annually in the United States, primarily a mild traumatic brain injury (mTBI) [6]. The mTBI incidence is rising worldwide due to greater access to motor vehicles in developing countries and ongoing wars [115]. mTBI has been described as the silent epidemic, as many patients are left with, among other symptoms, sleep–wake disturbances, chronic pain and mood disorders [48-49,66].

Sleep–wake disturbances are common consequences of traumatic brain injury. For example, excessive daytime sleepiness was found in patients with mild TBI [116-117]. High prevalence (72%) of sleep–wake disturbances was found in TBI patients during the first six months [58]. After three years, 67% of patients still reported sleep–wake disturbances (mainly hypersomnia and insomnia) [118]. Whereas the presence of sleep disorders did not correlate with trauma severity, conflicting results were found for depression and anxiety [47,49]. Sleep disorders reported in the mTBI population are referred to as sleep and wake disturbances because the major complaint is post-traumatic hypersomnia and the second major complaint is insomnia [46-48,119-120]. The etiology of sleep disorders in mTBI remains poorly understood.

Polysomnographic recording of biophysical changes of sleep parameters that occur during sleep (including sleep stages, arousals during sleep) were performed in mTBI patients. Few mTBI studies have obtained conflicting results on sleep stage 1 duration (lighter sleep), sleep efficiency, or REM sleep duration, or finding no differences from healthy controls

[49,56,61,117]. Therefore, polysomnographic recording could not confirm subjective sleep complaints.

Studies on quantitative electroencephalogram (qEEG) power during sleep also found conflicting results, mainly due to whether sleep was recorded in the acute or chronic post-trauma phase. QEEG power is a useful tool to extend the analysis of the EEG and to decompose the signal into a voltage and frequency power spectrum. Thus, mTBI patients showed lower delta (slow waves, important for sleep homeostasis) and higher alpha and beta power (faster bands that represent arousal) during non-REM sleep than controls one week post-trauma [56]. A significant power reduction in low qEEG frequency bands (0.5–9.75 Hz) during non-REM sleep was also found in adolescents post-mTBI in the acute phase [54]. Conversely, no differences in sleep qEEG were reported in a general sample of mTBI patients or in athletes with mTBI compared to healthy subjects and control athletes [55,63]. Even with a technique able to detect subtle differences that polysomnography is unable to detect, no conclusion could be reached regarding changes that occur during sleep following mTBI.

At one year post-trauma, excessive daytime sleepiness complaint was equivalent in TBI patients and patients with trauma other than in the brain, suggesting that bodily pain plays an important role in sleep–wake disturbances [121].

Pain is defined as an unpleasant sensory and emotional experience associated with actual or potential tissue damage, or described in terms of such damage (International Association for the Study of Pain [64]). Pain is a highly prevalent post-concussion symptom in mTBI

patients, occurring in more than 75% of cases [66]. Acute post-traumatic pain presents under several forms, mostly musculoskeletal pain, widespread pain, post-traumatic headache, or vascular, neurogenic, visceral, and iatrogenic pain [122]. Mild TBI patients reported more pain than severe TBI patients did [66,83].

Up to 70% of patients with pain reported poor sleep quality or unrefreshing sleep [82]. Studies have indicated that the pathways that regulate sleep, arousal, and nociception overlap and interact [123] Therefore, pain is important in sleep studies due to the linear relationship between the two states. The literature has associated pain with sleep problems in mTBI, based on questionnaires and reports [83-84]. The presence of pain was associated with twice as many complaints of insomnia in mTBI [83]. mTBI patients presenting sleep complaints reported more headaches at six weeks post-trauma [84]. Pain as well as post-traumatic stress disorders were also shown to contribute significantly to sleep problems known as the "polytrauma clinical triad" [124]. The presence of pain was crucial when interpreting patients' physical, psychological, and cognitive complaints following TBI [125]. In fact, in clinical settings, comorbidities, including anxiety, depression and pain catastrophizing are becoming very frequently assessed in patients suffering of pain [126]. A large study that included more than 400 mTBI patients, revealed that patients presenting sleep-wake disturbances also suffer from concomitant depressive symptoms and irritability [84]. mTBI patients with pain reported more depressive and anxiety symptoms [127-128]. Pain catastrophizing was also related to the threatening nature of pain following injury or

illness [129]. However, to our knowledge, no studies have investigated sleep changes in the presence of pain in an mTBI population with an attention for other comorbidities.

Because the exact nature of this deleterious interaction remains unknown, it was relevant to investigate the relationship between sleep and pain in mTBI patients. Although pain has been widely cited as an important factor affecting sleep in mTBI, no studies to date have investigated the contribution of pain to sleep parameters in this population. Moreover, due to the important impact of comorbidities on sleep and pain, anxiety, depression and pain catastrophizing will be considered in this paper.

In order to characterize how pain affects sleep in mTBI patients, two primary objectives and one secondary objective were proposed. Primary objectives were: (1) to compare subjective sleep, sleep architecture, and quantitative EEG between patients with mTBI and controls; and (2) to compare subjective sleep, sleep architecture, and quantitative EEG between mTBI patients with pain, mTBI patients without pain, and controls; and the secondary objective was: (3) to explore the effects of depression, anxiety, and pain catastrophizing on the relationship between pain and sleep in mTBI patients.

We hypothesized that qEEG pattern variability during sleep would be associated with pain in mTBI

Materials and methods

   a) *Study Sample*

Patients were screened from about 300 mTBI visits per year to the trauma unit of a tertiary hospital centre. We recruited 29 patients six weeks following trauma. The mTBI diagnosis was confirmed by a trauma neurosurgeon (author JFG) according to the 2004 WHO Task Force on mTBI [12]. Inclusion criteria were 1) score of 13–15 on the Glasgow Coma Scale; 2) loss of consciousness and post-traumatic amnesia for <30 min; 3) age 18–60 years; and 4) self-reported sleep complaints. Participants were also asked to undergo two nights of sleep recordings in the laboratory.

Patients were excluded for 1) gross cognitive or speech dysfunctions; 2) use of psychotropic medication or other drugs known to influence sleep or motor behaviour; 3) presence of major neurological or psychiatric disorders or alcohol abuse; 4) history of chronic pain or fibromyalgia before mTBI; and 5) history of sleep disorders, including circadian disruption. Because five patients refused the second night of sleep assessment, the final sample comprised 24 patients.

Eight mTBI patients were classified as mTBI with pain based on the following criteria: 1) persistent pain since the accident at different body sites (head, neck, back, or jaw); 2) report of trouble sleeping due to pain on the Pittsburgh Sleep Quality Index (PSQI), item 5, three or more times a week; and 3) moderate to severe intensity pain upon awakening on a scale of 0–4 (0 = no pain, 4 = severe pain). None of these subjects had sustained a new injury that caused pain after the initial traumatic injury. Sixteen mTBI patients who did not meet these pain criteria were classified as mTBI without pain.

Eighteen healthy subjects free of pain and sleep complaints were recruited as controls. All patients and controls provided written consent to participate. The study protocol was approved by the ethics board of the Hôpital du Sacré-Coeur de Montréal.

b) <u>Subjective sleep assessment</u>

All participants completed the Pittsburgh Sleep Quality Index (PSQI), a self-rated questionnaire that assesses subjective sleep quality over the past month. A global PSQI score greater than 5 indicates sleep disturbance, as previously validated in TBI patients [130-131]. Sleep quality upon awakening was reported on a 100-mm visual analogue scale (VAS) (0 = worst sleep, 100 = best sleep).

c) *<u>Polysomnography</u>*

All mTBI patients and controls slept for two consecutive nights in a light- and sound-attenuated room at the hospital's sleep laboratory. The first night was used for habituation and the second night for data collection and analysis.

Recordings were performed using a 32-channel Grass polysomnograph with 0.1–100 Hz filter bandpass. The montage comprised 11 leads for EEG derivations placed according to the international 10–20 system (C3-A2; C4-A1; F3-A2; F4-A1; F7-A2; F8-A1; Fp1-A2; Fp2-A1; Fz-A1; O1-A2; O2-A1); bilateral electrooculograms; chin, masseter, and tibialis electromyograms; and three electrocardiogram (ECG) derivations. Thoracic and abdominal straps, a nasal airflow cannula, and a pulse oximeter were used to monitor respiration. Signals were digitalized at a 256 Hz and 512 Hz sampling rate for EEG and ECG, respectively.

Continuous audio and infrared video recordings were performed to detect abnormal movements and behaviours during sleep.
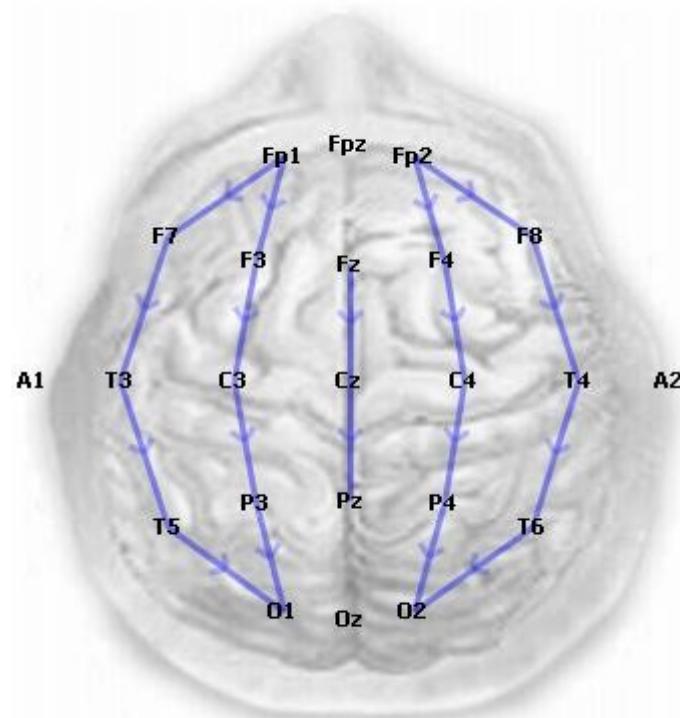
Sleep stages were visually scored off-line on 20-second epochs using Rechtschaffen and Kales' criteria [44]. Arousals, periodic leg movements, and respiratory disturbances were scored using standard criteria [50]. Sleep latency was determined as the time between bedtime and sleep stage 2, and persistent sleep latency as the time between bedtime and 10 consecutive minutes of uninterrupted sleep. REM sleep latency was defined as the time between bedtime and the first stage of REM sleep. Sleep duration is the number of minutes spent sleeping. Sleep efficiency is the percentage of time in bed that comprises sleep. The

percentage of time spent in each sleep stage (1, 2, 3&4 and REM sleep is provided. REM sleep efficiency is the percentage of REM sleep free of awakening. The micro-arousal index is the number of micro-arousals (<3 seconds) per hour. The index of periodic leg movements during sleep is the number of leg movements per hour of sleep. The apnea-hypopnea index is the number of sleep-disordered breathing events per hour of sleep.

d) qEEG Spectral Analysis

Spectra were analyzed off-line using IGOR Pro 6.12A (WaveMetrics) software on artefact- and wake-free signals for the 11 EEG derivations. Fast Fourier transform (FFT) was applied to consecutive 1-minute epochs over the night. Data were normalized to minutes per stage. Frequency bands were defined as follows: delta (0–4 Hz), theta (4–7 Hz), alpha (9–11 Hz), sigma (12–15 Hz), beta (16–30 Hz), and gamma (30–50 Hz) [132].

e) Self-report data

Questionnaires were administered to all participants to collect demographic, medical, and psychological data. Hospital medical records for mTBI patients were consulted to confirm diagnoses. Pain intensity was reported on a 100-mm visual analogue scale (VAS). Participants were asked to rate their current pain (0 = no pain, 100 = worst pain ever felt), using the question, "At this moment, how much pain do you feel?" We compiled scores on the Beck Depression Inventory (BDI-2), the Beck Anxiety Inventory (BAI), and the Pain Catastrophizing Scale. Each Beck questionnaire includes 21 items to assess depression and anxiety symptoms on a self-rated scale varying from 0–3. Higher total scores indicate more severe depression and anxiety symptoms [133-134]. Both Beck inventories have previously been used in subjects with mTBI [135]. The Pain Catastrophizing Scale (PCS) is composed

of 13 questions rated on a scale of 0 to 4 (0 = not at all 4 = all the time). It is widely used to assesses three components of catastrophizing: rumination, magnification, and helplessness[136]. These questionnaires were previously validated in whiplash patients [137].

f) *Statistical analysis*

Statistical analyses were performed with PASW Statistics 18 (SPSS Inc. Chicago, USA) with statistical significance at p <0.05. Results are reported as means with standard deviations (SD). Normality of data distribution was assessed with a Shapiro-Wilks test. A Student t-test was used to compare differences between mTBI and controls for normally distributed data and a Mann Whitney U-test to compare non-parametric data. A chi-square test with odds ratio determination and a 95% confidence interval (CI) were applied to PSQI scores. ANOVA were performed to compare qEEG between groups (mTBI with pain, mTBI without pain, Controls). A post-hoc Tukey's test was applied to the ANOVA to determine between-group differences. A simple and a multivariate stepwise regression were applied to the self-reported questionnaire data and the PSQI global score.

Results

Sample

Twenty-four mTBI patients (15M/9F) with mean age (± standard deviation) 38.33±11.39 years and with post-traumatic sleep complaints were recruited. Sleep was recorded at 48.71±22.69 days post-trauma on average (range 19–117 days). Hospitalization was less than 24 h for 79.2% of patients, with an average of 42 hours and a range of 05–456 hours. Injuries resulted from a fall in nine patients, a motor vehicle accident in eight patients, and a sports-related incident in four patients. Two patients were pedestrians hit by motor vehicles and one was involved in a fight. mTBI patients scored significantly higher on the Beck Depression Scale (mTBI:15.14 ± 10.0; Controls:1.23±1.96), the Beck Anxiety Scale (mTBI:9.89±10.50; Controls:1.08±1.44), and the Pain Catastrophizing Scale (mTBI:19.08±13.25; Controls:9.08±5.73) than controls (p<0.01 for all). All differences were statistically significant (Table 1a).

mTBI patients with pain rated pain intensity on a VAS scale at 49/100 (±30.98) compared to mTBI patients without pain at 19.38/100(±16.74) (p<0.001). Figure 1 shows painful body sites in mTBI patients with pain: 75% had back pain; 37.5% had frontal headache and neck and shoulder pain; 25% had sternum, knee, and pelvic pain; and 12.5% had occipital headache, hip, and foot pain. mTBI patients with pain scored at least twice as high on the BDI (mTBI with pain: 24.80±12.50; mTBI without pain: 12.13±7.13; p=0.01), the BAI (mTBI with pain: 19.50±11.21; mTBI without pain: 7.14±7.38; p=0.02), and the PCS (mTBI with pain: 30.57±9.22; mTBI without pain: 15.00±11.35; p=0.004) than mTBI without pain (Table 1b). All differences were statistically significant.

Subjective sleep

Upon awakening after laboratory recording, mTBI patients and controls reported their sleep quality on a 100-mm VAS. mTBI patients reported poorer sleep quality (44.96/100±23.96) compared to controls (72.53/100±15.70; p<0.001). No statistically significant difference on the sleep quality VAS was found between mTBI with pain (49.00/100±20.70) and mTBI without pain (43.19/100±25.70) (Table 1b).

mTBI patients reported sleep complaints based on the global PSQI score. 84.6% of mTBI patients scored greater than 5 on the global PSQI score. mTBI patients showed a statistically significant difference on the global score (p<0.001) and on all component scores (p<0.05) compared to controls, except for the use of sleep medication. Using the chi-square test (p=0.001), mTBI patients had an odds ratio (OR) of 38.5[CI: 4.7-318.5] to score greater than 5 on the global PSQI score (Table 2).

Sleep architecture

Sleep architecture showed statistically significant differences between mTBI patients and controls for sleep latency, persistent sleep latency, and sleep efficiency (p=0.03). Although mTBI patients presented sleep complaints, all the above sleep parameters fell within a clinically normal range according to AASM criteria [50]. No other sleep architecture parameters showed differences between mTBI patients and controls (Table 3a).

No statistically significant differences between mTBI with pain, mTBI without pain, and controls were found for all sleep architecture parameters (Table 3b).

Quantitative EEG

During REM sleep, lower delta frequency power was observed in mTBI (with and without pain combined) at C3 (mTBI=0.70μV^2; Controls=0.77μV^2; p=0.03) and at O2 (mTBI=0.56μV^2; Controls=0.69μV^2; p=0.02). Higher activity was observed at the F8 derivation during sleep stage 2 in mTBI for beta (mTBI=0.07μV^2; Controls=0.06μV^2; p=0.04) and gamma (mTBI=0.03μV^2; Controls=0.02μV^2; p=0.03) frequency bands. When these results were controlled for age and gender, neither co-variable affected the above-mentioned qEEG results (data not shown). Comparison between mTBI and controls showed no statistically significant differences in theta, alpha, or sigma bands for all non-REM and REM sleep stages (Figure 2a).

Overall, in a three-group comparison, mTBI with pain, mTBI without pain, and controls generally showed statistically significant differences on all derivations and sleep stages. mTBI with pain showed statistically significant larger spectrum values for theta, alpha, sigma, beta, and gamma frequencies than the two other groups on frontal, central, and occipital derivations. No statistically significant differences were found between mTBI without pain and controls, except for delta frequency bands during stage 2 at the C4 derivation and at O2 during slow wave and REM sleep. In the delta frequency band, both mTBI with and without pain showed statistically significant lower spectrum values than controls (p<0.04) (Figure 2b).

Psychological symptom scales and PSQI:

A simple regression showed that pain VAS (r^2= 0. 31, p < 0.01), pain catastrophizing (r^2=0.41, p < 0.01), and depression (r^2=0.57, p < 0.01), but not anxiety (r^2= 0.07, p = 0.23),

were related to the global PSQI score and subscores (r^2=0.14 to 0.62, $p < 0.05$). With multivariate stepwise regression, only depression explained global PSQI scores ($r^2 = 0.50$, $p < 0.001$).

Discussion

The main finding of this study was the association of pain with qEEG changes during sleep in mTBI patients. We first characterized an mTBI population with pain that reported more depression, anxiety, and catastrophizing than mTBI patients without pain. Although subjective sleep reports, the PSQI, and the VAS showed sleep disruption, sleep architecture in acute mTBI patients complaining of sleep–wake disturbances showed significant differences, but remained within normal range. Furthermore, qEEG did not differ between mTBI and controls, except for the delta frequency band at C3 and O2 during REM sleep and one derivation (F8) in the beta and gamma bands. However, in the presence of pain, spectral values increased in rapid EEG frequencies (alpha to gamma) during REM sleep, and delta spectral values decreased in both REM and nonREM sleep. mTBI patients with pain reported more depression, anxiety symptoms, and catastrophizing behaviour than mTBI patients without pain. Other results suggested that pain, depression, and pain catastrophizing were associated with poor sleep quality in mTBI patients, and there appeared to be interrelation with sleep quality, with depression showing a strong association with sleep complaints.

Our sleep architecture findings corroborated well with previous findings. Williams, Lazic, and Ogilvie (2008) found that mTBI patients had lower sleep efficiency and shorter REM sleep latency, whereas qEEG showed no significant results. Other studies also found no differences in qEEG during sleep after mTBI [55,138]. Another study found that trauma severity did not affect sleep architecture in chronic TBI cases, but mild TBI patients had more stage 1 sleep, increased sleep efficiency, and decreased wake time during sleep

compared to moderate and severe TBI patients [49]. The inclusion of other confounding factors such as pain and depression was proposed but not verified in these studies. In the present study, discrepancies were observed between perceptions of poor sleep and sleep architecture within normal limits. One possible explanation for these discrepancies is the presence of other cofactors, such as pain, depression, and a tendency to catastrophize in this population. This interpretation is consistent with our data, as our mTBI group reported more depression, catastrophizing, and anxiety than healthy controls, especially in the presence of pain. In addition, depression appears to be strongly associated with subjective sleep quality.

The relationship between sleep and pain has been demonstrated bidirectional: a night of poor sleep triggered more pain the next day, and intense pain was followed by frequent awakenings [82] A recent study found an interesting relationship between sleep and pain. Pre-sleep pain did not predict sleep quality, but poor sleep was a good predictor of next morning pain rating [139]. Similarly, mTBI patients with pain reported the same quality of subjective sleep as mTBI patients without pain. Even though the physiological interaction between sleep and pain in general, and in mild TBI in particular, remains unclear, it is possible that central thalamic processes play a role in both complaints, with diffuse nociceptive inhibitory control and inflammation suggested as potential mechanisms [140].The literature strongly associates pain with sleep problems in mTBI, based on questionnaires and reports [83-84]. Pain as well as post-traumatic stress disorder were also shown to be significantly associated with sleep problems, known as the "polytrauma clinical triad" [124]. As a first step towards understanding this relationship in this

population, we showed that pain in acute post-trauma affected both non-REM and REM sleep qEEG. This indicates that pain, along with depression, plays a physiological role in sleep disruption following mTBI.

Our qEEG results are in line with previous findings, showing that the increase in fast waves also persists during REM sleep. This may suggest lower sensory gating during sleep in MTBI patients with pain and an imbalance in the arousal system in insomnia conditions. It was previously suggested that hyperarousal during the sleep of insomnia patients may interfere with emotional regulation, mainly causing depression [141]. More specifically, REM instability was proposed to play a major role in non-refreshing sleep complaints, which were not shown in the sleep architecture [142]. This theory appears to fit with the data obtained in this population: mTBI patients with pain and presenting with sleep complaints show hyperarousal, mainly during REM sleep.

Change in qEEG activity leading to sleep disruption by physiological pain has been supported by previous experimental pain paradigms. One study using pregabalin as an analgesic identified slower brain oscillation as a biomarker of central analgesia [143]. Gamma oscillatory activity increased after nociceptive stimulus and was modulated by theta waves. Moreover, oscillations greater than 20 Hz were strongly related to pain perception rating, providing evidence that gamma oscillations play a role in pain perception. However, the exact role of gamma oscillations during slow wave sleep remains unknown, although they have been related to dreams in REM sleep [144-146]. Increased gamma band activity during REM sleep was also thought to be due to continuous sensory input [147].

Nociceptive experimental heat pain evoked moderate cortical arousal during sleep in healthy subjects [80,148]. Delta frequencies have also decreased in chronic widespread pain patients [81,149]. A noxious stimulus during slow wave sleep induced a decrease in delta and sigma bands but an increase in alpha and beta EEG frequencies [150]. Another study in depressed, chronic pain patients showed no difference in architecture but an increase in alpha and high beta frequency bands when compared to non-depressed patients and controls [151].

One limitation of this study is the absence of a group with chronic pain without mild traumatic brain injury. As shown in a previous study at our laboratory, qEEG changes during sleep in widespread pain conditions showed a decrease in delta waves during sleep [81]. Another limitation of this study is the absence of matching for age and gender, factors known to influence sleep structure. However, at each step of our analysis, we corrected for age and gender effects. Another limitation of this study is that the sample used is small regardless of our efforts of recruitment. This limitation is of major concern in many mTBI studies due to the heterogeneity of patients and high refusal rates [152]. Finally, although the long-term consequences of post-concussion symptoms remain poorly understood, they should be considered as interacting factors. The effects and interactions of other symptoms (headaches, cognitive deficits, personality changes, pain) on sleep–wake disturbances post-TBI remain to be clarified in a chronic TBI population. Future studies should address changes in sleep qEEG after administration of pain medications such as pregabalin or duloxetin, along with perceptions of sleep quality, in order to confirm causality between pain and qEEG changes post-MTBI.

In conclusion, our findings indicated that pain was associated with poor sleep quality and may be related to physiological qEEG changes during sleep. Pain treatment post-TBI may favour successful rehabilitation by treating not only the pain but also sleep problems, thereby contributing to long-term management of post-concussion symptoms.

<u>Acknowledgements</u>

Table 1

a)

Demographic, clinical, and psychological characteristics of mTBI and control groups

Variables	mTBI	Controls	p-Value
N	24	18	
Sex	15M/9F	6M/12F	
Age (yr.)	38.33 (11.39) [20;57]	29.11 (7.51) [18;41]	0.01
Education (yr.)	13.15 (2.15)	14.85 (1.52)	0.03
Days post-trauma	48.71 (22.69) [19;117]	----	
Hospitalization			
Less than 24h	n=19	----	
More than 24h	n=5	----	
Mechanism of injury			
Fall	n=9	----	
Motor vehicle	n=8	----	
Sports	n=4	----	
Pedestrian accident	n=2	----	
Fight	n=1	----	
Psychological			
Beck Depression Inventory II	15.14 (10.0)	1.23 (1.96)	0.00
Beck Anxiety Inventory	9.89 (10.50)	1.08 (1.44)	0.00
Pain Catastrophizing Scale	19.08 (13.25)	9.08 (5.73)	0.01

Values are given as mean (standard deviation); age range in brackets; A Student t-test was used to compare the two groups. mTBI=mild traumatic brain injury

b)

VAS and psychological characteristics of mTBI with pain, without pain, and control groups

	mTBI with pain	mTBI without pain	Controls	p-Value
VAS Pain	49.00(30.98)	19.38(16.74)	6.53(14.74)	<0.001
VAS Sleep	49.00(20.70)	43.19(25.70)	72.53(15.70)	ns
BDI	24.80(12.50)	12.13(7.13)	1.23 (1.96)	0.01
BAI	19.50(11.21)	7.14(7.38)	1.08 (1.44)	0.02
PCS	30.57(9.22)	15.00(11.35)	9.08 (5.73)	0.004

Values are given as mean (standard deviation); age range in brackets; ANOVA was used to compare the three groups. mTBI=mild traumatic brain injury; VAS=Visual Analog Scale; BDI=Beck Depression Inventory II; BAI=Beck Anxiety Inventory; PCS=Pain Catastrophizing Scale

<u>Table 2</u>
Pittsburgh Sleep Quality Index

	mTBI	Controls	p-Value
Global Score	9.35(4.82)	2.81(1.64)	0.00
Component Scores			
Sleep Quality	1.76(0.83)	0.56(0.51)	0.00
Sleep Onset Latency	1.59(1.33)	0.63(0.62)	0.04
Sleep Duration	1.29(1.21)	0.25(0.45)	0.05
Sleep Efficiency	1.41(1.42)	0.63(0.25)	0.00
Sleep Disturbance	1.41(0.71)	0.94(0.44)	0.03
Use of Sleep Medication	0.53(1.18)	0.63(0.25)	ns
Daytime Dysfunction	1.35(0.93)	0.31(0.48)	0.00

Data shown as mean (standard deviation); The Mann-Whitney U test was used to compare the two groups; mTBI=mild traumatic brain injury; ns=non-significant.

Table 3

Sleep architecture parameters for mTBI and control groups

Variables	mTBI	Controls	p-Value
Sleep latency (min)	17.79(15.27)	8.94(5.72)	0.03
Persistent sleep latency (min)	31.31(27.78)	15.46(10.22)	0.03
REM sleep latency (min)	84.01(28.39)	82.22(29.21)	ns
Sleep duration (min)	407.75(55.35)	434.94(49.69)	ns
Sleep efficiency (%)	89.20(7.50)	93.69(5.65)	0.03
Stage 1(%)	6.89(3.71)	5.60(2.73)	ns
Stage 2(%)	55.41(8.23)	53.83(5.89)	ns
Stages 3 and 4 (%)	18.25(9.86)	19.10(7.62)	ns
REM sleep (%)	19.45(3.89)	21.46(3.50)	ns
REM sleep efficiency (%)	80.91(10.83)	85.56(9.46)	ns
Micro-arousal index	14.90(5.33)	15.85(9.10)	ns
Periodic Leg Movement Index	8.83(11.19)	5.20(7.90)	ns
Apnea Hypopnea Index	6.25(6.52)	5.72(11.22)	ns

Values are given as mean ± standard deviation. A Student t-test was used to compare the two groups for variables with normal distribution and a Mann-Whitney U test when distribution was non-parametric; mTBI=mild traumatic brain injury; ns=non-significant; AHI=Apnea-Hypopnea Index.

Table 3b

Sleep architecture parameters for mTBI with pain, mTBI without pain and control groups

Variables	mTBI with pain	mTBI without pain	Controls	p-Value
Sleep latency (min)	20.00(9.11)	16.69(17.75)	8.94(5.72)	ns
Persistent sleep latency (min)	30.84(18.43)	31.54(32.01)	15.46(10.22)	ns
REM sleep latency (min)	72.92(18.39)	89.56(31.30)	82.22(29.21)	ns
Sleep duration (min)	413.38(56.30)	404.94(56.50)	434.94(49.69)	ns
Sleep efficiency (%)	90.35(5.28)	88.63(8.49)	93.69(5.65)	ns
Stage 1(%)	6.54(3.32)	7.06(3.99)	5.60(2.73)	ns
Stage 2(%)	57.29(11.64)	54.47(6.15)	53.83(5.89)	ns
Stages 3 and 4 (%)	15.28(13.01)	19.74(7.95)	19.10(7.62)	ns
REM sleep (%)	20.89(4.13)	18.73(3.68)	21.46(3.50)	ns
REM sleep efficiency (%)	84.42(11.56)	79.16(10.37)	85.56(9.46)	ns
Micro-arousal index	15.50 (6.70)	14.59(4.73)	15.85 (9.10)	ns
Periodic Leg Movement Index	9.29(8.41)	6.45(10.61)	5.20(7.90)	ns
Apnea Hypopnea Index	4.79(3.70)	6.88(7.44)	5.72(11.22)	ns

Values are given as mean ± standard deviation. An ANOVA with a Tukey's post-hoc test was used to compare the three groups; mTBI=mild traumatic brain injury; ns=non-significant; AHI=Apnea-Hypopnea Index.

Figure legends

Figure 1: A pain diagram where subjects were asked to mark painful body sites. Marked sections correspond to the sum of painful body sites indicated by mTBI patients with pain. Percentages represent the percentage of mTBI patients with pain who identified the area as painful.

Figure 2: Table a shows the EEG differences between MTBI patients and controls (C). Table b shows the EEG differences between the three groups. Histograms presented in Table c show examples of some derivations, revealing differences between the three groups.

C=Healthy controls; M=mTBI patients with no pain; P=mTBI patients with pain. ANOVA with a Tukey's correction was used to compare the three groups. The p-value shows the statistically significant difference between the highest and lowest group.

[a] The Pain group shows statistical differences from both the mTBI without pain and the Control group.

Figure 1:

Figure 2

a
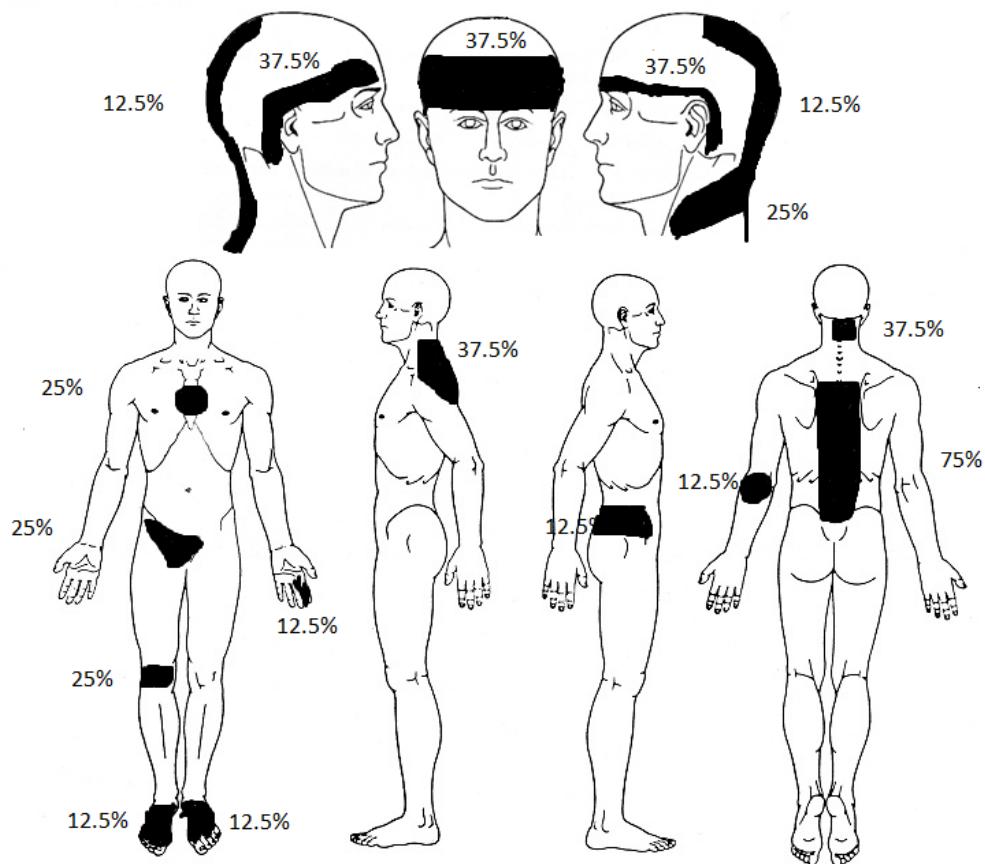
EEG differences between MTBI and Controls

	Sleep Stage 2	Slow Wave Sleep	REM Sleep
Delta			C3(p=0.03) MTBI=0.70µV²<C=0.77µV² O2(p=0.02) MTBI=0.56µV²<C=0.69µV²
Theta			
Alpha			
Sigma			
Beta	F8(p=0.04) MTBI=0.07µV²>C=0.06µV²		
Gamma	F8(p=0.03) MTBI=0.03µV²>C=0.02µV²		

b

EEG differences between MTBI (with and without pain)and Controls

	Sleep Stage 2	Slow Wave Sleep	REM Sleep
Delta	C4(p=0.03) M<P<C O1(p=0.04) M<C<P	O2(p=0.04) M<P<C	O2(p=0.03) M<P<C
Theta	F8(p=0.03) C<M<P		C4(p=0.04) M<C<P
Alpha			Fp2(p=0.01) C<M<P[a] F3(p=0.03) M<C<P F4(p=0.02) M<C<P[a] F7(p=0.04) M<C<P[a] F8(p=0.00) C<M<P[a] C3(p=0.03) M<C<P C4(p=0.02) M<C<P O2(p=0.03) M<C<P
Sigma		C4(p=0.04) M<C<P	Fp1(p=0.03) M<C<P[a] Fp2(p=0.00) C<M<P[a] F3(p=0.01) M<C<P[a] F4(p=0.00) M<C<P[a] F7(p=0.02) M<C<P[a] F8(p=0.00) C<M<P[a] C3(p=0.02) M<C<P C4(p=0.01) M<C<P O1(p=0.02) M<C<P O2(p=0.02) M<C<P[a]
Beta	Fp2(p=0.01) C<M<P F3(p=0.03) C<M<P[a] F4(p=0.01) C<M<P[a] F8 (p=0.00) C<M<P[a]	Fp1(p=0.04) C<M<P Fp2(p=0.01) C<M<P[a] F3(p=0.02) M<C<P[a] F4(p=0.01) C<M<P[a] F8(p=0.01) C<M<P[a] C4(p=0.03) M<C<P O1(p=0.05) C<M<P	Fp1(p=0.01) C<M<P[a] Fp2(p=0.01) C<M<P[a] F3(p=0.02) M<C<P[a] F4(p=0.01) M<C<P[a] F7(p=0.01) C<M<P[a] F8(p=0.00) C<M<P[a] C3(p=0.04) M<C<P C4(p=0.01) M<C<P[a]
Gamma	F8(p=0.03) C<M<P		F4(p=0.02) C<M<P F8(p=0.01) C<M<P[a]

c

Example of sigma values at the C4 position during slow wave sleep and REM sleep
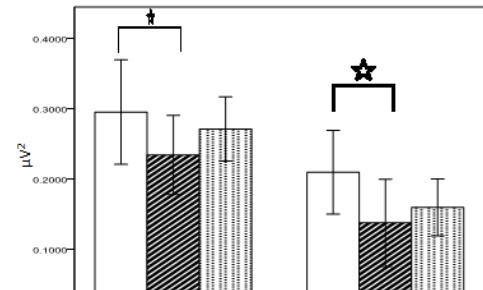
Example of **beta** values at the **F8** position during all sleep stages
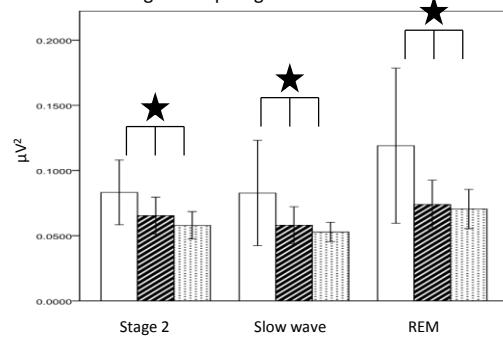
References

AASM (2005). International Classification of Sleep Disorders, 2nd edition: Diagnostic and Coding Manual. Westchester, IL, American Academy of Sleep Medicine.

Baglioni, C. and Riemann, D. (2012). Is chronic insomnia a precursor to major depression? Epidemiological and biological findings. Curr Psychiatry Rep Epub ahead of print.

Baumann, C.R., Werth, E., Stocker, R., Ludwig, S. and Bassetti, C.L. (2007). Sleep-wake disturbances 6 months after traumatic brain injury: a prospective study. Brain 130: 1873-1883.

Beck, A.T., Ward, C.H., Mendelson, M., Mock, J. and Erbaugh, J. (1961). An inventory for measuring depression. Arch Gen Psychiatry 4: 561-571.

Beck, A.T., Epstein, N., Brown, G. and Steer, R.A. (1998). An inventory for measuring clinical anxiety: psychometric properties. J Consult Clin Psychol 56: 893-897.

Beetar, J.T., Guilmette, T.J. and Sparadeo, F.R. (1996). Sleep and pain complaints in symptomatic traumatic brain injury and neurologic population. Arch Phys Med Rehabil 77: 1298-1302.

Bonica, J.J. (1979). The need of a taxonomy. Pain 6(3): 247-248.

Buysse, D.J., Reynold, C.F.r., Monk, T.H., Berman, S.R. and Kupfer, D.J. (1989). The Pittsburgh Sleep Quality Index: a new instrument for psychiatric practice and research. Psychiatry Res. 28(2): 193-213.

Carroll, L.J., Cassidy, J.D., Holm, L., Kraus, J. and Coronado, V.G. (2004). Methodological issues and research recommendations for mild traumatic brain injury: The WHO collaborating centre task force on mild traumatic brain injury. J Rehabil Med 36(Suppl. 43): 113-125.

Castriotta, R.J., Wilde, M.C., Lai, J.M., Atanasov, S., Masel, B.E. and Kuna, S.T. (2007). Prevalence and consequences of sleep disorders in traumatic brain injury. J Clin Sleep Med 3(4): 349-356.

Castriotta, R.J. and Murthy, J.N. (2010). Sleep disorders in patients with traumatic brain injury: a review. CNS Drugs 25(3): 175-185.

Chaput, G., Giguère, J.F., Chauny, J.M., Denis, R. and Lavigne, G.J. (2009). Relationship among subjective sleep complaints, headaches, and mood alterations following a mild traumatic brain injury. Sleep Med. 10(7): 713-716.

De Pascalis, V. and Cacace, I. (2005). Pain perception, obstructive imagery and phase-ordered gamma oscillations. Int J Psychophysiol. 56: 157-169.

Faul, M., Xu, L., Wald, M.M. and Coronado, V.G. (2010). Traumatic brain injury in the United States: emergency department visits, hospitalizations, and deaths 2002-2006. Atlanta (GA), Centers for Disease Control and Prevention. National Center for Injury Prevention and Control.

Fichtenberg, N., Putman, S., Mann, N., Zafonte, R. and Millard, A. (2001). Insomnia screening in postacute traumatic brain injury-utility and validity of the Pittsburgh Sleep Quality Index. Am J Phys Med Rehabil 80: 339-345.

Giamberardino, M.A. and Jensen, T.S. (2012). Pain comorbidities: Understanding and treating the complex patient. Seattle, WA, International Association for the Study of Pain.

Gosselin, N., Lassonde, M., Petit, D., Leclerc, S., Mongrain, V., Collie, A. and Montplaisir, J.Y. (2009). Sleep following sport-related concussions. Sleep Med. 10(1): 35-46.

Graversen, C., Olesen, S.S., Olesen, A.E., Steimle, K., Farina, D., Wilder-Smith, O.H.G., Bouwense, S.A.W., van Goor, H. and Drewes, A.M. (2012). The analgesic effect of pregabalin in patients with chronic pain is reflected by changes in pharmaco-EEG spectral indices. Br J Clin Pharmacol. 73(3): 363-372.

Guilleminault, C., Faull, K., Miles, L. and van den Hoed, J. (1983). Post-traumatic excessive daytime sleepiness: a review of 20 patients. Neurology 33: 1584-1589.

Harman, K., Pivik, R.T., D'Eon, J.L., Wilson, K.G., Swenson, J.R. and Matsunaga, L. (2002). Sleep in depressed and nondepressed participants with chronic low back pain: electroencephalographic and behaviour findings. Sleep 25(7): 775-783.

Hoffman, J.M., Pagulayan, K.F., Zawaideh, N., Dikmen, S., Temkin, N. and Bell, K.R. (2007). Understanding pain after traumatic brain injury: Impact on community participation. Am J Phys Med Rehabil 86(12): 962-969.

IFSECN (1974). A glossary of terms most commonly used by clinical electroencephalographers. Electroencephalogr Clin Neurophysiol 37(5): 538-548.

Iverson, G.L. and McCracken, L.M. (1997). 'Postconcussive' symptoms in persons with chronic pain. Brain Inj 11(11): 783-790.

Kempf, J., Werth, E., Kaiser, P.R., Bassetti, C.L. and Baumann, C.R. (2010). Sleep-wake disturbances 3 years after traumatic brain injury J Neurol Neurosurg Psychiatry 81: 1402-1405.

Klein, E., Koren, D., Arnon, I. and Lavie, P. (2003). Sleep complaints are not corroborated by objective sleep measures in post-traumatic stress disorder: a 1-year prospective study in survivors of motor vehicle crashes. J Sleep Res. 12: 35-41.

Lavigne, G.J., Zucconi, M., Castronovo, C., Manzini, C., Marchettini, P. and Smirne, S. (2000). Sleep arousal response to experimental thermal stimulation during sleep in human subjects free of pain and sleep problems. Pain 84(2): 283-290.

Lavigne, G.J., Brousseau, M., Kato, T., Mayer, P., Manzini, C., Guitard, F. and Montplaisir, J.Y. (2004). Experimental pain perception remains equally active over all sleep stages. Pain 110(3): 646-655.

Lavigne, G.J., Nashed, A., Manzini, C. and Carra, M.C. (2011). Does sleep differ among patients with common musculoskeletal pain disorders? Curr Rheumatol Rep 13(6): 535-542.

Lavigne, G.J., Okura, K., Abe, S., Colombo, R., Huynh, N., Montplaisir, J.Y., Marchand, S. and Lanfranchi, P.A. (2011). Gender specificity of the slow wave sleep lost in chronic widespread musculoskeletal pain. Sleep Med. 12(2): 179-185.

Lew, H.L., Pogoda, T.K., Hsu, P.T., Cohen, S., Amick, M.M., Baker, E., Meterko, M. and Vanderploeg, R.D. (2010). Impact of the ''polytrauma clinical triad'' on sleep

disturbance in a department of veterans affairs outpatient rehabilitation setting. Am J Phys Med Rehabil 89(6): 437-445.

Luoto, T.M., Tenovuo, O., Kataja, A., Brander, A., Ohman, J. and Iverson, G.L. (2012). Who gets recruited in mild traumatic brain injury research? J neurotrauma Epub ahead of print.

Masel, B.E., Scheibel, R.S., Kimbark, T. and Kuna, S.T. (2001). Excessive daytime sleepiness in adults with brain injuries. Arch Phys Med Rehabil 82: 1526-1532.

Miro, J., Nieto, R. and Huguet, A. (2008). The Catalan version of the Pain Catastrophizing Scale: a useful instrument to assess catastrophic thinking in whiplash patients. J Pain 9(5): 397-406.

Moldofsky, H. (2001). Sleep and pain. Sleep Med Rev 5(5): 385-396.

Nampiaparampil, D.E. (2008). Prevalence of chronic pain after traumatic brain injury: A systematic review. JAMA 300(6): 711-719.

Ouellet, M.C. and Morin, C.M. (2006). Subjective and objective measures of insomnia in the context of traumatic brain injury: a preliminary study. Sleep Med. 7(6): 486-497.

Parsons, L.C., Crosby, L.J., Perlis, M., Britt, T. and Jones, P. (1997). Longitudinal sleep EEG power spectral analysis studies in adolescents with minor head injuty. J Neurotrauma 14: 549-559.

Prigatano, G.P., Stahl, M.L., Orr, W.C. and Zeiner, H.K. (1982). Sleep and dreaming disturbances in closed head injury patients. J Neurol Neurosurg Psychiatry 45: 78-80.

Rao, V., Bergey, A., Hill, H., Efron, D. and McCann, U. (2011). Sleep disturbance after mild traumatic brain injury: indicator of injury? J Neuropsychiatry Clin Neurosci 23(2): 201-205.

Rechtschaffen, A. and Kales, A., Eds. (1968). A manual of standardized terminology, techniques and scoring system for sleep stages of human subjects. Los Angeles, Brain Information Service/Brain Research Institute, University of California.

Riemann, D., Spiegelhalder, K., Nissen, C., Hirscher, V., Baglioni, C. and Feige, B. (2012). REM sleep instability - A new pathway for insomnia? Pharmacopsychiatry 45(5): 167-176.

Smith, M.T. and Buenaver, L.F. (2007). Electroencephalographic correlates of pain and sleep interactions in humans. Sleep and Pain. G. J. Lavigne, B. J. Sessle, M. Choinière and P. Soja. Seattle, WA, IASP Press.

Smith, M.T., Quartana, P.J., Okonkwo, R.M. and Nasir, A. (2009). Mechanisms by which sleep disturbance contributes to oteoarthritis pain: A conceptual model. Curr Pain Headache R 6: 447-454.

Stalnacke, B.M. (2012). Postconcussion symptoms in patients with injury-related chronic pain. Rehabil Res Pract 2012: 528265.

Sullivan, M.J.L., Bishop, S. and Pivik, J. (1995). The pain catastrophizing scale: Development and validation. Psychol Assess 7: 524-532.

Tang, N.K.Y., Goodchild, C.E., Sanborn, A.N., Howard, J. and Salkovskis, P.M. (2012). Deciphering the temporal link between pain and sleep in a heterogeneous chronic pain patient sample: A multilevel daily process study. Sleep 35(5): 675-687.

Theodorou, A.A. and Rice, S.A. (2007). Is the silent epidemic keeping patients awake? J Clin Sleep Med 3(4): 347-348.

Timofeev, I., Bazhenov, M., Seigneur, J. and Sejnowski, T. (2012). Neuronal synchronization and thalamocortical rythms in sleep, wake and epilepsy. Jasper's basic mechanisms of the epilepsies. J. L. Noebels, M. Avoli, M. A. Rogawski, R. W. Olsen and A. V. Delgado-Escueta. Bethesda (MD), National Center for Biotechnology Information.

Trahan, D.E., Ross, C.E. and Trahan, S.L. (2001). Relationships among postconcussional-type symptoms, depression, and anxiety in neurologically normal young adults and victims of mild brain injury. Arch Clin Neuropsychol 16(5): 435-445.

Urbano, F.J., Kezunovic, N., Hyde, J., Simon, C., Beck, P. and Garcia-Rill, E. (2012). Gamma band activity in the reticular activating system. Front Neurol 3(6): In Press.

Vancleef, L.M. and Peters, M.L. (2006). Pain catastrophizing, but not injury/illness sensitivity or anxiety sensitivity, enhances attentional interference by pain. J Pain 7(1): 23-30.

Verma, A., Anand, V. and Verma, N.P. (2007). Sleep disorders in chronic traumatic brain injury. J Clin Sleep Med 3(4): 357-362.

Walker, W.C. (2004). Pain pathoetiology after TBI. J Head Trauma Rehabil 19(1): 72-81.

Wang, J., Li, D., Li, X., Liu, F.Y., Xing, G.G., Cai, J. and Wan, Y. (2011). Phase-amplitude coupling between theta and gamma oscillations during nociception in rat electroencephalography. Neuroscience Letters 499(2): 84-87.

Watson, N.F., Dikmen, S., Machamer, J., Doherty, M. and Temkin, N. (2007). Hypersomnia following traumatic brain injury. J Clin Sleep Med 3(4): 363-368.

Webster, J.B., Bell, K.R., Hussey, J.D., Natale, T.K. and Lakshminarayan, S. (2001). Sleep apnea in adults with traumatic brain injury: a preliminary investigation. Arch Phys Med Rehabil 82: 316-321.

WHO (2010). World Health Organization. Global status report on road safety. Department of Violence & Injury Prevention & Disability (VIP).

Williams, B.R., Lazic, S.E. and Ogilvie, R.D. (2008). Polysomnographic and quantitative EEG analysis of subjects with long-term insomnia complaints associated with mild traumatic brain injury. Clinical Neurophysiology 119: 429-438.

Worthington, A.D. and Melia, Y. (2006). Rehabilitation is compromised by arousal and sleep disorders: results of a survey of rehabilitation centres. Brain Inj 20: 327-332.

Article 2: BDNF polymorphism predicts the transition to chronic pain following mild

traumatic brain injury

Manuscrit en soumission

BDNF polymorphism predicts the transition to chronic pain following mild traumatic brain injury

Running title: Chronic pain following traumatic brain injury

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Indexing words: Mild traumatic brain injury, pain, BDNF

Abstract

Mild traumatic brain injury (mTBI) often causes chronic pain. This study determines whether post-traumatic pain, alterations in sensory perceptions, psychological or genetic factors in acute phase are predisposing factors for pain one year later. At the acute six-week phase, 94 mTBI patients were assessed for post-traumatic symptoms and pain and underwent quantitative sensory testing (QST) and psychomotor vigilance testing (PVT). Patients were genotyped for BDNF Val66Met and COMT Val158Met polymorphisms. Tests were repeated in 36 patients one year post-trauma. Predisposing factors for chronicity were determined with multiple stepwise regression. At acute post-trauma, 70% of patients who reported pain had worse psychological symptoms, were less able to return to work, and had slower mean reaction time, with no difference in sensory or pain detection than patients without pain. The BDNF val/val polymorphism was more frequent in mTBI with pain. One year later, 50% of 36 mTBI patients had pain with persistent psychological distress, slower reaction time, and less sensitivity to warmth. BDNF val/val genotype and anxiety in the acute phase predispose to chronic pain and post-concussion symptoms at one year. Three patients developed new-onset pain at one year post-trauma. mTBI patients with the BDNF val/val polymorphism had worse outcome one year post-trauma.

Introduction

Mild Traumatic Brain Injury (mTBI) refers to a blunt physical trauma to the head, mostly resulting from motor vehicle accidents, falls, and sport-related injuries. The annual incidence is 100–300/100 000 [153]. Historically, mild brain injury was considered a concussion that would heal in a relatively short time. More correctly, it is called the "silent epidemic," because it often leaves patients with disabilities such as chronic pain, headaches, mood disturbances, and deteriorated quality of life [7,40]. Although many mTBI patients recover after three months, some show chronic post-concussion symptoms, including chronic pain [154]. A recent systematic review reported chronic pain post-mTBI in 75% of patients [66]. Because pain can potentially influence recovery after a traumatic injury by exacerbating other post-concussive symptoms, it is crucial to characterize its components in this population [43]. The literature is extensive on the impact of pain in cohorts of whiplash injury patients. For example, in systematic reviews, patients have reported that initial pain intensity is linked to delayed functional recovery [155-157]. In another study, high intensity of initial pain also predicted poor outcome following whiplash, along with age, cold hyperalgesia, and moderate post-traumatic stress disorder [158]. Most studies have identified various pain parameters (passive pain coping, cold hyperalgesia, prior pain) as predictive factors for chronicity [159-161]. Others have examined predisposing factors for chronicity in a general pain population and after mTBI. No conclusions can be reached, however, as depressive symptoms, pre-trauma conditions, nature of the injury, and post-trauma treatment were all shown to influence outcome

[127,162-163]. Moreover, many symptoms are known to coexist. For example, 42% of war veterans with TBI have chronic pain, posttraumatic stress disorder, and post-concussion symptoms [164]. In experimental pain and sensory modality perception testing in mTBI patients with post-traumatic headache and post-traumatic stress disorder, thermal perceptions are altered [73,75]. In these experimental paradigms, other factors such as age, cognitive decline, and reaction time difference between individuals were not taken into account. A better outcome following mTBI implies brain plasticity, whereby brain structures and functions change in order to restore connexions that existed prior to the trauma. Synaptic changes are mediated by a key signaling molecule called the brain-derived neurotrophic factor (BDNF) [96]. Genetic susceptibility to BDNF regulation of brain plasticity may play an important role in post-trauma recovery [165]. A previous study showed that the Met allele is associated with poor cognitive performance following mTBI [98]. Polymorphisms in the BDNF may therefore be good candidates for predisposing factors. A common polymorphism of the BDNF gene is the Val66Met polymorphism (rs6265) that leads to a valine to methionine substitution at codon 66. Although this substitution does not affect transcription or translation of BDNF, it is linked to morphological changes in the hippocampus and the prefrontal cortex [166]. It is therefore important to hypothesize that this polymorphism is involved in the recovery following traumatic brain injury.

Pain processing is also known to be influenced by genetic factors. Neurotrophins, including BDNF, act as central modulators of pain [167], and along with Catechol-O-

methyltransferase (COMT), can augment cortical pain processing [168]. COMT polymorphisms affect pain perception and the ability to cope with pain [169-170].

In this study, psychological, physiological, and biological data on mTBI patients in the acute phase (less than three months) were assessed to identify predisposing factors for the transition to or new onset of chronic pain at one year post-trauma.

The hypothesis was that mTBI patients with pain in the acute phase would have worse outcome in the presence of comorbidities.

The study objectives were to: 1) to characterize pain, experimental heat pain perception, and the influence of other pain co-factors (vigilance, genotype) in acute mTBI;

and 2) to identify early predisposing factors in the acute post-trauma phase for the transition to or new onset of chronic pain.

Materials and Methods

1-Study population:

mTBI patients were screened from the Sacré-Coeur Hospital trauma center (n=102). mTBI diagnosis was confirmed by a trauma neurosurgeon (author JFG) based on the following 2004 WHO Task Force criteria [12]: a) Glasgow Coma Scale of 13–15 after 30 minutes post-injury; b) loss of consciousness for 30 minutes or less; c) post-traumatic amnesia for less than 24 hours; and d) age > 18 and < 65 years. Patients were excluded if they presented a) gross cognitive or speech dysfunctions; b) use of any psychotropic medications or other drugs known to influence pain perception; c) history of chronic pain or fibromyalgia before mTBI; or d) presence of major neurological or psychiatric disorders. Eight patients were excluded for the following reasons: not meeting the WHO task force inclusion criteria (n=2), depression (n=3), leukemia (n=1), chronic alcohol use (n=1), and collagenosis (n=1).

The final sample consisted of 94 mTBI patients. Patients filled out questionnaires, performed the Psychomotor Vigilance Test (PVT), and underwent quantitative sensory testing (QST) and blood sampling for genotyping. Patients were classified as mTBI patients with pain (n=65) if they responded "yes" to all pain-related questions during the interview and scored higher than "mild pain" on question 7 of the Medical Outcome Study short-form 36 (SF-36). This question states: How much bodily pain have you had during the past four weeks? Post-traumatic headache was also assessed, defined as a secondary headache that develops within seven days after head trauma or after regaining consciousness [171]. At the one-year post-trauma follow-up, all patients were contacted to participate in the follow-up protocol, of which 36 mTBI

(38%) agreed. The same experimental paradigm was repeated, except for genotyping. Pain was assessed as in the acute phase. All patients signed an informed consent form and the study was approved by the hospital's ethics board.

2-Quantitative Sensory Testing:

Quantitative Sensory Testing (QST) (TSA II; Medoc Ltd., Israel) was performed on the upper left arm using a Peltier-based computerized thermal stimulator (thermode). Base temperature was set at 32° C. Thermal threshold testing was performed for warmth, cold, and heat pain perception [74,172]. Temperature detection of the three thermal perceptions (warm, cold, and heat pain) was assessed in five trials for each perception, separated by 60 seconds. Patients were asked to detect warm and cold temperatures and to discriminate the first sensation of pain. Average recorded temperature was used for the analysis.

3-Psychomotor Vigilance Testing (PVT):

Psychomotor Vigilance Testing (PVT) is usually used to assess sleepiness and reaction time [173]. Sustained attention was assessed using the Psychomotor Vigilance Test (PVT-192: Ambulatory Monitoring Inc., Ardsley, NY) [174]. The 10-minute PVT trial was used. Patients were given a hand-held computerized device with a red light-emitting diode display of a three-digit millisecond counter and were instructed to press a response button as soon as possible when a visual stimulus appeared on the screen. Reaction times (RTs) were collected from each 10-minute trial. Patients were given pretest training to minimize the practice effect. Mean RT was used as a study variable.

4-Questionnaires:

Medical diagnoses and reports were used to confirm patient eligibility. General physical and emotional health questionnaires were administered at two time points: six weeks and one year post-trauma. Data on demographics, education, and return to work were also taken. Pain intensity was determined on a 100-mm Visual Analog Scale (VAS).

4.1-SF36:

To assess quality of life after mTBI, all patients received the standard Medical Outcome Study short-form 36 (SF-36). The SF-36 is a validated 36-item questionnaire divided into eight scales that can be aggregated into two summary scores on mental and physical aspects. A higher score indicates a better quality of life.

The eight scales are: 1) physical functional ability; 2) role limitation due to physical impairment; 3) bodily pain; 4) general health;5) vitality; 6) social functioning; 7) role limitation due to emotional impairment; and 8) mental health. Scores for the eight scales range from 0 to 100 [175-176].

4.2-Rivermead Post-Concussion Symptoms Questionnaire:

This is the most widely used validated questionnaire to measure post-concussive symptoms such as headache, dizziness, and concentration problems. It contains 16 items that assess the presence and severity of cognitive, emotional, and somatic complaints on a five-point scale (1=no problem to 5=severe problem). The overall score is obtained by summing all scores. Higher scores indicate more severe post-concussion symptoms [34].

4.3-Impact of Event Scale – Revised (IES-R)

This self-rate questionnaire assesses subjective trauma-related stress. Twenty-two questions address post-traumatic stress disorder: 8 measure intrusion symptoms, 8 avoidance symptoms, and 6 hyperarousal symptoms. Patients rate their perceived severity of PTSD symptoms on a five-point scale (0–4). Scores above 26 are classified as severe [177].

4.4-Beck Depression Inventory-II(BDI) and the Beck Anxiety Inventory(BAI):

These self-administered questionnaires contain 21 items that quantify depression and anxiety on a score from 0–3. Higher total score indicates more severe depressive and anxiety symptoms. Scores are classified as minimal, mild, moderate, and severe [133-134].

4.5-Pain Catastrophizing Scale:

The Pain Catastrophizing Scale is a validated tool to measure catastrophic pain-related thinking. It assesses the three components of catastrophizing: rumination, magnification, and helplessness, using 13 questions rated on a scale of 0–4. Higher scores represent greater catastrophizing [136].

4.6-Migraine Disability Assessment (MIDAS):

The MIDAS is a seven-item questionnaire. The first five assess the influence of headaches over the last three months in different domains: paid and school work, household work, and leisure activities with family or in social situations. The last two questions assess the total number of days with migraine attacks and the mean pain

intensity. The MIDAS score was calculated using the first five questions and the number of days in which migraine interfered with these activities. Disability scores were classified as follows: minimal (0–5), mild (6–10), moderate (11–20), and severe (>21) [178].

4.7-McGill Pain Questionnaire:

This questionnaire provides pain descriptors for the type of pain experienced. It has shown high agreement between subjects with different cultural, socioeconomic, and educational backgrounds. Pain descriptors are grouped to assess sensory, affective, and evaluative qualities of pain [179].

5-Genotyping:

Each participant gave informed consent and a blood sample was obtained. Genomic DNA was extracted from blood using the Puregene DNA kit using the manufacturer's protocol (Gentra System, USA). The genotyping of BDNF (rs6265) and COMT (rs4680) was performed using Sequenom IPLEX Gold technology, at McGill University and Génome Québec Innovation Center. BDNF and COMT genotypes were determined with the SNPs rs6265, rs4680 respectively. If rs6265 and rs4680 is a G, it encodes a Val, and if it is an A, it encodes for a Met in the BDNF Val66Met and COMT Val158Met polymorphisms, respectively.

6-Statistical analysis:

Statistical analyses were performed using SPSS (PASW 18) with significance set at 0.05. Data are reported as mean ± standard deviation (SD) unless otherwise specified. Group comparisons were performed using a student t-test, or a Mann-Whitney U test when data were abnormally distributed, and a chi-square test in categorical data cases. A repeated measure ANOVA was used to compare changes across repeated tests (questionnaire scores, PVT, QST) at the two time points. The predisposing variables in the acute phase and the variables at the one-year follow-up were assessed using multiple stepwise regression analysis, or for nominal data, by a separate binary logistic.

Results

Ninety-four mTBI patients (67 M, 27 F; mean age 37.7±12.7 yr) were assessed 46.1±22.6 days post-trauma. Thirty-six mTBI patients (23 M, 13 F; mean age 45.1±11.6 yr) agreed to participate at the one-year follow-up (429.9±59.4 days post-trauma). Traumatic brain injury was the consequence of a motor vehicle accident for 34 mTBI patients (36.1%), a fall for 26 mTBI patients (27.7%), a cycling accident for 19 mTBI patients (20.2%), an assault for 11 mTBI patients (11.7%), and a pedestrian hit for four mTBI patients (4.3%).

Objective 1: to characterize pain in mTBI patients

Psychological assessment

Twenty-one mTBI patients (22.3%) reported occasional pain prior to the traumatic brain injury. Eleven reported back pain, three headaches, two knee pain, two abdominal pain, three musculoskeletal pain, three shoulder pain, one foot pain, four neck pain, and one arm pain. None reported chronic pain. In all patients, except for abdominal pain and headaches, pain was related to work posture and exercise or sports. This pain was still present at the same intensity post-trauma, except for abdominal and muscular pain, which decreased. Sixty-five (69.2%) mTBI patients reported pain post-trauma at the acute phase and were classified as mTBI patients with pain. The most common form of pain was accident-related musculoskeletal pain. According to the McGill Pain Questionnaire, the descriptors most frequently reported to describe pain were in the affective category: tiring (27.7%), exhausting (22.3%), and wretched (20.2%), with annoying (20.2%) in the evaluative category, warm (23.4%) in the sensory category, and nagging (26.6%) in the miscellaneous category. Post-traumatic headache (PTHA) was also assessed with the MIDAS

Questionnaire, and 34 (37%) of patients reported PTHA with an average VAS of 60/100 lasting for an average of 10 days a month.

mTBI with pain patients reported a mean VAS of 51.3/100 (±24.9) compared with mTBI without pain patients, who reported a mean VAS=23.4/100 (±31.6); p<0.0001. The mTBI with pain group (n=65; 69.2%) reported worse symptoms on the Rivermead Post-Concussion Symptoms Questionnaire (with pain 22.0±12.7; without pain 13.2±9.9; p=0.003), worse quality of life on the SF-36 questionnaire (with pain 47.1±16.4; without pain 72.1±17.0; p<0.000), higher bodily pain scores on the SF-36 (with pain 34.4±17.8; without pain 77.0±19.7; p<0.000), higher overall and component scores on the Pain Catastrophizing Scale (with pain 15.2±12.3; without pain 9.9±7.5; p=0.05), higher scores for depression, anxiety, and post-traumatic stress symptoms on the BDI-2 (with pain 12.9±8.6; without pain 5.5±4.3; p<0.000), higher BAI scores (with pain 10.2±9.3; without pain 3.7±3.1; p=0.003), and higher trauma-related stress symptoms on the IESR (with pain 20.2±16.6; without pain 9.4±7.7; p=0.006) (Table 1). mTBI patients without pain were significantly more likely to be male (χ^2=4.6; d=1; p=0.05) and to return to work than mTBI with pain patients (χ^2=6.6; d=1; p=0.01), with OR=3.9 and 95% CI [1.32:11.63].

Sensory perceptions and reaction time

Quantitative sensory testing in the acute phase showed no differences in perceptions of heat, cold, or heat pain between mTBI with and without pain (Figure 1).

Moreover, mTBI patients with pain had slower reaction time (282.2±65.7) on a Psychomotor Vigilance Test (PVT) task than mTBI patients without pain (241.0±18.2) (p<0.0001).

Genotyping

Genotyping of common polymorphisms in the BDNFval66Met and the COMTval158met gene were performed. No statistically significant differences were found for the COMT genotypes. mTBI patients with pain were more likely to have the genotype BDNF val/val (χ^2=5.7; d=1; p=0.02). Because the genotype met/met was rare in our population (n=3), it was omitted from the analysis.

Objective 2: To identify early predisposing factors for the transition to or new onset of chronic pain.

At the one-year follow-up (mean days=429.9 ±59.4), 36 mTBI patients agreed to participate. At one year, 10 patients without pain at the acute phase remained pain-free and three developed new onset pain. Fifteen mTBI patients with pain at the acute phase still had (persistent) pain, and eight were free of pain. Overall, 50% of mTBI with pain patients in the acute phase still reported pain, with a mean pain VAS score of 36.2/100 (±19.5) compared to mTBI without pain patients, who had a mean VAS score of 6.3/100 (±15.4); p<0.0001. At the follow-up, the mTBI with pain group (n=18) reported worse symptoms on the Rivermead Post-Concussion Symptoms Questionnaire (mTBI with pain 17.9±11.9; mTBI without pain 7.1±6.8; p=0.007), worse quality of life on the SF-36 questionnaire

(mTBI with pain 67.0±19.6; mTBI without pain 88.2±6.9; p=0.005), more bodily pain on the SF-36 (mTBI with pain 52.9±25.4; mTBI without pain 77.9±20.4; p=0.01), higher scores for depression, anxiety, and post-traumatic stress symptoms on the BDI-2 (mTBI with pain 10.2±10.5; mTBI without pain 2.6±3.4; p=0.02), and higher BAI scores (mTBI with pain 8.7±12.2; mTBI without pain 2.2±3.6; p=0.04). The Impact of Event Scale and the overall Pain Catastrophizing Scale score showed no statistical differences between the two groups (Table 2).

Physiological data at one year showed no statistically significant differences between the pain and no-pain groups for mean reaction time on the PVT. However, on the QST, mTBI patients with pain were less sensitive to warm sensation than mTBI patients without pain (mTBI with pain 37.5°C±2.3; mTBI without pain 35.4°C±2.1; p=0.03) (Figure 2).

Changes in symptoms in mTBI patients with persistent or new-onset pain at one year post-trauma are illustrated in Figure 3. Overall, RPQ, PCS, BDI, and IESR scores decreased in both mTBI with pain and mTBI without pain. However, SF-36 and bodily pain SF-36 scores increased. These results indicate that mTBI patients with persistent or newly developed pain show fewer post-concussion symptoms, less depression, anxiety, less pain-related catastrophizing, less post-traumatic stress, and better quality of life than in the acute post-trauma stage. No differences were found on mean reaction time on the PVT, sensory perceptions, or heat pain detection between the acute and chronic phase for both groups. However, change in cold detection threshold differed between groups.

A correlational matrix between questionnaire scores and pain VAS at the acute phase shows that all variables are highly correlated (Table 3). In a multiple stepwise regression

model, the BAI score in the acute phase post-trauma explains 29% of the variability in the pain VAS, 42% of the variability in the Rivermead Post-Concussion score, 33% of the variability in the Pain Catastrophizing score, and 55% and 38% of the variability in the Beck Depression and Anxiety Inventory scores. In a logistic regression, the models that best differentiate between mTBI with and without chronic pain are summarized in Table 4. Excluding questionnaires, the independent variables BDNF and age best predict persistent or newly developed pain in mTBI independently of other variables, including sex, COMT genotype, pain VAS, return to work, mean reaction time, and warm temperature detection. The val/val genotype best predicts pain persistence or newly developed pain in mTBI (Table 5). When questionnaire variables are introduced into the model, regression cannot be established, because all these variables are interrelated (Table 3).

Discussion

The first objective of this study was to characterize pain in the acute phase following mTBI. Psychological factors such as depression, anxiety, and post-traumatic stress disorder were more prevalent in mTBI patients with pain than without pain. Post-concussion symptoms, measured using the Rivermead Post-Concussion Symptoms questionnaire, were more severe in the presence of pain. Moreover, due to pain, quality of life and return to work and usual activities were compromised in the acute phase following mTBI.

Paradoxically, psychophysiological pain testing and sensory perception (warm and cold) threshold determination did not differ between the presence or absence of pain. However, mTBI with pain patients were slower on a psychomotor vigilance task than mTBI without pain patients. Genetic polymorphism testing for BDNF showed that the majority of mTBI patients with pain had the val/val allele. No group difference was found for the COMT gene polymorphism.

Given the large number of mTBI patients seen in emergency departments and the cost for society of managing long-term post-concussion symptoms, it is critical to identify early predictors of chronic symptoms in order to facilitate screening for early intervention. The second objective of this study was to follow up mTBI patients at one year post-trauma. Nearly 40% agreed to participate in the follow-up, of whom 50% still suffered from pain. The results show that at one year post-trauma, mTBI patients with persistent or newly developed chronic pain also reported worse quality of life, persistent post-concussion symptoms, depression, and anxiety. More interestingly, three patients developed pain that was absent at the acute phase post-trauma. Of the possible explanations, we may posit that

pain at the acute phase was well controlled and therefore not reflected in the questionnaires or medical reports, or that traumatic brain injury did not play a role in pain development.

A high score on the Beck Anxiety Inventory during the acute phase predisposes to the persistence of pain, post-concussion symptoms, and depressive and anxiety symptoms at one year post-trauma. This interesting result underscores the importance of symptom management in the acute intervention window following traumatic brain injury. The most significant finding is that the BDNF val/val genotype is a predictor of persistent or newly developed chronic pain in an mTBI population. To our knowledge, this is the first study to demonstrate the BDNF role in chronic pain post-mTBI. Previous studies have reported that val/val polymorphism is associated with plasticity [96,165,180]. Neurotrophins, especially BDNF, play an important role in both the modulation of pain-related pathways and synaptic plasticity [96,167]. Its most known function is in the modulation of synaptic efficacy; i.e. long term potentiation; dendritic growth and synaptic formation and stabilization [165,181] One can therefore speculate a role for BDNF in synapses of the nociceptive pathway.

Other groups have performed quantitative sensory testing in traumatic brain injury patients, with conflicting results [73,75]. Heat pain stimulation by contact thermode is the most frequently used noxious stimulus in experimentally induced pain[182]. Defrin et al. found that trauma patients showed a lower heat sensation threshold when compared to healthy subjects. However, contrary to our results, they also found that TBI patients had higher cold and lower pain perception threshold [75]. These differences may be due to the age of the population studied and the severity of injuries. Moreover, compared to their study, our population presented mild traumatic brain injury only, with no moderate or severe injuries.

In addition, the PVT test sheds a revealing light on the interpretation of the results. Since quantitative sensory testing (QST) requires a relatively fast reaction time, mTBI patients with pain and a slower reaction time, might be more sensitive in detecting sensory perceptions and experimental heat pain.

A review of chronic pain after TBI concluded that the pain is independent of psychological disorders [39,66]. This controversial conclusion should be revisited, as chronic pain may be part of a larger family of post-concussion symptoms: rather than being isolated, it should be treated as part of a group of symptoms.

Limitations:

Our study has certain limitations that need to be addressed. Despite considerable efforts, only 38% of patients could be reached at the one-year follow-up. This low recruitment rate could have introduced a bias in the results, especially the percentage of patients with chronic pain. Moreover, we used the PVT to assess mean reaction time, although the PVT has been validated in sleep deprivation studies, and not in an mTBI population.

Conclusion:

More than 70% of mTBI patients suffer from pain at the acute phase post-trauma. The presence of pain appears to exacerbate psychological and mood disturbances such as depression, anxiety, catastrophizing, and post-traumatic stress disorder. Certain mTBI patients appear to have a genetic predisposition to develop pain following traumatic brain injury. Pain is also debilitating, because it prevents return to work and usual activities, and

it compromises quality of life. At one year post-trauma, pain remains in 50% of mTBI patients, which may be explained by high anxiety at the acute phase and BDNF polymorphism. Future investigations could focus on measures to mitigate anxiety, and on the consequences of better control of post-concussion symptoms in the acute phase post-trauma.

Table 1 : a) Psychological and questionnaires assessment at the acute phase

	mTBI with pain	mTBI without pain	p-value
Pain VAS	51.30 (24.87)	23.37 (31.63)	<0.000
Rivermead	21.98 (12.74)	13.17 (9.88)	0.003
SF-36	47.05 (16.42)	72.09 (16.96)	<0.000
Bodily pain SF-36	34.41 (17.76)	77.04 (19.68)	<0.000
Pain Catastrophizing Scale	15.21 (12.26)	9.88 (7.46)	0.05
BDI-II	12.91 (8.55)	5.48 (4.33)	<0.000
BAI	10.17 (9.28)	3.67 (3.14)	0.003
Impact of Event Scale	20.16 (16.58)	9.35 (7.71)	0.006

A student t-test comparison of pain VAS and questionnaires scores between mTBI with pain and mTBI without pain at the acute post-trauma phase (6 weeks). VAS : Visual Analog Scale. BDI-II: Beck Depression Inventory-II. BAI: Beck Anxiety Inventory.

b) <u>Genotype frequencies</u>

	Val/val	**Val/met**	**Met/met**
BDNF	43 (45.7%)	27 (28.7%)	3 (3.2%)
COMT	24 (25.5%)	30 (31.9%)	18 (19.1%)

*Genotype is unknown for 21 patients.

Table 2 : Psychological and questionnaires assessment at the chronic phase

	mTBI with pain	mTBI without pain	p-value
Pain VAS	36.18 (19.49)	6.25 (15.44)	<0.000
Rivermead	17.86 (11.88)	7.13 (6.80)	0.007
SF-36	67.00 (19.55)	88.18 (6.94)	0.005
Bodily pain SF-36	52.93 (25.37)	77.88 (20.42)	0.01
Pain Catastrophizing Scale	9.86 (9.35)	4.88 (5.01)	ns
BDI-II	10.22 (10.46)	2.55 (3.36)	0.02
BAI	8.67 (12.24)	2.18 (3.60)	0.04
Impact of Event Scale	11.78 (19.23)	5.36 (6.79)	ns

A student t-test comparison of pain VAS and questionnaires scores between mTBI with persistent and newly developed pain and mTBI without pain at one year post-trauma. VAS : Visual Analog Scale. BDI-II: Beck Depression Inventory-II. BAI: Beck Anxiety Inventory.

Table 3: Correlation matrix of variables in the acute phase (6 weeks)

		SF36	RPQ	BDI	BAI	IESR	PainVAS	MeanRT
SF36	Pearson Correlation	1	-.575	-.725	-.649	-.588	-.407	-.459
	Sig. (2-tailed)		.000	.000	.000	.000	.000	.000
RPQ	Pearson Correlation	-.575	1	.742	.709	.675	.183	.377
	Sig. (2-tailed)	.000		.000	.000	.000	.105	.001
BDI	Pearson Correlation	-.725	.742	1	.771	.690	.291	.485
	Sig. (2-tailed)	.000	.000		.000	.000	.010	.000
BAI	Pearson Correlation	-.649	.709	.771	1	.750	.253	.425
	Sig. (2-tailed)	.000	.000	.000		.000	.032	.000
IESR	Pearson Correlation	-.588	.675	.690	.750	1	.196	.426
	Sig. (2-tailed)	.000	.000	.000	.000		.086	.000
PainVAS	Pearson Correlation	-.407	.183	.291	.253	.196	1	.224
	Sig. (2-tailed)	.000	.105	.010	.032	.086		.058
MeanRT	Pearson Correlation	-.459	.377	.485	.425	.426	.224	1
	Sig. (2-tailed)	.000	.001	.000	.000	.000	.058	

RPQ: Rivermead post-concussion questionnaire. BDI: Beck Depression Inventory. BAI: Beck Anxiety Inventory. IESR: Impact of Event Scale Revised. VAS: Visual Analog Scale. Mean RT: Mean reaction time.

Table 4: Multiple regression for factors in acute phase that predispose to chronicity

	Model	Predictive variables	Beta	R square	p-value
Pain VAS at chronic	1	BAI	0.54	0.29	0.004
RPQ at chronic	1	BAI	0.65	0.42	<0.001
	2	BAI, warm	0.61; 0.45	0.62	<0.001
	3	BAI,warm, SF36	0.42; 0.46; 0.35	0.71	<0.001
	4	BAI, warm, SF36, sex	0.40; 0.36; 0.47; 0.29	0.77	<0.001
SF-36 at chronic	1	Bodily pain SF36	0.67	0.45	<0.001
	2	Bodily pain SF36, RTW	0.76; 0.45	0.65	<0.001
Bodily pain SF36 at chronic	1	Bodily pain SF36	0.54	0.29	0.007
	2	Bodily pain SF36, RTW	0.62; 0.39	0.44	0.002
	3	Bodily pain SF36, RTW, warm	0.60; 0.38; 0.36	0.56	<0.001
PCS at chronic	1	BAI	0.58	0.33	0.003
	2	BAI, sex	0.53; -0.38	0.47	<0.001
	3	BAI, sex, BDNF	0.64; -0.46; 0.34	0.57	<0.001
BDI at chronic	1	BAI	0.74	0.55	<0.001
	2	BAI, COMT	0.78; -0.32	0.64	<0.001
	3	BAI, COMT, warm	0.77; -0.41; 0.33	0.74	<0.001
BAI at chronic	1	BAI	0.62	0.38	<0.001
IESR at chronic	1	Warm	0.52	0.27	0.01

A multiple regression model of factors (predictive variables) in the acute post-trauma phase (6 weeks) that predispose to high scores on the pain VAS and questionnaires scores at one-year follow-up. VAS : Visual Analog Scale. BDI-II: Beck Depression Inventory-II. BAI: Beck Anxiety Inventory. COMT: Catechol-O-Methyl transferase. RPQ: Rivermead post-concussion questionnaire. PCS: Pain Catastrophizing Scale. IESR: Impact of Event Scale Revised. Warm: warm sensation temperature detection on the Quantitative Sensory Test. RTW: Return to work.

Table 5: Logistic regression for factors predisposing to persistent or new onset pain at follow-up

	Chi-square	Exp (Coef)	95% Lower	95% Upper	p-value
BDNF	4.97	0.03	0.001	0.65	0.03
Age	4.08	0.86	0.74	0.99	0.04
Sex (F)	3.00	94.75	0.55	16324.47	0.08
Pain VAS	2.34	1.08	0.98	1.18	0.13
RTW (yes)	2.19	0.04	0.0004	2.95	0.14
COMT	2.05	37.36	0.26	5328.34	0.15
Mean Reaction Time	0.13	0.99	0.95	1.03	0.72
Warm	0.78	0.92	0.52	1.63	0.77

A logistic binary regression model to predict factors in the acute post-trauma phase (6 weeks) that predispose to the persistence or new onset of pain at one-year follow-up. VAS :Visual Analog Scale. RTW: Return to work. Warm: warm sensation temperature detection on the Quantitative Sensory Test

Figure legends

Figure 1 : Quantitative sensory testing using the thermode at the acute phase of mTBI patients with and without pain. Three sensory modalities were tested: warm temperature detection, cold temperature detection and heat pain perception threshold detection on the upper left arm. Mean temperature detection represents the average temperature of five trials for each modality.

Figure 2 : Quantitative sensory testing using the thermode at the chronic phase of mTBI patients with and without pain. Three sensory modalities were tested: warm temperature detection, cold temperature detection and heat pain perception threshold detection on the upper left arm. Mean temperature detection represents the average temperature of five trials for each modality.

Figure 3: A comparison between acute and chronic questionnaire scores, pain VAS, mean reaction time on the PVT and temperature detection thresholds on the QST. A repeated measure ANOVA was used.

Figure 1 : Quantitative sensory testing at the acute phase (at 6 weeks)
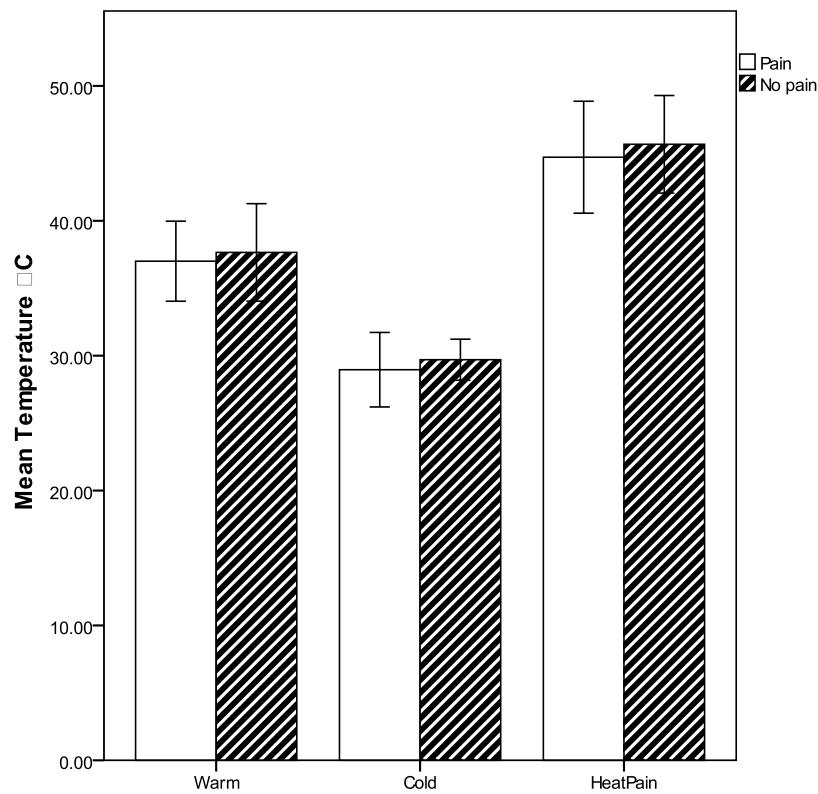
Figure 2: Quantitative sensory testing at the chronic phase (at one year)
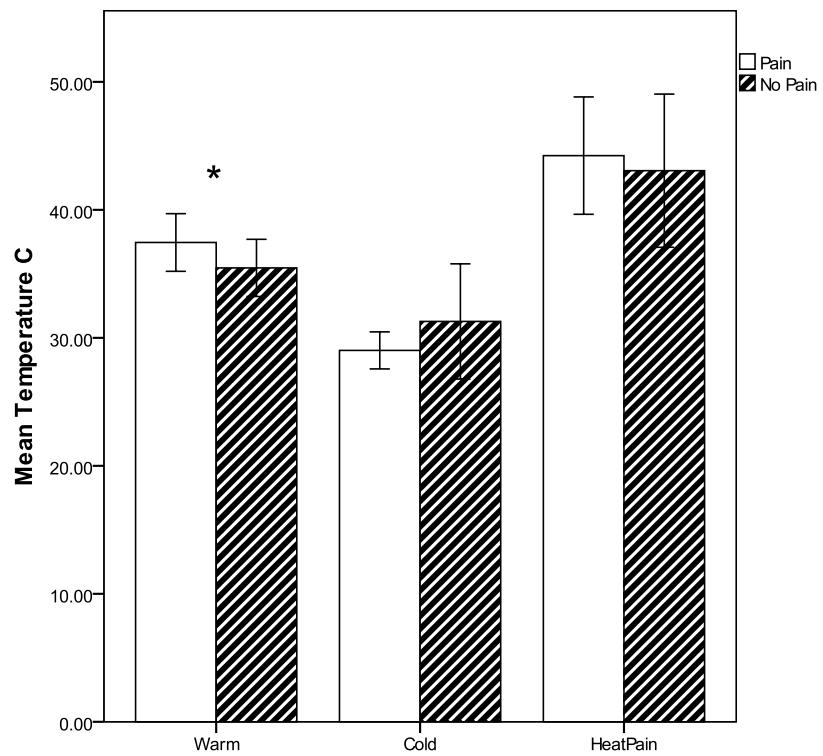
Figure 3 Evolution of questionnaire scores, Pain VAS, sensory modalities and reaction time

from the acute to chronic phase
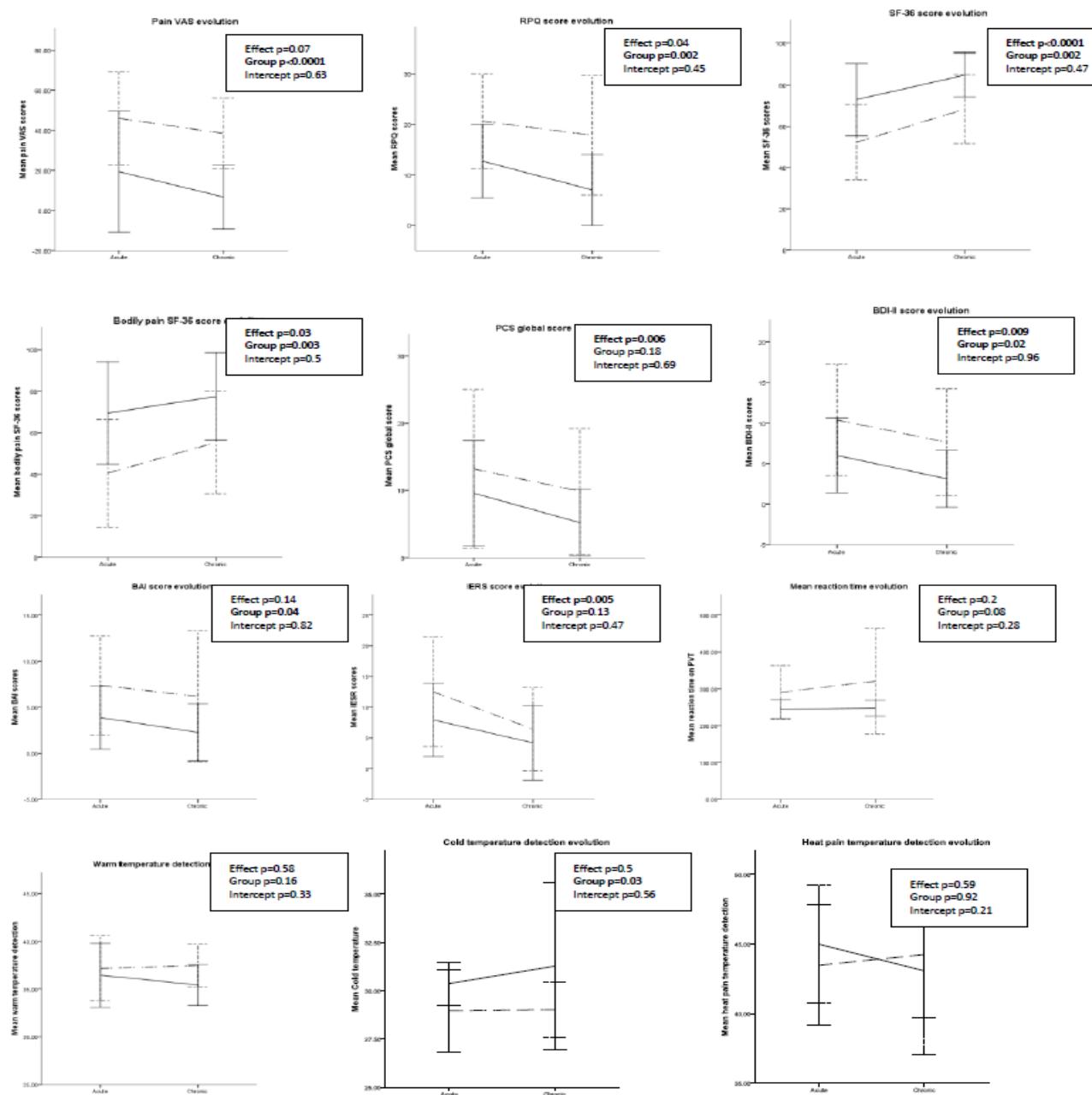
Reference list

1. Cassidy JD, Carroll LJ, Peloso PM*, et al. Incidence, risk factors and prevention of mild traumatic brain injury: Results of the WHO collaborating centre task force on mild traumatic brain injury.* J Rehabil Med, 2004. **36**(Suppl. 43): p. 28-60.

2. Feinstein A and Rapoport M. *Mild traumatic brain injury: the silent epidemic.* Can J Public Healt, 2000. **91**(5): p. 325-326.

3. Zumstein MA, Moser M, Mottini M*, et al. Long-term outcome in patients with mild traumatic brain injury: A prospective observational study.* J Trauma, 2011. **71**(1): p. 120-127.

4. Cancelliere C, Cassidy JD, Côté P*, et al. Protocol for a systematic review of prognosis after mild traumatic brain injury: an update of the WHO collaborating centre task force findings.* Syst Rev., 2012. **1**(1): p. 17.

5. Nampiaparampil DE. *Prevalence of chronic pain after traumatic brain injury: A systematic review.* JAMA, 2008. **300**(6): p. 711-719.

6. Otis JD, McGlinchey R, Vasterling JJ*, et al. Complicating factors associated with mild traumatic brain injury: Impact on pain and posttraumatic stress disorder treatment.* J Clin Psychol Med Settings, 2011. **18**: p. 145-154.

7. Côté P, Cassidy JD, Carroll LJ*, et al. A systematic review of the prognosis of acute whiplash and a new conceptual framework to synthesize the literature.* spine, 2001. **26**(19): p. E445-E458.

8. Scholten-Peeters GG, Verhagen AP, Bekkering GE*, et al. Prognostic factors of whiplash-associated disorders: a systematic review of prospective cohort studies.* Pain, 2003. **104**(1-2): p. 303-322.

9. Kamper SJ, Rebbeck TJ, Maher CG*, et al. Course and prognostic factors of whiplash: A systematic review and meta-analysis.* Pain, 2008. **138**: p. 617-629.

10. Sterling M, Jull G, and Kenardy J. *Physical and psychological factors maintain long-term predictive capacity post-whiplash injury.* Pain, 2006. **122**: p. 102-108.

11. Carroll LJ, Cassidy JD, and Côté P. *The role of pain coping strategies in prognosis after whiplash injury: Passive coping predicts slowed recovery.* Pain, 2006. **124**: p. 18-26.

12. Atherton K, Wiles NJ, Lecky FE*, et al. Predictors of persistent neck pain after whiplash injury.* Emerg Med J, 2006. **23**: p. 195-201.

13. Kasch H, Qerama E, Kongsted A*, et al. Deep muscle pain, tender points and recovery in acute whiplash patients: A 1-year follow-up study.* Pain, 2008. **140**: p. 65-73.

14. Young Casey C, Greenberg MA, Nicassio PM*, et al. Transition from acute to chronic pain and disability: A model including cognitive, affective, and trauma factors.* Pain, 2008. **134**(1): p. 69-79.

15. Stulemeijer M, van der Werf S, Borm GF*, et al. Early prediction of favourable recovery 6 months after mild traumatic brain injury.* J Neurol Neurosurg Psychiatry, 2008. **79**: p. 936-942.

16. Hoffman JM, Pagulayan KF, Zawaideh N*, et al. Understanding pain after traumatic brain injury: Impact on community participation.* Am J Phys Med Rehabil, 2007. **86**(12): p. 962-969.

17. Risdall JE and Menon DK. *Traumatic brain injury.* Philos Trans R Soc Lond B Biol Sci, 2011. **366**(1562): p. 241-250.

18. Defrin R, Ginzburg K, Solomon Z*, et al. Quantitative testing of pain perception in subjects with PTSD--implications for the mechanism of the coexistence between PTSD and chronic pain.* Pain, 2008. **138**(2): p. 450-459.

19. Defrin R, Gruener H, Schreiber S*, et al. Quantitative somatosensory testing of subjects with chronic post-traumatic headache: implications on its mechanisms.* Eur J Pain, 2010. **14**(9): p. 924-931.

20. Lu B. *BDNF and activity-dependent synaptic modulation.* Learn Mem, 2003. **10**(2): p. 86-98.

21. Kleim JA, Chan S, Pringle E, *et al. BDNF val66met polymorphism is associated with modified experience-dependent plasticity in human motor cortex.* Nat Neurosci, 2006. **9**(6): p. 735-737.

22. McAllister TW, Tyler AL, Flashman LA, *et al. Polymorphisms in the brain-derived neurotrophic factor gene influence memory and processing speed one month after brain injury.* J neurotrauma, 2012. **29**(6): p. 1111-1118.

23. Pezet S and McMahon SB. *Neurotrophins: Mediators and modulators of pain.* Annu Rev Neurosci, 2006. **29**: p. 507-538.

24. Vossen H, Kenis G, Rutten B, *et al. The genetic influence on the cortical processing of experimental pain and the moderating effect of pain status.* PLoS One, 2010. **5**: p. e13641.

25. Tammimaki A and Mannisto PT. *Catechol-O-methyltransferase gene polymorphism and chronic human pain: a systematic review and meta-analysis.* Pharmacogenet Genomics, 2012. **22**(9): p. 673-691.

26. Zubieta JK, Heitzeg MM, Smith YR, *et al. COMT val158met genotype affects u opioid neurotransmitter responses to a pain stressor.* Science, 2003. **299**: p. 1240-1243.

27. Carroll LJ, Cassidy JD, Holm L, *et al. Methodological issues and research recommendations for mild traumatic brain injury: The WHO collaborating centre task force on mild traumatic brain injury.* J Rehabil Med, 2004. **36**(Suppl. 43): p. 113-125.

28. ICHD. *The International Classification of Headache Disorders, 2nd edition.* Cephalalgia, 2004. **24**(Suppl 1).

29. Defrin R, Ohry A, Blumen N, *et al. Characterization of chronic pain and somatosensory function in spinal cord injury subjects.* Pain, 2001. **89**: p. 253-263.

30. Khoury S, Roy I, Giguère JF, *et al. Altered thermal heat sensation but not pain in mild traumatic brain injury patients: preliminary report on pre- and post-sleep assessments.* 2010. **Abstract IASP**(PM-039).

31.   Lim J and Dinges DF. *Sleep deprivation and vigilant attention.* Ann. N.Y. Acad. Sci., 2008. **1129**: p. 305-322.

32.   Dinges DF and Kribbs NB. *Performing while sleepy: effects of experimentally induced sleepiness*, in *Sleep, sleepiness and performance*, TH Monk, Editor. 1991, John Wiley: Winchester. p. 97-128.

33.   Ware JE, Snow KK, Kosinski M*, et al. The SF-36 health survey. Manual and interpretation guide.*, ed. Th institute. 1993, Boston, MA: New England Medical Center.

34.   Ware JE and Sherbourne CD. *The MOS 36-item short form health survey (SF-36). Conceptual framework and item selection.* Med Care, 1992. **30**: p. 473-483.

35.   King NS, Crawford S, Wenden FJ*, et al. The Rivermead Post Concussion Symptoms Questionnaire: a measure of symptoms commonly experienced after head injury and its reliability.* J Neurol, 1995. **242**: p. 587-592.

36.   Weiss DS and Marmar CR. *The impact of event scale-Revised*, in *Assessing Psychological Trauma and PTSD*, JP Wilson and TM Keane, Editors. 1997, Guilford Press: New York. p. 399-411.

37.   Beck AT, Ward CH, Mendelson M*, et al. An inventory for measuring depression.* Arch Gen Psychiatry, 1961. **4**: p. 561-571.

38.   Beck AT, Epstein N, Brown G*, et al. An inventory for measuring clinical anxiety: psychometric properties.* J Consult Clin Psychol, 1998. **56**: p. 893-897.

39.   Sullivan MJL, Bishop S, and Pivik J. *The pain catastrophizing scale: Development and validation.* Psychol Assess, 1995. **7**: p. 524-532.

40.   Stewart WF, Lipton RB, Whyte J*, et al. An international study to assess reliability of the Migraine Disability Assessment (MIDAS) score.* Neurology, 1999. **53**(5): p. 988-994.

41.   Melzack R. *The McGill Pain Questionnaire: Major properties and scoring methods.* Pain, 1975. **1**(277-299).

42. Cheeran B, Talelli P, Mori F*, et al. A common polymorphism in the brain-derived neurotrophic factor gene (BDNF) modulates human cortical plasticity and the response to rTMS.* J Physiol, 2008. **586**(23): p. 5717-5725.

43. Handwerker HO and Kobal G. *Psychophysiology of experimentally induced pain.* Physiol Rev, 1993. **73**(3): p. 639-671.

44. Hoge CW, McGurk D, Thomas JL*, et al. Mild traumatic brain injury in U.S. soldiers returning from Iraq.* N Engl J Med, 2008. **358**: p. 453-463.

Article 3: Mutations in BDNF gene affect recovery following mild traumatic brain injury

Manuscrit en préparation

Mutations in BDNF gene affect recovery following mild traumatic brain injury

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Abstract

Mild traumatic brain injury (mTBI) often leaves patients with post-concussion symptoms. Genetic predisposition is suggested to play a role in the poor prognosis following mTBI. The first aim of this study is to replicate findings of previous research on suggested gene polymorphisms known to play a role in poor outcome following mTBI in a homogeneous cohort of 83 mTBI patients and 193 controls. The second aim is to analyse protein expression in lymphoblastoid cell lines of mTBI patients for functional polymorphisms of the most conclusive gene.

Single nucleotide genotyping (SNP) was performed in 13 genes previously studied in the literature. A western blot was performed for protein expression. Amongst all SNPs genotyped, quantitative trait loci analysis showed that eight SNPs of the Brain-Derived Neurotrophic Factor (BDNF) were the most significantly associated with poor scores on the Rivermead Post-concussion symptoms questionnaire. BDNF protein expression was also modified in lymphoblastoid cell lines of mTBI patients with the met allele of the val66met polymorphism. BDNF protein expression in lymphoblastoid cell lines of mTBI patients and controls showed that polymorphisms in BDNF play an important role in the poor outcome following mTBI.

Introduction

Traumatic brain injury (TBI), even mild, is a significant burden on society, as patients are left with long term post-concussion symptoms [183-184]. The clinical course and prognosis of mTBI patients differs among individuals. Several mechanical, psychological and monetary compensation factors were previously shown to predispose to the chronicity of symptoms, however these factors alone couldn't provide an explanation to all cases with poor prognosis [163,185-186]. A genetic predisposition was suggested to explain variability in the outcome of individuals following mTBI. In earlier studies, numerous candidate genes were suggested as playing a predisposing role [90,187]. Clinical phenotypes following trauma were associated with several polymorphisms in apolipoprotein E (APOE), interleukin-6 (IL-6), catechol-O-methyltransferase (COMT), and brain-derived neurotrophic factor (BDNF) with conflicting results [92,105,109,112].

A better outcome following mild TBI implies that the brain undergoes, or is susceptible to undergo plasticity to restore connexions that existed prior to diffuse axonal injury, hence pre-trauma. BDNF is the most abundant neurotrophin produced in the brain. It is first produced as a precursor long molecule, proBDNF, which is then cleaved into mature BDNF. Both proteins are released at central synapses [96,188]. It is an interesting molecule in the context of TBI as it plays an important role in synaptic plasticity [189]. The BDNF gene, located on chromosome 11 bears the most commonly studied single nucleotide polymorphism (SNP) Val66Met (rs6265) where a guanine to adenosine substitution leads to a missense mutation, a valine to methionine substitution at codon 66

(Val66Met) in the 5′ proregion [97]. This substitution was reported to reduce intracellular trafficking and activity-dependent secretion of BDNF [97].

Many associations between BDNF polymorphisms and outcomes following TBI were investigated in previous studies [98-99,111-112]. Variations in BDNF was shown to explain general intelligence on cognitive and executive tasks many years after combat-related penetrating TBI [99,112]. Another study found an association between memory and processing speed following mTBI and polymorphisms in the BDNF gene [98]. Other literatures shows that the val/val genotype is associated with experience dependent plasticity in the motor cortex [165]. Carriers of the Met-allele showed significantly higher levels of BDNF in multiple sclerosis patients in comparison with healthy controls [190]. Therefore, genetic susceptibility in factors regulating brain plasticity may play an important role in recovery following mTBI.

The study hypothesis is that BDNF Val66Met polymorphism is an important predictor of poor prognosis following mTBI.

In this study, two objectives were set: The first objective is to verify whether known polymorphisms in previously studied genes predispose to a poor outcome following mTBI. The second objective is to test whether SNP variants in BDNF gene impacted outcome following mild TBI and if the val66met polymorphism affects gene expression in a mTBI population with poor prognosis.

Materials and Methods

Subjects

The study was approved by the ethics committee of the Sacré-Coeur hospital (wheresubjects were recruited) and the Centre Hospitalier de l'Université de Montréal (CHUM) (where samples were stored and analysed). All patients gave informed consent before data and blood were collected. A cohort of 83 mild TBI patients were recruited and accepted to take part of this study. Mild TBI was diagnosed according to the 2004 WHO task force criteria [12]. Post-concussive symptoms were assessed using the Rivermead post concussion symptoms questionnaire at the acute phase post-trauma (less than 3 month) [34]. This is the most widely used validated questionnaire to measure post-concussive symptoms such as headache, dizziness, and concentration problems. It contains 16 items that assess the presence and severity of cognitive, emotional, and somatic complaints on a five-point scale (1=no problem to 5=severe problem). The overall score is obtained by summing all scores. Higher scores indicate more severe post-concussion symptoms [34]. Control blood samples (n=193) from individuals with no history of TBI or other neurological diseases were used for comparison. Genomic DNA was extracted from blood using the Puregene DNA kit using the manufacturer's protocol (Gentra System, USA).

Gene search and genotyping

From the literature, a list of candidate genes were identified as being relevant to traumatic brain injury. This list included the apolipoprotein E (ApoE), ApoE promoter, Poly[ADP

ribose] polymerase 1 (PARP-1), the following interleukins: Il-1b, Il6, Il-1a, Angiotensin-converting enzyme (ACE), Catechol O-methyl transferase (COMT), p53, Dopamine receptor D2 (DRD2), brain-derived neurotrophic factor (BDNF), Neurofilament, heavy polypeptide (NEFH), and neuroglobin (NGB) (Table 1). Using HapMap Genome Browser release #28 based on data available on August 2010, SNP and tagSNP were identified for each gene. SNP and tagSNP were retained if they have a minor allelic frequency of at least 20% in the CEU population or if they were necessary to identify known mutations. Genotyping of all SNPs was performed using Sequenom IPLEX Gold technology, according to manufacturer instruction, at McGill University and Génome Québec Innovation Center.

Protein extraction and fractionation

Lymphoblast cell lines were derived using standard methods from human whole blood extracted from patients and controls. Cells were transferred to 15 mL falcon tubes and pelleted by centrifugation at 1500 rpm for 5 minutes at room temperature followed by a PBS wash under the same conditions, and a subsequent PBS wash and centrifugation in 1.5 mL eppendorf tubes. The pellet was re-suspended in 200µl SUB (4.8g urea; 250µL SDS 20%; 200µL β-mercaptoethanol, H_2O), chilled on ice for 20 minutes and then sonicated. Following this step, the tubes were centrifuged at 12000 rpm for 15 minutes at 4°C.

Western blot

A total of 25 µg of each protein sample was resolved on a 12% polyacrylamide gel and transferred to a Nitrocellulose membrane. Membranes were blocked in PBS 0.2% Tween

with 5% non-fat dry milk for three hours followed by incubation with the primary antibody (mouse monoclonal MM0109-3D44; Abcam) at a dilution of 1:1000 in PBS 0.2% Tween with non-fat dry milk overnight at 4°C. Following washing, the membranes were incubated with the secondary antibody, which is a donkey anti-mouse HRP (Jackson ImmunoResearch; 1:10,000) in PBS 0.2% Tween with non-fat dry milk 1 hour at room temperature. An ECL kit was used for detection followed by a 10-minutes exposure.

Statistical analysis

Association analysis was performed using the PLINK version 1.07-DOS (http://pngu.mgh.harvard.edu/~purcell/PLINK/) [191]. Single SNP p-values were obtained using χ^2 tests. A genomic-control correction was applied for multiple testing. A Quantitative Trait Loci (QTL) analysis was used for an association between SNP and the Rivermead post-concussion symptom scores.

Table 1: Gene list SNPs and polymorphisms genotyped

Genes	SNP/polymorphism	Chromosome	Location	MAF(%)	Ref
ApoE	rs429358	19	45411941	11.4	[87]
	rs7412	19	45412079	8.6	
ApoE promoter	rs405509	19	45408836	47.2	[104]
	rs449647	19	45408564	18.1	
PARP-1	rs1109032	1	226561403	17	[106]
	rs3219090	1	226564691	31.4	
	rs3219119	1	226556443	31.5	
	rs2271347	1	226549498	23.6	
Il1a	rs1800587	2	113542960	27.8	[91]
Il1b	rs1143634	2	113590390	22.4	[107]
	rs16944	2	113594867	31.6	
Il6	rs1800795	7	22766645	41.6	[92]
ACE	rs4318	17	61562373	0.55	[108]
	rs4364	17	61574662	0	
COMT	rs75012854	22	19950164	0	[109]
	rs4680	22	19951271	49.9	
p53	rs1042522	17	7579472	22.9	[110]
DRD2	rs1800497	11	113270828	18.2	[113]
BDNF	rs11030101	11	27680744	45.9	[98]
	rs11030102	11	27681596	23.1	
	rs11030104	11	27684517	21.7	
	rs11030107	11	27694835	23.1	
	rs7103411	11	27700125	21.3	
	rs7127507	11	27714884	30	
	rs6265	11	27679916	20.9	
	rs7124442	11	27677041	30	
	rs12273363	11	27744859	20.3	
	rs1519480	11	27675712	31.1	
	rs7934165	11	27731983	47.4	
	rs11030121	11	27736207	31.7	
	rs908867	11	27745764	8.4	
NEFH	rs165602	22	29886043	13.7	[114]
	rs3815335	22	29881468	30	
NGB	rs3783988	14	77734580	22.5	[192]

Results

Objective 1) Genotyping of polymorphisms in genes previously cited in the literature are presented in table 1. A total of 34 SNP from 13 genes were genotyped using sequenom IPLEX gold technology with an average call rate of 99%. SNPs chosen in the ACE gene (rs4318 and rs4364) and rs75012851 in the COMT gene failed as expected because their minor allelic frequency is close to zero in our population. We chose to keep them in the analysis as they were cited in previous literature. An mTBI and control group association between SNPs is presented in table 2. After adjustment for multiple testing, none of the SNPs showed a significant group association meaning that our two groups were homogeneous in genotype distribution.

A preliminary analysis of the association between SNP and category of mTBI patients with high and low Rivermead scores is presented in table 3. Eight SNP of the BDNF gene were shown to be statistically significant, however, after correction for multiple testing, only rs11030121 and rs12273363 were shown to be associated with high scores on the Rivermead questionnaire with an odds ratio of 2.4 and 2.3 respectively. A QTL analysis with continuous scores of the Rivermead confirmed previously cited results of the BDNF gene as eight SNP were significantly associated with poor outcome following mTBI after correction for multiple testing with $p<0.04$ (table 4). All SNPs were situated in a 69 Mbp distance of each other.

Objective 2) In the second part of the study, a western blot of BDNF protein expression from lymphoblastoid cell lines of mTBI and control subjects is shown in figure 1. For each group, a val/val, val/met and met/met sample was chosen. A 75, 50 and 30 KD

band appears for all subjects and phenotypes with different levels of expression. The val/val genotype also reveals a 25 KD band. In the mTBI subjects with the val/met and met/met genotype, and not in the mTBI val/val and controls, a faint band located at 20 KD position is present.

Table 2- Minor allelic frequencies of SNP in mTBI and control subjects

Gene	SNP	BP position	Allele 1	mTBI MAF	Controls MAF	Allele 2	Chi-Square	OR	P-value
PARP-1	rs2271347	226549498	T	0.25	0.2422	C	0.03732	1.043	0.8468
	rs3219119	226556443	A	0.3	0.3047	T	0.01174	0.978	0.9137
	rs1109032	226561403	A	0.1875	0.138	T	2.139	1.441	0.1436
	rs3219090	226564691	A	0.3	0.3047	G	0.01174	0.978	0.9137
Il1A	rs1800587	113542960	A	0.2625	0.2708	G	0.03994	0.9583	0.8416
Il-1B	rs1143634	113590390	A	0.2062	0.224	G	0.2072	0.9004	0.6489
	rs16944	113594867	A	0.2812	0.3099	G	0.4401	0.8714	0.5071
Il6*	**rs1800795**	**22766645**	**G**	**0.4875**	**0.3958**	**C**	**3.889**	**1.452**	**0.04861**
BDNF	rs1519480	27675712	C	0.3063	0.3229	T	0.1447	0.9256	0.7037
	rs7124442	27677041	C	0.2975	0.3115	T	0.1036	0.9358	0.7475
	rs6265	27679916	T	0.2375	0.1927	C	1.386	1.305	0.2391
	rs11030101	27680744	A	0.4313	0.4688	T	0.6399	0.8593	0.4238
	rs11030102	27681596	G	0.2437	0.237	C	0.02847	1.038	0.866
	rs11030104	27684517	C	0.2313	0.2005	T	0.6438	1.199	0.4223
	rs11030107	27694835	G	0.2388	0.2407	A	0.002029	0.9894	0.9641
	rs7103411	27700125	G	0.2313	0.1927	A	1.033	1.26	0.3095
	rs7127507	27714884	C	0.3125	0.3099	T	0.003577	1.012	0.9523
	rs7934165	27731983	T	0.4375	0.487	C	1.109	0.8194	0.2922
	rs11030121	27736207	T	0.3312	0.3272	C	0.008283	1.018	0.9275
	rs12273363	27744859	C	0.2125	0.2109	T	0.001654	1.009	0.9676
	rs908867	27745764	A	0.09375	0.08333	G	0.1553	1.138	0.6936
DRD2	rs1800497	113270828	A	0.1392	0.1885	G	1.885	0.6965	0.1698
NGB*	**rs3783988**	**77734580**	**G**	**0.2687**	**0.1953**	**A**	**3.586**	**1.514**	**0.05827**
p53	rs1042522	7579472	C	0.2375	0.2083	G	0.5656	1.184	0.452
ACE	rs4318	61562373	C	0	0.005208	T	0.8364	0	0.3604
	rs4364	61574662	0	0	0	C	NA	NA	NA
ApoE promoter	rs449647	45408564	A	0.141	0.1909	T	1.881	0.696	0.1702
	rs405509	45408836	G	0.5062	0.4766	T	0.3985	1.126	0.5279
ApoE	rs429358	45411941	C	0.1507	0.1114	T	1.472	1.415	0.225
	rs7412	45412079	T	0.08228	0.09511	C	0.2194	0.853	0.6395
COMT	rs75012854	19950164	0	0	0	A	NA	NA	NA
	rs4680	19951271	T	0.443	0.5079	C	1.879	0.7709	0.1704
NEFH	rs3815335	29881468	T	0.2625	0.3151	C	1.489	0.7736	0.2223
	rs165602	29886043	C	0.1562	0.1302	A	0.6444	1.237	0.4221

*After correction for multiple testing, no significance is found between the two groups for Il6 and NGB.
Highlighted results in red show the statistically significant results.
MAF: minor allelic frequency; SNP: Single nucleotide polymorphism; OR: odds ratio

Table 3- SNP association with high and low Rivermead scores

Gene	SNP	BP position	Allele 1	mTBI MAF	Controls MAF	Allele 2	Chi-square	OR	P-value	Adjusted
PARP-1	rs2271347	226549498	T	0.2632	0.2431	C	0.07712	1.112	0.7812	
	rs3219119	226556443	A	0.2632	0.3063	T	0.3117	0.8088	0.5767	
	rs1109032	226561403	A	0.1316	0.1542	T	0.1393	0.8314	0.709	
	rs3219090	226564691	A	0.2632	0.3063	G	0.3117	0.8088	0.5767	
Il1A	rs1800587	113542960	A	0.2105	0.2727	G	0.6965	0.7111	0.404	
Il-1B	rs1143634	113590390	A	0.1579	0.2233	G	0.8853	0.6521	0.3468	
	rs16944	113594867	A	0.2632	0.3043	G	0.2848	0.8163	0.5936	
Il6	rs1800795	22766645	G	0.5263	0.415	C	1.794	1.566	0.1804	
BDNF	rs1519480	27675712	C	0.4737	0.3063	T	4.565	2.038	0.03264	0.079
	rs7124442	27677041	C	0.4737	0.2948	T	5.308	2.153	0.02123	0.058
	rs6265	27679916	T	0.1579	0.2095	C	0.5754	0.7075	0.4481	
	rs11030101	27680744	A	0.3421	0.4664	T	2.2	0.5949	0.138	
	rs11030102	27681596	G	0.3947	0.2273	C	5.45	2.217	0.01956	0.055
	rs11030104	27684517	C	0.1579	0.2134	T	0.6583	0.691	0.4172	
	rs11030107	27694835	G	0.4062	0.2292	A	5.154	2.301	0.02319	0.062
	rs7103411	27700125	G	0.1579	0.2075	A	0.5357	0.7161	0.4642	
	rs7127507	27714884	C	0.4474	0.3004	T	3.565	1.885	0.059	0.12
	rs7934165	27731983	T	0.3158	0.4842	C	4.022	0.4917	0.04492	0.099
	rs11030121	**27736207**	**T**	**0.5263**	**0.3135**	**C**	**7.257**	**2.433**	**0.007064**	**0.027**
	rs12273363	**27744859**	**C**	**0.3684**	**0.1996**	**T**	**6.042**	**2.339**	**0.01397**	**0.043**
	rs908867	27745764	A	0.1316	0.083	G	1.057	1.674	0.304	
DRD2	rs1800497	113270828	A	0.1579	0.1753	G	0.07443	0.8821	0.785	
NGB	rs3783988	77734580	G	0.2368	0.2154	A	0.09554	1.13	0.7573	
p53	rs1042522	7579472	C	0.2105	0.2174	G	0.009807	0.96	0.9211	
ACE	rs4318	61562373	C	0	0.003953	T	0.1508	0	0.6978	
	rs4364	61574662	0	0	0	C	NA	NA	NA	
ApoE promoter	rs449647	45408564	A	0.1316	0.1796	T	0.5602	0.6921	0.4542	
	rs405509	45408836	G	0.3421	0.496	T	3.353	0.5283	0.06707	
ApoE	rs429358	45411941	C	0.1579	0.1201	T	0.465	1.374	0.4953	
	rs7412	45412079	T	0.02632	0.09631	C	2.083	0.2536	0.149	
COMT	rs75012854	19950164	0	0	0	A	NA	NA	NA	
	rs4680	19951271	T	0.4474	0.492	C	0.282	0.8357	0.5954	
NEFH	rs3815335	29881468	T	0.2368	0.3043	C	0.7675	0.7094	0.381	
	rs165602	29886043	C	0.1579	0.1364	A	0.1379	1.188	0.7104	

After correction for multiple testing, statistically significant results are highlighted in bold red.
MAF: minor allelic frequency; SNP: Single nucleotide polymorphism; OR: odds ratio

Table 4- QTL analysis of SNP and Rivermead scores in mTBI patients

Gene	SNP	BP position	BETA	SE	R^2	T	P-value
PARP-1	rs2271347	226549498	2.364	2.318	0.01223	1.02	0.3106
	rs3219119	226556443	-0.04779	2.226	5.49E-06	-0.02147	0.9829
	rs1109032	226561403	-1.105	2.802	0.001849	-0.3945	0.6942
	rs3219090	226564691	-0.04779	2.226	5.49E-06	-0.02147	0.9829
Il1A	rs1800587	113542960	1.413	2.469	0.003881	0.5721	0.5688
Il1B	rs1143634	113590390	1.883	2.504	0.006687	0.752	0.4542
	rs16944	113594867	-0.04526	2.403	4.22E-06	-0.01884	0.985
Il6	rs1800795	22766645	1.469	2.071	0.005957	0.7095	0.48
BDNF	**rs1519480**	**27675712**	**6.006**	**2.217**	**0.08038**	**2.71**	**0.008162**
	rs7124442	**27677041**	**6.564**	**2.26**	**0.09228**	**2.905**	**0.004709**
	rs6265	**27679916**	**-5.429**	**2.496**	**0.05334**	**-2.176**	**0.0324**
	rs11030101	27680744	-1.06	2.114	0.002985	-0.5015	0.6173
	rs11030102	**27681596**	**5.251**	**2.261**	**0.06035**	**2.323**	**0.02261**
	rs11030104	**27684517**	**-5.804**	**2.485**	**0.06097**	**-2.335**	**0.02191**
	rs11030107	**27694835**	**5.052**	**2.441**	**0.05769**	**2.07**	**0.04213**
	rs7103411	**27700125**	**-6.272**	**2.301**	**0.08124**	**-2.725**	**0.007814**
	rs7127507	**27714884**	**6.684**	**2.331**	**0.08917**	**2.868**	**0.005226**
	rs7934165	27731983	-1.911	2.093	0.009824	-0.9129	0.3639
	rs11030121	**27736207**	**6.749**	**2.102**	**0.1093**	**3.211**	**0.001873**
	rs12273363	**27744859**	**6.001**	**2.49**	**0.06465**	**2.41**	**0.01815**
	rs908867	27745764	5.365	3.535	0.0267	1.518	0.1328
DRD2	rs1800497	113270828	2.488	2.852	0.009082	0.8722	0.3856
NGB	rs3783988	77734580	0.3521	2.33	0.0002717	0.1511	0.8803
p53	rs1042522	7579472	-0.2582	2.773	0.0001032	-0.09313	0.926
ACE	rs4318	61562373	NA	NA	NA	NA	NA
	rs4364	61574662	NA	NA	NA	NA	NA
APOE promoter	rs449647	45408564	-1.059	2.944	0.001575	-0.3597	0.72
	rs405509	45408836	0.6364	2.092	0.0011	0.3041	0.7618
APOE	rs429358	45411941	-1.487	3.066	0.003126	-0.485	0.6291
	rs7412	45412079	-2.391	3.968	0.004463	-0.6026	0.5484
COMT	rs75012854	19950164	NA	NA	NA	NA	NA
	rs4680	19951271	0.02083	2.016	1.29E-06	0.01034	0.9918
NEFH	rs3815335	29881468	-3.303	2.242	0.02518	-1.473	0.1445
	rs165602	29886043	-1.188	3.193	0.001646	-0.3722	0.7107

Results highlighted in bold red are statistically significant after correction for multiple testing.
SE: Standard error; SNP: Single nucleotide polymorphism

Figure 1 – Western blot of BDNF expression in lymphoblasts
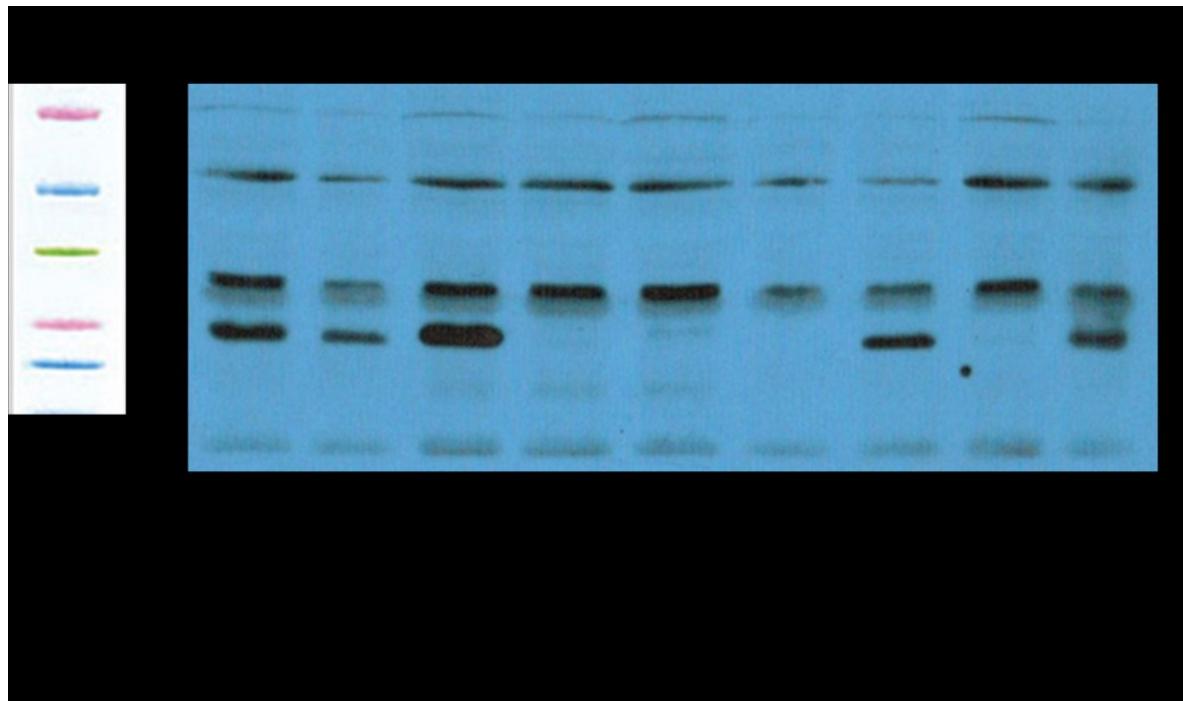
Legend: The first 5 columns are derived from lymphoblasts of mTBI patients, the last 4 columns are from controls. The molecular masses used 20, 25, 37, 50 and 75 starting from the bottom.

Discussion

In this study, the BDNF gene seems to be the only gene to contain polymorphisms important for predicting outcome following mild TBI. All the other genes listed above and previously investigated in the literature were not significantly related to poor outcome in our population. Although previous studies have found a role for the val66met polymorphism in cognitive, learning and memory functions following mild TBI, other SNPs in the same gene also seem important [98,112]. The SNPs found to be related to a poor outcome following mild TBI as defined by a high Rivermead score are close in position. It is still to be determined if they code for functional domains of the protein BDNF. This finding is consistent with our hypothesis that BDNF plays an important role in the outcome following mild TBI.

BDNF is a 12.4 kDa protein expressed in neurones but also outside of the nervous system. The role of the BDNF protein in the poor outcome following mild TBI is plausible as it is involved in many of the post-concussion symptoms, for example sleep disturbances and mood disorders. It was previously suggested that homeostatic SWA in subsequent sleep is proportional to cortical BDNF expression and prior learning during wakefulness [193]. Lower serum and hippocampal BDNF concentrations were found in rats who develop depressive behavior when subjected to stressors [194].

As the val66met polymorphism was shown to be important in many previous studies in TBI, a western blot of BDNF with different polymorphisms val/val, val/met and met/met was performed in lymphoblastoid cell lines of mTBI and control subjects. The result clearly shows a band in the val/met and met/met of mTBI cells that is not present in

the others. The interpretation of this finding is interesting as a previous unpublished study from our groups has shown that the mild TBI patients carrying the val/val polymorphism have a worse long-term outcome than the val/met or met/met. The BDNF met allele was previously suggested as being protective against psychiatric disorders by acting through a modified proBDNF [189]. The met phenotype is expressed differently in pathological complex conditions and under normal circumstances [99]. Therefore, this expressed protein might be playing a neuroprotective role and is only expressed following trauma.

Another possiblility is that this polymorphism affects the proBDNF protein. ProBDNF is expressed not only intracellularly but also extracellularly. It has the ability to bind to p75 receptor and induce long-term depression, whereas mature BDNF binds to TrkB inducing long-term potentiation and cell survival [195].

In conclusion, other experiments aiming to identify the role and nature of this protein will follow. Moreover, the SNP polymorphisms found in BDNF need to be further characterized in order to determine if they affect protein expression.

References

Ariza, M., M. D. Matarin, et al. (2006). "Influence of Angiotensin-converting enzyme polymorphism on neuropsychological subacute performance in moderate and severe traumatic brain injury." J Neuropsychiatry Clin Neurosci **18**(1): 39-44.

Ariza, M., R. Pueyo, et al. (2006). "Influence of APOE polymorphisme on cognitive and behavioural outcome in moderate and severe traumatic brain injury." J Neurol Neurosurg Psychiatry **77**: 1191-1193.

Bagnato, S., L. Minafra, et al. (2012). "Brain-Derived Neurotrophic Factor (Val66Met) polymorphism does not influence recovery from a post-traumatic vegetative state: a blinded retrospective multi-centric study." J neurotrauma **29**(11): 2050-2059.

Blugeot, A., C. Rivat, et al. (2011). "Vulnerability to depression: from brain neuroplasticity to identification of biomarkers." J Neurosc **31**(36): 12889-12899.

Carroll, L. J., J. D. Cassidy, et al. (2004). "Methodological issues and research recommendations for mild traumatic brain injury: The WHO collaborating centre task force on mild traumatic brain injury." J Rehabil Med **36**(Suppl. 43): 113-125.

Dardiotis, E., S. Grigoriadis, et al. (2012). "Genetic factors influencing outcome from neurotrauma." Curr Opin Psychiatry **25**(3): 231-238.

Dellavalle, B., C. Hempel, et al. (2010). "In vivo expression of neuroglobin in reactive astrocytes during neuropathology in murine models of traumatic brain injury, cerebral malaria, and autoimmune encephalitis." Glia **58**: 1220-1227.

Egan, M. F., M. Kojima, et al. (2003). "The BDNF Val66Met polymorphism affects activity-dependent secretion of BDNF and human memory and hippocampal function." Cell **112**: 257-269.

Huber, R., G. Tononi, et al. (2006). "Exploratory behavior, cortical BDNF, and sleep homeostasis." Sleep **30**(2): 129-139.

Ingebrigtsen, T., K. Waterloo, et al. (1998). "Quantification of post-concussion symptoms 3 months after minor head injury n 100 consecutive patients." J Neurol **245**(9): 609-612.

King, N. S., S. Crawford, et al. (1995). "The Rivermead Post Concussion Symptoms Questionnaire: a measure of symptoms commonly experienced after head injury and its reliability." J Neurol **242**: 587-592.

Kleim, J. A., S. Chan, et al. (2006). "BDNF val66met polymorphism is associated with modified experience-dependent plasticity in human motor cortex." <u>Nat Neurosci</u> **9**(6): 735-737.

Krueger, F., M. Pardini, et al. (2011). "The role of the Met66 brain-derived neurotrophic factor allele in the recovery of executive functioning after combat-related traumatic brain injury." <u>J Neurosc</u> **31**(2): 598-606.

Leibrock, J., F. Lottspeich, et al. (1989). "Molecular cloning and expression of brain-derived neurotrophic factor." <u>Nature</u> **341**(6238): 149-152.

Liguori, M., F. Fera, et al. (2009). "A longitudinal observation of brain-derived neurotrophic factor mRNA levels in patients with relapsing-remitting multiple sclerosis." <u>Brain Res</u> **1256**: 123-128.

Lipsky, R. H. and A. M. Marini (2007). "Brain-derived neurotrophic factor in neuronal survival and behavior-related plasticity." <u>Ann N Y Acad Sci</u> **1122**: 130-143.

Lipsky, R. H., M. B. Sparling, et al. (2005). "Association of COMT Val158Met genotype with executive functioning following traumatic brain injury." <u>J Neuropsychiatry Clin Neurosci</u> **17**(4): 465-471.

Lu, B. (2003). "BDNF and activity-dependent synaptic modulation." <u>Learn Mem</u> **10**(2): 86-98.

Martinez-Lucas, P., J. Moreno-Cuesta, et al. (2005). "Relationship between the Arg72Pro polymorphism of p53 and outcome for patients with traumatic brain injury." <u>Intensive Care Med</u> **31**: 1168-1173.

McAllister, T. W. (2010). "Genetic factors modulating outcome after neurotrauma." <u>PM&R</u> **2**(Suppl 12): S241-S252.

McAllister, T. W., L. A. Flashman, et al. (2008). "Single nucleotide polymorphisms in ANKK1 and dopamine D2 receptor gene affect cognitive outcome shortly after traumatic brain injury: a replication and extension study." <u>Brain Inj</u> **22**(9): 705-714.

McAllister, T. W., A. L. Tyler, et al. (2012). "Polymorphisms in the brain-derived neurotrophic factor gene influence memory and processing speed one month after brain injury." <u>J neurotrauma</u> **29**(6): 1111-1118.

McDevitt, J. K., R. T. Tierney, et al. (2011). "Neuronal structure protein polymorphism and concussion in college athletes." <u>Brain Inj</u> **25**(11): 1108-1113.

Minambres, E., A. Cemborain, et al. (2003). "Correlation between transcranial interleukin-6 gradient and outcome in patients with acute brain injury." <u>Crit Care Med</u> **31**(3): 933-938.

Mott, T. F., M. L. McConnon, et al. (2012). "Subacute to chronic mild traumatic brain injury." <u>Am Fam Physician</u> **86**(11): 1045-1051.

Purcell, S., B. Neale, et al. (2007). "PLINK: A tool set for whole-genome association and population-based linkage analyses." <u>Am J Hum Genet</u> **81**(3): 559-575.

Radanov, B. P., G. di Stefano, et al. (1991). "Role of psychosocial stress in recovery from common whiplash." <u>Lancet</u> **338**(8769): 712-715.

Rostami, E., F. Krueger, et al. (2011). "BDNF polymorphism predicts general intelligence after penetrating traumatic brain injury." <u>PLoS One</u> **6**(11): e27389.

Sarnaik, A. A., Y. P. Conley, et al. (2010). "Influence of PARP-1 polymorphisms in patients after traumatic brain injury." <u>J neurotrauma</u> **27**(3): 465-471.

Silverberg, N. D. and G. L. Iverson (2011). "Etiology of the post-concussion syndrome: Physiogenesis and psychogenesis revisited." <u>Neurorehabil</u> **29**(4): 317-329.

Stulemeijer, M., S. van der Werf, et al. (2008). "Early prediction of favourable recovery 6 months after mild traumatic brain injury." <u>J Neurol Neurosurg Psychiatry</u> **79**: 936-942.

Tanriverdi, T., M. Uzan, et al. (2006). "Lack of association between the IL1A (-889) polymorphism and outcome after head injury." <u>Surg Neurol</u> **65**: 7-10.

Terrell, T. R., R. M. Bostick, et al. (2008). "APOE, APOE promoter, and Tau genotypes and risk for concussion in college athletes." <u>Clin J Sport Med</u> **18**(1): 10-17.

Tierney, R. T., J. L. Mansell, et al. (2010). "Apolipoprotein E genotype and concussion in college athletes." <u>Clin J Sport Med</u> **20**(6): 464-468.

Uzan, M., T. Tanriverdi, et al. (2005). "Association between interleukin-1 beta (IL-1b) gene polymorphism and outcome after head injury: an early report." <u>Acta Neurochir</u> **147**: 715-720.

Woo, N. H., H. K. Teng, et al. (2005). "Activation of p75NTR by proBDNF facilitates hippocampal long-term depression." <u>Nat Neurosci</u> **8**(8): 1069-1077.

Discussion

Dans le premier article de la présente thèse, le sommeil suite à un TCCL en phase aigue (de moins de trois mois) a été étudié en laboratoire du sommeil. Tel qu'attendu on a répliqué les résultats des études précédentes, soit que la macrostructure (architecture) du sommeil est conservée même si les sujets rapportent des plaintes de mauvais sommeil et donc la persistance du paradoxe. Nous avons cherché à comprendre cette dichotomie. L'analyse quantitative de l'électroencéphalographie (qEEG) nous a permis dans un premier temps de mettre en évidence des différences mineures entre les patients ayant subi un TCCL et les sujets sains, même si elle étaient statistiquement significatives. Soit, une baisse d'ondes delta sur les dérivations centrales gauches et occipitales droites durant le sommeil paradoxal ainsi qu'une augmentation en stade 2 sur la dérivation frontale droite des ondes béta et gamma. La signification clinique de ces changements reste nébuleuse car ces résultats n'ont pas été répliqués dans des études précédentes. Compte tenu de la forte prévalence de douleur chez les victimes de TCCL et que la douleur est connue comme pouvant altérer le sommeil [66,83], nous avons tenté de départager l'influence de la douleur sur les variables physiologiques du sommeil.

L'originalité de la présente étude réside en l'introduction de la douleur comme facteur important qui influence la microstructure du sommeil dans une population ayant subi un TCCL. D'abord, cette étude a permis de mettre en évidence que les patients ayant subi un TCCL et qui souffrent de douleur présentent aussi de l'anxiété, de la dépression ainsi que du catastrophisme face à la douleur. La douleur n'interfère pas dans l'architecture du sommeil, cependant elle semble jouer un rôle important dans la microstructure du

sommeil, surtout le sommeil paradoxal. En effet, durant le sommeil paradoxal des patients ayant subi un TCCL et qui souffrent de douleur, il y a une augmentation marquée des ondes rapides (alpha à gamma). Même si les ondes rapides, plus particulièrement les ondes alpha, ont longtemps été associées aux plaintes de mauvais sommeil et aussi à la douleur, nous avons observé que ce sont surtout dans les bandes béta et gamma qui distinguaient les TCCL avec et sans douleur. Les oscillations gamma enregistrées sur le cortex somato-sensoriel primaire sont corrélées avec la perception subjective nociceptive [196]. Dans une autre étude, le couplage des ondes gamma (et beta) avec la perception de la douleur se passe dans les zones sensori-motrices [197-198]. Le rôle suggéré des oscillations gamma est l'intégration corticale des perceptions nociceptives [199]. À notre connaissance, aucune étude précédente n'a enregistré des ondes gamma durant le sommeil d'un patient souffrant de douleur ou suite à une stimulation nociceptive. Dans cette étude, la douleur est qualifiée d'aigue car elle est apparue suite au TCCL, de moins de trois mois. La présence d'ondes rapides durant le sommeil confère donc une signature physiologique précoce de la douleur durant le sommeil. De plus, une diminution des ondes lentes delta, associée au sommeil dit récupérateur, est observée durant tous les stades de sommeil chez les patients TCCL avec douleur aigue. L'intensité des ondes delta durant le sommeil étant associée avec l'homéostasie du sommeil, on peut spéculer que ces ondes lentes jouent un rôle aussi important que le gamma dans la plainte subjective de sommeil. Cependant, il ne faut pas négliger la littérature provenant du domaine de l'insomnie qui spécule que la non-récupération du sommeil réside en la présence d'ondes béta durant le sommeil paradoxal [142].

Avec ces résultats d'EEG durant le sommeil paradoxal, on ne peut que constater la similarité avec l'instabilité du sommeil paradoxal en insomnie [141-142]. Cette similarité rend la relation douleur/sommeil encore plus stimulante car il existerait, peut-être, des substrats physiologiques entre la présence de douleur et le développement d'un trouble de sommeil. Pour cette raison, dans une analyse (non présenté dans cette thèse) subséquente réalisée chez la même cohorte, nous essayons de nous attarder sur la nature de la plainte de sommeil (insomnie, hypersomnie et troubles circadiens).

Un autre point soulevé dans l'article est que les symptômes dépressifs expliqueraient en partie la plainte subjective de sommeil. En effet, une autre étude récente vient d'arriver à la même conclusion chez une population de militaires ayant subi un TCCL [200]. Il reste à démontrer si les plaintes concomitantes de troubles du sommeil sont des variables explicatives indépendantes et si un ou des liens de causalité peuvent être identifiés. Les symptômes post-commotionnels ainsi que les troubles de sommeil sont intimement liés et ne devraient pas être traités comme des états indépendants.

Malgré les constatations intéressantes de cette étude, il est difficile de spéculer sur une relation de cause à effet. Des études subséquentes devraient d'abord enregistrer le sommeil de patients ayant eu un TCCL en phase chronique avec douleur persistante pour voir si la présence d'ondes rapides se maintient. Ensuite, il faudrait refaire des études polysomnographiques chez des patients ayant subi un TCCL et chez qui la douleur a été traitée ou contrôlée pour en déduire les effets sur le sommeil. Finalement, nous avons tenté de répliquer cette étude chez une population de traumatisés sans atteinte cérébrale (blessés orthopédiques graves) pour pouvoir distinguer si ce phénomène est propre à la douleur ou

bien est renforcé par l'atteinte cérébrale. Pour des raisons de logistique de recrutement, cette étude fut difficile à réaliser.

Voyant l'importance de la douleur au sein des autres symptômes post-commotionnels, le deuxième article de cette thèse s'attarde sur l'interaction de la douleur en phase aigue en présence des autres symptômes ainsi que les facteurs prédisposants à la douleur chronique suite à un TCCL.

La première partie du second article traite de la douleur en phase aigue suite à un TCCL et la deuxième partie examine la phase chronique, soit un an post-trauma. En phase aigue, la douleur post-traumatique est fréquente et est accompagnée de plaintes psychologiques, de manque de retour aux activités et au travail, et un temps de réaction ralenti. Cependant, les tests de modalités sensoriels n'ont pas révélé de différences entre les douloureux et les non-douloureux. La non-sensibilité aux modalités sensorielles et à la douleur expérimentale chez les TCCL s'avère donc un moyen intéressant d'étudier les mécanismes impliqués dans la douleur, mécanismes qui semblent paradoxaux. En effet, bien que les sujets se plaignent de douleur celle-ci n'est pas objectivée avec les outils sensoriels quantitatifs usuels ce qui peut suggérer que la douleur présente n'est pas de type neuropathique. Les nouvelles directions de la recherche devraient comporter l'usage de meilleurs outils pour mettre en évidence un déficit sensoriel ou comportemental à exprimer la douleur pour pouvoir mettre en évidence une douleur de type musculo-squelettique [201]. Se pourrait-il que les sujets présentent une dissociation sensorielle, à savoir que la plainte est plus affective ou liée à des mécanisme d'attention, de vigilance et de la mémoire

du traumatisme que sensorielle? Une analyse plus poussée en psychophysiologie est requise pour mieux comprendre la dichotomie entre les plaintes et les mesures de la douleur.

Un autre point semble bien surprenant, même si une douleur de type musculo-squelettique est souvent rapportée suite à un TCCL, l'International Association for the Study of Pain (IASP) n'inclue pas de définition ou de classification pour ce type de douleur. La seule définition présentée est celle des céphalées post-traumatiques [65].

L'intensité de la douleur semble être aussi un bon indice pour prédire la chronicité des symptômes post-commotionnels [185,202]. Dans notre étude, même si l'intensité de la douleur était élevée dans le groupe ayant développé une douleur chronique, elle n'a pas été la variable explicative pour prédire la chronicité. La variable majeur explicative pour la persistance de la douleur chez les patients ayant subi un TCCL est la présence du polymorphisme val/val du gène BDNF. Même si quelques études précédentes se sont intéressées au polymorphisme du gène BDNF en traumatologie (tableau 2 en Introduction), la performance, la cognition, l'intelligence et le développement d'un état végétatif étaient les fonctions d'intérêt. La douleur n'a jamais été l'objet d'étude chez cette population. Comme le rôle du BDNF est principalement lié à la plasticité synaptique, il est difficile de spéculer sur sa fonction dans la nociception. Certaines études lui confèrent un rôle dans la potentialisation de la sensibilité à la douleur lorsqu'il est sécrété par la microglie [203-204]. L'étude du polymorphisme val/met du BDNF à une douleur expérimentale a montré une différence dans les potentiels évoqués de la réponse corticale. Les sujets porteurs du polymorphisme val/val montrent moins de réponses à une stimulation douloureuse [205].

Cependant, le rôle du BDNF ne semble pas être spécifique à la douleur. En contrepartie, tel que mentionné dans le deuxième article, on ne devrait pas isoler la douleur des autres symptômes post-commotionnels, tous étant intimement liés. Serait-il possible qu'en effet, le rôle de la plasticité synaptique agirait non seulement dans la matrice de la douleur mais aussi dans d'autres structures ayant subi des dommages axonaux lors du TCCL? C'est pour cette raison que dans le troisième article de cette thèse, nous avons choisi d'utiliser le score du questionnaire Rivermead qui traite de la présence de symptômes post-commotionnels en général.

La prédisposition génétique à la chronicité des symptômes post-commotionnels suite à un TCC a été suggéré dans plusieurs études précédentes [90,187]. Nous avions donc émis l'hypothèse que des polymorphismes jouent un rôle important dans le mauvais pronostic des patients ayant subi un TCCL. Pour tester cette hypothèse, nous avons d'abord validé grâce à une liste de polymorphismes déjà cités dans la littérature leur rôle au sein de notre population. Après correction pour tests multiples, seul des polymorphismes du BDNF prédisposaient à une augmentation des symptômes post-commotionnels. Des études d'expressions de la protéine BDNF dans des cellules de lymphoblastes de patients ayant subi un TCCL et de sujets sains ont permis d'identifier une protéine de 20 kD qui serait exprimée uniquement chez les TCCL ayant le génotype met. Si l'on se fie aux résultats présentés dans l'étude sur la douleur, les patients ayant le génotype met sont moins à risque de développer de la douleur chronique, donc l'on peut spéculer que cette protéine pourrait avoir un rôle neuroprotecteur. Des analyses subséquentes sont nécessaires pour confirmer la nature et le rôle de cette protéine.

Les études présentées dans cette thèse présentent des limites. D'abord, pour pouvoir entreprendre des études d'une telle envergure, le nombre de patients recrutés se doit d'être élevé. Or, dans nos études, nous n'avons pu recruter qu'une centaine de patients. Cette limite est une réalité non-négligeable en traumatologie, à tel point qu'un article spécial a été publié pour traiter de la question [152]. En effet, les patient ayant subi un traumatisme sont réticent à retourner au centre hospitalier et sont souvent en attente d'un litige ou d'une compensation financière des agents payeurs. Ensuite, nos études auraient pu bénéficier d'un groupe de patients ayant subi un traumatisme sans atteinte cérébrale pour pouvoir avoir un groupe de contrôle positif. Ce groupe nous aurait permis de distinguer la contribution de la douleur musculo-squelettique et celle des atteintes ou lésions cérébrales.

Finalement, en ce qui concerne notre étude génétique, nos cellules proviennent de cultures de lymphoblastes. Notre étude aurait été plus complète si l'on avait des échantillons de tissus cérébraux pour étudier l'expression du BDNF. Cependant, pour des raisons éthiques, nous n'avons pas pu prélever des échantillons de tissus cérébraux.

Conclusion

La douleur suite à un traumatisme crânien cérébral léger joue un rôle majeur dans la persistance de symptômes post-commotionnels. La douleur influence l'homéostasie et la profondeur du sommeil. La présence de douleur exacerbe les plaintes physiques et psychologiques et sa chronicité peut être expliquée par une prédisposition génétique. Le polymorphisme du BDNF aurait un rôle majeur dans le mauvais pronostic suite à un TCCL qui pourrait être expliqué par un effet neuroprotecteur en présence du polymorphisme met/met. De futures investigations seront nécessaires pour élucider le mécanisme exact joué par le BDNF dans la chronicité des symptômes post-commotionnels ainsi que les méthodes de renversement de ces mécanismes.

Bibliographie

1.   Thurman DJ, Alverson CA, Dunn KA*, et al. Traumatic brain injury in the United States: a public health perspective.* **J Head Trauma Rehabil, 1999. 14: p. 602-615.**

2.   Thurman DJ and Guerrero J. *Trends in hospitalization associated with traumatic brain injury.* **JAMA, 1999. 282(10): p. 954-957.**

3.   Langlois JA, Rutland-Brown W, and Thomas KE. *Traumatic brain injury in the United States: emergency department visits, hospitalizations, and deaths.* **2006, Centers for Disease Control and Prevention, National Center for Injury Prevention and Control: Atlanta.**

4.   Thurman DJ. *Epidemiology and economics of head trauma*, in *Head trauma: basic, preclinical, and clinical directions*, L Miler and R Hayes, Editors. 2001, Wiley and Sons: New York. p. 1193-1202.

5.   Nathens AB, Rivara FP, Jurkovich GJ*, et al. Management of the injured patient: Identification of research topics for systematic review using the Delphi technique.* **J Trauma, 2003. 54: p. 595-601.**

6.   Yoganandan N, Gennarelli TA, Zhang J*, et al. Association of contact loading in diffuse axonal injuries from motor vehicle crashes.* **J Trauma, 2009. 66: p. 309-315.**

7.   Maas AIR, Stocchietti N, and Bullock R. *Moderate and severe traumatic brain injury in adults.* **Lancet Neurol, 2008. 7: p. 728-741.**

8.   Gennarelli TA and Meaney DF. *Mechanisms of primary head injury*, in *Neurosurgery*, R Wilkins and S Rengachary, Editors. 1996, McGraw Hill: New York. p. 2611-2621.

9.   Faul M, Xu L, Wald MM*, et al. Traumatic brain injury in the United States: emergency department visits, hospitalizations, and deaths 2002-2006*. 2010, Centers for Disease Control and Prevention. National Center for Injury Prevention and Control: Atlanta (GA).

10.   Feinstein A and Rapoport M. *Mild traumatic brain injury: the silent epidemic.* **Can J Public Healt, 2000. 91(5): p. 325-326.**

11.   Okie S. *Traumatic brain injury in the war zone.* **N Engl J Med, 2005. 352: p. 2043-2047.**

12. **Carroll LJ, Cassidy JD, Holm L*, et al. Methodological issues and research recommendations for mild traumatic brain injury: The WHO collaborating centre task force on mild traumatic brain injury.* J Rehabil Med, 2004. 36(Suppl. 43): p. 113-125.**

13. **Menon DK, Schwab K, Wright DW*, et al. Position statement: Definition of traumatic brain injury.* Arch Phys Med Rehabil, 2010. 91: p. 1637-1640.**

14. **Jasper HH, Kershman J, and Elvidge A.** *Electroencephalographic studies of injury to the head.* **Arch Neurol, 1940. 44: p. 328-348.**

15. **Glaser MA and Sjaardema H.** *The value of electroencephalogram in craniocerebral injuries.* **West J Surg Obstet Gynecol, 1940. 48: p. 689-695.**

16. **Graham DI, McIntosh TK, Maxwell WL*, et al. Recent advances in neurotrauma.* J Neuropathol Exp Neurol, 2000. 59(8): p. 641-651.**

17. **Marshall LF.** *Recent past, present and future.* **Neurosurgery, 2000. 47(3): p. 546-561.**

18. **Rothman SM and Olney JW.** *Excitotoxicity and the NMDA receptor.* **Trends Neurosci, 1987. 10: p. 299-302.**

19. **Kermer P, Klocker N, and Bahr M.** *Neuronal death after brain injury. Models, mechanisms, and therapeutic strategies in vivo.* **Cell Tissue Res, 1999. 298: p. 383-395.**

20. **Juurlink BH and Paterson PG.** *Review of oxidative stress in brain and spinal cord injury: suggestions for pharmacological and nutritional management strategies.* **J Spinal Cord Med, 1998. 21(4): p. 309-334.**

21. **McCall JM, Braughler JM, and Hall ED.** *Lipid peroxidation and the role of oxygen radicals in CNS injury.* **Acta Anaesthesiol Belg, 1987. 38: p. 373-379.**

22. **Hall ED and Braughler JM.** *Central nervous system trauma and stroke. II. Physiological and pharmacological evidence for involvement of oxygen radicals and lipid peroxidation.* **Free Radic Biol Med, 1989. 6(3): p. 303-313.**

23. **Hall ED.** *Free radicals and CNS injury.* **Crit Care Clin, 1989. 5(4): p. 793-805.**

24. **Simonian NA and Coyle JT.** *Oxidative stress in neurodegenerative diseases.* **Annu Rev Pharmacol Toxicol, 1996. 36: p. 83-106.**

25. **Kampfl A, Posmantur RM, Zhao X*, et al. Mechanisms of calpain proteolysis following traumatic brain injury: Implication for pathology and therapy -***

Implications for pathology and therapy: A review and update. **J Neurotrauma, 1997. 14: p. 121-134.**

26. **Bramlett HM, Green EJ, and Dietrich WD.** *Hippocampally dependant and independant chronic spatial navigational deficits following parasagittal fluid percussion brain injury in the rat.* **Brain Res, 1997. 762: p. 195-202.**

27. **Levin HS.** *Cognitive function outcomes after traumatic brain injury.* **Curr Opin Neurol, 1998. 11: p. 643-646.**

28. **Wu A, Ying Z, and Gomez-Pinilla F.** *Dietary curcumin counteracts the outcome of traumatic brain injury on oxidative stress, synaptic plasticity, and cognition.* **Exp Neurol, 2006. 197: p. 309-317.**

29. **Adibhatla RM and Hatcher JF.** *Phospholipase A2, reactive oxygen species, and lipid peroxidation in CNS pathologies.* **BMB reports, 2008. 41(8): p. 560-567.**

30. **Amat JA, Ishiguro H, Nakamura K***, et al. Phenotypic diversity and kinetics of proliferating microglia and astrocytes following cortical stab wounds.* **Glia, 1996. 16: p. 368-382.**

31. **Barkhoudarian G, Hovda DA, and Giza CC.** *The molecular pathophysiology of concussive brain injury.* **Clin Sports Med, 2011. 30: p. 33-48.**

32. **Association AP.** *Diagnostic and Statistical Manual of Mental Disorders*, **ed. t ed. 1994, Washington, DC.**

33. **King NS, Crawford S, Wenden FJ***, et al. The Rivermead Post Concussion Symptoms Questionnaire: a measure of symptoms commonly experienced after head injury and its reliability.* **J Neurol, 1995. 242: p. 587-592.**

34. **Ponsford JL, Willmott C, Rothwell A***, et al. Factors influencing outcome following mild traumatic brain injury in adults.* **J Int Neuropsychol Soc, 2000. 6(5): p. 568-579.**

35. **Zhang S, Carroll LJ, Cassidy JD***, et al. Factors influencing self-rated health in traffic-related mild traumatic brain injury.* **J Rehabil Med, 2009. 41(13): p. 1062-1067.**

36. **Paniak C, Reynold S, Toller-Lobe G***, et al. A longitudinal study of the relationship between financial compensation and symptoms after treated mild traumatic brain injury.* **J Clin Exp Neuropsychol, 2002. 24(2): p. 187-193.**

37. **Cassidy JD, Carroll LJ, Côté P***, et al. Mild traumatic brain injury after traffi collisions: a population-based inception cohort study.* **J Rehabil Med, 2004. Suppl. 43: p. 15-21.**

38.   Hoge CW, McGurk D, Thomas JL, *et al. Mild traumatic brain injury in U.S. soldiers returning from Iraq.* N Engl J Med, 2008. 358: p. 453-463.

39.   Zumstein MA, Moser M, Mottini M, *et al. Long-term outcome in patients with mild traumatic brain injury: A prospective observational study.* J Trauma, 2011. 71(1): p. 120-127.

40.   Rechtschaffen A and Kales A, eds. *A manual of standardized terminology, techniques and scoring system for sleep stages of human subjects*. 1968, Brain Information Service/Brain Research Institute, University of California: Los Angeles.

41.   Theodorou AA and Rice SA. *Is the silent epidemic keeping patients awake?* J Clin Sleep Med, 2007. 3(4): p. 347-348.

42.   Verma A, Anand V, and Verma NP. *Sleep disorders in chronic traumatic brain injury.* J Clin Sleep Med, 2007. 3(4): p. 357-362.

43.   Zeitzer JM, Friedman L, and O'Hara R. *Insomnia in the context of traumatic brain injury.* J Rehabil Res Dev, 2009. 46(6): p. 827-836.

44.   Parsons LC, Crosby LJ, Perlis M, *et al. Longitudinal sleep EEG power spectral analysis studies in adolescents with minor head injuty.* J Neurotrauma, 1997. 14: p. 549-559.

45.   Gosselin N, Lassonde M, Petit D, *et al. Sleep following sport-related concussions.* Sleep Med., 2009. 10(1): p. 35-46.

46.   Rao V, Bergey A, Hill H, *et al. Sleep disturbance after mild traumatic brain injury: indicator of injury?* J Neuropsychiatry Clin Neurosci, 2011. 23(2): p. 201-205.

47.   Cohen M, Oksenberg A, Snir D, *et al. Temporally related changes of sleep complaints in traumatic brain injured patients.* J Neurol Neurosurg Psychiatry, 1992. 55: p. 313-315.

48.   Baumann CR, Werth E, Stocker R, *et al. Sleep-wake disturbances 6 months after traumatic brain injury: a prospective study.* Brain, 2007. 130: p. 1873-1883.

49.   Ayalon L, Borodkin K, Dishon L, *et al. Circadian rhythm sleep disorders following mild traumatic brain injury.* Neurology, 2007. 68: p. 1136-1140.

50.   Kaufman Y, Tzischinsky O, Epstein R, *et al. Long-term sleep disturbances in adolescents after minor head injury.* Pediatr Neurol, 2001. 24: p. 129-134.

51.   Ouellet MC and Morin CM. *Subjective and objective measures of insomnia in the context of traumatic brain injury: a preliminary study.* Sleep Med., 2006. 7(6): p. 486-497.

52.   Schreiber S, Barkai G, Gur-Hartman T*, et al. Long-lasting sleep patterns of adult patients with minor traumatic brain injury (mTBI) and non-mTBI subjects.* Sleep Med, 2008. 9(5): p. 481-487.

53.   Williams BR, Lazic SE, and Ogilvie RD. *Polysomnographic and quantitative EEG analysis of subjects with long-term insomnia complaints associated with mild traumatic brain injury.* Clinical Neurophysiology, 2008. 119: p. 429-438.

54.   Bonica JJ. *The need of a taxonomy.* Pain, 1979. 6(3): p. 247-248.

55.   taxonomy Itf. *Classification of chronic pain*, ed. H Merskey and N Bogduk. 2012: IASP Press.

56.   Nampiaparampil DE. *Prevalence of chronic pain after traumatic brain injury: A systematic review.* JAMA, 2008. 300(6): p. 711-719.

57.   Dikmen S, Machamer J, Fann JR*, et al. Rates of symptom reporting following traumatic brain injury.* J Int Neuropsychol Soc, 2010. 16: p. 401-411.

58.   Jensen OK and Nielsen FF. *The influence of sex and pre-traumatic HA on the incidence and severity of HA after head injury.* Cephalalgia, 1990. 10(6): p. 285-293.

59.   Landy PJ. *Neurological sequelae of minor head and neck injuries.* Injury, 1998. 29(3): p. 199-206.

60.   Geisser ME, Roth RS, Bachman JE*, et al. The relationship between symptoms of post-traumatic stress disorder and pain, affective disturbance, and disability among patients with accident and non-accident related pain.* Pain, 1996. 66(2-3): p. 207-214.

61.   Alexander MP. *Mild traumatic brain injury: Pathophysiology, natural history, and clinical management* Neurology, 1995. 45: p. 1253-1260.

62.   Dean PJ, O`Neill D, and Sterr A. *Post-concussion syndrome: prevalence after mild traumatic brain injury in comparison with a sample without head injury.* Brain Inj, 2012. 26(1): p. 14-26.

63.   Defrin R, Gruener H, Schreiber S*, et al. Quantitative somatosensory testing of subjects with chronic post-traumatic headache: implications on its mechanisms.* Eur J Pain, 2010. 14(9): p. 924-931.

64. Khoury S, Roy I, Giguère JF, *et al. Altered thermal heat sensation but not pain in mild traumatic brain injury patients: preliminary report on pre- and post-sleep assessments.* 2010. Abstract IASP(PM-039).

65. Defrin R, Ginzburg K, Solomon Z, *et al. Quantitative testing of pain perception in subjects with PTSD--implications for the mechanism of the coexistence between PTSD and chronic pain.* Pain, 2008. 138(2): p. 450-459.

66. Borsook D. *Neurological disease and pain.* Brain, 2012. 135(Pt2): p. 320-344.

67. Yarnitsky D. *Quantitative sensory testing.* Muscle Nerve, 1997. 20: p. 198-204.

68. Jennum P, Ole Hein H, Suadicani P, *et al. Cognitive function and snoring.* Sleep, 1993. 16(8): p. S62-S64.

69. Drewes AM, Nielsen KD, Arendt-Nielsen L, *et al. The effect of cutaneous and deep pain on the electroencephalogram during sleep--an experimental study.* Sleep, 1997. 20(8): p. 632-640.

70. Lavigne GJ, Zucconi M, Castronovo C, *et al. Sleep arousal response to experimental thermal stimulation during sleep in human subjects free of pain and sleep problems.* Pain, 2000. 84(2): p. 283-290.

71. Lavigne GJ, Okura K, Abe S, *et al. Gender specificity of the slow wave sleep lost in chronic widespread musculoskeletal pain.* Sleep Med., 2011. 12(2): p. 179-185.

72. Lavigne GJ, Nashed A, Manzini C, *et al. Does sleep differ among patients with common musculoskeletal pain disorders?* Curr Rheumatol Rep, 2011. 13(6): p. 535-542.

73. Beetar JT, Guilmette TJ, and Sparadeo FR. *Sleep and pain complaints in symptomatic traumatic brain injury and neurologic population.* Arch Phys Med Rehabil, 1996. 77: p. 1298-1302.

74. Chaput G, Giguère JF, Chauny JM, *et al. Relationship among subjective sleep complaints, headaches, and mood alterations following a mild traumatic brain injury.* Sleep Med., 2009. 10(7): p. 713-716.

75. Teasdale GM, Nicoll JA, Murray G, *et al. Association of apolipoprotein E polymorphism with outcome of head injury.* Lancet, 1997. 350: p. 1069-1071.

76. Nathoo N, Chetry R, van Dellen JR, *et al. Apolipoprotein E polymorphism and outcome after closed traumatic brain injury: Influence of ethnic and regional differences.* J Neurosurg, 2003. 98: p. 302-306.

77.   **Ariza M, Pueyo R, Matarin MdM, *et al. Influence of APOE polymorphisme on cognitive and behavioural outcome in moderate and severe traumatic brain injury.* J Neurol Neurosurg Psychiatry, 2006. 77: p. 1191-1193.**

78.   **Mahley RW. *Apolipoprotein E: cholesterol transport protein with expanding role in cell biology.* Science, 1988. 240: p. 622-630.**

79.   **Chen Y, Lomnitski L, Michaelson DM, *et al. Motor and cognitive deficits in apolipoprotein E-deficient mice after closed head injury.* Neuroscience, 1997. 80: p. 1255-1262.**

80.   **Dardiotis E, Grigoriadis S, and Hadjigeorgiou GM. *Genetic factors influencing outcome from neurotrauma.* Curr Opin Psychiatry, 2012. 25(3): p. 231-238.**

81.   **Tanriverdi T, Uzan M, Sanus GZ, *et al. Lack of association between the IL1A (-889) polymorphism and outcome after head injury.* Surg Neurol, 2006. 65: p. 7-10.**

82.   **Minambres E, Cemborain A, Sanchez-Velasco P, *et al. Correlation between transcranial interleukin-6 gradient and outcome in patients with acute brain injury.* Crit Care Med, 2003. 31(3): p. 933-938.**

83.   **Chao MV. *Neurotrophins and their receptors: A convergence point for many signalling pathways.* Nat Rev Neurosci, 2003. 4: p. 299-309.**

84.   **Skaper S. *The biology of neurotrophins, signalling pathways, functional peptide mimetics of neurotrophins and their receptors.* CNS Neurol Disorders Drug Targets, 2008. 7: p. 46-62.**

85.   **Ray S, Dixon C, and Banik N. *Molecular mechanisms in the pathogenesis of traumatic brain injury.* Histol Histopathol, 2002. 17: p. 1137-1152.**

86.   **Lu B. *BDNF and activity-dependent synaptic modulation.* Learn Mem, 2003. 10(2): p. 86-98.**

87.   **Egan MF, Kojima M, Callicott JH, *et al. The BDNF Val66Met polymorphism affects activity-dependent secretion of BDNF and human memory and hippocampal function.* Cell, 2003. 112: p. 257-269.**

88.   **McAllister TW, Tyler AL, Flashman LA, *et al. Polymorphisms in the brain-derived neurotrophic factor gene influence memory and processing speed one month after brain injury.* J neurotrauma, 2012. 29(6): p. 1111-1118.**

89.   **Krueger F, Pardini M, Huey ED, *et al. The role of the Met66 brain-derived neurotrophic factor allele in the recovery of executive functioning after combat-related traumatic brain injury.* J Neurosc, 2011. 31(2): p. 598-606.**

90. Jiang L, Yin X, Yin C, *et al. Different quantitative EEG alterations induced by TBI among patients with different APOE genotypes.* Neurosci Lett, 2011. 505(2): p. 160-164.

91. Chamelian L, Reis M, and Feinstein A. *Six-month recovery from mild to moderate traumatic brain injury: the role of APOE-epsilon4 allele.* Brain, 2004. 127(Pt 12): p. 2621-2628.

92. Terrell TR, Bostick RM, Abramson R, *et al. APOE, APOE promoter, and Tau genotypes and risk for concussion in college athletes.* Clin J Sport Med, 2008. 18(1): p. 10-17.

93. Tierney RT, Mansell JL, Higgins M, *et al. Apolipoprotein E genotype and concussion in college athletes.* Clin J Sport Med, 2010. 20(6): p. 464-468.

94. Sarnaik AA, Conley YP, Okonkwo DO, *et al. Influence of PARP-1 polymorphisms in patients after traumatic brain injury.* J neurotrauma, 2010. 27(3): p. 465-471.

95. Uzan M, Tanriverdi T, Baykara O, *et al. Association between interleukin-1 beta (IL-1b) gene polymorphism and outcome after head injury: an early report.* Acta Neurochir, 2005. 147: p. 715-720.

96. Ariza M, Matarin MD, Junque C, *et al. Influence of Angiotensin-converting enzyme polymorphism on neuropsychological subacute performance in moderate and severe traumatic brain injury.* J Neuropsychiatry Clin Neurosci, 2006. 18(1): p. 39-44.

97. Lipsky RH, Sparling MB, Ryan LM, *et al. Association of COMT Val158Met genotype with executive functioning following traumatic brain injury.* J Neuropsychiatry Clin Neurosci, 2005. 17(4): p. 465-471.

98. Martinez-Lucas P, Moreno-Cuesta J, Garcia-Olmo DC, *et al. Relationship between the Arg72Pro polymorphism of p53 and outcome for patients with traumatic brain injury.* Intensive Care Med, 2005. 31: p. 1168-1173.

99. Bagnato S, Minafra L, Bravata V, *et al. Brain-Derived Neurotrophic Factor (Val66Met) polymorphism does not influence recovery from a post-traumatic vegetative state: a blinded retrospective multi-centric study.* J neurotrauma, 2012. 29(11): p. 2050-2059.

100. Rostami E, Krueger F, Zoubak S, *et al. BDNF polymorphism predicts general intelligence after penetrating traumatic brain injury.* PLoS One, 2011. 6(11): p. e27389.

101.	McAllister TW, Flashman LA, Harker Rhodes C*, et al. Single nucleotide polymorphisms in ANKK1 and dopamine D2 receptor gene affect cognitive outcome shortly after traumatic brain injury: a replication and extension study.* Brain Inj, 2008. 22(9): p. 705-714.

102.	McDevitt JK, Tierney RT, Mansell JL*, et al. Neuronal structure protein polymorphism and concussion in college athletes.* Brain Inj, 2011. 25(11): p. 1108-1113.

103.	WHO. *World Health Organization. Global status report on road safety.* 2010, Department of Violence & Injury Prevention & Disability (VIP).

104.	Guilleminault C, Faull K, Miles L*, et al. Post-traumatic excessive daytime sleepiness: a review of 20 patients.* Neurology, 1983. 33: p. 1584-1589.

105.	Prigatano GP, Stahl ML, Orr WC*, et al. Sleep and dreaming disturbances in closed head injury patients.* J Neurol Neurosurg Psychiatry, 1982. 45: p. 78-80.

106.	Kempf J, Werth E, Kaiser PR*, et al. Sleep-wake disturbances 3 years after traumatic brain injury* J Neurol Neurosurg Psychiatry, 2010. 81: p. 1402-1405.

107.	Castriotta RJ, Wilde MC, Lai JM*, et al. Prevalence and consequences of sleep disorders in traumatic brain injury.* J Clin Sleep Med, 2007. 3(4): p. 349-356.

108.	Castriotta RJ and Murthy JN. *Sleep disorders in patients with traumatic brain injury: a review.* CNS Drugs, 2010. 25(3): p. 175-185.

109.	Masel BE, Scheibel RS, Kimbark T*, et al. Excessive daytime sleepiness in adults with brain injuries.* Arch Phys Med Rehabil, 2001. 82: p. 1526-1532.

110.	Webster JB, Bell KR, Hussey JD*, et al. Sleep apnea in adults with traumatic brain injury: a preliminary investigation.* Arch Phys Med Rehabil, 2001. 82: p. 316-321.

111.	Watson NF, Dikmen S, Machamer J*, et al. Hypersomnia following traumatic brain injury.* J Clin Sleep Med, 2007. 3(4): p. 363-368.

112.	Walker WC. *Pain pathoetiology after TBI.* J Head Trauma Rehabil, 2004. 19(1): p. 72-81.

113.	Smith MT and Buenaver LF. *Electroencephalographic correlates of pain and sleep interactions in humans,* in *Sleep and Pain,* GJ Lavigne, et al., Editors. 2007, IASP Press: Seattle, WA.

114.   Lew HL, Pogoda TK, Hsu PT, *et al. Impact of the "polytrauma clinical triad" on sleep disturbance in a department of veterans affairs outpatient rehabilitation setting.* Am J Phys Med Rehabil, 2010. 89(6): p. 437-445.

115.   Iverson GL and McCracken LM. *'Postconcussive' symptoms in persons with chronic pain.* Brain Inj, 1997. 11(11): p. 783-790.

116.   Giamberardino MA and Jensen TS. *Pain comorbidities: Understanding and treating the complex patient*, ed. I Press. 2012, Seattle, WA: International Association for the Study of Pain.

117.   Hoffman JM, Pagulayan KF, Zawaideh N, *et al. Understanding pain after traumatic brain injury: Impact on community participation.* Am J Phys Med Rehabil, 2007. 86(12): p. 962-969.

118.   Stalnacke BM. *Postconcussion symptoms in patients with injury-related chronic pain.* Rehabil Res Pract, 2012. 2012: 528265.

119.   Vancleef LM and Peters ML. *Pain catastrophizing, but not injury/illness sensitivity or anxiety sensitivity, enhances attentional interference by pain.* J Pain, 2006. 7(1): p. 23-30.

120.   Buysse DJ, Reynold CFr, Monk TH, *et al. The Pittsburgh Sleep Quality Index: a new instrument for psychiatric practice and research.* Psychiatry Res., 1989. 28(2): p. 193-213.

121.   Fichtenberg N, Putman S, Mann N, *et al. Insomnia screening in postacute traumatic brain injury-utility and validity of the Pittsburgh Sleep Quality Index.* Am J Phys Med Rehabil, 2001. 80: p. 339-345.

122.   AASM. *International Classification of Sleep Disorders, 2nd edition: Diagnostic and Coding Manual.* 2005, American Academy of Sleep Medicine: Westchester, IL.

123.   IFSECN. *A glossary of terms most commonly used by clinical electroencephalographers.* Electroencephalogr Clin Neurophysiol, 1974. 37(5): p. 538-548.

124.   Beck AT, Ward CH, Mendelson M, *et al. An inventory for measuring depression.* Arch Gen Psychiatry, 1961. 4: p. 561-571.

125.   Beck AT, Epstein N, Brown G, *et al. An inventory for measuring clinical anxiety: psychometric properties.* J Consult Clin Psychol, 1998. 56: p. 893-897.

126.   Trahan DE, Ross CE, and Trahan SL. *Relationships among postconcussional-type symptoms, depression, and anxiety in neurologically normal young adults*

and victims of mild brain injury. Arch Clin Neuropsychol, 2001. 16(5): p. 435-445.

127.   Sullivan MJL, Bishop S, and Pivik J. *The pain catastrophizing scale: Development and validation.* Psychol Assess, 1995. 7: p. 524-532.

128.   Miro J, Nieto R, and Huguet A. *The Catalan version of the Pain Catastrophizing Scale: a useful instrument to assess catastrophic thinking in whiplash patients.* J Pain, 2008. 9(5): p. 397-406.

129.   Klein E, Koren D, Arnon I*, et al. Sleep complaints are not corroborated by objective sleep measures in post-traumatic stress disorder: a 1-year prospective study in survivors of motor vehicle crashes.* J Sleep Res., 2003. 12: p. 35-41.

130.   Tang NKY, Goodchild CE, Sanborn AN*, et al. Deciphering the temporal link between pain and sleep in a heterogeneous chronic pain patient sample: A multilevel daily process study.* Sleep, 2012. 35(5): p. 675-687.

131.   Smith MT, Quartana PJ, Okonkwo RM*, et al. Mechanisms by which sleep disturbance contributes to oteoarthritis pain: A conceptual model.* Curr Pain Headache R, 2009. 6: p. 447-454.

132.   Baglioni C and Riemann D. *Is chronic insomnia a precursor to major depression? Epidemiological and biological findings.* Curr Psychiatry Rep, 2012. Epub ahead of print.

133.   Riemann D, Spiegelhalder K, Nissen C*, et al. REM sleep instability - A new pathway for insomnia?* Pharmacopsychiatry, 2012. 45(5): p. 167-176.

134.   Graversen C, Olesen SS, Olesen AE*, et al. The analgesic effect of pregabalin in patients with chronic pain is reflected by changes in pharmaco-EEG spectral indices.* Br J Clin Pharmacol., 2012. 73(3): p. 363-372.

135.   Wang J, Li D, Li X*, et al. Phase-amplitude coupling between theta and gamma oscillations during nociception in rat electroencephalography.* Neuroscience Letters, 2011. 499(2): p. 84-87.

136.   De Pascalis V and Cacace I. *Pain perception, obstructive imagery and phase-ordered gamma oscillations.* Int J Psychophysiol., 2005. 56: p. 157-169.

137.   Timofeev I, Bazhenov M, Seigneur J*, et al. Neuronal synchronization and thalamocortical rythms in sleep, wake and epilepsy*, in *Jasper's basic mechanisms of the epilepsies*, JL Noebels, et al., Editors. 2012, National Center for Biotechnology Information: Bethesda (MD).

138.   Urbano FJ, Kezunovic N, Hyde J, *et al. Gamma band activity in the reticular activating system.* **Front Neurol, 2012. 3(6): p. In Press.**

139.   Lavigne GJ, Brousseau M, Kato T, *et al. Experimental pain perception remains equally active over all sleep stages.* **Pain, 2004. 110(3): p. 646-655.**

140.   Worthington AD and Melia Y. *Rehabilitation is compromised by arousal and sleep disorders: results of a survey of rehabilitation centres.* **Brain Inj, 2006. 20: p. 327-332.**

141.   Moldofsky H. *Sleep and pain.* **Sleep Med Rev, 2001. 5(5): p. 385-396.**

142.   Harman K, Pivik RT, D'Eon JL, *et al. Sleep in depressed and nondepressed participants with chronic low back pain: electroencephalographic and behaviour findings.* **Sleep, 2002. 25(7): p. 775-783.**

143.   Luoto TM, Tenovuo O, Kataja A, *et al. Who gets recruited in mild traumatic brain injury research?* **J neurotrauma, 2012. Epub ahead of print.**

144.   Cassidy JD, Carroll LJ, Peloso PM, *et al. Incidence, risk factors and prevention of mild traumatic brain injury: Results of the WHO collaborating centre task force on mild traumatic brain injury.* **J Rehabil Med, 2004. 36(Suppl. 43): p. 28-60.**

145.   Cancelliere C, Cassidy JD, Côté P, *et al. Protocol for a systematic review of prognosis after mild traumatic brain injury: an update of the WHO collaborating centre task force findings.* **Syst Rev., 2012. 1(1): p. 17.**

146.   Otis JD, McGlinchey R, Vasterling JJ, *et al. Complicating factors associated with mild traumatic brain injury: Impact on pain and posttraumatic stress disorder treatment.* **J Clin Psychol Med Settings, 2011. 18: p. 145-154.**

147.   Côté P, Cassidy JD, Carroll LJ, *et al. A systematic review of the prognosis of acute whiplash and a new conceptual framework to synthesize the litterature.* **spine, 2001. 26(19): p. E445-E458.**

148.   Scholten-Peeters GG, Verhagen AP, Bekkering GE, *et al. Prognostic factors of whiplash-associated disorders: a systematic review of prospective cohort studies.* **Pain, 2003. 104(1-2): p. 303-322.**

149.   Kamper SJ, Rebbeck TJ, Maher CG, *et al. Course and prognostic factors of whiplash: A systematic review and meta-analysis.* **Pain, 2008. 138: p. 617-629.**

150.   Sterling M, Jull G, and Kenardy J. *Physical and psychological factors maintain long-term predictive capacity post-whiplash injury.* **Pain, 2006. 122: p. 102-108.**

151. Carroll LJ, Cassidy JD, and Côté P. *The role of pain coping strategies in prognosis after whiplash injury: Passive coping predicts slowed recovery.* Pain, 2006. 124: p. 18-26.

152. Atherton K, Wiles NJ, Lecky FE*, et al. Predictors of persistent neck pain after whiplash injury.* Emerg Med J, 2006. 23: p. 195-201.

153. Kasch H, Qerama E, Kongsted A*, et al. Deep muscle pain, tender points and recovery in acute whiplash patients: A 1-year follow-up study.* Pain, 2008. 140: p. 65-73.

154. Young Casey C, Greenberg MA, Nicassio PM*, et al. Transition from acute to chronic pain and disability: A model including cognitive, affective, and trauma factors.* Pain, 2008. 134(1): p. 69-79.

155. Stulemeijer M, van der Werf S, Borm GF*, et al. Early prediction of favourable recovery 6 months after mild traumatic brain injury.* J Neurol Neurosurg Psychiatry, 2008. 79: p. 936-942.

156. Risdall JE and Menon DK. *Traumatic brain injury.* Philos Trans R Soc Lond B Biol Sci, 2011. 366(1562): p. 241-250.

157. Kleim JA, Chan S, Pringle E*, et al. BDNF val66met polymorphism is associated with modified experience-dependent plasticity in human motor cortex.* Nat Neurosci, 2006. 9(6): p. 735-737.

158. Pezet S and McMahon SB. *Neurotrophins: Mediators and modulators of pain.* Annu Rev Neurosci, 2006. 29: p. 507-538.

159. Vossen H, Kenis G, Rutten B*, et al. The genetic influence on the cortical processing of experimental pain and the moderating effect of pain status.* PLoS One, 2010. 5: p. e13641.

160. Tammimaki A and Mannisto PT. *Catechol-O-methyltransferase gene polymorphism and chronic human pain: a systematic review and meta-analysis.* Pharmacogenet Genomics, 2012. 22(9): p. 673-691.

161. Zubieta JK, Heitzeg MM, Smith YR*, et al. COMT val158met genotype affects u opioid neurotransmitter responses to a pain stressor.* Science, 2003. 299: p. 1240-1243.

162. ICHD. *The International Classification of Headache Disorders, 2nd edition.* Cephalalgia, 2004. 24(Suppl 1).

163. Defrin R, Ohry A, Blumen N*, et al. Characterization of chronic pain and somatosensory function in spinal cord injury subjects.* Pain, 2001. 89: p. 253-263.

164. Lim J and Dinges DF. *Sleep deprivation and vigilant attention.* Ann. N.Y. Acad. Sci., 2008. 1129: p. 305-322.

165. Dinges DF and Kribbs NB. *Performing while sleepy: effects of experimentally induced sleepiness*, in *Sleep, sleepiness and performance*, TH Monk, Editor. 1991, John Wiley: Winchester. p. 97-128.

166. Ware JE, Snow KK, Kosinski M*, et al. The SF-36 health survey. Manual and interpretation guide.*, ed. Th institute. 1993, Boston, MA: New England Medical Center.

167. Ware JE and Sherbourne CD. *The MOS 36-item short form health survey (SF-36). Conceptual framework and item selection.* Med Care, 1992. 30: p. 473-483.

168. Weiss DS and Marmar CR. *The impact of event scale-Revised*, in *Assessing Psychological Trauma and PTSD*, JP Wilson and TM Keane, Editors. 1997, Guilford Press: New York. p. 399-411.

169. Stewart WF, Lipton RB, Whyte J*, et al. An international study to assess reliability of the Migraine Disability Assessment (MIDAS) score.* Neurology, 1999. 53(5): p. 988-994.

170. Melzack R. *The McGill Pain Questionnaire: Major properties and scoring methods.* Pain, 1975. 1(277-299).

171. Cheeran B, Talelli P, Mori F*, et al. A common polymorphism in the brain-derived neurotrophic factor gene (BDNF) modulates human cortical plasticity and the response to rTMS.* J Physiol, 2008. 586(23): p. 5717-5725.

172. Handwerker HO and Kobal G. *Psychophysiology of experimentally induced pain.* Physiol Rev, 1993. 73(3): p. 639-671.

173. Silverberg ND and Iverson GL. *Etiology of the post-concussion syndrome: Physiogenesis and psychogenesis revisited.* Neurorehabil, 2011. 29(4): p. 317-329.

174. Ingebrigtsen T, Waterloo K, Marup-Jensen S*, et al. Quantification of post-concussion symptoms 3 months after minor head injury n 100 consecutive patients.* J Neurol, 1998. 245(9): p. 609-612.

175. Radanov BP, di Stefano G, Schnidrig A*, et al. Role of psychosocial stress in recovery from common whiplash.* Lancet, 1991. 338(8769): p. 712-715.

176. Mott TF, McConnon ML, and Rieger BP. *Subacute to chronic mild traumatic brain injury.* Am Fam Physician, 2012. 86(11): p. 1045-1051.

177. **McAllister TW.** *Genetic factors modulating outcome after neurotrauma.* **PM&R, 2010. 2(Suppl 12): p. S241-S252.**

178. **Leibrock J, Lottspeich F, Hohn A,** *et al. Molecular cloning and expression of brain-derived neurotrophic factor.* **Nature, 1989. 341(6238): p. 149-152.**

179. **Lipsky RH and Marini AM.** *Brain-derived neurotrophic factor in neuronal survival and behavior-related plasticity.* **Ann N Y Acad Sci, 2007. 1122: p. 130-143.**

180. **Liguori M, Fera F, Patitucci A,** *et al. A longitudinal observation of brain-derived neurotrophic factor mRNA levels in patients with relapsing-remitting multiple sclerosis.* **Brain Res, 2009. 1256: p. 123-128.**

181. **Purcell S, Neale B, Todd-Brown K,** *et al. PLINK: A tool set for whole-genome association and population-based linkage analyses.* **Am J Hum Genet, 2007. 81(3): p. 559-575.**

182. **Dellavalle B, Hempel C, Kurtzhals JAL,** *et al. In vivo expression of neuroglobin in reactive astrocytes during neuropathology in murine models of traumatic brain injury, cerebral malaria, and autoimmune encephalitis.* **Glia, 2010. 58: p. 1220-1227.**

183. **Huber R, Tononi G, and Cirelli C.** *Exploratory behavior, cortical BDNF, and sleep homeostasis.* **Sleep, 2006. 30(2): p. 129-139.**

184. **Blugeot A, Rivat C, Bouvier E,** *et al. Vulnerability to depression: from brain neuroplasticity to identification of biomarkers.* **J Neurosc, 2011. 31(36): p. 12889-12899.**

185. **Woo NH, Teng HK, Siao CJ,** *et al. Activation of p75NTR by proBDNF facilitates hippocampal long-term depression.* **Nat Neurosci, 2005. 8(8): p. 1069-1077.**

186. **Zhang ZG, Hu L, Hung YS,** *et al. Gamma-band oscillations in the primary somatosensory cortex --a direct and obligatory correlate of subjective pain intensity.* **J Neurosc, 2012. 32(22): p. 7429-7438.**

187. **Schulz E, Tiemann L, Witkovsky V,** *et al. Gamma oscillations are involved in the sensorimotor transformation of pain.* **J Neurophysiol, 2012. 108(4): p. 1025-1031.**

188. **Senkowski D, Kautz J, Hauck M,** *et al. Emotional facial expressions modulate pain-induced beta and gamma oscillations in sensorimotor cortex.* **J Neurosc, 2011. 31(41): p. 14542-14550.**

189.   Rodriguez E, George N, Lachaux JP, *et al. Perception's shadow: long-distance synchronization of human brain activity.* Nature, 1999. 397: p. 430-433.

190.   Waldron-Perrine B, McGuire AP, Spencer RJ, *et al. The influence of sleep and mood on cognition functioning among veterans being evaluated for mild traumatic brain injury.* Mil Med, 2012. 177(11): p. 1293-1301.

191.   Lavigne GJ, Khoury S, Laverdure-Dupont D, *et al. Tools and Methodological Issues in the Investigation of Sleep and Pain Interactions*, in *Sleep and Pain*, GJ Lavigne, et al., Editors. 2007, IASP Press: Seattle, U.S.A. p. 235-266.

192.   Ettlin TM, Kischka U, Reichmann S, *et al. Cerebral symptoms after whiplash injury of the neck: a prospective clinical and neuropsychological study of whiplash injury.* J Neurol Neurosurg Psychiatry, 1992. 55(10): p. 943-948.

193.   Tender GC, Li YY, and Cui JG. *Brain-derived neurotrophic factor redistribution in the dorsal root ganglia correlates with neuropathic pain inhibition after resiniferatoxin treatment.* Spine J, 2010. 10(8): p. 715-720.

194.   Lu VB, Ballanyi K, Colmers WF, *et al. Neuron type-specific effects of brain-derived neurotrophic factor in rat superficial dorsal horn and their relevance to "central sensitization".* J Physiol, 2007. 584: p. 543-563.

195.   Di Lorenzo C, Di Lorenzo G, Daverio A, *et al. The val66met polymorphism of the BDNF gene influences trigeminal pain-related evoked responses.* J of Pain, 2012. 13(9): p. 866-873.

196.   Kraus JF and Nourjah P. *The epidemiology of mild, uncomplicated brain injury.* J Trauma, 1988. 28(12): p. 1637-1643.

197.   Langlois JA, Marr A, Mitchko J, *et al. Tracking the silent epidemic and educating the public: CDC's traumatic brain injury-associated activities under the TBI Act of 1996 and the Children's Health Act of 2000.* J Head Trauma Rehabil, 2005. 20: p. 196-204.

198.   Hungerbuhler JP. *Les traumatismes cranio-cérébraux mineurs.* Douleur et analgésie, 2004. 4: p. 191-203.

199.   Ryan LM and Warden DL. *Post concussion syndrome.* Int Rev Psychiatry, 2003. 15(4): p. 310-316.

200.   Collen J, Orr N, Lettieri CJ, *et al. Sleep disturbances among soldiers with combat related traumatic brain injury.* Chest, 2012. In Press.

201. **Gosselin N and Tellier M.** *Patients with traumatic brain injury are at high risk of developing sleep-wake disturbances.* **J Neurol Neurosurg Psychiatry, 2010. 81(12): p. 1297.**

202. **Ouellet MC, Beaulieu-Bonneau S, and Morin CM.** *Insomnia in patients with traumatic brain injury: frequency, characteristics, and risk factors.* **J Head Trauma Rehabil, 2006. 21: p. 199-212.**

203. **Ponsford JL, Ziino C, Parcell DL***, et al. Fatigue and sleep disturbance following traumatic brain injury-their nature, causes, and potential treatments.* **J Head Trauma Rehabil, 2012. 27(3): p. 224-233.**

204. **Khoury S, Amzica F, Giguère JF***, et al. Sleep characteristics in Mild Traumatic Brain Injury patients.* **WFSRS, 2011(PO-1-226).**

205. **Chouchou F, Khoury S, Roy I***, et al. Subjective sleep complaints in traumatic brain injury patients are explained by the influence by depression, presence of pain and pain catastrophizing.* **Eur Soc of Res on Sleep, 2012. Paris, France(Submitted).**

Perturbation du sommeil par la douleur chez les traumatisés crâniens légers

Sleep and pain interaction in Mild Traumatic Brain Injury patients

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Mots clés: Traumatisme crânien, sommeil, douleur

Résumé
La prévalence des troubles de sommeil et de douleur chronique est élevée chez le patient ayant subi un traumatisme craniocérébral léger (TCCL). Ces deux troubles peuvent interagir de manière bidirectionnelle : un mauvais sommeil engendre une sensibilité accrue à la douleur et un douloureux chronique se plaint d'un mauvais sommeil. L'interaction entre ces plaintes est suggérée chez les TCCL, mais son étiologie reste encore peu connue. De nouvelles pistes de recherche suggèrent que les TCCL qui souffrent de douleur ont une modification des ondes cérébrales durant leur sommeil qui pourrait expliquer en partie comment les deux symptômes interagissent.

Summary
Mild Traumatic Brain Injury (MTBI) is a major public health concern as patients are left, amongst other symptoms, with sleep complaints and chronic pain. An interaction between the two is suggested and is thought of as a bidirectional relationship. A night of poor sleep is followed by hypersensitivity to pain and chronic pain leads to sleep complaints. New research shows that MTBI patients with pain have more rapid EEG waves during sleep than those without pain. This may suggest that there is an intrinsic relationship between the two complaints. In this review, we suggest new research avenues in sleep-wake disturbances post-MTBI that consider pain and other post-concussion symptoms as important, interrelated factors.

Le traumatisme craniocérébral (TCC) se définit par une force mécanique extérieure à la tête résultant et s'exprimant par un signe de dysfonction cérébrale [13]. Un diagnostic de TCC léger (TCCL) est posé lorsqu'un score de 13 à 15 sur l'échelle de Glasgow est observé et généralement lorsqu'il y a une perte de conscience de moins de 30 minutes, une amnésie rétro- ou antérograde de l'accident d'au plus 24 heures, un état confusionnel ou une lésion cérébrale traumatique aiguë [12].

Le *Center for Disease Control and Prevention* (CDC) a été mandaté par le gouvernement américain pour étudier la prévalence des traumatismes crâniens, craignant un problème de santé publique [6]. Le rapport 2002-2006 a déterminé que l'incidence annuelle est de 600 cas par 100 000 personnes et que 1.7 millions d'individus souffrent d'un traumatisme crânien chaque année, dont 75% sont des TCCL [206]. Même si le TCCL n'est pas fatal, il impose au patient de nombreuses séquelles physiques et psychologiques pouvant s'étaler sur plusieurs années; d'où sa qualification d'épidémie silencieuse [207]. La majorité des individus victimes d'un TCCL sont des hommes âgés entre 25 et 34 ans ou bien les enfants de moins de 4 ans. Les principales causes sont les chutes (35%), les accidents de la route (17%), les coups lors d'activités sportives (16%) et les agressions (10%) [6]. De plus, la guerre actuelle en Irak et en Afghanistan vient d'ajouter une nouvelle catégorie de TCCL, soit celle due à des explosions qui s'avère être un fléau chez 22% des vétérans. Même si la mortalité a baissé dû à des casques de guerre plus robustes, une majorité des soldats vétérans en subissent encore les conséquences [8].

Bien que la majorité des patients TCCL ne présentent pas de séquelles à long terme, une année plus tard, 15% restent symptomatiques [208]. Les symptômes post-commotionnels sont multiples – principalement des céphalées, des troubles de l'humeur (dépression, anxiété), une baisse de concentration, de l'étourdissement ainsi que de la douleur et des troubles de sommeil [209]. Les principaux indicateurs pour prédire la persistances des symptômes post-

commotionnels sont le statut socio-économique bas, des TCC répétés, l'attente ou la réception d'une compensation monétaire [71]. Cependant, il semble que les facteurs autant physiques que psychologiques contribuent à la chronicité des symptômes. En fait, les troubles du sommeil et la douleur (dont la céphalée post-traumatique, les cervicalgies et les douleurs musculosquelettiques) sont parmi les symptômes qui seraient les plus susceptibles de contribuer à l'amplification et/ou la chronicisation des symptômes post-commotionnels [84]. Toutefois, l'interrelation entre les troubles du sommeil et la présence de douleur reste à être déchiffrée car la concomitance est fréquente en clinique et ses impacts sont mal compris tant au niveau de l'étiologie, des facteurs de risques ou la gestion des TCCL. Cette revue vise à décrire l'interaction entre les troubles du sommeil et la douleur chez le sujet victime d'un TCCL.

Les troubles de sommeil post-TCCL :

Les troubles du sommeil et de l'éveil rapportés par la population TCCL est suffisamment importante pour que l'on s'y attarde. Plus de 97% des vétérans de guerre ayant subi un TCCL rapportent des troubles de sommeil [210]. Les troubles du sommeil les plus communs post-TCCL sont l'hypersomnie (allongement de la durée de sommeil), l'insomnie (difficulté d'endormissement et de maintien du sommeil), et le délai de phase (se coucher plus tard et se lever plus tard) [58]. Ces troubles apparaissent rapidement, environ 3 jours après le traumatisme et ce en parallèle avec les troubles de l'humeur telles les conditions de type dépression et anxiété [202].

Le principal trouble du sommeil et de l'éveil suite à un TCC est la somnolence diurne excessive [48]. Ce trouble est surtout présent en phase aiguë, soit dans les trois premiers mois. En phase chronique, soit de 3 mois à 2 ans post-trauma, la somnolence diurne reste présente dans 50% des cas, l'insomnie et les parasomnies (manifestation physique des rêves, cauchemars, paralysie du sommeil…) persistent chez 25% respectivement [49]. Trois (3) ans après un TCC, deux patients sur trois souffrent encore de ces troubles du sommeil et de l'éveil

[118]. Ces plaintes ne seraient pas liées à la sévérité du TCC, mais seraient secondaires ou en cause avec les symptômes dépressifs, résultats controversés dans d'autres études [47,49,58,211].

Toutes les plaintes et troubles du sommeil mentionnées ci-haut ont fait l'objet d'enregistrement du sommeil en laboratoire ou polysomnographie. Le but principal est de détecter une anomalie dans l'architecture du sommeil. Une étude réalisée auprès de jeunes athlètes ayant subi une commotion cérébrale n'a pas montré de différence dans la macrostructure (i.e. pourcentage de chacun des stades de sommeil) du sommeil en comparaison avec des athlètes sans commotion cérébrale, malgré le fait que les athlètes avec commotion rapportaient une mauvaise qualité de sommeil [55]. Une autre étude a montré une diminution du stade 1 (endormissement), une augmentation de l'efficacité du sommeil ainsi qu'une diminution d'éveil durant le sommeil chez les TCC légers en comparaison aux TCC sévères [49]. Chez des adolescents ayant subi un TCCL, une étude montre une diminution de l'efficacité du sommeil, alors qu'une autre n'en trouve aucune [54,60]. La table 1 résume les études polysomnographiques sur les TCCL. Tous ces résultats ne semblent pas se confirmer d'une étude à l'autre; les plaintes de sommeil subjectives ne seraient pas expliquées par un dérèglement de l'architecture du sommeil.

La douleur post-traumatique :

Selon un article de revue récent, 75% des patients ayant subi un TCCL souffrent de douleur chronique et, de façon surprenante, les patients avec TCC léger rapportent plus de douleur que les sujets ayant subi un TCC sévère [66]. Les céphalées post-traumatiques seraient la douleur la plus fréquente et représenterait près de 60% des symptômes post-commotionnels rapportés [67]. Le facteur principal associé à un plus haut risque de présenter des douleurs chroniques suivant le TCCL est la présence de céphalées avant le traumatisme [68-69]. Il a été démontré que les patients TCCL présentant un syndrome de stress post-traumatique et des symptômes associés à la dépression sont plus à risque de

rapporter de la douleur [70-71]. L'intensité de la douleur semble être aussi un bon indice pour prédire de la chronicité des symptômes post-commotionnels [185,202].La chronicité de la douleur ne semble pas être spécifique au TCCL, elle est aussi un fardeau chez les blessés orthopédiques sans atteintes cérébrales [72].

De multiples outils sont disponibles en laboratoire pour étudier la douleur expérimentale ainsi que la détection de modalités sensorielles tels que la température (chaud, froid), la pression, la vibration et la discrimination des surfaces. L'outil de détection thermique consiste en une plaque que l'on pose sur l'avant-bras, qui se réchauffe ou se refroidit, dans un créneau de températures cibles. Cet outil nous permet de mesurer le seuil de détection de la température ainsi que la perception d'une douleur expérimentale causé, soit par la chaleur ou par le froid. L'utilisation de ce paradigme expérimental pour tester les modalités sensorielles n'a pas mis en évidence des différences significatives dans la détection du froid et de la douleur chez les patients TCCL. Toutefois, tel que rapporté, nous avons aussi observé (non publié) que le seuil de détection de la chaleur est sensiblement plus élevé que chez les TCCL qui souffrent de céphalées et de syndromes de stress post-traumatique que chez les sujets sains [73-75]. Suite à ces observations, l'on pourrait se questionner sur l'étiologie centrale de la douleur chez les TCCL [76]. La sensibilité aux modalités sensorielles et non à la douleur expérimentale(au chaud et au froid) chez les TCCL s'avère donc un moyen intéressant d'étudier les mécanismes impliqués dans la douleur, mécanismes qui semblent paradoxaux. En effet, bien que les sujets se plaignent de douleur, celle-ci n'est pas objectivée avec les outils sensoriels quantitatifs usuels. Les nouvelles directions de la recherche devraient comporter l'usage de meilleurs outils (e.g., résonnance magnétique fonctionnelle, contrôle nociceptif diffus inhibiteur, test de perception de la douleur mécanique) pour mettre en évidence un déficit sensoriel (d'origine centrale/périphérique) ou comportemental à exprimer la douleur [201].Une question demeure non-répondue. Le fait que les

patients rapportent de la douleur qui ne peut être «objectivée» à l'aide de tests sensoriels suggérerait-il que d'autres variables soient en cause? Parmi celle-ci, citons la méthode (qu'aurait donné des tests à la vibration, aiguilles, pression, etc.?), les mécanismes psychophysiologiques (attention et vigilance, mémoire de la douleur, dissociation sensorielle et émotive). Une analyse plus poussée en psychophysiologie est requise pour mieux comprendre la dichotomie entre les plaintes et les mesures de la douleur.

Concomitance douleurs et troubles du sommeil chez le sujet TCCL

Une corrélation entre les troubles du sommeil et la douleur chez les TCCL est également rapportée [83-84]. Il est même connu que les TCCL rapportent plus de troubles de sommeil et de douleur que les TCC modérés et sévères [66,212]. Il a été montré que les patients ayant subi un TCC rapportent plus de troubles de sommeil et de douleur que ceux ayant d'autres troubles neurologiques [83]. De plus, dans cette population, la douleur est intimement liée au rapport subjectif d'un trouble de sommeil, plus ils rapportent de la douleur, plus le rapport subjectif de sommeil est mauvais [83]. La fatigue contribue aussi aux troubles de sommeil post-TCC et a été associé à l'anxiété, à la dépression ainsi qu'à la douleur [213]. Une étude de dossier chez les TCCL de notre centre tertiaire hospitalier en traumatologie a révélé que les patients souffrant d'un trouble de sommeil sont plus à risque de rapporter des céphalées, une humeur dépressive et de l'irritabilité à 6 semaines post-trauma [84]. Une autre étude est arrivée à la même conclusion auprès de patients présentant un syndrome de stress post-traumatique [124]. La douleur, les troubles de sommeil et le syndrome post-traumatique sont devenus une triade dominante dite *polytraumatique*.

Une des limites des études actuelles est qu'elles sont basées sur un rapport subjectif des troubles de sommeil au professionnel traitant. En effet, bien que la nature descriptive de ces analyses de cohortes offre une identification des facteurs de risques et de prédictions, elle ne permet pas d'investiguer les liens de cause à effet.

Une étude a été réalisée dans notre laboratoire visant à investiguer les bases physiologiques des observations mentionnées ci-haut sur la relation physiologique entre le sommeil et la douleur. Le sommeil a été étudié de façon quantitative en laboratoire chez 24 patients à environ 6 semaines post-TCCL. En comparaison au sommeil de sujets sains et de sujets TCCL sans douleur, l'analyse quantitative de l'électroencéphalogramme a révélé que les sujets TCCL qui souffrent de douleur modérée à intense présentent une augmentation des ondes cérébrales rapides (p.ex., alpha, beta et gamma) durant leur sommeil paradoxal [214]. Ces résultats mettent en évidence un sommeil paradoxal plus intense, pouvant être plus instable en activité cérébrale. Ce type d'instabilité peut influencer la qualité subjective du sommeil rapportée par les patients TCCL [142]. Ceci laisse suggéré qu`il n'y a pas de dissociation corticale lors du sommeil et que le message sensoriel douloureux traverse la barrière thalamique durant le sommeil. De plus, les TCCL souffrant de douleur rapportent 10 fois plus de symptômes dépressifs et 15 fois plus de catastrophisme face à la douleur que les TCCL qui n'en souffrent pas. Ces facteurs seraient donc aussi importants dans l'interprétation des données de sommeil. Une analyse plus poussée a montré que la dépression expliquerait la plainte de mauvais sommeil [215].

En conclusion, les mécanismes d'interaction entre la douleur et le sommeil chez cette population restent à décortiquer. L'interprétation de ces premiers résultats reste complexe lorsqu'on tient compte de la présence de signes et symptômes multiples (troubles de l'humeur et du sommeil concomitant), l'absence d'hypersensibilité à la douleur expérimentale, ceci malgré des plaintes cliniques importantes. Les actions délétères de la douleur et des troubles du sommeil sont-elles indépendantes, en liens de causalité, dues à un dysfonctionnement cérébral suite au TCCL ou dues à une prédisposition génétique ? Toutes ces pistes restent à explorer. De plus, lors d'études futures en sommeil chez les TCCL, la douleur, devrait être considérée comme un facteur crucial du tableau clinique.

Parsons et al.[54]	1997	8 adolescents	72h 6 semaines 12 semaines	-Pas de différence en macrostructure -↓delta, thêta et alpha1 avec le temps.
Kaufmann et al.[60]	2001	19 adolescents	3 ans	-↓efficacité de sommeil -↑éveils durant le sommeil
Ouellet et al. [61]	2006	14 adultes	21 mois	-Insomnie
Verma et al.[49]	2007	24 adultes	3 mois à 2 ans	-↑ efficacité du sommeil -↓ stade 1 -↓éveils durant le sommeil
Schreiber et al.[62]	2008	26 adultes	12 mois à 21 ans	-↑stade 2 -↓ sommeil paradoxal
Williams et al.[63]	2008	9 adultes	28 mois	-↓efficacité de sommeil -↓latence au sommeil paradoxal
Gosselin et al.[55]	2009	10 athlètes	1 an	- pas de différence en macrostructure - pas de différence spectrale
Rao et al.[56]	2011	7 adultes	1 semaine	- pas de différence en macrostructure
Khoury et al.[214]	2012	24 adultes	45 jours	-↓efficacité de sommeil -↓ondes delta en sommeil paradoxal

Table 1 : Tableau récapitulatif de la littérature sur la polysomnographie de patients TCCL.

Références :

Voir ci-haut