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Cerebrospinal Fluid Taurine after Traumatic Brain Injury

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In the experimental setting, taurine is known to be released from swollen cells to reestablish their normal volume. However, its clinical relevance has not been fully understood. This study was undertaken to reveal changes in cerebrospinal fluid (CSF) amino acids concentration in patients with severe traumatic brain injury (TBI). The study included eight patients, in whom a ventricular catheter was inserted to measure intracranial pressure and obtain CSF samples for 5 days. CSF obtained from patients with normal pressure hydrocephalus served as a control. CSF taurine concentration increased 1.8 times control ($P < 0.05$) after TBI and returned to control value approximately 67 h after injury. Taurine decreased further and remained lower than control thereafter. Phosphoethanolamine showed similar increase, whereas glutamine decreased transiently and arginine remained close to control value. The present data support the period of astrocytic swelling observed after TBI in other morphological studies. The mechanism and consequences of CSF taurine decrease in the subacute stage of TBI need to be elucidated.

KEY WORDS: Cytotoxic edema; regulatory volume decrease; taurine; traumatic brain injury.

INTRODUCTION

Cytotoxic edema is known to occur soon after traumatic brain injury (TBI) mainly in astrocytes (1,2) due to increased extracellular K^+ concentration (3,4) and lactic acidosis (5). It may contribute to secondary brain damage causing ischemia or excitotoxicity (1–3). In the experimental setting, it has been demonstrated that osmolytes including amino acids such as taurine or phosphoethanolamine are released from neural cells in response to swelling to reestablish their normal volume, and this mechanism is termed as regulatory volume decrease (4,6–9). However, in the clinical setting like TBI, specific release of such amino acids in the injured brain has not been demonstrated clearly. The aim of the present study is to measure intraventricular CSF amino acids concentration in patients with severe TBI.

EXPERIMENTAL PROCEDURE

Patients with a severe TBI who were admitted to the Nagoya Daini Red Cross Hospital between October 1998 and October 2000 were considered for this study, which was approved by the Hospital Ethical Committee. Patients were excluded from this study if a family member refused to consent to it. Selected patients were those with age between 16 and 80 years and a Glasgow Coma Scale score of 8 or less on admission. Ten patients were found to be eligible for this study, but two of them who died within 48 h after admission were excluded from data analysis. All patients were managed according to standard protocol and received aggressive treatment including rapid removal of large intracranial hematomas, intracranial pressure (ICP) monitoring with use of an intraventricular sensor (110-4HMT, Camiono NeuroCare, Inc., San Diego, CA), and careful management of ICP by means of moderate hyperventilaton ($PaCO_2 \approx 35$),

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mannitol, ventricular drainage, and barbiturate infusion if necessary. Brain temperature was measured continuously and kept close to 37°C. One patient who had uncontrolled high ICP required moderate hypothermia. Patients received parenteral neutrition containing 5 mg of vitamin B6 and at least 780 mg of methionine and 200 mg of cysteine daily starting within 60 h after operation. Taurine was not given in any patient. CSF samples were obtained through a ventricular catheter during operation and 12, 36, 60, 84, and 108 h after operation. These samples were centrifuged, and the supernatant was stored frozen at −40°C. Control ventricular CSF samples were obtained from patients with normal pressure hydrocephalus after subarachnoid hemorrhage at the time of ventriculoperitoneal shunt ($n = 9$). Patients who had brain infarction greater than 2 cm in diameter on computed tomograms were excluded from the control group. CSF albumin concentration was determined by the BCG method, and CSF lactate dehydrogenase (LDH) by the Wroblewski–LaDue method.

Measurement of Amino Acids. Amino acids were analyzed by use of precolumn *o*-phthaldialdehyde derivatization, separation by reverse-phase high-performance liquid chromatography and fluorometric detection, essentially as previously described (10).

Statistical Analysis. All data in the text are presented as mean ± SD. Comparisons of CSF amino acids concentration between the control and the sample obtained from patients with TBI were made using the Mann–Whitney *U* test. To determine a correlation between each of CSF taurine and phosphoethanolamine concentration and CSF albumin and LDH concentration, generalized multivariate linear regression model was fit. A $P < 0.05$ was considered significant.

RESULTS

The mean age of patients was 39 ± 22 years old. The mean score of the Glasgow Coma Scale on admission was 6 ± 2. The mean time interval from the onset of TBI to operation was 6.6 ± 3.7 h. Patient data are summarized in Table I. No patient had bacterial meningitis.

Amino Acids Changes

As there were some patterns in the time course of amino acid concentration, representative amino acids (i.e., glutamine and arginine) were presented in addition to taurine in Fig. 1. Phosphoethanolamine, another osmolyte released in response to cell swelling, and methionine, one of substrates of taurine, were also presented. CSF taurine and phosphoethanolamine concentration increased significantly soon after injury, and decreased close to control levels (8.0 ± 1.7 and 6.3 ± 2.0 nmol/ml, respectively)(Fig. 1a and b) 60 h after operation ($n = 8$). CSF taurine concentration decreased further and remained significantly lower than the control level thereafter. CSF glutamine decreased significantly soon after injury and gradually returned to the control level (618.9 ± 120.6 nmol/ml) (Fig. 1c). CSF arginine remained close to the control level (25.2 ± 3.4 nmol/ml) throughout the study period (Fig. 1d). CSF mechionine showed a trend to decrease transiently soon after injury (Fig. 1e). CSF glutamate increased to 6.7 ± 3.3 nmol/ml soon after injury and decreased to less than 4 nmol/ml by 60 h after operation.

Changes in taurine and phosphoethanolamine concentration expressed as individual results are shown in Fig. 2. In most of cases taurine and phosphoethanolamine showed similar time course. There was a trend that patients having increasing taurine concentraion after ventricular catheter installation (intra-op) and therefore having higher levels of taurine at 36 h had unfavorable clinical outcomes.

CSF Albumin and LDH Changes

Control CSF albumin concentration was 0 g/dl, and control CSF LDH concentration was 4.1 ± 4.0 IU/l. CSF albumin and LDH concentration increased after TBI (Fig. 3). Using a generalized multivariate linear regression model, CSF taurine was found to have some correlation with CSF albumin ($P = 0.007$, adjusted $R^2 = 0.16$), whereas it did

Table I. Patient Data

Case	Age (year)	Sex	Initial GCS	Radiological diagnosis	Craniotomy	GOS at 3 months
1	16	M	7	SDH, contusion	Yes	MD
2	22	M	6	Traumatic ICH	Yes	SD
3	34	M	8	SDH, traumatic SAH, contusion	Yes	SD
4	26	M	8	Traumatic SAH, contusion	Yes	GR
5	21	M	3	DAI	No	GR
6	57	M	6	Contusion, diffuse brain swelling	No	VS
7	77	M	8	SDH, contusion	Yes	SD
8	58	M	5	SDH	Yes	D

ICH, intracerebral hematoma; DAI, diffuse axonal injury; SAH, subarachnoid hemorrhage; SDH, subdural hematoma; GCS, Glasgow Coma Scale score; GOS, Glasgow Outcome Score; GR, good recovery; MD, moderate disability; SD, severe disability; VS, vegetative state; D, death.

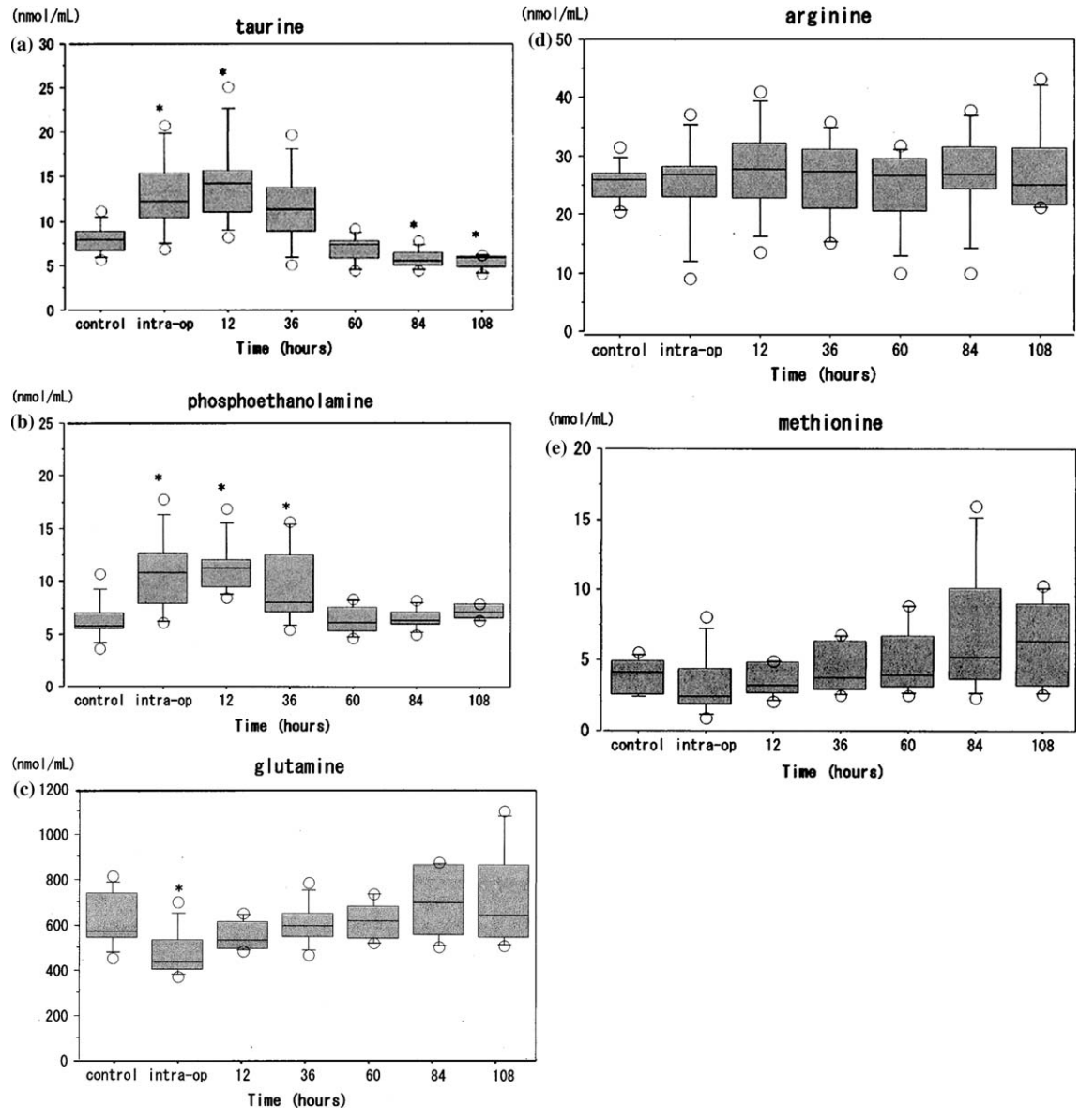

Fig. 1. Box–whisker plots showing the time course for ventricular cerebrospinal fluid tautine (a), phosphoethanolamine (b), glutamine (c), arginine (d) and methionine (e) concentration after severe traumatic brain injury in humans ($n = 8$). *$P < 0.05$ vs. control by the Mann–Whitney U test. Control samples were obtained from patients with normal pressure hydrocephalus ($n = 9$). Numbers on the time axis indicate h after ventricular catheter installation.

not have significant correlation with CSF LDH ($P = 0.21$). Neither CSF albumin nor CSF LDH showed significant correlation with CSF phosphoethanolamine ($P = 0.64$ and 0.81, respectively).

DISCUSSION

In essentially all animal cells including astrocytes and neurons, the mechanism to adjust cell volume in response to osmolarity change has been preserved to prevent disruption of the cytoarchitecture and to keep normal cell functions (7). After swelling cultured primary astrocytes reestablish their normal volume through a process termed regulatory volume decrease by extrusion of intracellular ions such as K^+, Cl^-, and HCO_3^- and amino acids via volume-sensitive organic osmolyte/anion channel (2,3,11). Amino acids released from astrocytes and neurons in response to cell swelling include taurine,

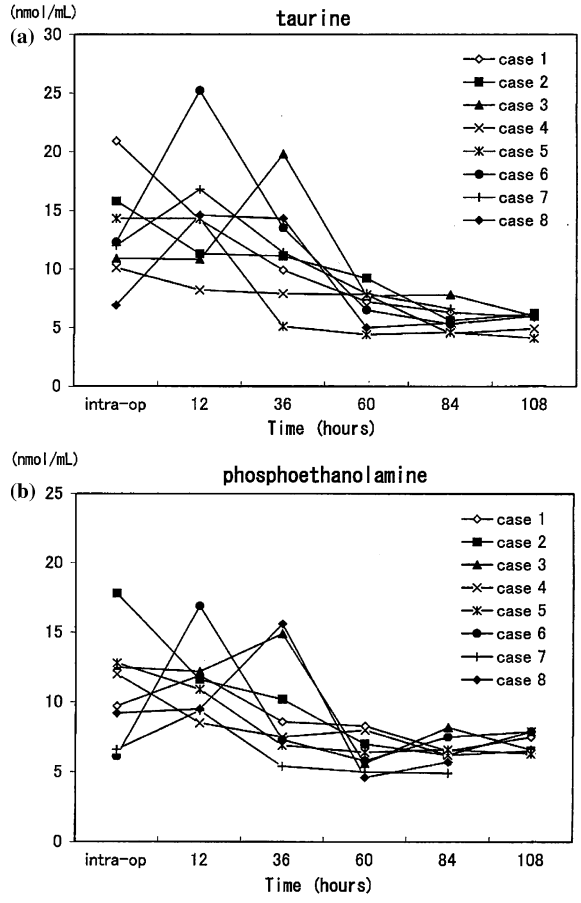

Fig. 2. Changes in cerebrospinal fluid taurine (a) and phosphoethanolamine (b) concentration expressed as individual results. Case numbers correspond to those in Table I. Two results are missing at 108 h, one due to patient death and another sampling error.

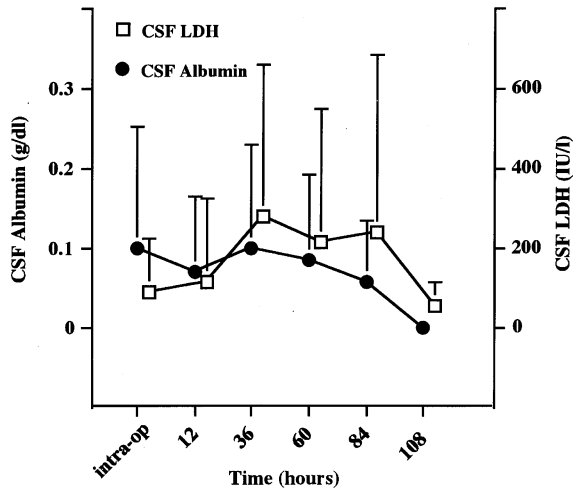

Fig. 3. Changes in cerebrospinal fluid albumin and LDH concentration after severe traumatic brain injury in humans ($n = 8$). All data are presented as mean ± SD. Numbers on the time axis indicate h after ventricular catheter installation.

phosphoethanolamine, alanine, glycine, glutamate and aspartate (2–4,6,9), and taurine has the lowest efflux threshold suggesting its major role in cell volume regulation (4,6,9). In the cytosol, taurine is found either free or in some simple peptides suggesting its role as an osmolyte (7,12,13). High extracellular K^+ concentration, the condition observed in concussive brain injury (14), has also been shown to produce cell swelling *in vitro* (2–4), and this K^+-induced astrocytic swelling is also associated with progressive taurine release (8). In TBI, such cell swelling is known as cytotoxic edema. In the present study, CSF taurine and phosphoethanolamine concentration increased significantly soon after the onset of TBI and remained elevated for approximately 67 h. This time course is in accord with the duration of cytotoxic edema observed morphologically in human patients with TBI (1,15). Bullock et al. (1) demonstrated that massive astrocytic swelling lasted from 3 h to 3 days after TBI. The present data suggest that osmolytes such as taurine and phosphoethanolamine are released from neural cells following the development of cytotoxic edema in TBI. Interestingly, Stover et al. (16) have reported that ventricular CSF glutamate and taurine concentration remain elevated up to day 14 after severe TBI, which is inconsistent with our data. The reason for these conflicting data remains unclear.

In an animal experiment, it has been shown that transport in the extracellular space of the brain is primarily by diffusion in the gray matter and by both diffusion and bulk flow in the white matter, and that about 30% of CSF production comes from bulk flow of brain extracellular fluid (17). In a cat model of global brain ischemia, Shimada et al. showed that amino acids diffused slowly from the gray matter to the adjacent white matter and into CSF (18). For example, glutamate increased 25, 4, and 2.4 times control 2 h after ischemia induction in the cortex, in the internal capsule, and in CSF, respectively. These findings suggest that amino acids concentration in ventricular CSF reflects their extracellular concentration to some extent. In the present study, taurine and phosphoethanolamine did not show significant correlation with CSF LDH. Taurine showed a weak correlation with CSF albumin, but phosphoethanolamine did not. These findings suggest that none of the following conditions, cell

lysis, blood brain barrier breakdown or blood contamination into CSF is major contributor to increase of these amino acids in CSF. Actually in two cases, CSF taurine increased whereas CSF albumin was 0 g/dl.

In the present study, elevated CSF taurine concentration decreased to the control level 67 h after TBI, and remained lower than control through the rest of the study period. Taurine is synthesized from methionine and cysteine primarily in the liver and the brain. In this metabolism pyridoxal phosphate (vitamin B6) is required by the essential enzyme cysteinesulfinic acid decarboxylase (12,13). Since adult human is known to have low synthetic capacity for taurine (19), an inadequate biosynthesis may be responsible for CSF taurine decrease even though patients received parenteral nutrition containing methionine, cystein and vitamin B6. Plasma taurine concentration is reported to decrease after surgical injury, sepsis and in critically injured patients probably due to increased urinary excretion or increased uptake by neutrophils (13,20). Such plasma taurine decrease may also be responsible for CSF taurine decrease after TBI. On the other hand, there is a possibility that CSF taurine decrease in the subacute stage is not real provided that the control taurine level obtained from patients with normal pressure hydrocephalus is higher than that from healthy individuals. However, to the best of our knowledge, CSF taurine increase in normal pressure hydrocephalus has not been reported. CSF taurine concentration obtained from subjects free of central nervous system pathology varies from 3.9 to 12 μmol/l (16,21,22). Control taurine and phosphoethanolamine levels in the present study are quite similar to those of normal individuals reported by McGale et al. (21) (7.6 ± 2.1 and 5.4 ± 2.3 μmol/l, respectively). Evidence supporting cytoprotective and immunomodulatory actions of taurine is accumulating (13,23–25). It needs to be elucidated if taurine supplementation to patients with severe TBI is beneficial.

CONCLUSION

Intraventricular CSF concentration of taurine, a major osmolyte in cell volume regulation, increased from very early to 67 h after TBI supporting the period of morphologically observed astrocytic swelling (1,15).

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