Theta burst stimulation to characterize changes in brain plasticity following mild traumatic brain injury: A proof-of-principle study

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Abstract.
Purpose: Recent studies investigating the effects of mild traumatic brain injury (mTBI) suggest the presence of unbalanced excitatory and inhibitory mechanisms within primary motor cortex (M1). Whether these abnormalities are associated with impaired synaptic plasticity remains unknown.
Methods: The effects of continuous theta burst stimulation (cTBS) on transcranial magnetic stimulation-induced motor evoked potentials (MEPs) were assessed on average two weeks and six weeks following mTBI in five individuals.
Results: The procedure was well-tolerated by all participants. Continuous TBS failed to induce a significant reduction of MEP amplitudes two weeks after the injury, but response to cTBS normalized six weeks following injury, as a majority of patients became asymptomatic.
Conclusions: These preliminary results suggest that cTBS can be used to assess M1 synaptic plasticity in subacute phase following mTBI and may provide insights into neurobiological substrates of symptoms and consequences of mTBI.

Keywords: Traumatic brain injury, concussion, transcranial magnetic stimulation, plasticity, motor cortex

1. Introduction

The Centers for Disease Control and Prevention estimate that between 1.4 and 3.8 millions of mild traumatic brain injuries (mTBI) occur annually in the USA (Rutland-Brown et al., 2006). Although mTBI has been long considered a short-lasting “minor” injury, current literature suggests that it may involve a clinically silent pathological process that is related to subclinical neurophysiologic and neurometabolic changes. An increasing number of studies have revealed the long term impact of mTBI or concussion since the discovery of a possible link between multiple mTBIs and the development of neurodegenerative diseases (Bazarian et al., 2009), such as Alzheimer's disease (Guskiewicz et al., 2005; McCrory, 2011; Mortimer et al., 1985; Plassman et al., 2000), chronic traumatic encephalopathy (Cantu, 2007; McCrory et al., 2007) and amyotrophic lateral sclerosis (Piazza et al.,...
2. Methods

2.1. Participants

Case 1. This 44-year-old right-handed man was playing soccer when he sustained a head-on collision with another player and then hit the ground with his head. There was loss of consciousness (LOC) for about 90 seconds, followed by confusion, blurred vision, agitation and about 1-2 minutes of retrograde and 3-4 minutes of anterograde post-traumatic amnesia (PTA). The symptoms resolved approximately 20 minutes after the event at which point his physical and neurological exams were normal, and he remained alert. He was diagnosed with a Grade 3 concussion according to the American Academy of Neurology classification (1997). For two weeks following the accident, he complained of fatigue and poor concentration, memory problems, mild headaches and some dizziness. These symptoms had markedly improved by week 6, although he still complained of mild headaches, slight fatigue and intermittent memory difficulties. He was not taking any drugs known to alter brain excitability, plasticity, or excitability/inhibition balance. A 6-week period of rehabilitation was recommended.

Case 2. This is a 24-year-old right-handed woman who was involved in a collision with a skateboarder while she was on her bike. Following the impact, she fell over the handle bar. The front of her helmet broke and she sustained a left pre-occipital ecchymosis. The duration of the LOC is unknown. She experienced confusion and retrograde PTA for about 30 sec and anterograde amnesia for approximately 30 min. She has a past medical history of migraines. During a few days following the accident, she experienced some word finding difficulties but she did not report any increase in the frequency of her migraines or changes in her concentration. Her physical and neurological examinations were normal. She was diagnosed with a Grade 3 concussion according to the American Academy of Neurology classification (1997). She was not taking any drugs known to alter brain excitability, plasticity, or excitability/inhibition balance. Her past medical history, review of system and family history were negative.

Case 4. This is a 28-year-old right-handed woman who was involved in a car-pedestrian collision. The presence of LOC is unknown and there was no report of anterograde/retrograde PTA. Following the incident, the patient experienced increased intensity and frequency of headaches. Her neurological and physical exams were normal. She was diagnosed with a Grade 2 concussion according to the American Academy of Neurology classification (1997). A computed-tomography brain (CT-scan) of her head revealed a small right peritrigonal subgaleal hematoma along the vertex with no underlying fracture. She was not taking any drugs known to alter brain excitability, plasticity, or excitability/inhibition balance. Her past medical history, review of system and family history were negative.

2.2. Procedure

All participants were seen approximately two weeks post-mTBI (M ± 14 ± 3 days) and again approximately six weeks post-injury (separated by 61 ± 19 days). All participants were first seen by a neurologist and had to meet the concussion criteria of the American Academy of Neurology (1997).

All participants completed the TMS safety questionnaire (Rossi et al., 2011) prior to testing to screen for possible contraindications. On visit 1, a brain magnetic resonance imaging (MRI) exam was performed, followed by baseline measures of TMS and the cTBS procedure. On visit 2, TMS and cTBS procedures were repeated. All participants gave their written informed consent for the study, which had been approved by the Institutional Review Board of Beth Israel Deaconess Medical Center.

2.3. TMS recordings

All participants underwent an anatomical brain MRI, using a 3-Tesla GE scanner, to rule out structural lesions and to generate matching baseline images to guide magnetic stimulation. For single-pulse, a Nexstim stimulator (Nexstim Ltd, Helsinki, Finland) was used, delivering biphasic pulses with a current flowing in the brain with an antero-posterior and then a postero-anterior (AP-PA) direction. For repetitive TMS, i.e. cTBS, a MagPro stimulator (MagVenture A/S, Farum, Denmark) was used, delivering biphasic pulses with the current flowing in an AP-PA direction. In order to ensure stable coil positioning over the stimulation site during the experiment and to ensure that the exact same cortical location was
targeted within each study session as defined by each individual’s brain MRI, a Nexstim eXimia Neuromarker system was used. During stimulation, surface electromyography (EMG) was recorded and each TMS pulse and to contract the FDI at approximately 20% of maximal voluntary contraction. In order to control for prior motor contraction during the measurement of AMT, participants were asked to contract the FDI muscle for approximately 2 s prior to each TMS pulse and to relax it about 1 s after each TMS pulse, for at least 3 s. The eTBS protocol was applied approximately 1 min after the end of the AMT measurement procedure; the experiments monitored the relaxation of hand muscles continuously during and after the stimulation.

2.5. eTBS protocol

Continuous TBS was applied using parameters similar to those used by Huang et al. (Huang et al., 2005): three pulses at 50 Hz, with an interval of 200 ms between the last pulse of a triplet and the first pulse of a second triplet (i.e., with an interstimulus interval of 240 ms), for a total number of 600 pulses. Thus, in the present eTBS paradigm, the triplet repetition rate was about 4.17 Hz instead of 5 Hz; both frequencies being included in the theta band. The intensity was fixed at 80% of AMT. This paradigm was recently shown to induce significant suppression of MEPs in healthy controls (see Vernet et al., 2014). Before eTBS, two to three batches of 20 to 30 MEPs (60 in total) were acquired in response to stimulation over the optimal FDI location, at an intensity of 120% of RMT and a rate of approximately 0.1 Hz (a random jitter of ±1 s was introduced to avoid any training effects). Such measures allowed verifying for stability of the pre-eTBS measure of excitability; moreover, the second batch was used as the baseline to which the post-eTBS measures of excitability were compared. Following eTBS, a single batch of MEPs was measured immediately after (T0) and then at 3, 10, 20, 30, 40, 50, 60, 75, and 90 min following eTBS to track changes in amplitude over time.

2.6. Data analysis

MEP peak-to-peak amplitude was automatically determined using the Nexstim Neuromarker Analysis software and then visually inspected. Mean right M1 MEP peak-to-peak amplitudes for each of the time points were used for analysis. Paired-sample t-tests were conducted to assess the reproducibility of baseline MEP amplitude. A within-subjects repeated measure multivariate analysis of variance (MANOVA) was used to compare the impact of eTBS on MEP amplitude over time, using session (session 1 and 2) and MEP measures (11 time points) as within-group factors.

Paired-sample t-tests were used to identify the effect of eTBS at the different time points in comparison to the baseline MEP measure. The critical p-value was set to 0.05. Because of the very small sample and the exploratory nature of the present case report, no correction for multiple comparisons was applied. The participant (case 4) did not complete the second session. For statistical analyses, the missing data were replaced by the average data from the 4 other cases. All analyses were performed on raw TMS data. All statistical tests were two-tailed and performed using the Statistical Package for the Social Sciences (version 21).

3. Results

A questionnaire was used at the beginning and at the end of each session to evaluate the presence of pain and discomfort. Two patients reported the presence of mild discomfort during the procedure. Case 2 reported, at the beginning of session 2, mild headache, for which acetaminophen was given and, at the end of session 2, mild neck pain. Again, at the beginning of session 2, Case 2 reported mild headache and trouble concentrating and, at the end of session 2, a mild neck pain in addition to those symptoms. Case 3 reported mild neck pain at the beginning and at the end of both sessions. Thus, the only side-effect associated with the procedure was a mild neck pain for Case 2.

Raw tracings showing average MEP at both time points following concussion in a patient are shown in Fig. 1. A paired-sample t-test revealed no significant difference between baseline MEP measures from both sessions (t(14) = 0.7, p = .44). The MEP response profiles in the two sessions were not parallel as indicated by a significant [session $\times$ time] interaction (F = 2.23, df = 10, p = 0.035) (Fig. 2). Subsequent paired-sample t-tests revealed no significant reduction in the MEPs size compared to baseline at all time points for session 1 (Table 1). A significant inhibition of the MEPs compared to baseline at T0, T5, T20, T30, T40, T50 and T90 was observed for session 2 (Table 2). Individual data are shown in Fig. 3.

4. Discussion

The goal of this proof-of-principle study was to investigate the feasibility of using eTBS to evaluate
Table 1

<table>
<thead>
<tr>
<th>Time point</th>
<th>Mean difference (SD)</th>
<th>t value</th>
<th>p value</th>
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<tbody>
<tr>
<td>T0</td>
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<td>-4.62</td>
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<tr>
<td>T1</td>
<td>5.06 ± 0.27</td>
<td>-4.62</td>
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<td>T2</td>
<td>6.06 ± 0.27</td>
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<td>T3</td>
<td>7.06 ± 0.27</td>
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<td>T4</td>
<td>8.06 ± 0.27</td>
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<tr>
<td>T5</td>
<td>9.06 ± 0.27</td>
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<td>T6</td>
<td>10.06 ± 0.27</td>
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<tr>
<td>T7</td>
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<tr>
<td>T8</td>
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<tr>
<td>T9</td>
<td>13.06 ± 0.27</td>
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Fig. 2. Mean MEP amplitude and standard deviations following cTBS over the different points and for session 1 and 2. Error bars show standard deviations. No significant reductions are observed in MEP amplitudes for the first session, although a small trend is observed with the last time point. Significant reductions of MEP amplitudes are observed for sessions two and three for 7 out of the 10 time points. *p < 0.05, **p < 0.01.

Fig. 3. A) Individual mean MEP amplitudes for the first cTBS session. High variability is observed between subjects for each subject and therefore no clear inhibitory pattern can be visually observed. B) Individual mean MEP amplitudes for the second cTBS session. Significant reductions in MEP amplitudes are observed for sessions two and three for 7 out of the 10 time points. *p < 0.05, **p < 0.01.

Table 2

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Note: *p < 0.05, **p < 0.01.

patients. Preliminary results suggest the presence of altered plasticity changes in the acute phase of mTBI, failure to detect the change in MEPs in the acute phase, which could reflect altered M1 LTD-like mechanisms. Significant cTBS-related suppression of MEPs was observed in 5 patients with mTBI who did not have a history of mTBI and were subjected to recurrent sub-concussive blows through contact sports. Additionally, the age of participants in the present study ranged from 22 to 44 years, which might be a confounding factor. Indeed, it has been shown that cTBS effects are modulated by age, where less cortex plasticity is observed in older individuals (Pearce et al., 2011). More specifically, increased intracortical inhibition (Christyakov et al., 2001; Pearce et al., 2014) and increased intracortical facilitation (Powers et al., 2014) have been reported in the acute and subacute phases of mTBI. Despite strong evidence suggesting inhibitory/excitatory imbalance in the primary motor cortex of individuals with mTBI, the duration of such effects is unclear. Pearce and collaborators (2014b) found increased GABA-related inhibition in 44 and 56 patients with mTBI, which indicates that there is a change in GABA-related inhibition in 44 and 56 patients with mTBI. In our study, we observed similar changes in GABA-related inhibition which lasted 4 months after the concussive event. Intracortical inhibition has also been reported to be increased 1-4 weeks (Powers et al., 2014) and 9 months after a concussion (Tremblay et al., 2017) and within normal values 41 months post-injury (Tremblay et al., 2014).

In the present study, we show reduced synaptic plasticity in the subacute phase as indexed by the response to cTBS, and that this abnormality persists for weeks post-injury. An association between abnormal intracortical excitability and aberrant synaptic plasticity has been previously shown in concussed athletes on average 14 months post-injury. De Beaumont et al. (2012) reported that increased silent period durations in concussed athletes, presumably reflecting faulty GABAergic transmission, were negatively correlated with the level of synaptic plasticity induced with paired associative stimulation. In the present study, the hyperexcitability or hypoinhibitory state of M1 intracortical networks could prevent the injured brain from responding adequately to the effects of cTBS and therefore be an accurate marker of early abnormal plasticity. The inability of the injured brain to respond to cTBS appears short-lived, however, with results consistent with the previous study by De Beaumont and collaborators (2012) who showed persistent motor cortex LTD and LTP-like deficits in the chronic phase following sport concussion. This discrepancy could be explained by the fact that the current sample included 4 patients with mTBI who did not have a history of mTBI and that were not subjected to recurrent sub-concussive blows through contact sports. Additionally, the age of participants in the present study ranged from 22 to 44 years, which might be a confounding factor. Indeed, it has been shown that cTBS effects are modulated by age, where less cortex plasticity is observed in older individuals (Pearce et al., 2011). More specifically, increased intracortical inhibition (Christyakov et al., 2001; Pearce et al., 2014) and increased intracortical facilitation (Powers et al., 2014) have been reported in the acute and subacute phases of mTBI. Despite strong evidence suggesting inhibitory/excitatory imbalance in the primary motor cortex of individuals with mTBI, the duration of such effects is unclear. Pearce and collaborators (2014b) found increased GABA-related inhibition in 44 and 56 patients with mTBI, which indicates that there is a change in GABA-related inhibition in 44 and 56 patients with mTBI. In our study, we observed similar changes in GABA-related inhibition which lasted 4 months after the concussive event. Intracortical inhibition has also been reported to be increased 1-4 weeks (Powers et al., 2014) and 9 months after a concussion (Tremblay et al., 2017) and within normal values 41 months post-injury (Tremblay et al., 2014).

Continuous TBS has been used with various populations to non-invasively probe synaptic plasticity in the conscious human brain. This method has many advantages over other techniques as its applicability (on and off) and that are not subjected to recurrent sub-concussive blows through contact sports. Additionally, the age of participants in the present study ranged from 22 to 44 years, which might be a confounding factor. Indeed, it has been shown that cTBS effects are modulated by age, where less cortex plasticity is observed in older individuals (Pearce et al., 2011). More specifically, increased intracortical inhibition (Christyakov et al., 2001; Pearce et al., 2014b; Miller et al., 2014; Powers et al., 2014) and increased intracortical excitability following intermittent TMS (Tremblay et al., 2017; 56 healthy participants. A similar absence of significant modulations of excitability was reported for iTBS in a sample of 52 healthy patients. As a whole, this study provides evidence suggesting that TBS can be used safely to assess motor cortex plasticity in individuals with TBI. Studies with larger and more homogeneous samples are needed to determine the clinical usefulness of TBS for evaluating plastic changes related to TBI.

Animal studies have shown that bursts of 5-5 pulses at 50 Hz induce LTD/PLD when applied to the motor cortex or hippocampus (Hess & Donoghue, 1996; Larson et al., 1986). While the exact mechanism underling the effects of cTBS on the human brain is still unknown, it has been suggested that TBS suppression following stimulation could be related to long-term depression (LTD)-like processes mediated by N-methyl-D-aspartate receptors (NMDA-R), as NMDA-R antagonist memantine was shown to block the after effects of cTBS (Huang et al., 2007). Modulation of GABA receptors (Thickbroom, 2007; 2007; 2008; and glutamate receptors (Glu-R) Huang et al., 2007) has also been proposed as a possible mechanism explaining excitability changes following TBS. TBS could therefore target both excitatory and inhibitory networks within the human motor cortex (Cádiz-Morales et al., 2010). The present data are in line with this hypothesis since M1 alterations in glanuates (Babikian et al., 2006; Henry et al., 2010; Shutter et al., 2004) and abnormal interactions between M1 GABA and glutamate (Tremblay et al., 2014) have been shown in acute/subacute and chronic phases of TBI and sport-related mTBI using magnetic resonance spectroscopy. Abnormal GABA and glutamate transmission could therefore partly explain the inhibitory/excitatory imbalance found in the motor cortex of individuals with mTBI and its associated effects on synaptic plasticity.
Conflict of interest statement


References


References


