

# Conventional Diet Therapy for Hyperammonemia is Risky in the Treatment of Hepatic Encephalopathy Associated with Citrin Deficiency

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## Abstract

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Citrin deficiency caused by *SLC25A13* gene mutations develops into adult-onset type II citrullinemia (CTLN2) presenting with hepatic encephalopathy. Recent studies have suggested that excessive loading of carbohydrates is harmful in citrin-deficient individuals. Here we report a CTLN2 patient who showed further deterioration of encephalopathy after the employment of conventional low-protein diet therapy for chronic liver failure. Owing to the high carbohydrate content, the conventional low-protein diet therapy should be avoided in patients with hepatic encephalopathy associated with citrin deficiency. In addition, our observation may suggest that carbohydrate-restricted diet in which the content of carbohydrate is below 50% of daily energy intake can have therapeutic efficacy in CTLN2 patients.

**Key words:** citrin deficiency, CTLN2, low protein diet, carbohydrate

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## Introduction

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Citrin is a liver-type mitochondrial aspartate (Asp)-glutamate (Glu) carrier (AGC) (1, 2), and citrin deficiency, caused by a mutation of the *SLC25A13* gene, is an autosomal recessive disorder and leads to neonatal intrahepatic cholestasis caused by citrin deficiency (NICCD) and adult-onset type II citrullinemia (CTLN2) (1, 3). Patients with CTLN2 present with intractable hepatic encephalopathy, showing various neuropsychotic manifestations (3). One of the distinct features of CTLN2 is that most patients have a peculiar fondness for protein- and fat-rich foods such as beans and peanuts, and an aversion to carbohydrate-rich foods such as rice and sweets (3). It is assumed that their unusual food preferences may be directly related to the underlying pathophysiology (3). Here we report a CTLN2 pa-

tient who showed a deterioration of consciousness levels accompanied by highly elevated ammonia after conventional low-protein diet therapy for chronic liver failure. His hepatic encephalopathy was gradually ameliorated in parallel with a reduction in the carbohydrate content of his diet.

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## Case Report

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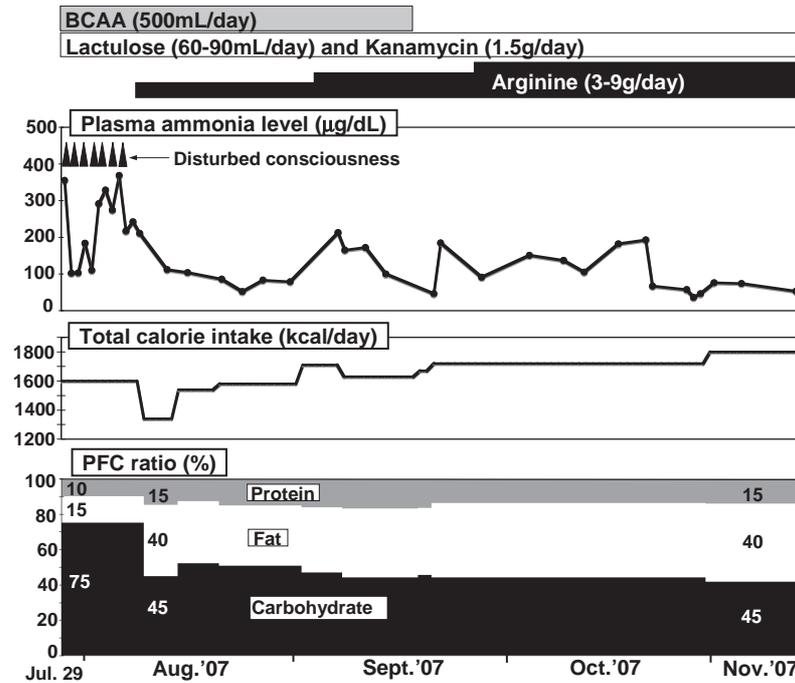
A 51-year-old Japanese man was emergently transferred to a local hospital because of consciousness disturbance in late July, 2007. An elevated plasma level of ammonia was revealed (355 µg/dL, normal <70 µg/dL). He was regarded as having hepatic encephalopathy and was treated with an infusion of branched amino acids followed by a conventional protein-restricted diet (total calories 1,600 kcal/day, protein 40 g/day, protein, fat, and carbohydrate (PFC) ratio 10:15:75%) (Fig. 1). In addition, oral administration of lac-

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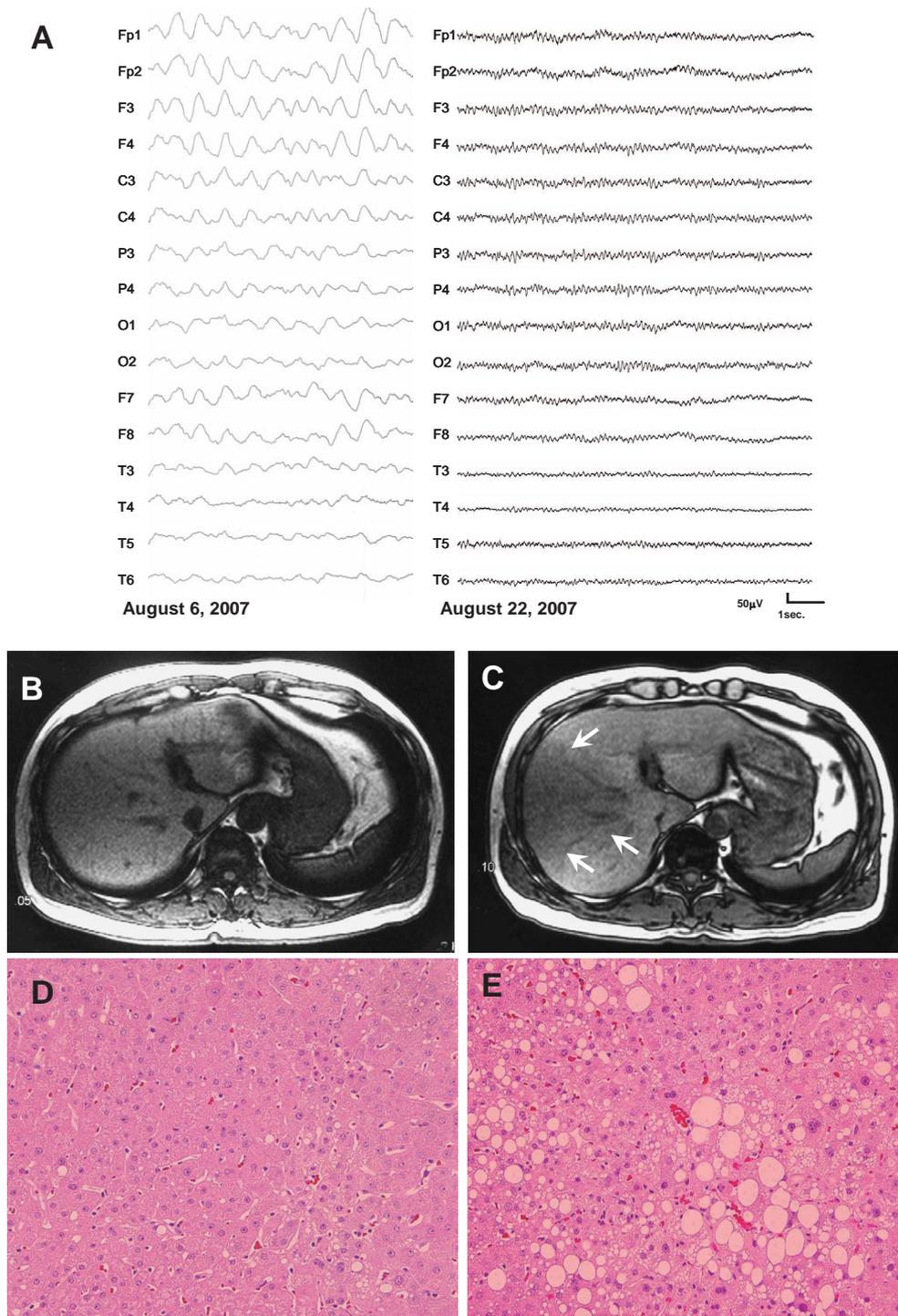
**Figure 1.** Clinical course of the patient showing changes in daily dietary calories and the proportion of dietary energy ratios. BCAA: branched chain amino acid fluid

tulose (60-90 mL/day) and kanamycin (1.5 g/day) was begun. After this treatment, attacks of disturbed consciousness frequently occurred (Fig. 1). An EEG showed diffuse slow waves with the appearance of triphasic waves (Fig. 2A). As his plasma levels of citrulline and arginine were raised (408.9 nmol/mL: normal <40 nmol/mL, and 186.4 nmol/mL: normal <120 nmol/mL, respectively), he was thought to have CTLN2 and referred to our hospital in early August, 2007. He had a fondness for peanuts, milk, meat and fish, and had disliked sweets from childhood. On neurological examination, he was highly irritable and confused, and showed flapping tremor in his hands. His serum transaminases were slightly high (aspartate aminotransferase: 52 IU/L, normal <37 IU/L; alanine aminotransferase: 96 IU/L, normal <45 IU/L) and the serum level of  $\gamma$ -glutamyltransferase was moderately elevated to 138 IU/L (normal <50 IU/L). The levels of total bilirubin, albumin, and total cholesterol, and the hepaplastin test were within normal values. The serum pancreatic secretory trypsin inhibitor (PSTI) level (4) was elevated to 40 ng/mL (normal <20 ng/mL). There was no serological evidence of hepatitis-related viral infection. The abdominal CT and MR images demonstrated no liver cirrhosis or extrahepatic portovenous shunt. There were no remarkable findings suggestive of hepatic steatosis on MR images (Fig. 2B). DNA analysis of the *SLC25A13* gene demonstrated that he was a compound heterozygote for the mutations of 851del4 and IVS13+1 G>A (1), and he was, therefore, diagnosed as having CTLN2. He was started on arginine (3 g/day) (5) and a carbohydrate-restricted diet with a high fat content (total calories 1,340 kcal/day, protein 50 g/day, carbohydrate 150 g/day, PFC 15%:40%:45%) from mid-August. His consciousness level gradually ameliorated

and his plasma ammonia level also decreased (Fig. 1). The EEG recording on August 22 showed an almost normal appearance (Fig. 2A). After that, the daily dose of arginine was increased to 9.0 g/day and total dietary calories were also gradually increased to 1,800 kcal by November 2007 with the ratio of carbohydrate in the total dietary calories restricted to approximately 45% (protein 70 g/day, carbohydrate 200 g/day) (Fig. 1). The MR images in November 2007 showed mild progression of hepatic steatosis (Fig. 2C) and liver biopsy in November 2007 demonstrated relatively mild steatosis (Figs. 2D, 2E). He was discharged in November 2007 and returned to his previous work.

## Discussion

A recent study has disclosed that carbohydrate intake was selectively reduced in the diet of most citrin-deficient subjects, compared to that of the general Japanese population (6). While the PFC ratio of the general Japanese population was 14-15%: 25-30%: 54-58%, that of the citrin-deficient subjects was 19±2%: 44±5%: 37±7% (6). This carbohydrate aversion in citrin deficiency is quite unique in contrast to the protein aversion in other urea cycle enzyme deficiencies (6). In citrin deficiency, the cytosolic NADH/NAD<sup>+</sup> ratio in the hepatocytes can be significantly increased in accordance with the carbohydrate metabolism, resulting in inhibition of ureagenesis by limitation of the supply of Asp for the urea cycle (3). Accumulating data have suggested that the toxicity of high carbohydrate intake, and indeed, intravenous infusion of a high glucose solution or the administration of a glycerol solution resulted in severe hyperammonemia or rapid deterioration of encephalopathy leading to



**Figure 2.** Serial EEG recordings (A), MR images (T1-weighted, out-of-phase images) (B,C) and histopathology of the biopsied liver (D,E) in the patient. Serial EEG recordings show a marked decrease in slow waves in the two weeks between early August, 2007 (PFC: 10%:15%:75%) and mid-August, 2007 (PFC:15%:35%:50%) (A). On MR imaging, mild progression of hepatic steatosis (white arrows) was seen between MR imaging performed in mid-August, 2007 (B) and mid-November, 2007 (C). The histopathology of the liver, which was biopsied in mid-November, 2007, demonstrated mild steatosis (D) with scattered moderate fat infiltration (E) (Hematoxylin and Eosin staining  $\times 100$ ).

death in many CTLN2 patients (7). Also, it has been found that oral sucrose administration exacerbated hyperammonemia in citrin/mitochondrial glycerol 3-phosphate dehydrogenase double-knockout mice, which are an animal model of

human citrin deficiency (8). Therefore, high carbohydrate intake can deteriorate hepatic encephalopathy in CTLN2 patients.

Protein restriction has classically been considered a main-

stay of treatment in hepatic encephalopathy to reduce the nitrogen load (9). Usually, in patients with chronic liver disease, especially in liver cirrhosis, protein intake is restricted to 0.8 to 1.0 g/kg/day (9). In the conventional diet for chronic liver disorders in our institution, dairy protein intake is commonly restricted to 40 g (total daily calories 1,400 kcal). However, carbohydrate intake is inevitably increased (270 g/day) and thus the PFC ratio of this diet is 12%:11%:77%, which is the almost same as the patient's initial hospital diet (Fig. 1). Therefore, in our patient, the deterioration of consciousness with highly elevated plasma ammonia appeared to be closely associated with the protein-restricted diet with high carbohydrate ratio. Interestingly our patient's condition gradually improved after starting a low carbohydrate and high fat diet (Fig. 1). The efficacy of a carbohydrate-restricted diet in citrin deficiency has been reported in a few patients (5, 10). Imamura et al described a 37-year-old CTLN2 patient who had amelioration of hypertriglyceridemia and ketogenesis impairment with a reduction of carbohydrate content from 70% to 60% in the PFC ratio (5). In addition, Dimmock et al reported a 10-month-old patient with citrin deficiency presenting with failure to thrive, which improved after starting a high protein and low carbohydrate diet (PFC ratio 15%:50%:35%) (10).

Hepatic steatosis or steatohepatitis is one of the cardinal manifestations in CTLN2 patients and often severe fatty liver is also seen (11, 12). So far, over 10 patients with CTLN2 have undergone liver transplantation at our institution (13), and rapid progression of steatosis often occurred in some patients with CTLN2 who were given a low-protein diet (30-40 g/day), and/or administered hyperalimentation fluid over several months while waiting for liver transplantation (13, 14). Particularly, in our previously reported 32-year-old Japanese man with CTLN2 (14, 15), significant progression of steatosis, probably associated with the low-protein diet and infusion of hyperalimentation fluid, was observed in just three weeks. In the past, this progression of steatosis was thought to be due to the malnutrition caused by the excessively protein-restricted diet. However, several reviews have recently suggested that it could be directly associated with glucose metabolism in citrin deficiency: an increase of the cytosolic NADH/NAD<sup>+</sup> ratio following carbo-

hydrate metabolism could activate the citrate-malate shuttle in compensation for the hepatic AGC (citrin) insufficiency, resulting in the overproduction of fatty acids in the hepatocytes (3). In the present patient, it was of particular interest that hepatic steatosis was relatively mild at biopsy and the rapid progression of hepatic steatosis was not observed in the serial MR images (Figs. 2B, 2C). This may suggest both the risk of the conventional low-protein diet and the efficacy of a carbohydrate-restricted diet as described previously (5).

Thus, since an excessively high-carbohydrate diet can have a harmful effect on CTLN2 patients, it is concluded that the conventional protein-restricted diet for hepatic encephalopathy, leading to a high carbohydrate intake of over 60-70% in the PFC ratio, should be avoided, at least in patients with CTLN2. In addition, a carbohydrate-restricted diet in which the content of carbohydrate is below 50% of daily energy intake may have therapeutic efficacy in CTLN2 patients. It remains unclear whether protein-restriction in the diet is necessary or not in addition to carbohydrate-restriction in CTLN2 patients. However, because most CTLN2 patients have urea cycle deficiency due to the decrease of hepatic argininosuccinate synthetase, high protein diet may lead to excessive nitrogen load to the urea cycle. In our patient's diet, therefore, in addition to carbohydrate-restriction, the daily protein was restricted (50 g-70 g/day).

In East Asian countries, the frequency of heterozygotes with the mutated *SLC25A13* gene is approximately 1 in 70, suggesting that over 80,000 East Asians may be homozygotes (16), thus large numbers of patients may still be undiscovered. In patients with hepatic encephalopathy, particularly those who have a peculiar food preference with a dislike of alcohol, the possibility of citrin deficiency should be considered and special care is necessary in this unique inherited hepatocerebral disorder.

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