Maple Syrup Urine Disease

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This article is a revision of the previous edition article by Dean J. Danner, volume 3, pp 34–39, © 2003, Elsevier Inc.

Introduction

Inborn errors of metabolism (IEM) most often result from rare single-gene defects that follow Mendelian patterns for an autosomal-recessive trait. This implies that both alleles at a single-gene locus harbor a mutation. By definition, parents of the affected individual are obligate carriers for a single mutant allele but they express no phenotype of the disease. Left untreated, many of these disorders present with neurological complications, resulting in impaired function of the brain and central nervous system that can lead to death. By understanding the affected metabolic pathway, treatments have been designed that correct or greatly minimize the neurological pathologies for many of these disorders. Maple syrup urine disease (MSUD) serves as an example of such a single-gene trait in which the use of protein-modified diets (PMDs) has allowed most of the affected individuals to become productive members of society. However, the possibility of a severe neurological crisis remains a constant threat to even the most well-managed MSUD patient. Therefore, individuals with MSUD require greater clinical vigilance than do patients with phenylketonuria, the better-known inborn error of amino acid metabolism. Thus, management of the MSUD patient presents a constant challenge to the dietitian and physician. Orthotopic liver transplantation has come to the forefront of therapy.

History of Maple Syrup Urine Disease

In the mid-1950s, John Menkes was intrigued by a family whose child had severe physical and mental complications and died in infancy. The family history revealed that previous siblings had died in infancy of unknown cause, but all emitted a sweet odor from their body fluids. These findings brought Menkes to the realization that he was observing the consequences of an IEM. He went on to identify the source of the odor, which he likened to maple syrup, as resulting from the accumulation of the branched-chain amino acids (BCAAs) (leucine, isoleucine, and valine) and their transaminated branched-chain ketoacid derivatives (BCKAs) in body tissues and fluids. In 1954, Menkes, Hurst, and Craig described, in the *Journal of Pediatrics*, four patients with neonatal ketoacidosis, progressive neurological demise, and an odor suggestive of maple syrup. Hence the name coined for this inborn error of BCAA metabolism was MSUD. Further studies confirmed the inherited nature of MSUD with transmission following Mendelian patterns of an autosomal-recessive trait placing each pregnancy at risk for MSUD with a 25% chance of having a child with the disease. Heterozygous parents tolerate high-protein diets without clinical consequences. MSUD is known to exist in all ethnic groups, with an overall incidence of approximately 1/120 000 to 1/500 000 live births. The prevalence is higher in certain ethnic groups, such as the Mennonite population of Pennsylvania, and is attributed to a founder effect. The founder effect is based on families in a defined population tracing their origin to a common ancestor, thereby increasing the probability for inheriting the mutant allele.

By the early 1960s, the metabolic block was found to be due to the defective function of the mitochondrial branched-chain α-ketoacid dehydrogenase (BCKD) complex. This enzyme catalyzes the irreversible oxidative decarboxylation of BCKAs, the second step in BCAA catabolism, and commits them to their degradation (Figure 1). Because BCAAs are essential amino acids, they must be supplied by dietary protein or through autophagy of cellular protein. The ability of cells to sense the concentrations of the BCAAs plays important roles in maintaining normal cellular homeostasis. When cellular leucine concentration is maintained within normal limits by dietary intake, autophagy of cellular protein turnover (catabolism) is low. A reduction in cellular leucine concentration prompts endogenous protein degradation to reestablish the norm. The importance of maintaining BCAA concentration is further evidenced in conditions in which their oxidation is increased. Cachexia is associated with many chronic diseases, such as cancer and acquired immunodeficiency syndrome (AIDS), and the normal process of aging is associated with increased BCAA oxidation rates. Because BCKD is the only pathway for BCAA oxidation, increased BCKD activity must be occurring in this condition. When cellular isoleucine concentration is low, the cell cycle is arrested at the G1 stage and results in disorders of the hair, skin, and eyes. In the brain, BCAAs are the major source of nitrogen for formation of glutamate as a neural transmitter. The reason relates to the rapid access BCAAs have in crossing the blood–brain barrier. Likewise, transaminated BCKA, in turn, serves as the nitrogen acceptor in glutamate conversion back to α-ketoglutarate. The rapid formation and breakdown of glutamate is critical for proper synaptic function. Severe neurological consequences occur with

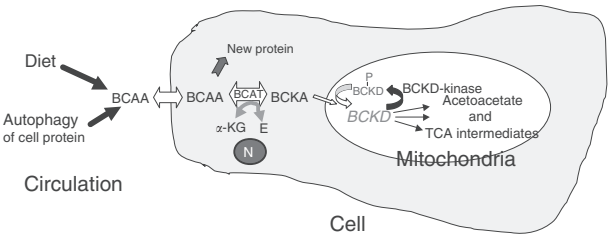

Figure 1 Schematic representation of branched-chain amino acid (BCAA) metabolism. BCAT, branched-chain amino transferase; BCKA, branched-chain ketoacids; BCKD, branched-chain ketoacid dehydrogenase; E, glutamate; α-KG, α-ketoglutarate; N, nucleus; TCA, tricarboxylic acid cycle.

 doi:10.1016/B978-0-12-385157-4.00076-2

increased BCAA as found in untreated MSUD, which can result in death. Thus, the importance of maintaining a cellular balance of BCAA appears to be the main reason for all cells retaining the capacity to metabolize the BCAAs.

Because irreversible loss of the BCAAs can only occur by the action of BCKD, the activity state of BCKD serves to control the cellular concentration of these amino acids. However, BCKD is not functioning in individuals with MSUD; therefore, control of BCAA concentration must rely on dietary intake of protein and supplementation of BCAAs. Enough BCAAs must be supplied so that synthesis of new protein can occur in the rapidly developing newborn. BCAAs must also be supplied to the brain for normal synaptic activity. The fact that PMDs are successful in treating MSUD provided the emphasis to include measurements of plasma leucine in newborn-screening programs used in many areas throughout the world. These programs have proved successful in providing early identification and intervention to minimize complications of the physical and mental development for a number of IEMs in addition to MSUD. Commercially available BCAA-free infant formulas are supplemented with appropriate amounts of BCAA. In older children and adults with MSUD, natural foods low in BCAAs are included in the diet, and MSUD formula is used only to balance the nutritional requirements not met by natural foods. Plasma BCAA is monitored on a regular basis to obtain optimal growth based on height and weight charts. Diet therapy, with the addition of isoleucine, valine, and essential amino acids, remains the primary treatment and must be continued throughout life. Orthotopic liver transplantation is now considered as an effective long-term treatment for classic MSUD. Despite the liver contributing only 9% of total-body BKCD activity, it arrests brain damage, controls BCAA levels, and allows up to 10-fold increase in oral leucine tolerance. Unfortunately, it will not reverse brain damage already present at the time of transplant. Of the 54 patients receiving liver transplantation in the United States from 2004–09, 98% have survived and 96% had engraftment of the liver as of 2012. It is, however, recognized that transplant may only trade one set of complications for another.

The Branched-Chain α-Ketoacid Dehydrogenase Complex

It was not until the late 1970s that the BCKD complex was isolated, purified, and shown to be required for all three BCKAs. Deficiency of BCKD – a multienzyme complex of three catalytic components, E1, decarboxylase composed of 2a and 2b units; E2, transacylase; and E3, a dehydrogenase – causes MSUD. These reactions are diagrammed in (Figure 2). Decarboxylation activity resides in a 2a2b tetramer, which requires thiamine pyrophosphate (vitamin B_1 derivative) as a cofactor. The E1a and E1b subunits are products of two separate genes located on human chromosomes 19 and 6, respectively. Transfer of the resulting branched-chain acyl group to coenzyme A (CoA) occurs by the action of a lipoate-containing acyltransferase (E2), which forms the scaffold core of the complex. The two sulfur residues in lipoate are reduced during the process and must be oxidized by the flavoprotein dihydrolipoamide dehydrogenase (E3). Nicotinamide adenine dinucleotide serves as the ultimate acceptor of the hydrogen molecules. The gene for E2 is found on chromosome 1 and that for E3 on chromosome 7. BCKD activity commits the BCKAs to their catabolic fate, with each CoA ester now following a separate substrate-specific pathway to acetoacetate from leucine, acetoacetate and succinyl-CoA from isoleucine, or succinyl-CoA from valine.

The fact that all mammalian cells with mitochondria have the BCKD complex means that in order to prevent uncontrolled catabolism of the BCAAs, the activity of BCKD must be regulated; two regulatory enzymes, a kinase and phosphorylase, have been identified. The complex-specific kinase (BCKD kinase) resides on chromosome 16. Activity of BCKD kinase phosphorylates two serine residues in the E1a protein, blocking

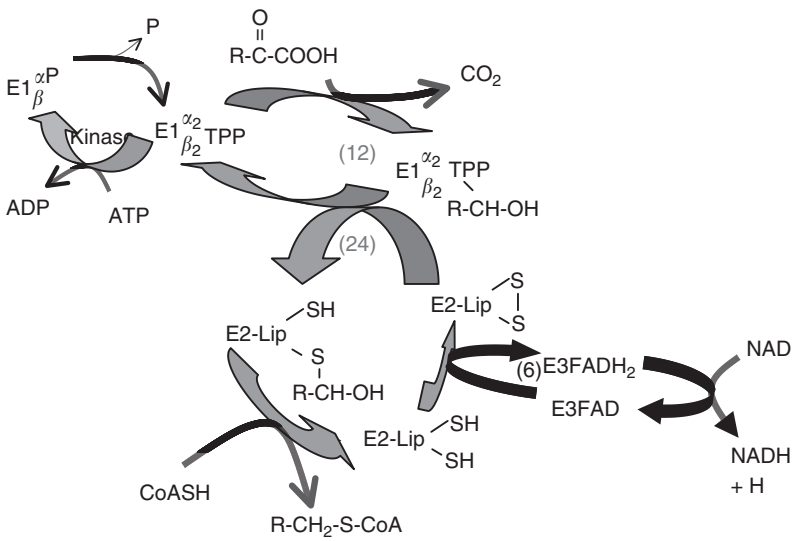

Figure 2 Reactions of BCKD. E1 decarboxylase is an $\alpha_2\beta_2$ tetramer using thiamin PP (TPP) as a cofactor. E2 acyltransferase forms the core and E3 dihydrolipoamide dehydrogenase is a homodimer. The numbers of subunits in the complex are shown in parentheses.

the substrate binding site, thus effectively stopping the overall function of BCKDs. Tissue-specific control of BCKD activity by the kinase is best illustrated by comparing kinase expression in skeletal muscle and liver. The high-protein content of skeletal muscle must be maintained to prevent wasting as found in cachexia. Therefore, BCKD activity must remain low, a feat accomplished by a high expression of the BCKD kinase. In contrast, hepatocytes express low amounts of BCKD kinase, rendering most of the BCKD active for ready catabolism of BCKAs brought to the liver from peripheral tissues such as muscle. Tissues such as the brain and kidney have intermediate levels of kinase expression, but very little is known about the importance of BCKD activity in brain. Hormones and nutritional state will increase or decrease the expression of BCKD kinase with the appropriate concomitant changes in the activity of BCKD. Our current understanding is that regulation of BCKD activity in the various tissues is controlled by BCKD kinase expression. A countering BCKD phosphatase that dephosphorylates E1a with apparent reactivation of the complex has been reported.

Mutations Causing Maple Syrup Urine Disease

In 1985, the first complementary deoxyribonucleic acid (cDNA) clone for E2 of the BCKD was isolated and characterized. Genomic and cDNA clones have been prepared and studied for all components of the BCKD complex. DNA mutations in the genes for E1a, E1b, and E2 have been shown as causative of MSUD. The E3 flavoprotein also functions in three other mitochondrial multienzyme complexes; therefore, mutations in this gene present with a phenotype distinct from MSUD, which is lethal in the neonatal period. Attempted treatments for individuals with mutations in the E3 gene have met with failure. No patients have been described with mutations in the kinase or phosphorylase components of the BCKD complex. All types of DNA alterations have been reported, including genomic deletions, base insertions and deletions, and base substitutions that change the codons or splice site junctions. More than 150 different mutant alleles have been described among the three genes. Amino acid substitutions over the entire coding region of each gene product have been demonstrated as disruptive of enzyme function. Before MSUD is expressed as an autosomal-recessive trait, mutations in one allele for two different genes of the BCKD complex do not result in disease. Synergistic heterozygosity does not appear to be present nor play a role in any patient with MSUD. Both alleles at a single locus must be mutated to present the clinical phenotype. No dominant negative alleles have been identified. Only the 1325 T>A transition in the cDNA for E1a, which results in a Y438N substitution in the protein, is found with any frequency in the general population. This mutation accounts for approximately 100% of the MSUD alleles found in the Mennonite population. However, in most families without consanguinity, each parent holds a different mutant allele. Progeny of these individuals are therefore compound heterozygotes having two different mutant alleles at the single-gene locus. In several ethnic groups, consanguinity is encouraged, thus increasing the probability for expression of rare disorders such as MSUD. Recent data indicate that mutation frequency is approximately the same for each of the three genes.

The identification of mutations within the three genes has allowed a new classification for MSUD as Ia, IIb, II, and III, for mutations in E1a, E1b, E2, and E3, respectively. A clinical definition of varying phenotypes for MSUD is still found in the literature, though this now appears to be a continuum. The 'classic' form presents in the first 4–7 days of life with lethargy, poor tone, poor feeding, and ketoacidosis as a manifestation of the encephalopathy. Seizure and coma with death ensues if not treated. Plasma leucine levels range from 1000 to >5000 μM (normal 100–140 μM) and BCKD activity is 0–2% of control values. The 'intermediate' form presents from infancy to young adulthood with plasma leucine levels between 400 and 2000 μM and BCKD activity less than 25%. The 'intermittent' form becomes evident in childhood to young adulthood usually after a high-protein insult. Plasma leucine concentrations are between 150 and 1000 μM with less than 20% activity of control values. A single case of a thiamine-responsive form was due to the reduced affinity of the mutant of the BCKD thiamine pyrophosphate. These patients have 30–40% of control BCKD activity. Unfortunately, clearly defined genotype–phenotype correlations have not been forthcoming. The most important factor with respect to outcome appears to be early identification and initiation of a PMD.

Many factors contribute to the lack of a clear relationship between genotype and phenotype. First, BCKD is a multiprotein complex, and most individuals with MSUD are compound heterozygotes. This condition allows for extensive variation among components forming the complex, which can result in functional differences. Both the catalytic and protein interactions can be altered. Second, the use of PMDs minimizes or eliminates the full clinical phenotype. Third, some MSUD patients will tolerate a higher protein diet when provided with a pharmacological dose of thiamine. Lastly, the genetic background of all humans, except identical twins, is unique to the individuals and, therefore, will influence the expression of a single-gene trait. Together, these factors, and likely others, make it necessary to tailor the treatment and management of each patient, even for affected siblings within a single family.

Pathophysiology

Newborn infants with MSUD are full-term products of an uncomplicated pregnancy. Newborn-screening programs that test for elevated plasma leucine concentrations should detect infants at risk for classic MSUD. Infants with classic MSUD present in the newborn period with irritability, hypertonicity, poor feeding, and can progress rapidly into seizures, coma, and death. The clinical key to the attending physician is the sweet odor easily detected in the ear wax, and to the parent, the same odor in the urine or sweat. Whether identified by the newborn screen or an alert pediatrician, the infant should be tested with a complete quantitative plasma amino acid profile. The presence of allo-isoleucine in plasma is the most sensitive and specific marker and is considered pathognomonic for the diagnosis. It is present in all classic MSUD patients and many

variants. Confirming diagnosis may be made by an enzyme assay for BCKD activity using isolated peripheral white cells or cultured fibroblasts or lymphoblasts prepared from the patient.

Leucine is the most detrimental among the three BCAAs and can reach concentrations greater than 5000 μM. Normal plasma concentrations are 100–140 μM, and clinical signs can appear with concentrations higher than 400 μM. Children identified by the newborn screen often have no symptoms and only mild elevation of the BCKAs. It is critical that leucine concentrations be reduced as rapidly as possible, which has been accomplished through the use of hemodialysis or peritoneal dialysis or administration of total parenteral nutrition without BCAAs. The choice depends mainly on the stage of decompensation and the age of the child. Care must be taken to monitor the isoleucine and valine levels because additional complications can result if these are not present in adequate amounts. These two BCAAs do not usually reach the concentrations observed for leucine and therefore can rapidly decrease to zero by this treatment. As a consequence of low valine and/or isoleucine, there have been reports of ophthalmic, hair, and dermal lesions. Thus, supplementation with isoleucine and valine are often initiated early in therapy. There is some early evidence that oxidative stress may play a role in the pathophysiology of this disease. Supplementation with L-carnitine or phenylbutyrate may avert these outcomes, but further long-term studies are required before determining efficacy.

With the advent of newborn screening, early recognition of elevated plasma leucine has enabled treatment of the infant with MSUD within the first 3–10 days of life. The use of tandem mass spectrometry in newborn screening, with its speed, sensitivity, and accuracy, has greatly enhanced the early diagnosis.

In the early years after discovery of MSUD, autopsies were done to seek the pathophysiological basis of the disease and identify markers unique to the phenotype. Brain tissue showed status spongiosis, reduced myelination, decreased lipid content, and increased cerebral water content. The naturally occurring animal model, the Poll Hereford cow, has authentically replicated these findings in cow brain. Both white and gray matter are affected, but none of these findings are different from those seen in many other conditions that result in brain edema. These findings do explain the vomiting, lethargy, seizures, coma, and death seen in the untreated newborn.

Identification and treatment with PMDs in the newborn period have resulted in near-normal development in the MSUD patient. However, there remain individuals whose mental development is compromised. The IQ values in the MSUD population range from less than 75 to greater than 130.

Another dilemma remains. Even patients in excellent metabolic control can experience decompensation and a rapid progression to a life-threatening condition. This occurs most often with viral or bacterial infections that trigger cellular breakdown or endogenous protein (catabolism). Immunizations, exercise, or physiological stress may also be precipitants of catabolism. The initial signs are often ataxia, vomiting, and dehydration that bring about the need for hospitalization. After the initial presentation, the condition parallels the untreated newborn. Unfortunately, even the best treatment with rapid decrease in BCAAs may still not be effective and may result in death. In all cases that have been examined by cranial magnetic resonance imaging (MRI), the brain shows extensive edema unique to MSUD and characterized as MSUD edema. It is now thought to represent intramyelinic edema and may be reversible in patients. MR spectrometry has revealed that during exacerbation, elevated lactate and decreased *N*-acetyl aspartate (NAA)/choline ratio indicate mitochondrial dysfunction. Both lactate and NAA were found to normalize by 3 months. Methyl species of BCAA were also elevated and remained visible despite clinical resolution of the decompensation even at 3 months. It is suggested that repeated insults have a cumulative effect, resulting in compromised brain function. Only one longitudinal study covering 20 years and 26 patients followed by electroencephalography (EEG) monitoring has been reported. The results showed no direct correlation between BCAA plasma levels and the EEG pattern.

One possible explanation for pathophysiology that is actively being investigated is the glutamate–GABA neurotransmitter balance in neuron function. BCAAs serve as the major contributor of nitrogen for the formation of glutamate from α-ketoglutarate. In turn, the BCAAs can interfere with this process, and this disturbance may be responsible for the MSUD phenotype.

Future Concerns

Despite the success of newborn-screening programs in identification and the use of PMDs in the management of the individual with MSUD, many challenges remain. Altered homeostasis, protein insults, infection, exercise, injury, and physiological stress still hold the threat of coma and death. Constant vigilance is needed, and the health care provider must be aware of the special needs of these patients. Maternal MSUD is an additional challenge to be faced. Several children have been born to mothers with MSUD; careful postdelivery monitoring is paramount to prevent catabolism and coma. The adult patient with MSUD will face new challenges, many as yet unrecognized. Movement disorders appear to be present in 70% of survivors with MSUD. Long-term follow up of the natural history of MSUD is required to further understand the pathophysiology, treatment, and future complications of the disorder.

See also: Menkes Disease. Micturition. Organic Acid Disorders

Further Reading

Chuang DT and Shih VE (1995) Disorders of branched chain amino acid and keto acid metabolism. In: Scriver CR, Beaudet AL, Sly WS, and Valle D (eds.) *The Metabolic Basis of Inherited Disease*, vol. 1, pp. 1239–1277. New York: McGraw-Hill.

Danner DJ and Doering CB (1998) Human mutations affecting branched chain α-ketoacid dehydrogenase. *Frontiers in Bioscience* 3: 517–524.

Ellerine NP, Herring WJ, Elsas LJ, *et al.* (1993) Thiamine responsive maple syrup urine disease in a patient antigenically missing dihydrolipoamide acyltransferase. *Biochemical Medicine and Metabolic Biology* 49: 363–374.

Fisher CR, Chuang JL, Cox RP, *et al.* (1991) Maple syrup urine disease in Mennonites: Evidence that the Y393N mutation in E1 alpha impedes assembly

of the E1 component of branched-chain α-ketoacid dehydrogenase complex. *Journal of Clinical Investigation* 88: 1034–1037.

Grunewald S, Hinrichs F, and Wendel U (1998) Pregnancy in a woman with maple syrup urine disease. *Journal of Inherited Metabolic Disease* 21: 89–94.

Heindel W, Kugel H, Wendel U, Roth B, and Benz-Bohm G (1995) Proton magnetic resonance spectroscopy reflects metabolic decompensation in maple syrup urine disease. *Pediatric Radiology* 25: 296–299.

Korein J, Sansaricq C, Kalmijn M, Honig J, and Lange B (1994) Maple syrup urine disease: Clinical, EEG, and plasma amino acid correlations with a theoretical mechanism of acute neurotoxicity. *International Journal of Neuroscience* 79: 21–45.

Mazariegos GV, Morton DH, Sindhi R, *et al.* (2012) Liver transplantation for classical MSUD: Long term follow-up in 37 patients and comparative united network for organ sharing. *Journal of Pediatrics* 160: 116–121.

Nellis MM and Danner DJ (2001) Gene preference in maple syrup urine disease. *American Journal of Human Genetics* 68: 232–237.

Peinemann F and Danner DJ (1994) Maple syrup urine disease, 1954 to 1993. *Journal of Inherited Metabolic Disease* 17: 3–15.

Relevant Website

http://www.omim.org

Online Mendelian Inheritance in Man, An Online Catalog of Human Genes and Genetic Disorders.