Encephalopathy in acute liver failure resulting from acetaminophen intoxication: New observations with potential therapy*

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Objective: **Hyperammonemia is a major contributing factor to the encephalopathy associated with liver disease. It is now generally accepted that hyperammonemia leads to toxic levels of glutamine in astrocytes. However, the mechanism by which excessive glutamine is toxic to astrocytes is controversial. Nevertheless, there is strong evidence that glutamine-induced osmotic swelling, especially in acute liver failure, is a contributing factor: the osmotic gliopathy theory. The object of the current communication is to present evidence for the osmotic gliopathy theory in a hyperammonemic patient who overdosed on acetaminophen.**

Design: **Case report.**

Setting: **Johns Hopkins Hospital.**

Patient: **A 22-yr-old woman who, 36 hrs before admission, ingested 15 g acetaminophen was admitted to the Johns Hopkins Hospital. She was treated with *N*-acetylcysteine. Physical examination was unremarkable; her mental status was within normal limits and remained so until approximately 72 hrs after ingestion when she became confused, irritable, and agitated.**

Interventions: **She was intubated, ventilated, and placed on lactulose. Shortly thereafter, she was noncommunicative, unresponsive to painful stimuli, and exhibited decerebrate posturing. A clinical diagnosis of cerebral edema and increased intracranial pressure was made. She improved very slowly until 180 hrs after ingestion when she moved all extremities. She woke up shortly thereafter.**

Measurements and Main Results: **Despite the fact that hyperammonemia is a major contributing factor to the encephalopathy observed in acute liver failure, the patient's plasma ammonia peaked when she exhibited no obvious neurologic deficit. Thereafter, her plasma ammonia decreased precipitously in parallel with a worsening neurologic status. She was deeply encephalopathic during a period when her liver function and plasma ammonia had normalized. Plasma glutamine levels in this patient were high but began to normalize several hours after plasma ammonia had returned to normal. The patient only started to recover as her plasma glutamine began to return to normal.**

Conclusions: **We suggest that the biochemical data are consistent with the osmotic gliopathy theory—high plasma ammonia leads to high plasma glutamine—an indicator of excess glutamine in astrocytes (the site of brain glutamine synthesis). This excess glutamine leads to osmotic stress in these cells. The lag in recovery of brain function presumably reflects time taken for the astrocyte glutamine concentration to return to normal. We hypothesize that an inhibitor of brain glutamine synthesis may be an effective treatment modality for acute liver failure. (Crit Care Med 2011; 39:2550–2553)**

KEY WORDS: acetaminophen poisoning; acute liver failure; astrocytes; encephalopathy; hyperammonemia; intracranial pressure; plasma ammonia; plasma glutamine; osmotic stress

Acute liver failure (ALF) is an uncommon but nevertheless serious disorder. In 2005, ALF accounted for 6% of liver-related deaths and 6% of orthotopic liver transplants in the United States (1). Viral hepatitis was previously reported to be the leading cause of ALF in the United States, but acetaminophen overdose and idiosyncratic drug reactions are now a more common cause (1). For example, in 2004, data from the US Acute Liver Failure Study Group registry of >700 patients with ALF implicated acetaminophen poisoning in nearly 50% of all cases of ALF in the United States (2).

Hyperammonemia and the attendant increase in brain glutamine are major factors in the encephalopathy associated with ALF (3). In addition to ALF, it is apparent that ammonia-induced encephalopathy is a contributing factor to an increasingly recognized spectrum of disorders, including the more common, chronic form of liver disease (4). For example, hyperammonemia-induced encephalopathy is a prominent feature of inborn errors of the urea cycle (5), idiopathic hyperammonemic syndrome after immunosuppression or cytotoxic therapy (6, 7), ureolysis in stagnant urine (8, 9), valproate therapy (10, 11), and essential amino acid total parenteral nutrition (12). Ammonia enters the brain mostly by nonsaturable diffusion of the free base (NH_3) (4, 13), where it is metabolized almost exclusively to glutamine in a reaction catalyzed by glutamine synthetase (14, 15). In the brain, glutamine synthetase is localized largely, if not exclusively, in astrocytes (16, 17), and astrocytes are compromised in liver disease/hyperammonemia (18–23). It has been proposed that hyperammonemic encephalopathy in any clinical setting (24) is attributable, at least in part, to increased intracranial pressure, resulting from glutamine-induced osmotic stress in astrocytes—the osmotic gliopathy theory (25). Several magnetic resonance imaging and spectroscopic studies are fully in accord with

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this hypothesis (26). Indeed, based on recent studies, the US Acute Liver Failure Study Group attributes the encephalopathy of hyperammonemia in ALF largely to the synthesis in astrocytes of osmotically active glutamine, resulting in astrocyte swelling, which in turn contributes to the cerebral edema and encephalopathy (27).

The increasing awareness of the importance of glutamine in ALF prompted us to describe sequential plasma ammonia and glutamine levels in relation to neurologic status in a patient with severe acetaminophen toxicity (data not previously reported). This case allows us to speculate on a possible new treatment for ALF.

CASE REPORT

This was the first Johns Hopkins Hospital admission of a 22-yr-old woman who, 36 hrs before admission, ingested 15 g acetaminophen. Shortly after admission, her plasma acetaminophen level was 179 mg/dL, for which she was treated with *N*-acetylcysteine. Apart from mild nausea she was asymptomatic. Her plasma aspartate aminotransferase, alanine aminotransferase, and lactic dehydrogenase values (IU/L) were, respectively, 7090, 6710, 3110, and her total and direct bilirubins (mg/dL) were 12.9 and 6.3, respectively. Blood pH and pCO_2 were 7.50 and 34 torr, respectively. The serum phosphate level was 1.8 mg/dL (normal value, approximately 2.5 mg/dL). Although it is well known that hyperventilation causes hypophosphatemia (28), that explanation has not routinely been applied to the hyperventilation regularly seen early in hyperammonemia. Her physical examination was unremarkable; her mental status was within normal limits and remained so until approximately 72 hrs after ingestion when she became confused, irritable, and agitated. She was intubated, ventilated, and placed on lactulose. Shortly thereafter, she was noncommunicative, unresponsive to painful stimuli, and exhibited decerebrate posturing. Her intracranial pressure was 17–25 mm Hg as measured through an epidural monitor (reference values 7–15 mm Hg). A clinical diagnosis of cerebral edema and increased intracranial pressure was made and she advanced to Stage IVA on the liver transplant list; 80 hrs after the ingestion of acetaminophen, her coma score was 6. She was unchanged for approximately 24 hrs when it was noted that she was responsive to painful stimuli. She improved very slowly until 180 hrs after ingestion when she moved all extremities; she woke up shortly thereafter.

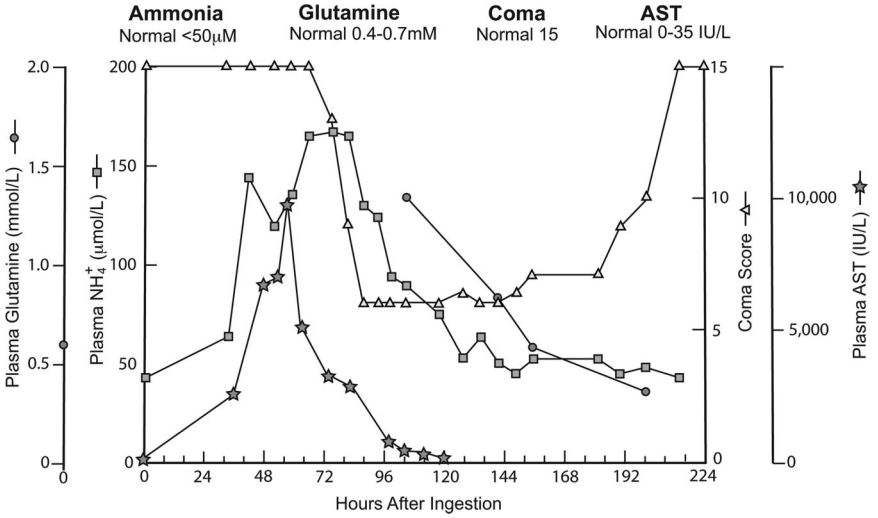

Figure 1. Relationship among coma score (*triangles*), liver function (as assessed by plasma aspartate transaminase [aminotransferase] activity [*AST*]; *stars*), and plasma biochemistries ($NH4^+$, *squares*; glutamine, *circles*). The zero time (preingestion) values of plasma ammonia, glutamine, AST, and coma score were assigned normal reference values. Ammonia was measured in the hospital laboratory using the glutamate dehydrogenase method in the Dupont Automatic Clinical Analyzer. The coma scores (Glasgow scale) were measured by the attending nurse. Plasma glutamine was measured by automated amino acid chromatography. Note that the plasma glutamine was not measured before 102 hrs. Plasma glutamine probably reached its maximum value coincident with or several hours later than the maximum plasma ammonia level.

This patient revealed an informative relationship among neurologic status, plasma aspartate aminotransferase level, plasma ammonia, and plasma glutamine during and after a single uncomplicated 72-hr episode of acute liver failure (Fig. 1). A brief summary of the time course of the patient's symptoms and recovery is given in Table 1.

DISCUSSION

Plasma Glutamine and Encephalopathy. Inspection of the relationship between plasma glutamine and neurologic status reveals that the patient's encephalopathy coincided with increased plasma glutamine concentration and only abated when the plasma glutamine level returned to normal (Fig. 1). Plasma glutamine was not measured in this patient at admission, but the zero time preingestion value is plotted in this figure as a normal reference value (approximately 0.6 mmol/L). It is clear from Figure 1 that she was hyperglutaminemic before the first measurement of the plasma glutamine (approximately 1.3 mmol/L) at 102 hrs after ingestion, a value three times that measured at 200 hrs after ingestion (approximately 0.4 mmol/L). At 102 hrs, the patient was comatose, whereas at 200 hrs, she was rapidly regaining normal neurologic function.

Brain Glutamine: Estimation Using Plasma Levels as a Proxy. Although not measured in the patient, her brain glutamine concentration was undoubtedly greatly elevated for a time as a result of the hyperammonemia. In experimental animals, almost all blood-derived ammonia metabolically trapped in the brain is converted to glutamine, even under severely hyperammonemic conditions (15). Furthermore, high ammonia levels inhibit the glutaminase reaction, the main route for metabolism of glutamine in the brain (29). These factors result in glutamine accumulation in the brains of hyperammonemic experimental animals (4, 14, 15, 30). Increased brain glutamine is also well established for hyperammonemic patients. For example, glutamine is elevated 2.5-fold compared with controls in frontal cortex biopsies of hyperammonemic patients dying with ALF, and this is accompanied by a 3.8-fold increase in blood glutamine (31). Noninvasive proton magnetic resonance spectroscopy studies of patients with hepatic encephalopathy have shown the presence of increased brain glutamine (26). Furthermore, glutamine is markedly increased in

Table 1. Chronology of the patient's illness and recovery

Time after Ingestion of Acetaminophen	Comments
64 hrs	The patient was neurologically normal, yet her plasma ammonia was 168 μmol/L (more than four times normal), just below its maximum value of 169 μmol/L
72 hrs	Her neurologic status suddenly and rapidly deteriorated in concert with a decrease in the plasma ammonia concentration and an inferred very high plasma glutamine concentration
110 hrs	Her deep encephalopathy persisted despite normal plasma ammonia levels but with plasma glutamine levels two to three times normal; increased brain glutamine is inferred at this time by exploiting 1) plasma glutamine as a surrogate for brain glutamine; 2) the known increase in brain glutamine in hyperammonemic animal models; and 3) prior studies showing increased brain glutamine in patients with acute liver failure by means of magnetic resonance spectroscopy techniques and by amino acid analysis of brain biopsies (see the text)
150 hrs	She showed signs of neurologic improvement coincident with a decrease of plasma glutamine concentration toward normal or near normal and an inferred decrease of brain glutamine
212 hrs	She was judged to be neurologically normal

cerebrospinal fluid in ALF (31, 32) and arginase-deficient (33) patients. Finally, a net negative arteriovenous difference (i.e., net output) has been reported for glutamine across the brain in healthy, adult human volunteers (34, 35) and in experimental animals (36). This output is likely to increase in hyperammonemic patients with ALF.

The blood–brain barrier and choroid plexus contain several glutamine transporters (37). Thus, a possible factor contributing to hyperammonemia-induced increased brain glutamine is stimulation of glutamine uptake. However, the arrangement and properties of these transporters is such that it is probable the transporters are involved in the net output of glutamine from the brain as a mechanism contributing to nitrogen homeostasis (37). Evidently, however, the efflux rate is not sufficient to keep pace with glutamine synthesis at high levels of ammonia in the astrocyte compartment. Thus, glutamine in the brain accumulates until the astrocyte ammonia levels subside and rate of synthesis is slowed sufficiently to keep pace with glutamine efflux. Taken together, the accumulated findings are consistent with the hypothesis that the plasma glutamine concentration is a likely proxy for glutamine synthesis and output from (rather than input into) the brain, notwithstanding the production of glutamine by other tissues (38). Thus, the correlation between plasma glutamine and neurologic status suggests that the encephalopathy in the patient was dependent on increased brain glutamine, particularly in the astrocytic compartment.

If the encephalopathy was glutamine-dependent, it may reasonably be asked why the patient did not become fully encephalopathic earlier than 72 hrs, especially because it is apparent that she was hyperglutaminemic and her plasma glutamine likely reached its peak before the first glutamine measurement at 102 hrs. The absence of symptoms in the early stages of cerebral edema may be accounted for by the well-known phenomenon of brain compliance in which compression of nonneuronal intracerebral tissue mitigates the early effects of increased volume, thus minimizing the deleterious effects of an increase in intracranial pressure (39, 40).

In summary, the order of recovery of biologic parameters in the patient was: liver function (as deduced from plasma aspartate aminotransferase levels) <plasma ammonia <plasma glutamine <coma score (Fig. 1). Although proton magnetic resonance spectroscopy analysis of brain glutamine was not carried out on this patient, one can infer, as noted previously, that plasma glutamine is a good surrogate for brain glutamine.

The rapid decline of the patient's mental status occurred at a time (64–88 hrs after ingestion of acetaminophen) when plasma ammonia was declining but still above normal. Presumably, during this phase, the concentration of brain glutamine continued to increase because, as a result of persistent increased flux of ammonia into brain, glutamine synthesis continued at an accelerated rate in the astrocytic compartment, and glutamine breakdown may have been slowed by the still high ammonia level. As noted, compliance will protect the brain against astrocyte swelling to some extent. However, this compliance was no longer effective when the increase in astrocyte glutamine reached a critical level and the patient became encephalopathic. Only when brain glutamine levels began to normalize (and relieve osmotic stress) at approximately160 hrs did the patient begin to recover, at which time plasma glutamine levels had normalized.

Our studies underscore the need in patients with ALF for more correlative studies among blood/plasma parameters (ammonia, glutamine, cerebral arteriovenous difference for glutamine, pH, and liver function indices) and intracranial pressure. It is anticipated that positron emission tomography studies and noninvasive magnetic resonance spectroscopy and magnetic resonance imaging techniques for measuring multiple brain parameters will be useful adjuncts to such studies.

Brain Glutamine Synthetase Inhibitor as a Potential Therapeutic Target. If excess cerebral glutamine is a major problem in ALF, then interfering with its synthesis in the brain may be of clinical use. We have suggested that L-methionine-*S*,*R*-sulfoximine, a potent glutamine synthetase inhibitor (41), may be useful (24).

The Johns Hopkins University School of Medicine institutional review board has approved the publication of this case report.

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