대한 유전성 대사질환 : 제 1 권 제 1 호, pp23~27, 2001

A Korean patient with Fanconi-Bickel Syndrome Presenting with Transient Neonatal Diabetes Mellitus and Galactosemia : Identification of a Novel Mutation in the GLUT2 Gene

Han-Wook Yoo[1, 3], Eul-Ju Seo[1, 2], Gu-Hwan Kim[3]

Department of Pediatrics[1], Asan Medical Center, Ulsan University College of Medicine, Seoul, Korea
Department of Clinical Laboratory Medicine[2], Asan Medical Center, Ulsan University College of Medicine, Seoul, Korea
Medical Genetics Clinic & Laboratory[3], Asan Medical Center, Seoul, Korea

INTRODUCTION

Fanconi-Bickel syndrome(FBS)(Mckusick 227810) is a rare, well-characterized clinical entitiy, inherited in an autosomal recessive mode. It is manifested by postprandial hyperglycemia and hypergalactosemia, hepatorenal glycogen accumulation, and proximal renal tubular dysfunctions. Recently, its molecular defect has been elucidated in the glucose transporter 2 gene (GLUT2), which encodes one of facilitative glucose transporter expressed in liver, kidney, intestine and pancreatic islet cells. Both an impaired glucose tolerance and a hypergalactosemia may be resulted from a defect of the facilitative glucose transporter 2 (Glut2).

We describe clinical features and an identification of a molecular defect of the GLUT2 gene in a Korean patient with FBS, initially presenting diabetes mellitus and hypergalactosemia during neonatal period, subsequently succumbed to severe rickets and liver cirrhosis at 10 month of age.

CLINICAL REPORT

1. Clinical presentation

The patient was the second preterm female newborn to normal healthy parents with nonconsanguinous marriage with the birth weight of 2.0 kg at the 34th week of gestation. The pregnancy of the second gravid 31 year old mother had been complicated by oligohydramnios and breech presentation. The newborn was delivered by cesarean section. Although she was not dysmorphic, she was emaciated with little subcutaneous fat. She was initially cared at neonatal intensive care unit because of pneumothorax and hyperglycemia (320 mg%) detected at 6 days after birth on formula milk feeding. Blood glucose level had been remarkably fluctuating before and after being fed. Serum C-peptide level was 0.19 ng/mL (0.5-2.0), 24 hours urine C-peptide 2.8 ug/day (44-116). Neonatal metabolic screening for galactosemia performed at 3days after birth revealed hypergalactosemia (135 mg/dL). Enzyme activities of galactose-1-phosphate uridyltransferase, galactokinase, and UDP-galactose epimerase were within normal range. The serum level of total alkaline phosphatase was markedly

elevated (6520 IU/L) (66-220). It turned out to be mostly from bone origin based on alkaline phosphatase isoenzyme isoelectrofocusing. Total calcium was 8.7 mg/dL, phosphorus 1.9 mg/dL, sodium 131 mEq/L, potassium 3.8 mEq/L, chloride 112 mEq/L, bicarbonate 13 mEq/L. The renal tubular reabsorption rate of phosphate was 25%. Urine analysis revealed both a glycosuria and a galactosuria. There was no detectable ketonuria. The aminoacid analysis in urine suggested massive generalized aminoaciduria. She had been managed with lactose free milk, intermittent insulin injection, and administration of electrolyte solutions (Joulie's solution, Shol's solution) and $1,25(OH)_2$ vitamin D_3. However, she profoundly failed to grow and her liver had been progressively enlarged, palpable 10 cm below right costal margin. Her cheeks were chubby and craniotabes was found at both parietotemporal regions of her skull. Her liver functions were progressively deteriorated with increased prothrombin time. Liver biopsy done at age 9 months indicated micronodular cirrhosis with marked fatty changes. She succubmed to hepatic failiure with pneumonia at 10 months of age.

2. Mutation analysis of the GLUT2 gene

Since a defect in glucose-galactose transport was presumed, the diagnosis of Fanconi-Bickel syndrome (FBS) was made clinically. In order to confirm the diagnosis at molecular level, a mutation analysis of the GLUT2 gene was conducted using genomic DNA isolated from the patient's peripheral leukocytes. All 11 coding exons and their intron-exon boundaries were amplified by PCR. The PCR products were analyzed by a single stranded conformational polymorphism (SSCP) method, using silver staining for visualization of the DNA strands. Subsequently, the PCR product showing mobility shift was directly sequenced. We identified a novel nonsense mutation at the 5th codon; Lysine5 Stop(K5X) in exon 1, which was resulted from an A to T base substitution at cDNA nt. 322. It leads to the premature termination, causing the enormous truncation of the Glut2 protein. The patient was a homozygote for this mutation, and her parents were heterozygote for the same mutation.

DISCUSSION

The concurrence of both hyperglycemia and hypergalactosemia is the main sign of Fanconi-Bickel syndrome (FBS), a rare autosomal recessive metabolic disorders characterized by hepatorenal glycogen accumulation, proximal renal tubular dysfunction, and impaired utilization of glucose and galactose. Fanconi and Bickel were the first who described the combination of a generalized proximal renal tubular dysfunction and histologically proven glycogen storage disease. Although more than 80 cases have been reported as FBS during past 50 years, the exact prevalence of this disease is not known. Since the first description of a molecular defect in the GLUT2 gene in 1997, 14 cases have been confirmed by the identification of mutations in the GLUT2 gene. Most of them were from European, Arabian, Jewish or African descent. Our patient is the first Asian patient with FBS confirmed by a mutation detection in the GLUT2 gene.

It is unusual that patients with FBS manifest full blown clinical pictures in early infancy as well as die of hepatic failure like in this case. The common first symptoms are fever, vomiting, growth failure, and hypophospahatemic rickets at the age of 3-10 months. As neonatal screening for galactosemia is available, a few cases have been detected during neonatal period. With renal tubular defects, generalized hyperaminoaciduria, hyperphosphaturia, and hypercalciuria are constant findings. Also renal bicarbonate threshold is de-

creased and leads to metabolic acidosis. Later the patients presented with dwarfism, protuberant abdomen, hepatomegaly, moon-shaped face. The striking feature is short stature. Rickets and osteoporosis later in life were frequently reported resulting in pathological fractures in a few cases. Our patient manifested severe rickets and failure to thrive, eventually complicated by pneumonia.

The liver appears normal or slightly increased at birth and typically becomes greatly enlarged during infancy. In general, the glycogen content of liver tissue is significantly elevated. However, hepatic adenoma or malignancies, cirrhosis as reported for other types of glycogen storage disease have never been reported. In this regards, this patient is unique since a liver biopsy done at 9 months of age revealed cirrhotic pattern and clinically she developed hepatic faliure, which she was succumbed to.

This patient presented with transient neonatal diabetes mellitus and a galactosemia, which were caused by a molecular defect of the facilitative glucose transporter 2. The glucose transporter 2 is expressed in hepatocytes, pancreatic beta cells, and the basolateral membranes of intestinal and renal epithelial cells. It mediates the gradient-dependent facilitative bidirectional transport of glucose and galactose across cell membranes. Hyperglycemia and hypergalactosemia in the fed state can be explained by a decreased monosaccharide uptake by liver. In the pancreatic beta cell, glucose sensing is so defective that blood glucose concentrations are fluctuating. Hyperglycemia in Fanconi-Bickel syndrome also seems to be enhanced by an inappropriately low insulin secretion. In this case, 24 hours urine c-peptide level was reduced.

The cDNA of the human GLU2 gene was cloned in 1988 by Fukumoto et al., subsequently mapped on human chromosome 3q26.1-26.3. The genomic DNA structure of the GLU2 gene has been described by Takeda et al in 1993. It consists of 11 exons spanning approximately 30 kb. This provided the basis for the identification of a molecular defect in the GLU2 gene. Since both glucose and galactose metabolism are impaired in FBS, Santer et al. (1997) hypothesized a primary defect in monosaccharide transport across membranes and subsequently investigated mutations in the GLUT2 gene. He described three different mutations in three FBS families. Interestingly all the mutations were homozygous nonsense or frame shift mutations, resulting in truncated protein. Functional monosaccharide transport activity is not obviously expected. Out patient was also carrying a homozygous nonsense mutation (K5X), her parents were heterozygous for the mutation. This nonsense mutation gives rise to the markedly truncated protein with only 4 out of 524 aminoacids, which can explain severe clinical features shown in this patient.

There is no known effective therapy for FBS. Patients are managed only symptomatically. It includes replacement of water and electrolytes, alkalinization with Shohl or bicarbonate solutions, supplementation of vitamin D and phosphate, restriction of galactose and a diabetes mellitus like diet with small frequent meals with adequate caloric intake. Use of uncooked corn starch or fructose for an alternate carbohydrate source is suggested to be beneficial. Our patient has been on lactose free milk, intermittent insulin injection, electrolyte solutions (Joulie's solution, Shol's solution) and $1,25(OH)_2$ vitamin D_3. However, she failed to respond, her symptoms were progressively aggravated.

Overall prognosis of FBS is reported as favorable. Many cases survived in adulthood. However, this patient presented her symptoms at birth She might be affected even during intrauterine period since she was prematurely born with small birth weight for gestational age and oligohydramnios.

Severe clinical presentations and bad prognosis must be correlated with her genotype in this patient.

REFERENCS

1) Fukumoto H, Seino S, Imura H, SeinoY, Eddy RL, Fukushima Y, Byers MG, Shows TB, Bell G I. Sequence, tissue distribution, and chromosomal localization of mRNA encoding a human glucose transporter-like protein. Proc Nat Acad Sci 1998; 85:5434-5438.
2) Muller D, Santer R, Krawinkel M, Christiansen B, Schaub J. Fanconi-Bickel syndrome presenting in neonatal screening for galactosaemia. J Inherit Metab Dis 1997;20:607-608
3) Santer R, Schneppenheim R, Dombrowski A, Gotze H, Steinmann B, Schaub J. Mutations in GLUT2, the gene for the liver-type glucose transporter, in patients with Fanconi-Bickel syndrome. Nature Genet 1997;17:324-326.
4) Santer R, Schneppenheim R, Suter D, Schaub J, Steinmann B. Fanconi-Bickel syndrome - the original patient and his natural history, historical steps leading to the primary defect, and a review of the literature. Eur J Pediat 1988;157:783-797
5) Matsutani A, Hing A, Steinbrueck T, Janssen R, Weber J, Permutt MA, Donis-Keller H. Mapping the human liver/islet glucose transporter (GLUT2) gene within a genetic linkage map of chromosome 3q using a (CA)n dinucleotide repeat polymorphism and characterization of the polymorphism in three racial groups. Genomics 1992;13: 495-501

= **Abstract** =

A Korean patient with Fanconi-Bickel Syndrome Presenting with Transient Neonatal Diabetes Mellitus and Galactosemia : Identification of a Novel Mutation in the GLUT2 Gene

Han-Wook Yoo[1,3], Eul-Ju Seo1[2] and Gu-Hwan Kim[3]

Department of Pediatrics[1], Asan Medical Center, Ulsan University College of Medicine, Seoul, Korea
Department of Clinical Laboratory Medicine[2], Asan Medical Center, Ulsan University College of Medicine, Seoul, Korea
Medical Genetics Clinic & Laboratory[3], Asan Medical Center, Seoul, Korea

Fanconi-Bickel Syndrome (FBS) is a rare autosomal recessive disorder of carbohydrate metabolism recently demonstrated to be caused by mutations in the GLUT 2 gene for the glucose transporter protein 2 expressed in liver, pancreas, intestine, and kidney. This disease is characterized by hepatorenal glycogen accumulation, both fasting hypoglycemia as well as postprandial hyperglycemia and hyperglactosemia, and generalized proximal renal tubular dysfunctions. We report the first Korean patient with FBS diagnosed based on clinical manifestations and identification of a novel mutation in the GLUT 2 gene. She was initially diagnosed having a neonatal diabetes mellitus due to hyperglycemia and glycosuria at 3 days after birth. In addition, newborn screening for galactosemia revealed hypergalactosemia. Thereafter, she has been managed with lactose free milk, insulin therapy. However, she failed to grow and her liver has been progressively enlarging. Her liver functions were progressively deteriorated with increased prothrombin time. Liver biopsy done at age 9 months indicated micronodular cirrhosis with marked fatty changes. She succubmed to hepatic failiure with pneumonia at 10 months of age. Laboratory tests indicated she had generalized proximal renal tubular dysfuctions; renal tubular acidosis, hypophosphatemic rickets, and generalized aminoaciduria. Given aforementioned findings, the diagnosis of FBS was appreciated at age of 2 months. The DNA sequencing analysis of the GLUT 2 gene using her genomic DNA showed a novel mutation at 5th codon; Lysine5 Stop (K5X).

Key Words : Fanconi-Bickel syndrome, Transient diabetes mellitus, Galactosemia, GLUT2 gene, novel mutation, Proximal renal tubular dysfunction