Role of Magnetic Resonance Spectroscopy for Prognosis of Patients with Traumatic Brain Injury

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Abstract

Background Traumatic brain injury (TBI) is one of the leading causes of death worldwide. Long-term clinical outcome following TBI can be difficult to predict. Evaluation of the degree of severity of injury and prediction of outcome are important for the management of these patients.

Objective To evaluate whether degree of severity of injury and outcome in moderate to severe TBI is possible by proton magnetic resonance spectroscopy (1H-MRS).

Materials and Methods Patients with moderate (Glasgow coma scale [GCS] 9–13) and severe head injury (GCS: 5–8), within 1 week of trauma with their normal computed tomographic (CT) scan findings, their magnetic resonance imaging (MRI) finding, and neurologic status were investigated with single-voxel proton MRS (^{1}H-MRS). The study included 51 patients and 24 controls.

Result The MRS study revealed lower ratio of *N*-acetylaspartate (NAA)/choline (Cho) and NAA/creatine (Cr) and higher ratio of Cho/Cr and lactate level compared with the control group. The ratio of NAA/Cr, NAA/Cho, and Cho/Cr were statistically significant with initial GCS (p = < 0.00001, r = 0.7595; p = < 0.00001, r = 0.7506; and p < 0.00001, r = −0.5923, respectively), and these ratios were also statistically significant with Glasgow outcome scale (GOS) (p < 0.00001, r = 0.8498, p < 0.00001, r =0.9323, p < 0.00001, r = −0.9082, respectively). The ratio of NAA/Cr, NAA/Cho, and Cho/Cr were also statistically significant with severity of injury (p < 0.0001).

Conclusion MRS can quantify damage after TBI and may be a method of assessing severity and outcome in TBI.

Keywords
- proton magnetic resonance spectroscopy
- traumatic brain injury
- N-acetylaspartate
- choline
- creatinine

Introduction

Traumatic brain injury (TBI) is a leading cause of mortality and long-term disability, particularly affecting young people. It has been called a "silent epidemic." TBI is the most frequent etiology of severe brain damage among young and middle-aged adults (as compared with stroke and tumors in old-aged patients; Katz, 1997). Efforts have been made to develop tools that aid in detecting injury severity and outcomes. The more commonly used indicators of the severity of TBI include Glasgow coma scale (GCS) scores,[1] duration of impaired consciousness and posttraumatic amnesia,[2] presence of nonreactive pupils,[3] and brain imaging techniques.[4] Unfortunately, these indicators of severity have not proved sufficiently accurate in predicting outcomes.

Magnetic resonance imaging (MRI) is more sensitive than computed tomography (CT) at detecting stroke in the early phase, subtle abnormalities related to anoxic/hypoxic encephalopathy, and diffuse axonal injury (DAI) in TBI patients. However, in approximately 5 to 10% of these TBI patients, anatomical lesions detected by classical morphological MRI (sequences such as T2*, fluid-attenuated inversion recovery [FLAIR], and diffusion) are unable to explain their clinical status and to give clue about their chance of recovery. These

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patients present significant problems concerning diagnosis and misdiagnosis, prognosis, and therapy (Andrews et al, 1996; Childs and Mercer, 1996; Schnakers et al, 2009).

Current imaging does not permit quantification of neural injury after TBI and therefore limits both the development of new treatment and the appropriate counseling of patients concerning prognosis. Advances in neuroimaging techniques may allow the development of new treatments and refinement of existing therapies aimed at preventing neuronal injury and ultimately improving functional outcome for patients.

Proton magnetic resonance spectroscopy (MRS) imaging is a powerful method that combines MRI and MRS for examining brain metabolism.[5] MRS differs from MRI in that the signal from water protons is suppressed, thus allowing signals obtained from other metabolites containing protons to be measured. The protons on each metabolite resonate at a particular frequency, referred to as the "chemical shift." This frequency is determined by the local magnetic environment and varies slightly from water protons.

The proton signal from each metabolite is plotted as a peak on a magnetic resonance spectrum at a known chemical shift on the horizontal axis (measured in parts per million [ppm]) with the vertical scale representing relative metabolite concentrations. Several key brain metabolites are measured with proton MRS, using both short (i.e., TE = 20–40 milliseconds) and long (i.e., TE = 135–270 milliseconds) echo time techniques. Although MRS can detect many metabolites, there are seven commonly measured metabolites in the clinical setting (►**Fig. 1**). Metabolite information is measured on a regional basis from specific volumes, or "voxels." Single voxel spectroscopy (SVS) allows acquisition of a single spectrum from one volume element (voxel) at approximately 8 cc or more. MRS obtains chemical signals, or metabolites, from a region of interest (ROI or voxel).

A spectrum of peaks is generated whereby each peak is reflective of a chemical that resonates at a specific frequency, and the height of the peak reflects the concentration of the specific marker in the brain.[5] It has been postulated that a temporary decrease in *N*-acetylaspartate (NAA) levels after brain injury may be caused by accelerated lipid synthesis involved in myelin repair or may be attributable to NAA providing a temporary source of cellular energy locally at the site of axonal injury, which would produce a transient decrease that might precede any loss of NAA as a result of axonal death.[6]

As a choline (Cho) derivative, the spikes of Cho are composed of several compounds, mainly free Cho, choline glycerophosphatide, and phosphocholine.[7] The synthesis and decomposition of Cho cause cytoplasmic membrane metabolic changes. Cho increases indicate incision injury of the myelin sheath and cytoplasmic membrane rupture.

Lactate (La) due to impaired aerobic glycolysis is a specific marker for posttraumatic secondary hypoxic and ischemic injury.[8]

Creatine (Cr) is a marker for intact brain energy metabolism. It is found in glia and neurons and serves as a point of reference because its level is believed to be stable.

The aim of the current study was to determine whether there was a correlation between early reduction in NAA and increase in Cho and La level with the severity of injury or neurologic outcome of the patients.

Materials and Methods

Patient Population

After obtaining signed consent, as approved by our institutional review board, this was a prospective observational study of TBI patients admitted to our SSKM Hospital, Kolkata.

This study was conducted between November 2013 and December 2015 and a total 51 patients with head injury (male: 38, female: 13) and 24 controls (male: 18, female: 6) were included in the study. If required, sedation was maintained with the propofol infusion under the guidance of the neuroanesthetist, which is present in the magnet room throughout the MRI examination. Causes of injury included road traffic accident (RTA, 37, fall from height (10), physical traumas (1), and others (3). The follow-up periods were at the third and sixth

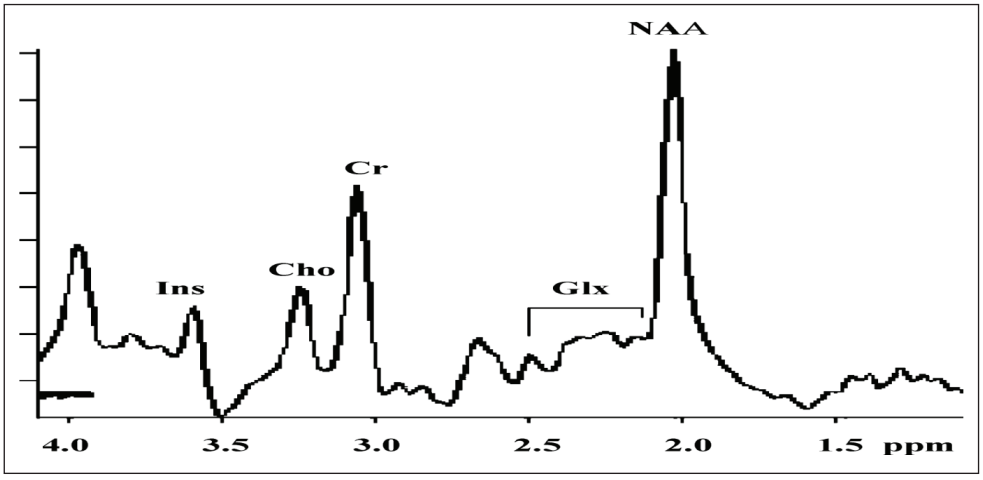

Fig. 1 Representative spectrum of proton MRS (STEAM; TR/TE/TM = 3000/20/13 milliseconds) from occipital gray matter of the adult human brain. Each peak is labeled with the associated molecule. Note that lactate and lipid are not observed in a healthy brain but are provided for reference information (inositol [Ins], 3.56; choline [Cho], 3.22; creatine [Cr], 3.03; glutamate [Glx], 2.05–2.5; *N*-acetylaspartate [NAA], 2.02; lactate [Lac], 1.33; lipids, 0.9–1.2). The chemical shift information in parts per million (ppm) is also provided for each metabolite.

months from the date of discharge of the recruited patients that ended in December 2015.

Inclusion criteria included patients presenting with nonpenetrating TBI, age range 5 to 55 years (mean age: 21.9 years), having moderate head injury (32 patients) and severe head injury (19 patients) based on GCS score (GCS 5–13)[9] within 1 week of head injury who underwent clinical and CT examination, and discrepancy between the apparently normal CT scan findings and their neurologic status were studied with MRI and MRS findings. Age range of control was 8 to 55 years with mean age of 24.5 years.

Exclusion criteria included patients having penetrating head injury; age group below 5 years and above 55 years; having obvious CT scan finding, such as contusion, extradural hemorrhage (EDH), acute subdural hemorrhage (SDH), intraventricular hemorrhage (IVH), subarachnoid hemorrhage (SAH), brainstem involvement having minimal and mild head injury (GCS > 13), and critical head injury (GCS < 5) based on GCS score; and polytrauma that caused unstable hemodynamic status, spine injury, and injury involving multiorgan involvement.

Outcome Measures

Clinical variables assessed included age, demographic data and medical history, cause of brain injury, GCS scores, presence of associated injuries, hemodynamic status, laboratory and radiologic data, and the number of days after the insult when MRS was performed. Neurologic assessments were performed daily during the period of hospital stay, at discharge, and at 3 and 6 months after injury. Clinical outcomes were determined using the Glasgow outcome score (GOS) (Jennett and Bond, 1975; Wilson et al, 1998). The GOS attributes a patient with a score in the range 1 to 5, whereby a patient who dies scores 1, a patient who remains in a vegetative state scores 2, a patient who is with a severe disability scores 3, a patient who has moderate disability scores 4, and a patient who makes a good recovery scores 5. The populations of the two cohorts were statistically comparable. Of the 51 patients, 11 patients had good recovery, 10 patients had moderate disability, 15 patients had severe disability, 6 patients were in a persistent vegetative state, and 9 were dead.

Analysis

MRI and MRS were performed using 3-T MRI scanner (Siemens verio dot 3T; Siemens AG). Proton spectra were acquired from the posterior aspect of one of the frontal lobe, containing predominantly deep white matter tract, corpus callosum region. The voxel was carefully positioned to avoid any areas of T1 and T2 abnormality. Localization of the signal was performed using T2-weighted transaxial, coronal, and sagittal (three planes), with echo time (TE) 135 and repetition time (TR) 1,700 to 2,000. A single voxel acquisition (voxel size 2×2×2 cm^3; number of acquisition 128 acquisition time 4.1 minutes) was used. Spectra data were autoanalyzed using the Siemens verio dot syngo platform. Postprocessing done on syngo main console platform and software to automatically obtain the ratio of NAA/Cr, NAA/Cho, and Cho/Cr. Image analyses were completed by one senior radiologist and one senior neurosurgeon.

Statistical Methods

For statistical analysis, data were entered into a Microsoft Excel spreadsheet and then analyzed by SPSS 20.0.1 (IBM Inc.) and GraphPad Prism version 5 (GraphPad Software, Inc.). Data have been summarized as mean and standard deviation (SD) for numerical variables and count and percentages for categorical variables. The median and the interquartile range have been stated for numerical variables that are not normally distributed. Student's independent sample's *t*-test was applied to compare normally distributed numerical variables between groups. Unpaired proportions were compared by chi-square test or Fischer's exact test, as appropriate.

Explicit expressions that can be used to carry out various *t*-tests are given in the following text. In each case, the formula for a test statistic that either exactly follows or closely approximates a *t*-distribution under the null hypothesis is given. Also, the appropriate degrees of freedom are given in each case. Each of these statistics can be used to carry out either a one- or a two-tailed test.

Once a *t* value is determined, a *p* value can be found using a table of values from Student's *t*-distribution. If the calculated *p* value is below the threshold chosen for statistical significance (usually the 0.10, the 0.05, or 0.01 level), the null hypothesis is rejected in favor of the alternative hypothesis.

To test for differences in the values of spectrum between the control group and TBI group, analysis of variance (ANOVA) test was applied to correlate the GCS and GOS scores with the metabolite ratios. The statistician was blind to the two cohorts (►**Table 1**).

Table 1 Acquisition parameters of different scan type

	TR (ms)	TE (ms)	Average	Section thickness (mm)	Intersection gap (mm)	Acquisition time(s)	Field of view (cm^2)
T1-spin echo	450	20	2–3	5	5	120	220 × 220
T2-TSE	4,000	Within 90	2–3	5	5	120	220 × 220
T2-FLAIR	8,800	90	2	5	5	120	220 × 220
T2-GRE	500–600	Within 20	2–3	5	5	120	220 × 220
MRS	1,700–2,000	135	2–3			246	

Abbreviations: FLAIR, fluid-attenuated inversion recovery; GRE, gradient-echo; MRS, magnetic resonance spectroscopy; TE, echo time; TR, repetition time; TSE, turbo spin echo.

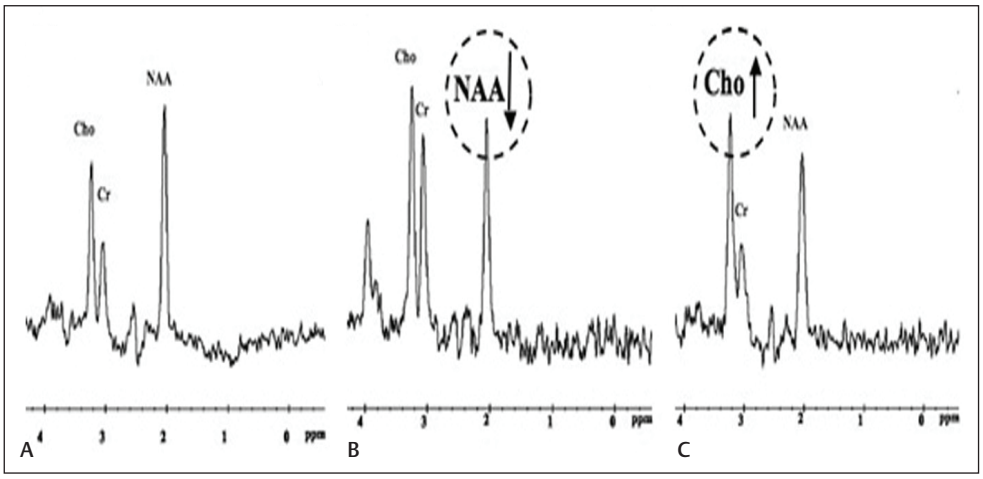

Fig. 2 (**A**) normal profile (the peak of *N*-acetylaspartate [NAA] is higher than the peaks of choline [Cho] and creatine [Cr]), and neuronal loss profile (**B**) and (**C**). The NAA peak is decreased and increased Cho peak with no change in the Cr.

Results

MRS profile of the pons after TBI from a controls and a patient is shown in ►**Fig. 2**: (a) normal profile (the peak of NAA is higher than the peaks of Cho and Cr) and neuronal loss profile (b) and (c). The NAA peak is decreased, and there is increased Cho peak with no change in the Cr.

The metabolite ratios for NAA/Cr, NAA/Cho, and Cho/Cr in patients and control are given in ►**Table 2**. Considering the patients as a single group, there was a significant reduction in the NAA/Cr ratio (mean ± SD, 1.39 ± 0.16) relative to controls (1.70 ± 0.07, $p = 0.0001$). This is in keeping with the marked reduction in NAA in the patients, assuming that the Cr level was stable. The Cho/Cr ratio was significantly increased in patients (0.99 ± 0.06) relative to controls (0.92 ± 0.04, $p < 0.0001$), suggestive of an increase in Cho in patients after TBI. The NAA/Cho ratio was reduced in the patients (1.40 ± 0.24) compared with controls (1.82 ± 0.10, $p < 0.0001$). This is likely to have been caused by both reduction in NAA and increase in Cho level.

The La peak was increased in seven patients they were either expired or in persistent vegetative state or in severe disability. There was no La visible in the spectra of rest of the patients and control patients.

Using ANOVA test, the metabolic ratios of NAA/Cr, NAA/Cho, and Cho/Cr were significantly correlated with changes of GCS ($p \leq 0.00001$, $r = 0.7595$; $p \leq 0.00001$, $r = 0.7506$; $p \leq 0.00001$, $r = -0.5923$, respectively) (►**Fig. 3**) and with clinical outcome of the patients (GOS) ($p \leq 0.00001$, $r = 0.8498$; $p \leq 0.00001$, $r = 0.9323$; $p \leq 0.00001$, $r = -0.9082$, respectively).

The ratio of NAA/Cr (moderate [mean ± SD], 1.57 ± 0.07; severe [mean ± SD] 1.27 ± 0.05; $p < 0.0001$) and NAA/Cho (moderate (mean ± SD), 1.67 ± 0.08; severe (mean ± SD) 1.22 ± 0.09; $p < 0.0001$) is decreased in both patients with moderate and with severe head injury (►**Table 3**), but it is more significantly decreased in severe head injury group and ratio of Cho/Cr (moderate [mean ± SD], 0.93 ± 0.02; severe [mean ± SD] 1.04 ± 0.05; $p < 0.0001$) increased in both moderate and severe head injury group (►**Table 3**), but it is more significantly increased in severe head injury group that was statistically significant.

Discussion

Being rapid, sensitive, and noninvasive, the ^{1}H-MRS technique can identify and quantify the biochemical compounds and explore metabolic and biochemical changes in TBI. Peaks in Cho, Cr, and NAA, La, glucose, myo-inositol (MI), and other substances appear in the proton spectrum. Recently, ^{1}H-MRS

Table 2 Mean (SD) values for NAA/Cr, NAA/Cho, and Cho /Cr for the patients and control are shown

	NAA/Cr	NAA/Cho	Cho/Cr
Patients group ($n = 51$)	1.39 (0.16)[a]	1.40 (0.24)[a]	0.99 (0.06)[a]
Control group ($n = 24$)	1.70 (0.07)	1.82 (0.10)	0.92 (0.04)

Abbreviations: Cho, choline; Cr, creatine; NAA, N-acetylaspartate; SD, standard deviation.

[a]$p < 0.0001$.

Table 3 Mean (SD) values for NAA/Cr, NAA/Cho, and Cho /Cr ratios are shown in moderate and severe head injury group

	NAA/Cr	NAA/Cho	Cho/Cr
Moderate group ($n = 21$)	1.57 (0.07)[a]	1.67 (0.08)[a]	0.93 (0.02)[a]
Severe group ($n = 30$)	1.27 (0.05)[a]	1.22 (0.09)[a]	1.04 (0.05)[a]

Abbreviations: Cho, choline; Cr, creatine; NAA, N-acetylaspartate; SD, standard deviation.

[a]$p < 0.0001$.

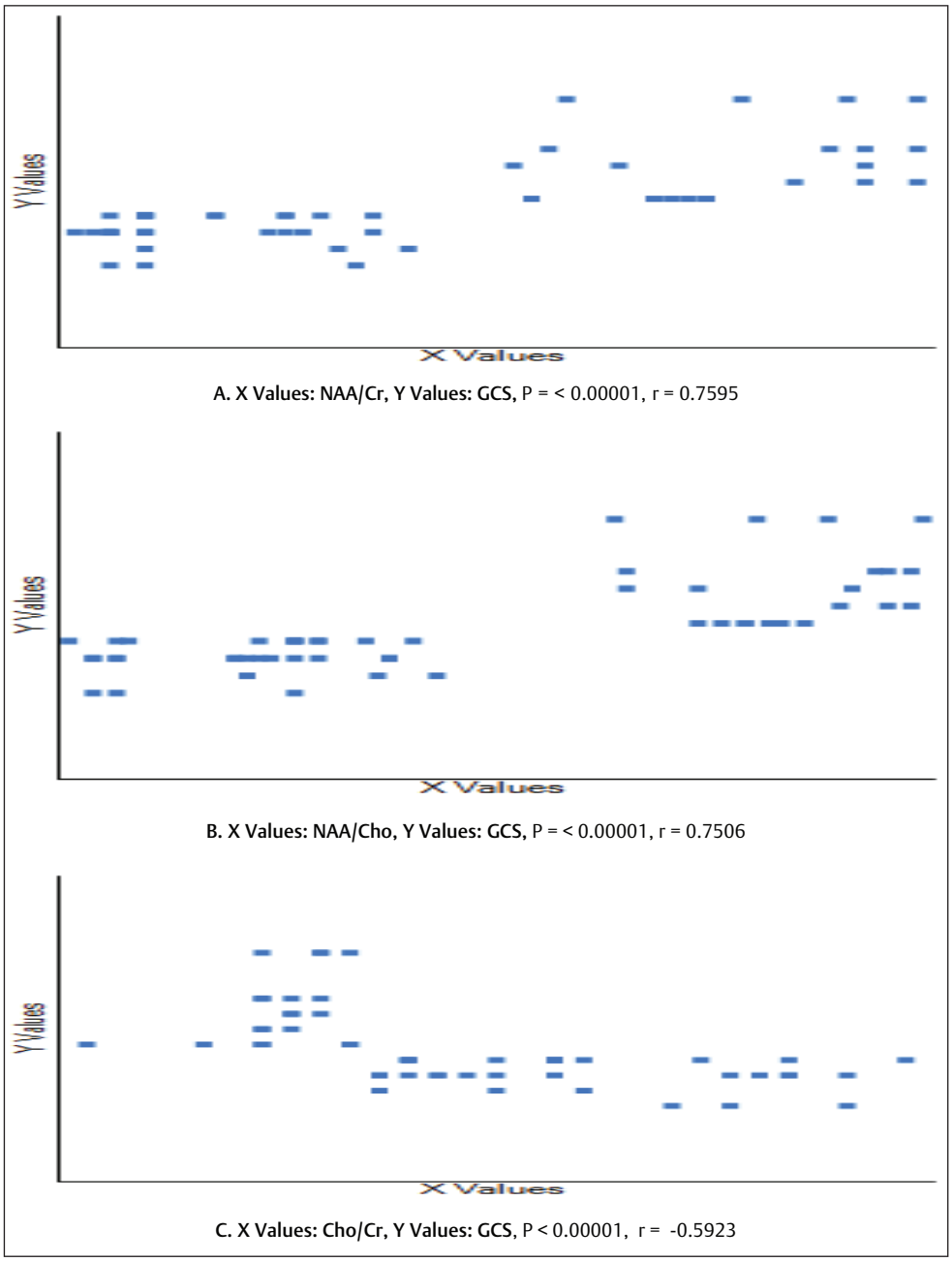

Fig. 3 Data from 51 patients show that (**A**) there was a significant correlation between the ratios of *N*-acetylaspartate/creatine (NAA/Cr) and Glasgow coma scale (GCS) score ($p = < 0.00001$, $r = 0.7595$); (**B**) between *N*-acetylaspartate/choline (NAA/Cho) and the GCS score ($p = < 0.00001$, $r = 0.7506$); and (**C**) between Cho/Cr and GCS score ($p < 0.00001$, $r = -0.5923$), respectively.

has been applied in several studies of TBI (Choe et al, 1995; Ricci et al, 1997; Cecil et al, 1998; Friedman et al, 1998, 1999; Ross et al, 1998) and has demonstrated abnormalities in regions of normal-appearing brain.

A reduction in NAA has been found previously in patients after TBI in both the gray matter (Ricci et al, 1997; Ross et al, 1998; Friedman et al, 1999) and the white matter (Choe et al, 1995; Cecil et al, 1998; Friedman et al, 1998; Ross et al, 1998), but these papers made no reference to the severity of injury. Furthermore, the previous studies have generally performed the MRS investigations in the later stages (6 weeks to several years) after TBI. An acute (within 1 day) reduction in NAA has only been described in patients with severe head injury in areas of visible contusion (Condon et al, 1998), and there was an early (within 1 month) reduction in NAA in a cohort of pediatric and adult patients (Ross et al, 1998). The present study performed the MRS investigation in the early stages after TBI, that is, within 7 days after injury. Our results are in keeping with these previous studies in finding a reduction in the NAA levels in the early stages after TBI, but in addition, there was a significant correlation of reduced NAA and increase in Cho level with the severity of head injury.

The anatomical area investigated for this study was normal-appearing white matter in one (or both) of the frontal lobes. DAI in the lobar white matter tends to be primarily microscopic and hence undetectable using conventional neuroimaging techniques (Mittl et al, 1994). The reduction in

NAA (reduced NAA/Cr ratio) in these areas was statistically significant in head injury patients assessed using the GCS and GOS (►**Fig. 3**). This is in keeping with substantial cellular injury in patients after TBI. The apparent reduction in NAA in these areas of normal-appearing white matter supports the hypothesis that regions of the brain remote from the focal lesion are involved in the pathology of head injury. This could result from shearing damage at the time of the impact, subsequent ischemia, or wallerian degeneration of the axons that connect to the areas of focal damage. Wallerian degeneration has been regarded as the cause of the reduction in NAA level that has been found in white matter tracts at a distance from areas of pathology in stroke (Pendlebury et al, 1999) and multiple sclerosis (De Stefano et al, 1998).

NAA is known to be a mitochondrial product (Bates et al, 1996). Consequently, a reduction in NAA could be caused by impaired mitochondrial energy production rather than cellular loss (Bates et al, 1996). It has been postulated that a temporary decrease in NAA levels after brain injury may be caused by accelerated lipid synthesis involved in myelin repair or may be attributable to NAA providing a temporary source of cellular energy locally at the site of axonal injury, which would produce a transient decrease that might precede any loss of NAA as a result of axonal death.[6]

In addition to evidence of decreased NAA and increased Cho, we found a diffusely high signal of La resonance intensity in the 7 patients, out of 51 patients with acute TBI. As the intracerebral La concentration is below the detection limit for the ^{1}H-MRS method used in this study, the presence of the trace of La in the brains must be an expression of pathologic conditions occurring in the brain parenchyma[10] and cerebrospinal fluid.[11] These include impairment of the oxidative metabolism or infiltration of macrophages and inflammatory cells, all conditions that occur after a TBI.[7]

Although increases in brain parenchymal La level are often found in children,[12] most of the spectroscopic studies on adults with injured brains either do not mention it or report focal La increases when the mechanism of injury is clearly ischemic.[12] The scarce evidence for increases in brain La level in previous spectroscopic studies of adult TBI may be a function of the time span between the initial insult and the imaging examination. In previous studies, the imaging examination was usually performed several days or even months after the trauma, when La increases might have disappeared. In agreement with this, experimental work has shown that cerebral La increases progressively soon after the brain injury and decreases after 1 per week.[13] It should be mentioned, however, that in the few reported cases with ^{1}H MRS[14] performed early after the trauma, increases in brain La were found in only two patients,[14] and that this was found only in 7 patients of our 51 patient population. Despite these limitations, this study results suggest that ^{1}H-MRS can provide, in some circumstances, an accurate estimation of the impairment of the energy metabolism occurring in the brains of patients with TBI, and give support to the importance of controlling and possibly reducing the time between the injury and the spectroscopic examination.

La due to impaired aerobic glycolysis is a specific marker for posttraumatic secondary hypoxic and ischemic injury. Several studies have shown that an increase in La in the normal-appearing white matter area of patients with severe TBI could suggest poor prognosis (death or severe disability) after posttraumatic secondary ischemic and hypoxic injury.[15] La presence has been related to multiple factors, including excessive release of glutamate, disordered mitochondrial and oxidative metabolism, and systemic responses to trauma.[16] In our study, La peaks appeared in seven patients with severe TBI (GCS 5–8). In these patients, one patient was dead (GOS = 1), two patients were in persistent vegetative state (GOS = 2), and four patients were in severe disability (GOS = 3). It has been demonstrated that increase in La concentrations indicates poor prognosis in patients with severe TBI.

In the current study, the apparent level of Cho (Cho/Cr ratio) in normal-appearing white matter was increased in patients (►**Table 2**) compared with control. The ratios of Cho/Cr were statistically correlated with GOS of the patients, and there was a moderately negative correlation between Cho/Cr ratio and GCS of the patients (►**Fig. 3**), which shows as the ratio of increase in Cho/Cr and decrease in GCS of the patients (severity of head injury increase). Previous studies have demonstrated an increase in Cho in the gray matter of patients in the chronic stages after TBI (Ricci et al, 1997; Friedman et al, 1998). The Cho peak consists of several compounds, the principle ones being phosphocholine and glycerophosphocholine, together with some free Cho (Miller et al, 1996). The increase in Cho could be due to membrane degradation following cellular damage. However, significant cell disruption would be expected to be associated with abnormalities particularly on T_2-weighted images, or with evidence of blood-brain barrier breakdown, neither of which was evident in the areas from which the ^{1}H-MRS were acquired. Alternatively, the elevation in Cho could be consistent with an increase in membrane turnover similar to that found in tumors (McBride et al, 1995). An increase in membrane turnover may indicate the potential repair of cells within this area.

The Cho levels could also be affected by reduction in the activity of Cho acetyltransferase, which has been reported in patients after head injury (Murdoch et al, 1998). High-resolution ^{1}H-MRS of extracts of brain demonstrates that the Cho peak consists of free Cho, phosphocholine, and glycerophosphocholine (Miller et al, 1996). It is therefore possible that the observed difference between the ratios of patients and controls could be explained by changes in the activity of Cho acetyltransferase.

There were a significant increase in the Cho/Cr ratio (►**Table 2**) and a significant reduction in the NAA/Cr (►**Table 2**) and NAA/Cho (►**Table 2**) ratio compared with the controls, confirming our hypothesis that white matter that appears normal on conventional imaging may show metabolic abnormalities when ^{1}H-MRS is used. The relationship between the biochemical changes and the severity of injury provides a metabolic basis for the neurologic disability seen in these patients. As discussed earlier, the mechanisms responsible for these abnormalities are currently unclear, but they may provide a target for therapeutic intervention in the future.

The results used in this study were expressed on the assumption that Cr was present at a constant level. Cr is present in slightly higher concentrations in gray matter than in white matter (Pouwels and Frahm, 1998) and is relatively refractive to change. An alteration in Cr levels could not account for the decrease in the NAA/Cr ratio together with the increase in the Cho/Cr ratio. Alternatively, the apparent differences in the ratios in the patients and controls could be explained by a change in the relaxation properties of the metabolites in the patients. Trauma disrupts the normal cellular environment and thus could theoretically alter the T_1 and T_2 relaxation properties of the metabolites. This, in turn, would alter the apparent metabolite concentrations and hence the ratios. Assuming a T_1 value in the cerebral white matter of 1,500 milliseconds (Frahm et al, 1989*b*), it would not be possible to explain an increase in Cho by alteration of the T_1 or T_2 values. Furthermore, it would require a 55% increase in T_1 or a 55% decrease in T_2 to explain the reduction in NAA observed in this study, which is unlikely.

At early points (within 7 days, mean = 4.5 days), we found that NAA/Cr and NAA/Cho ratios were significantly reduced ($p < 0.0001$) and Cho/Cr ratio was significantly increased ($p < 0.0001$). We conclude that there is a sustained alteration in NAA, Cho, and Cr. Similar findings have been reported by other MRS studies in TBI, which focused on metabolite ratios rather than absolute concentration levels. Govindaraju and colleagues[17] assessed ratio scores (NAA/Cr and NAA/Cho) from 25 single predominantly gray- or white-matter voxels, and reported that both ratios were reduced in TBI patients for two of the white-matter voxels. Similarly, Vagnozzi and associates[18] recently reported reduced NAA/Cr in TBI patients in anterior periventricular white matter. Significant differences were observed between severe TBI patients and matched controls for several ^{1}H-MRS metabolites. Ricci et al (1997) examined 14 vegetative brain-injured patients with proton magnetic resonance single-volume spectroscopy performed 1 to 90 months after the injury. Cho/Cr was significantly higher, whereas NAA/Cho and NAA/Cr were markedly lower than in those controls, indicating that sensitivity of ^{1}H-MRS to metabolite alterations might serve as biomarkers of severe TBI.

We have found that not only did the ratios of NAA/Cr, NAA/Cho, and Cho/Cr significantly correlate with changes in GCS, but they also correlated with the clinical outcome (GOS). Shutter et al[19] performing a prospective longitudinal MRS study in adult TBI patients noted that NAA/Cr ratio from the corpus callosum was most useful for long-term outcome prediction, whereas Nakabayashi et al[20] and Signoretti et al[21] showed the recovery of NAA levels in patients with favorable outcomes. NAA levels seem to discriminate patients who recovered from coma from those who died or remained in a persistent vegetative state,[22] indicating that ^{1}H-MRS could reflect the degree of nerve tissue injury objectively and is a reliable and innovative means to evaluate the trauma levels and prognosis of TBI patients. These findings provide evidence for cellular injury not visible by conventional techniques. This may be relevant to understanding the extent of disability following TBI.

Paper Presented

The study was presented at the fifth Asian Australasian Society of Neurological Surgeons Neurotrauma Committee and second WFNS-Military Neurosurgeons Meeting on October 9–11, 2015, Jaipur, India.

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Conflict of Interest

None.

References

1 Jennett B, Bond M. Assessment of outcome after severe brain damage. Lancet 1975;1(7905):480–484
2 Broman SH, Michel ME. Traumatic Head Injury in Children. New York, NY: Oxford University Press; 1995
3 Levin HS, Aldrich EF, Saydjari C, et al. Severe head injury in children: experience of the traumatic coma data bank. Neurosurgery 1992;31(3):435–443, discussion 443–444
4 van der Naalt J, Hew JM, van Zomeren AH, Sluiter WJ, Minderhoud JM. Computed tomography and magnetic resonance imaging in mild to moderate head injury: early and late imaging related to outcome. Ann Neurol 1999;46(1):70–78
5 Lin AP, Liao HJ, Merugumala SK, Prabhu SP, Meehan WP III, Ross BD. Metabolic imaging of mild traumatic brain injury. Brain Imaging Behav 2012;6(2):208–223
6 Cecil KM, Hills EC, Sandel ME, et al. Proton magnetic resonance spectroscopy for detection of axonal injury in the splenium of the corpus callosum of brain-injured patients. J Neurosurg 1998;88(5):795–801
7 Brooks WM, Friedman SD, Gasparovic C. Magnetic resonance spectroscopy in traumatic brain injury. J Head Trauma Rehabil 2001;16(2):149–164
8 Signoretti S, Marmarou A, Fatouros P, et al. Application of chemical shift imaging for measurement of NAA in head injured patients. Acta Neurochir Suppl (Wien) 2002;81:373–375
9 Stein SC, Narayan RK, Wilberger JE, Povlishock JT. In: Classification of head injury. Neurotrauma. New York, NY: McGraw-Hill; 1996:31–41
10 Prichard JW. What the clinician can learn from MRS lactate measurements. NMR Biomed 1991;4(2):99–102
11 DeSalles AA, Kontos HA, Ward JD, Marmarou A, Becker DP. Brain tissue pH in severely head-injured patients: a report of three cases. Neurosurgery 1987;20(2):297–301
12 Holshouser BA, Ashwal S, Luh GY, et al. Proton MR spectroscopy after acute central nervous system injury: outcome prediction in neonates, infants, and children. Radiology 1997;202(2):487–496
13 Schuhmann MU, Stiller D, Skardelly M, et al. Metabolic changes in the vicinity of brain contusions: a proton magnetic resonance spectroscopy and histology study. J Neurotrauma 2003;20(8):725–743
14 Condon B, Oluoch-Olunya D, Hadley D, Teasdale G, Wagstaff A. Early 1H magnetic resonance spectroscopy of acute head injury: four cases. J Neurotrauma 1998;15(8):563–571
15 Holshouser BA, Tong KA, Ashwal S. Proton MR spectroscopic imaging depicts diffuse axonal injury in children with traumatic brain injury. AJNR Am J Neuroradiol 2005;26(5):1276–1285

16 Krishnappa IK, Contant CF, Robertson CS. Regional changes in cerebral extracellular glucose and lactate concentrations following severe cortical impact injury and secondary ischemia in rats. J Neurotrauma 1999;16(3):213–224
17 Govindaraju V, Gauger GE, Manley GT, Ebel A, Meeker M, Maudsley AA. Volumetric proton spectroscopic imaging of mild traumatic brain injury. AJNR Am J Neuroradiol 2004;25(5):730–737
18 Vagnozzi R, Signoretti S, Tavazzi B, et al. Temporal window of metabolic brain vulnerability to concussion: a pilot 1H-magnetic resonance spectroscopic study in concussed athletes—part III. Neurosurgery 2008;62(6):1286–1295, discussion 1295–1296
19 Shutter L, Tong KA, Lee A, Holshouser BA. Prognostic role of proton magnetic resonance spectroscopy in acute traumatic brain injury. J Head Trauma Rehabil 2006;21(4):334–349
20 Nakabayashi M, Suzaki S, Tomita H. Neural injury and recovery near cortical contusions: a clinical magnetic resonance spectroscopy study. J Neurosurg 2007;106(3):370–377
21 Signoretti S, Marmarou A, Aygok GA, Fatouros PP, Portella G, Bullock RM. Assessment of mitochondrial impairment in traumatic brain injury using high-resolution proton magnetic resonance spectroscopy. J Neurosurg 2008;108(1):42–52
22 Ricci R, Barbarella G, Musi P, Boldrini P, Trevisan C, Basaglia N. Localised proton MR spectroscopy of brain metabolism changes in vegetative patients. Neuroradiology 1997;39(5):313–319