Medium-Chain Acyl Coenzyme A Dehydrogenase Deficiency

Occurrence in an Infant and His Father

Marc Bodman, BS; David Smith; William L. Nyhan, MD, PhD; Robert K. Naviaux, MD, PhD

Background: Autosomal recessive inborn errors of metabolism often present as life-threatening disease in infancy and have adverse effects on the nervous system. Parents are usually heterozygotes. This is true of most disorders of fatty acid oxidation, which are rare and present with hypoketotic hypoglycemia. However, the gene for medium-chain acyl coenzyme A dehydrogenase (MCAD) deficiency is common in white people, raising the possibility that a parent may be homozygous.

Objective: To document the occurrence of MCAD deficiency in a 12-month-old boy and his father, both of whom were homozygous for the A985G mutation.

Design: Clinical observations and definitive biochemical testing.

Setting: Children’s hospital and university laboratory.

Participants: One child and one adult.

Interventions: Diagnosis and treatment.

Main Outcome Measures: Clinical outcome; analysis results of plasma and urine for carnitine and organic acids.

Results: An infant admitted with an acute illness requiring intensive care was found to have carnitine deficiency and dicarboxylic aciduria; MCAD deficiency was diagnosed by assay of his DNA for the common mutation. Test results of the father revealed him also to be homozygous.

Conclusion: In MCAD deficiency, as opposed to the usual rare autosomal recessive metabolic disease, a parent may also be an affected homozygote.

Arch Neurol. 2001;58:811-814

MEDIUM-CHAIN acyl coenzyme A dehydrogenase (MCAD) (EC 1.3.99.3) catalyzes the β-oxidation of straight-chain acyl-coenzyme A esters from C4 to C12 units in length. An MCAD deficiency is an inborn error of metabolism in which fatty acid oxidation is impaired. It is an autosomal recessive disorder affecting approximately 1 in 10,000 white people. The most frequent cause of MCAD deficiency is a point mutation that changes an adenosine (A) to a guanine (G) at nucleotide 985, which results in a lysine to glutamic acid change in position 329 of the protein. Clinical manifestations of the disorder are typically hypoglycemia with impaired ketogenesis and secondary deficiency of carnitine. It is the purpose of this report to describe a boy with MCAD deficiency who was homozygous for the A985G mutation and whose father was also homozygous for this mutation.

CASE 1

A 12-month-old white infant was admitted to the pediatric intensive care unit at Children’s Hospital and Health Center in San Diego, Calif, because of diarrhea, an upper respiratory tract infection, and lethargy. Parents were not consanguineous. For the first 9 months of his life, his mother had awakened him at 2 AM for a feeding, but at age nine months he began to sleep through the night. At about this time the mother began to incorporate meals of solid food and to develop more of a 3-meals-a-day program. Nine days prior to admission he was treated with an antibiotic because of a diagnosis of otitis media, but this treatment was discontinued after 3 days. Vomiting and diarrhea began 2 days prior to admission. He was brought to the emergency department where a barium enema was admin-
METHODS

Organic acid analysis was performed using gas chromatography–mass spectrometry.³ Urine and plasma levels of carnitine were determined with a radioisotopic enzyme assay.³ ³

Analysis of DNA for the A985G mutation was carried out by polymerase chain reaction to amplify a 63-base pair (bp) fragment with mismatched oligonucleotide primers.³ ³ The product was digested with the restriction endonuclease NcoI and analyzed by electrophoresis on 12% polyacrylamide gel. With the mismatched primer, the mutant DNA yielded a 43-bp fragment that could be distinguished from the normal 63-bp fragment.

istered because the presence of Hemoccult-positive (Beckman Coulter Inc, Fullerton, Calif) stool and lethargy suggested the possibility of intussusception. Urine output was thought to be reduced.

On admission he appeared sleepy but was rousable. He could track with his eyes but had little interaction with the examiner. His temperature was 36.7°C; pulse, 105 beats/min; respirations, 30 breaths/min; and blood pressure, 96/40 mm Hg. His extremities were cool and blue, and slow capillary refill indicated poor perfusion. Nasal mucous membranes were inflamed. He appeared acutely and seriously ill. The liver was palpable 4 cm below the costal margin on the right and had a firm and smooth contour. Deep tendon reflexes were normal, and there was no clonus. Findings of physical examination were otherwise unremarkable.

Laboratory findings included the following values in millimoles per liter: sodium, 136; potassium, 3.3; chloride, 104; and bicarbonate ions, 14. The leukocyte count was 18.3 × 10⁹/L with 56% segmented polymorphonuclear cells and 28% band forms. The hemoglobin level was 104 g/L; hematocrit, 0.31; and platelet count 436 × 10⁹/L. Serum activity of creatine phospho kinase was elevated at 796 U/L and rose to 1077 U/L the next day. The serum alanine aminotransferase reading was 167 U/L; aspartate aminotransferase reading was 167 U/L; aldolase, 120 U/L; and γ-glutamyltransferase, 16 U/L. The serum urea nitrogen was 15 mmol/L (40 mg/dL) and creatinine, 44 µmol/L. The initial blood glucose level was 1.83 mmol/L (33 mg/dL) rising to 16.65 mmol/L (300 mg/dL) after intravenous administration of a 2 mL/kg bolus of 10% glucose. The albumin level was 33 g/L; calcium, 2.03 mmol/L (8.1 mg/dL); glucose in the cerebrospinal fluid, 5.38 mmol/L; and ammonia, 56.9 µmol/L (97 µg/dL). Findings of the urine test for ketones revealed a trace amount. The uric acid level was not measured until day 3, at which time it was 172.5 µmol/L. Culture findings of the blood, urine, and cerebrospinal fluid were negative. Results of viral studies were positive for rotavirus and respiratory syncytial virus.

The patient was treated with parenteral fluids containing glucose, saline, and potassium acetate, along with added sodium bicarbonate until the serum bicarbonate became normal on day 2. In addition, ceftriaxone sodium was given intravenously for 24 hours.

Atrial bigeminy developed on day 2 after an initial period of stability, and a 2:1 block was noted. Findings of an echocardiogram revealed normal cardiac anatomy. Cardiac output was low, and the patient was treated with dopamine hydrochloride, dobutamine hydrochloride, atropine sulfate, and isoproterenol hydrochloride. The arrhythmia resolved promptly and findings of electrocardiography became normal. Peripheral perfusion and mental status were considered to be normal after the initial hydration. The creatine kinase level was 174 U/L on day 4.

Urine was analyzed for the pattern of organic acid excretion and carnitine, and plasma for amino acid and carnitine (Table). The patient was discharged after 5 days in the hospital. Once a diagnosis of MCAD deficiency was made, the parents were counseled about the avoidance of fasting, a cornstarch supplement was prescribed at bedtime, and 1 g of carnitine was given daily.

CASE 2

When patient 2 (the 29-year-old father of the patient in case 1) and his wife were told of the diagnosis of their son’s illness and its nature and treatment, the history emerged that patient 2 at age 11 months had developed what was called hypoglycemic shock after a period of extended fasting. This resolved, but he experienced a second episode soon after that. Thereafter he had studiously avoided fasting. He would eat at other than normal mealtimes and would often wake up from sleep to eat. In addition, throughout his childhood, family members were encouraged to give him carbohydrates to prevent a recurrence. Blood was sent for molecular study, and he was found to be homozygous for the A985G mutation. Carnitine therapy was initiated. Patient 2 has one sibling to whom testing for the mutation has been offered.

The findings of analysis of concentrations of amino acids in the plasma of case 1 were notable for a concentration of alanine of 642 µmol/L. Analysis of organic acids of the urine was carried out the day prior to discharge when he was clinically well, and the results revealed that the suberylglycine level was 12 mmol per mole of cre-* Unless otherwise indicated, data are micromoles per liter.
† Unless otherwise indicated, data are millimoles per mole of creatinine.
In dealing with families of patients with inherited metabolic disease, it is generally expected that the parents are heterozygotes and asymptomatic. This is because most disorders are the result of rare autosomal recessive genes. However, MCAD deficiency is different in that the gene is relatively common in white populations. Results of a literature search showed that this situation has occurred previously. Hepinstall et al described in 1995 a family in which the father and his proband daughter were both homozygous for the A985G mutation. In this family the proband presented at age 6 months with a life-threatening hypoglycemic episode, while the father was asymptomatic. He had always had a predilection for sweet food. A brother of the proband was found to be homozygous by analysis of cord blood. Nevertheless, the possibility that a parent and child are both homozygous is not generally considered.

In our kindred, direct transmission of the mutant alleles from each parent to case 1 is the logical explanation. The alternative uniparental disomy is unlikely. The importance of these experiences is the utility of investigation for homozygosity in a parent of a child with a prevalent mutation. This is particularly relevant to MCAD deficiency, a disorder in which sudden death can be an initial presentation at any age.

An MCAD deficiency usually presents first in infants between ages 6 months and 2 years. In the usual situation, acute intercurrent infection is followed by decreased oral intake and vomiting. Fatty acid oxidation is required when glycogen stores are depleted, and this usually occurs after 12 hours or more of fasting. At the same time, it is clear that some individuals can live many years without clinical manifestation of the disease. The excretion of esterified carnitine in the urine is high. The esterified carnitine–free carnitine ratios were high.

The findings of the organic acid analysis in the urine of patient 2 were unremarkable. His plasma free carnitine level was low (Table). In the urine, the esterified carnitine–free carnitine ratio was high.

Analysis of genomic DNA extracted from blood and amplified prior to restriction enzyme digestion revealed the 43-bp fragmentation in each patient and the expected 63-bp fragment in controls, indicating that both patients were homozygous for the A985G mutation (Figure). In the brother and the mother of patient 1, the presence of both the 63-bp and 43-bp fragments indicated heterozygosity.

In a study from the Netherlands, the incidence of the common A985G mutation in 6195 newborn screening cards gave a carrier frequency of 1 in 55 persons. Calculations based on Hardy-Weinberg equilibrium predict a homozygote frequency of 1 in 12000. The odds that a parent might be a homozygote would doubtless be lower because there are no data on the proportion of homozygotes surviving to a childbearing age. In a prospective surveillance report from England, Pollit and Leonard found a disease frequency of 1 in 22,222. There are few data on other ethnic groups. Among gypsies in the submission of the urine sample for organic acid analysis, which led to the diagnosis. Classic MCAD deficiency presents with hypoketotic hypoglycemia. Initial urinalysis indicated a trace amount of ketones, and even a small amount can be misleading. The elevated creatine kinase levels could have served as an alerting marker. Increased levels of creatine kinase and uric acid characterize acute episodes of metabolic imbalance in disorders of fatty acid oxidation. Findings of uric acid status were not available immediately; most clinical chemical analysis panels in children’s hospitals do not include uric acid. The significance of the elevated creatine kinase level is highlighted by the occurrence of the cardiac arrhythmia. This is another way in which disorders of fatty acid oxidation may become life-threatening. In the index patient, all of the clinical manifestations of acute disease resolved promptly, and he was sent home.

The dicarboxylic aciduria of MCAD-deficient individuals is characteristically evident only at times of acute illness. The father’s urine was unrevealing. The pattern includes elevated amounts of adipic (C6), suberic (C8), and sebacic (C10) acids and the glycine conjugates hexanoylglycine and suberylglycine. Phenylpropionylglycine excretion is helpful when present, but in most acutely ill patients in the intensive care unit, this compound is absent because of antibiotic therapy. Both patients were found to be deficient in plasma free carnitine and to have increased amounts of esterified carnitine in the urine. The accumulation of coenzyme A esters of carboxylic acids results in esterification with carnitine and preferential excretion in the urine. The consequent depletion of body stores of free carnitine may further exacerbate the disordered fatty acid oxidation in this disease.

The relative frequency of MCAD deficiency makes the assembly of rigorous epidemiological data possible. In a study from the Netherlands, the incidence of the common A985G mutation in 6195 newborn screening cards gave a carrier frequency of 1 in 55 persons. Calculations based on Hardy-Weinberg equilibrium predict a homozygote frequency of 1 in 12000. The odds that a parent might be a homozygote would doubtless be lower because there are no data on the proportion of homozygotes surviving to a childbearing age. In a prospective surveillance report from England, Pollit and Leonard found a disease frequency of 1 in 22,222. There are few data on other ethnic groups. Among gypsies in
Spain, a carrier frequency for the common mutant allele was 1 in 17, which led to an estimated frequency of homozygosity of 1 in 1156.

Accepted for publication June 12, 2000.

Partial funding from this work came through grant M01 RR008727 from the General Clinical Research Center Program, National Center for Research Resources, National Institutes of Health, Bethesda, Md, and a grant from the Lennox Foundation, Dallas, Tex.

Reprints: William L. Nyhan, MD, PhD, Department of Pediatric/Biochemical Genetics, University of California, San Diego, 9500 Gilman Dr, La Jolla, CA 92039-0830 (e-mail: wnyhan@ucsd.edu).

REFERENCES