

A Suspected Case of Ornithine Transcarbamylase Deficiency in a Cat

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ABSTRACT. An 18 month-old, intact female American Shorthair cat was presented for evaluation of stunted growth and postprandial depression. Fasting serum ammonia and serum bile acid concentrations were above reference ranges at 396 $\mu\text{g/dl}$ and 6.5 $\mu\text{mol/l}$ and their postprandial concentrations were 785 $\mu\text{g/dl}$ and 9.5 $\mu\text{mol/l}$, respectively. The initial tentative diagnosis of a portosystemic shunt was excluded by mesenteric portography and histopathology of the liver. The cat was then suspected of a urea cycle enzyme deficiency and its urine was analyzed by gas chromatography-mass spectrometry. A presumptive diagnosis of ornithine transcarbamylase deficiency was made on the basis of the detection of orotic acid and uracil.

KEY WORDS: feline, ornithine transcarbamylase, urea cycle.

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Ornithine transcarbamylase (OTC) deficiency is the most common urea cycle enzyme deficiency in humans and is inherited as a sex-linked dominant trait. The clinical features of OTC deficiency range from an acute sickness resulting in early death to remaining completely asymptomatic [5].

Neonatal screening for the early diagnosis of OTC deficiency and other inherited diseases of metabolism has been implemented in human medicine for the early detection, management and forestall the development of deleterious clinical symptoms. Gas chromatography-mass spectrometry (GC/MS) is often used for the initial diagnostic screen of urine in cases of suspected inherited diseases. This report describes the use of GC/MS for the analysis of urinary metabolites in a cat suspected of an OTC deficiency.

An 18 month-old, intact female American Shorthair cat weighing 1.9 kg was presented to a private veterinary hospital for evaluation of stunted growth and postprandial depression. On physical examination the cat was alert, afebrile and in a normal state of hydration. No physical signs of overt disease were observed. Thoracic and abdominal radiographs were unremarkable. The results of a complete blood count were within reference ranges. In the serum chemistry profile, only the alanine amino transferase (ALT, reference range, 10-109 U/l) and aspartate amino transferase (AST, reference range, 13-85 U/l) activities were elevated at 367 U/l and 131 U/l, respectively. Fasting serum ammonia (reference range < 50 $\mu\text{g/dl}$) and serum bile acid (reference range < 10 $\mu\text{mol/l}$) concentrations were 396 $\mu\text{g/dl}$ and 6.5 $\mu\text{mol/l}$, respectively, and their postprandial concentrations were 785 $\mu\text{g/dl}$ and 9.5 $\mu\text{mol/l}$, respectively. The cat was fed Hills feline k/d and l/d (Hills Pet Products, Topeka, KS, U.S.A.), both protein restricted diets and was treated with lactulose and ursodeoxycholic acid. ALT and AST activities gradually decreased to 255 U/l and 61 U/l, respectively over a 2 months period whereas serum ammonia concentra-

tions remained consistently higher than 300 $\mu\text{g/dl}$.

Suspecting a portosystemic shunt, the referring veterinarian performed a positive-contrast portography and obtained a wedge biopsy during an exploratory laparotomy. No shunt vessels were observed on the radiographs and there were no histopathological findings such as sinusoidal distension, parenchymal atrophy, or an increased number of small caliber vessels to suggest a portosystemic shunt [2]. The case was then referred to Nippon Veterinary and Animal Science University Veterinary Medical Teaching Hospital. GC/MS analysis was performed to elucidate the possible metabolic disturbances.

Precise procedure for GC/MS analysis of urine has been reported previously [4]. Briefly, 100 μl of urine was treated with 15 units of urease, deproteinized with ethanol, evaporated to dryness under reduced pressure, trimethylsilylated and injected into a GC/MS. GC/MS was carried out using a Shimadzu QP 5050A instrument on an ultra alloy capillary column. The temperature of the GC was maintained at 60°C for 1 min, and then increased to 350°C at a rate of 17°C/min. Mass spectra were recorded between m/z 50 to m/z 650 every 0.25 sec. The data were analyzed by a computer-assisted program. This GC/MS system for analyzing urinary metabolites enables detection of most of the congenital metabolic disturbances of human neonates causing hyperammonemia.

GC/MS analysis of urine revealed the presence of orotic acid (peak number 27) and uracil (peak number 13). These two metabolites are consistently identified by GC/MS in the urine of human patients with OTC deficiency. There were no additional metabolites suggestive of other metabolic diseases which would result in hyperammonemia in the urine of this cat. The cat's urine was analyzed three times by GC/MS; at initial presentation, at one month and at twelve months after presentation. Similar results were obtained each time. Orotic acid and uracil are not present in the urine

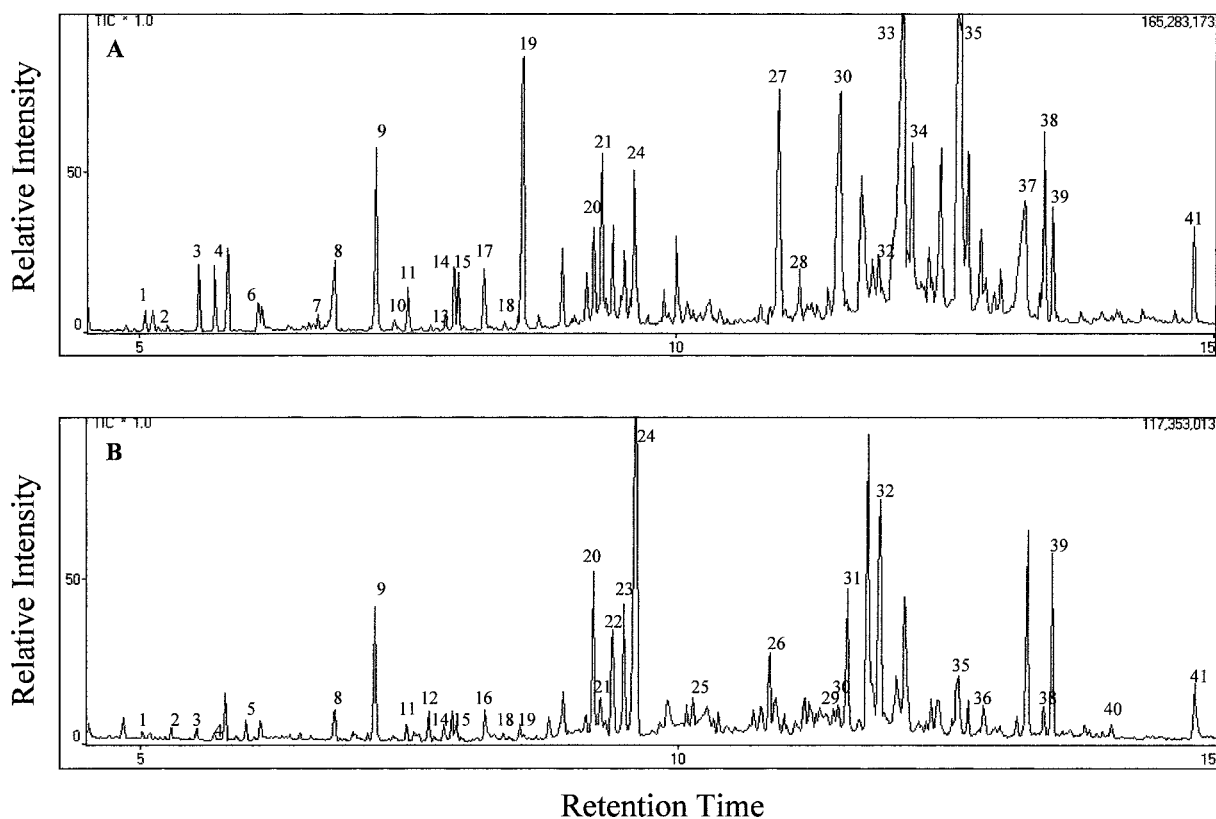


Fig. 1. Reconstructed ion chromatogram of the urinary trimethylsilylated organic compounds of the patient (A) and normal cat (B). Orotic acid (peak number 27) and uracil (peak number 13) are identified only in the urine of the patient. The peaks are: 1, lactate; 2, glycolate; 3, alanine; 4, glycine-1 (glycine-2-trimethylsilylated); 5, cresol; 6, 3-hydroxybutyrate; 7, valine; , urea; 9, phosphate; 10, isoleucine; 11, glycine-2 (glycine-3-trimethylsilylated); 12, glycerate; 13, uracil; 14, 4-deoxytetronate; 15, serine; 16, catechol; 17, threonine; 18, 3-deoxytetronate; 19, 2-deoxytetronate; 20, erythritol; 21, pyroglutamate; 22, thermal decomposite from allantoin; 23, erythronate; 24, creatinine; 25, 4-hydroxyphenylalanine; 26, arabitol; 27, orotic acid; 28, glutamine; 29, fructose; 30, citrate; 31, hippurate; 32, phenylacetylglucine; 33, α -glucose; 34, histidine; 35, β -glucose; 36, gluconate; 37, kynurenate; 38, *myo*-inositol; 39, internal standard (*n*-heptadecanoate); 40, 3-hydroxyhippurate; 41, pseudouridine. Peaks without numbers are unknown peaks.

of healthy cats, hyperammonemic cats with portosystemic shunt or in those with a hepatic insufficiency.

In OTC deficiency, orotic acid concentrations increase as a result of the diversion of accumulated mitochondrial carbamoyl phosphate to the cytosolic pyrimidine synthetic pathway. Other pyrimidines such as uracil and uridine are also found in the urine of human patients with OTC deficiency [8]. OTC is the second enzyme in the urea cycle and catalyzes the reaction of carbamoyl phosphate and ornithine to synthesize citrulline. In OTC deficiency, ammonia, the precursor of carbamoyl phosphate is generally very high in the blood [7].

It has been reported that the liver enzymes are frequently elevated in human patients with OTC deficiency and not limited to episodes of hyperammonemia [3]. Liver enzyme activities of this cat were high at the first presentation and gradually decreased during the first 2 months and then fluctuated thereafter. The cat has remained essentially free of clinical signs since being placed on a protein restricted diet and medication. During this time, however, blood ammonia

concentration has always remained above the reference range. Similar to human patients, liver enzyme activities did not correlate with blood ammonia concentration in this cat.

Hyperammonemia can develop due to several different mechanisms, including: 1) portosystemic shunt, 2) hepatic insufficiency, 3) reduced activity of urea cycle enzymes, 4) metabolic disorders of organic acids. In the presence of a persistently high concentration of blood ammonia, these animals may demonstrate neurobehavioral abnormalities. When young animals are presented with hyperammonemia, a portosystemic shunt is an important differential because its incidence rate is relatively high in dogs and cats. However, if pre- and postprandial bile acid concentrations are not elevated, inherited metabolic diseases must be considered. Few reports concerning urea cycle enzyme deficiencies resulting in hyperammonemia have been published in animals [6]. In general, direct measurement of the deficient enzyme activity in liver tissue is considered essential for the definitive diagnosis of a urea cycle enzyme deficiency. In

this study, the liver OTC activity could not determined because a second biopsy of the liver was not permitted.

In humans, OTC deficiency is the most frequently encountered inherited urea cycle enzymopathy and its incidence is reported to be approximately 1 per 14,000 [1]. It is possible to hypothesize that there are more cases of inherited metabolic diseases in companion animals than in humans because inbreeding among purebreds is common. Screening analysis of the urine by GC/MS is a noninvasive, accurate and relatively rapid procedure. It can be a very useful tool to screen animals suspected of having metabolic disorders such as OTC deficiency. In the present case, a presumptive diagnosis of OTC deficiency was made on the basis of the detection of orotic acid and uracil in the cat's urine by GC/MS.

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