Original Investigation

Neurometabolites Alteration in the Acute Phase of Mild Traumatic Brain Injury (mTBI): An In Vivo Proton Magnetic Resonance Spectroscopy (1H-MRS) Study

Vigneswaran Veeramuthu, PhD*, Pohchoo Seow, MSc*, Vairavan Narayanan, FRCS, Jeannie Hsiu Ding Wong, PhD, Li Kuo Tan, MEng, Aditya Tri Hernowo, PhD, Norlisah Ramli, FRCR, MBBS

Rationale and Objectives: Magnetic resonance spectroscopy is a noninvasive imaging technique that allows for reliable assessment of microscopic changes in brain cytoarchitecture, neuronal injuries, and neurochemical changes resultant from traumatic insults. We aimed to evaluate the acute alteration of neurometabolites in complicated and uncomplicated mild traumatic brain injury (mTBI) patients in comparison to control subjects using proton magnetic resonance spectroscopy (1H magnetic resonance spectroscopy).

Material and Methods: Forty-eight subjects (23 complicated mTBI [cmTBI] patients, 12 uncomplicated mTBI [umTBI] patients, and 13 controls) underwent magnetic resonance imaging scan with additional single voxel spectroscopy sequence. Magnetic resonance imaging scans for patients were done at an average of 10 hours (standard deviation 4.26) post injury. The single voxel spectroscopy adjacent to side of injury and noninjury regions were analysed to obtain absolute concentrations and ratio relative to creatine of the neurometabolites. One-way analysis of variance was performed to compare neurometabolite concentrations of the three groups, and a correlation study was done between the neurometabolite concentration and Glasgow Coma Scale.

Results: Significant difference was found in ratio of *N*-acetylaspartate to creatine (NAA/Cr + PCr) ($\chi^2(2) = 0.22$, $P < .05$) between the groups. The sum of NAA and *N*-acetylaspartylglutamate (NAAG) also shows significant differences in both the absolute concentration (NAA + NAAG) and ratio to creatine (NAA + NAAG/Cr + PCr) between groups ($\chi^2(2) = 4.03$, $P < .05$and ($\chi^2(2) = 0.79$, $P < .05$)). NAA values were lower in cmTBI and umTBI compared to control group. A moderate weak positive correlation were found between Glasgow Coma Scale with NAA/Cr + PCr ($\rho = 0.36$, $P < .05$ and NAA + NAAG/Cr + PCr ($\rho = 0.45$, $P < .05$)), whereas a moderate correlation was seen with NAA + NAAG ($\rho = 0.38$, $P < .05$).

Conclusion: Neurometabolite alterations were already apparent at onset of both complicated and uncomplicated traumatic brain injury. The ratio of NAA and NAAG has potential to serve as a biomarker reflecting injury severity in a quantifiable manner as it discriminates between the complicated and uncomplicated cases of mTBI.

Key Words: Neurometabolite; mild Traumatic Brain Injury (TBI); Magnetic Resonance Spectroscopy (MRS); *N*-acetylaspartate (NAA); Glasgow Coma Scale (GCS).

Acad Radiol 2018; ■:■■–■■

From the Division of Neurosurgery, Department of Surgery, Faculty of Medicine, University of Malaya, Kuala Lumpur (V.V., V.N., A.T.H.); Brain and Cognition Recovery Centre, Gleneagles Medini Hospital, Iskandar Puteri, Johor, Malaysia (V.V.); Department of Psychology, University of Reading Malaysia, Persiaran Graduan Kota Ilmu, Educity, Johor, Malaysia (V.V.); Department of Biomedical Imaging, Faculty of Medicine, University of Malaya, Jalan Universiti, 50603, Kuala Lumpur, Wilayah Persekutuan Kuala Lumpur, Malaysia (S.P., J.W.H.D., T.L.K., N.R.); University of Malaya Research Imaging Centre, Faculty of Medicine, University of Malaya, Kuala Lumpur, Malaysia (S.P., J.W.H.D., T.L.K., N.R.). Received November 13, 2017; revised December 18, 2017; accepted January 3, 2018. *Equal first authors (Vigneswaran Veeramuthu & Seow Pohchoo have equal contributions to this manuscript). **Address correspondence to:** N.R. e-mail: norlisahramli@gmail.com

https://doi.org/10.1016/j.acra.2018.01.005

INTRODUCTION

Deranged metabolites-induced cellular energy crisis is a common occurrence in traumatic head injury (1). The cascading events post trauma may lead to terminal membrane depolarization with excessive release of excitatory neurotransmitters (2), lysis of the cell membranes and apoptosis, disrupting various neural connectivity networks and consequentially affecting neurocognitive function or performance (1). Injury severity in mild traumatic brain injury (mTBI) could be categorized into complicated or

uncomplicated. A complicated mTBI is differentiated from an uncomplicated mTBI by the presence of a closed depressed skull fracture or trauma-related intracranial abnormality or lesion (3,4).

The advent of advanced magnetic resonance imaging (MRI) techniques in recent years have enabled reliable assessment of microscopic changes in brain cytoarchitecture, neuronal injuries, and neurochemical changes resultant from traumatic insults. While conventional computed tomography scans and structural magnetic resonance sequences are usually unable to detect such physiological and biochemical changes occurring at cellular level (5), magnetic resonance spectroscopy (MRS) is capable of evaluating metabolic perturbation associated with mTBI in vivo.

Several MRS studies showed differences between the semiacute, subacute, and chronic stages of mTBI. A study of semiacute mTBI with matched controls found elevated concentrations of glutamate plus glutamine (Gln) signal in the white matter but reduced gray matter (GM) concentrations of Gln at grey matter (GM), 13 days postinjury (6). The reduced GM Gln suggested reduced neural activity. The study also reported elevated WM concentrations of total creatine (Cr), believed to be consistent with an upregulation of WM metabolic activity and ionic balance restoration of high-energy phosphates pool for cellular repair.

Between the early subacute and chronic stage of mTBI, George et al. (2014) reported decreasing trends of thalamic NAA/Cr (*N*-acetylaspartate/creatine) and Cho/Cr (choline/creatine) levels measured in thalamus and centrum semiovale, corresponding with cognitive measures (5), whereas Kierans et al. (2014) found increased putaminal myoinositol (mI), myoinositol/creatine (mI/Cr) and total glutamine/creatine (Gln/Cr) that reflected complex glial and excitatory response to injury in mTBI patients compared to control (7). At chronic stage of 6 months post trauma, decreased NAA/Cr and NAA/Cho in white matter (WM) regions were reported in the parietal lobe and occipital lobe, respectively, together with increased Cho/Cr in the occipital GM (8). There is a scarcity of data on MRS at the acute stage of mTBI, which can, in principle, provide useful information on the very early proton MRS changes. This study aim to investigate the immediate and early neurometabolic changes, in the immediate aftermath of mTBI (10 hours post trauma). Absolute and the more clinically available ratio of metabolites from the brain spectra of patients with complicated mTBI (cmTBI), uncomplicated mTBI (umTBI), and healthy controls were also explored.

METHODS

Participant Recruitment

This prospective study of 48 subjects is composed of 23 cmTBI patients, 12 umTBI patients, and 13 healthy age-matched subjects as the control group. mTBI patients were recruited from the emergency department of the hospital for the duration of 11 consecutive months from April 2013 to March 2014. Informed consent was obtained from the patients and control groups. Local institutional review board (UM/EC. 949.15) ethical approval was obtained for this study. We adopted the Center for Disease Control's (USA) definition of mTBI (9). Mild TBI is generally characterized by one or more of the following: (1) confusion or disorientation, (2) loss of consciousness for less than 30 minutes, (3) transient focal neurological signs, and (4) Glasgow Coma Scale (GCS) of 13–15 upon clinical evaluation. The grouping of injury complexity as complicated (cmTBI) and uncomplicated (umTBI) groups was determined by the presence of intracranial lesion and/or skull fracture, as evaluated by two blinded clinicians, NR (a neuroradiologist) and VN (a neurosurgeon). Patients with a normal brain computed tomography scan (no fracture and no intracranial injury) were classified as umTBI. Details of the inclusion and exclusion criteria of the study are presented in Figure 1.

Age matched healthy controls were recruited on voluntary basis. They were either staff members of the hospital or patients' next of kin. Stringent exclusion criteria as of the patients were used in the recruitment effort.

Magnetic Resonance Imaging Acquisition

MRI scan was performed on cmTBI and umTBI patients upon admission using a 3T MRI scanner (Signa HDx, GE Healthcare, Harvey, IL) with a dedicated 8-channel head coil followed by single voxel spectroscopy (SVS) sequence. The standard imaging protocol and parameters were as follows: (1) axial T1-weighted three-dimensional fast spoiled gradient-recalled echo (FSPGR) with imaging parameters: repetition time (TR) = 6.7 ms, echo time (TE) = minimum 1.9 ms, field of view (FOV) = 31 mm, matrix = 256 × 256, slice thickness = 1.2 mm, slice overlap = 0.6 mm with image scan time of 3 minutes and 48 seconds; and (2) axial T2-weighted fast spin echo (TR = 4240 ms, TE = 102 ms, FOV = 24 mm, matrix = 512 × 384, thickness = 5 mm, spacing = 1.5 mm, and image scan time of 2 minutes and 30 seconds. MRS technique was performed using a point-resolved single-voxel spectroscopy probe-p sequence with the following parameters: TR = 1500 ms, TE = 35 ms, voxel size = 19 × 19 × 19 mm, slice thickness = 20 mm, number of excitation = 128, and scan time of 7 minutes. Axial T1W/T2 weighted were used to position SVS. For cmTBI, SVSs were placed at the white matter adjacent to injury site and the contralateral normal appearing white matter (Fig 2c). For umTBI and control, the SVS were placed in frontal or occipital white matter (Fig 3c and 4c). The SVS placement was carefully placed so as to avoid contact with the subcutaneous fat, skull, vasculature, arachnoid space, and cerebrospinal fluid to mitigate shimming and water suppression effects. Outer volume saturation bands were placed over the MRS volume over anatomy of interest to suppress signal from fat-containing scalp that might contaminate the spectrum. No higher order shimming was used in our study.