Abstract

Homocystinuria (HCU) is a rare metabolic disease, and untreated HCU may cause life-threatening venous thrombosis. This case study describes the presentation, diagnosis, and nutritional care of a 17 year-old male with lens subluxation and deep vein thrombosis of the leg. He struggled to achieve good metabolic control (with elevated plasma HCY and recurrent thrombosis as the chief concerns) and required vascular surgery to remove the second clot. With the aid of information provided by the patient and his parents, this report illustrates the medical complications of HCU, the rationale underlying dietary treatment, and recommendations for nutritional management of the late-diagnosed HCU patient. This report serves to increase awareness and understanding of this disease and reinforce the importance of nutritional management in resolving and avoiding future clinical complications of HCU. The implications are important as this patient’s difficulty achieving metabolic control and the consequent recurrent thrombosis are common in other patients with late diagnosed HCU.

Introduction

Metabolic disorders involving sulfur amino acids vary widely from benign clinical phenotypes to severe phenotypes that include profound developmental delay, corneal dislocation, and thromboembolism (Carson & Neill). The most common inborn error in methionine (MET) metabolism is cystathionine-B-synthase (CBS) deficiency, which results in the accumulation of homocysteine (HCY). The accumulation of HCY, as opposed to MET is believed to be the primary source of toxicity in homocystinuria. The clinical symptoms associated with delayed diagnosis are referred to as “classical homocystinuria.”

Homocystinuria (HCU) is rare: the estimated prevalence of CBS deficiency is 1/300,000 births, and in Ireland the estimated prevalence is 1/65,000. HCU is caused by a genetic mutation inherited from both parents (ONIM). In North Carolina newborns screening is completed about
24 hours after birth through a blood sample analysis, which is used to identify close to 30 metabolic disorders. Because screening at birth using a blood spot does not measure HCY concentration, it is not possible to identify all neonates with HCU on the basis of screening for hypermethionemia (Peterschmidt et al.). Further, unlike many inborn errors of amino acid metabolism, no characteristic signs or symptoms are present if HCU remains undiagnosed during infancy. Early detection followed by diagnosis and nutrition intervention have improved the clinical symptoms associated with CBS deficiency; therefore, the metabolic dietitian has the potential to reverse and subsequently prevent adverse clinical outcomes related to untreated HCU through continued dietary management.

The sulfur amino acids (SAAs), methionine and cysteine are linked via the methylation cycle. Methionine is required not only for protein synthesis, but also is converted to S-adenosylmethionine (SAM) by methionine adenosyltransferase. SAM is the primary methyl donor for a wide range of transmethylation reactions, including the synthesis of creatine, phosphatidylcholine, and DNA and RNA intermediates (Mudd et al.). Transsulfuration, the irreversible conversion of HCY and serine to cystathionine and CYS, requires cystathionine-B-synthase and gamma cystathionase, which are both pyridoxine-dependent enzymes (Fowler et al.). Cysteine is required for protein synthesis, and is a methyl donor for the remethylation of HCY. Since transmethylation of HCY to cystathionine is completely or partially blocked, CYS becomes an essential amino acid in CBS deficiency (Mudd et al.). CBS requires pyridoxal 5’ phosphate and heme as cofactors and is activated by SAM. A large number of mutations in different regions of human CBS have been found in patients with homocystinuria, with about half of these mutations resulting in a clinical phenotype that is responsive to pyridoxine (B6), although responsiveness is highly variable among patients (Kraus et al.).
Case presentation

The patient, a 17-year-old male, with HCU was diagnosed in 2011. He came to the UNC Genetics and Metabolism clinic for continued management of his condition. He was a healthy child except for a reportedly “sour” body odor,. The patient began wearing eye glasses at age four, and at age thirteen, lens subluxation in his left eye was diagnosed. Although Marfan syndrome was considered, no further work-up was completed. In November 2011, left leg pain developed and a thrombosis was attributed to a basketball injury. Three months after the clot was removed with a vein bypass graft, a second clot recurred. His plasma HCY level was 197 umol/L and a diagnosis of HCU was made following enzyme assay using skin fibroblast cultures. The patient’s HCY levels did not improve with B6 supplementation (25 mg of B6/day), and he was treated with Hominex-II, a MET-free metabolic formula, betaine, vitamins, and a protein-restricted diet. Surgery to remove the second clot was deferred until plasma HCY levels were less than 20 mg/dL.

From a developmental standpoint the patient was a polite, physically fit, 17-year-old, who was academically motivated and applying to college. His parents reported that his thought and behavior patterns seemed normal, and denied any abnormalities. . His growth and development had followed along the greater than the 95th percentile for height, weight and BMI most of his life. At the time of his initial visit to UNC, he weighed 98.1 kg (> 97%), was 188.5 cm (97%) tall, and had a BMI of 27.6 (90-95%). The following medications were prescribed: a low dose ASA EC 81 mg once daily; betaine 6 gm twice daily Metanx (2 mg methyl B12/ 3 mg L-methylfolate/ 35 mg B6 phosphate) twice daily; and Plavix 75 mg once daily.

Newborn Screening and Barriers to Diagnosis

When CBS deficiency is diagnosed based on clinical symptoms, roughly half of those diagnosed will be considered to have the B6-nonresponsive form. Because the B6-responsive
forms have residual enzyme activity, they are rarely detected with newborn screening and some nonresponsive cases have been missed (Peterschmidt et al.). Reducing the screening cutoff for blood MET concentration from 2 mg/dL to 1 mg/dL improved the detection of infants with B6-nonresponsive CBS deficiency; however, it did not result in increased diagnosis in the B6-responsive population (Peterschmidt et al.). In fact, it has been estimated that screening misses at least one in every five infants with CBS deficiency; others estimate a false negative rate to be even greater (Whiteman et al.). MS/MS screening is expected to improve specificity for detection in disorders associated with methionine metabolism. However, because use of MS/MS screening is not yet widespread, its effect on detection rates of patients with CBS deficiency has not been determined (Chace et al.). One suggestion for screening is to measure elevated HCY in blood or urine. However, no such method is available at this time and affected infants may not accumulate sufficient HCY by 24 to 48 hours of age for detection, because MET levels often rise more slowly than other plasma amino acids associated with inborn errors, such as phenylalanine in PKU (Snyderman et al.).

Diagnosis is determined by analyzing plasma amino acid and HCY concentrations (Mudd et al.). In a standard amino acid panel, the normal MET concentration is less than 35 umol/L (Mudd et al.). HCY exists in plasma in several forms: a thiol homocysteine (HCYH), a disulfide combining two HCY molecules (HCY-HCY), and a group of mixed disulfides, such as CYS-HCY (Picker et al.). The disulfide forms are measured in standard amino acid analyses and labeled as “free HCY.” Therefore, measurement of total plasma HCY is the preferred method of diagnosis for HCU; total HCY in a neonate with classical HCU is typically greater than 50 umol/L or 0.7 mg/dL (McDowell et al.).

Verifying a diagnosis of CBS deficiency is typically done using fibroblast cultures to measure the level of residual enzyme activity. Molecular genetic testing can then be completed
with targeted mutation analysis of the more common alleles, followed by sequence analysis of the entire coding region, if necessary (Picker et al.). Once the diagnosis of CBS deficiency is confirmed, a vitamin B6 challenge is required to determine B6-responsiveness. There is a strong relationship between responsiveness and the presence of residual CBS activity in the liver; however, the amount of activity of CBS can be highly variable in responsive patients (Picker et al.). Once responsiveness is determined, some experts advocate for determining the lowest dose of vitamin B6 necessary for a positive metabolic response because excess levels cause neuropathy, although some symptoms involving coordination have also been reported (Fowler et al.). Overall nutrition status can affect the results of a B6 trial. Folate deficiency may prevent accurate determination of responsiveness, because the remethylation of HCY to MET will be diminished. If this component of remethylation is compromised, it can contribute to the excess concentration of HCY and affect the amount of B6 required to increase CBS activity.

**Outcomes if Untreated**

Age of diagnosis, achievement of metabolic control, and degree of metabolic control play important roles in determining a HCU patient’s clinical outcome. The age of onset and severity of clinical manifestations vary widely among affected individuals. Untreated HCU often results in ectopic lentis and/or severe myopia, thromboembolism, skeletal abnormalities, such as scoliosis, and mental retardation prior to detection (Mudd et al.). Less common features included seizures, psychiatric problems, dystonia, and hypopigmentation (Mudd et al.). The etiology of central nervous system (CNS) abnormalities is unclear, but it has been suggested that such abnormalities may contribute to the neurological difficulties and developmental delay (Mudd et al.). For untreated patients with CBS deficiency, myopia and ocular dislocations are often the first clinical sign of this disorder and can be detected as early as 1 to 2 years of age (Mudd et al.). Ectopic lentis can progress despite tight biochemical control in late-detected
cases, and the later a diagnosis is made and treatment started, the worse the prognosis for normal vision tends to be. Similar to CNS involvement, the etiology of ocular deterioration in patients with CBS deficiency is not well characterized; however, evidence suggests that it is related to elevated plasma HCY levels and low CYS concentrations (Cross et al.). Lens zonules normally have a high CYS content and the deficiency of CYS may affect normal development (Cross et al.). Additionally, HCY inhibits cross-linkage in collagen, which may facilitate the deterioration of zonule fiber and lead to the spherical deformation of the lens (Cross et al.).

Mudd et al. found that 27% of untreated B6-nonresponsive patients develop a clinically detectable thromboembolic event by the age of 15 years. The mechanism for HCY toxicity on vascular connective tissue remains poorly defined. Yap summarized various animal studies that revealed that exposure to SAAs and HCY caused myointimal hyperplasia, accumulation of extracellular matrix and fibrils, and fragmentation of elastic lamellae and the internal elastic membrane. Animal studies conducted by Tsai et al. established that plasma HCY at concentrations similar to patients with CBS deficiency enhances smooth-muscle cell proliferation. High plasma HCY collects in the arterial wall and initiates a cascade of inflammatory mediators and inflammatory transcription factors, further aggravating endothelial dysfunction (Ling et al.). These findings are similar to those observed in patients with atherosclerotic lesions, where endothelial dysfunction increases the risk of thrombosis and adverse cardiovascular events (Yap et al.).

**Treatment of Homocystinuria**

If HCU is diagnosed by newborn screening, it can be treated in a relatively short period of time, and there is a potential for normal growth, development, and life span. Consistent HCY levels within the treatment range of 20 to 40 mg/dL lead to better clinical outcomes than do variable HCY levels (Mudd et al.). Also, continuing the HCU diet into adulthood prevents the
negative consequences of early termination: neurological deterioration, mental abnormalities, physical changes to the vasculature, psychiatric problems, and ocular disturbances (Wilcken et al.). Overall, continuing a protein-restricted diet throughout life promotes optimal physiological function.

Treatment for HCU is primarily dietary; a MET-free metabolic formula and low-protein foods are required throughout life. The metabolic formula contains carbohydrate, fat, the RDA of vitamins/ minerals, and free amino acids. All amino acids but MET are in HCU formulas, and a variety of formulas are available, depending on the patient’s individual preferences. Because CYS becomes an essential amino acid in this disorder, medical foods designed to treat homocystinuria are supplemented with CYS. Most of the reported CYS requirements are increased beyond minimum needs to ensure an adequate supply of CYS to spare MET as a methyl donor and allow for the lowest MET intake for protein synthesis (Acosta and Yannicelli). Apart from diet, supplementation with L-CYS, folic acid, B12, B6, and betaine is beneficial in helping to maintain metabolic control in patients with CBS deficiency (Acosta and Yannicelli). The rationale for supplementation with vitamins, and the betaine medication is to increase the re-methylation cycle of HCY to form MET, and reduce the substrate for MET through dietary protein restriction.

Betaine significantly lowers plasma HCY concentrations in patients with B6-nonresponsive HCU and serves as an effective therapeutic addition to the MET-restricted diet (Wilken et al.). Further, betaine treatment may prevent thromboembolic events by significantly reducing the total HCY concentrations in the plasma (Wilcken et al.). Although betaine treatment causes plasma MET concentrations to increase, MET contributes less to the pathophysiology of CBS deficiency than does HCY itself (Wilcken et al.). After HCU is diagnosed, the use of a MET-free metabolic formula plus betaine and the necessary cofactors
should allow the patient’s HCY levels to return to normal. Then with adequate calories, protein, and nutrients in the diet and normalized plasma HCY levels, normal physical growth and brain development should proceed; however, cognitive delay may be irreversible.

From diagnosis and throughout life, protein and betaine intake must be adjusted frequently to control HCY levels. The HCU diet prohibits protein-rich foods: meat, poultry, fish, eggs, dairy products, nuts, and beans. The diet includes very small amounts of grains and starchy vegetables, more of very low protein foods like fruits and vegetables, fats, and sugars, and modified low-protein foods. Without modified low protein versions of foods like bread, burgers, cheese, and peanut butter, the variety of the HCU diet is severely limited. Diet records for the three days preceding a clinic visit and blood draw are obtained, and growth patterns (for length, weight, and head circumference for children less than – years old) help the dietitian update dietary prescriptions as needed. Blood is collected to measure total protein, prealbumin, free amino acids, and total plasma HCY. Ideally, the diet should keep plasma free HCY levels within a treatment range of 20-40 umol/L (Acosta & Yannicelli). However, infection and fever cause muscle catabolism and high plasma HCY levels. This situation requires increased carbohydrates and fluids. Excessive natural protein can also elevate plasma MET, as can the catabolism resulting from sub-optimal natural protein and/or calorie intake. Achieving stable HCY concentration requires regular blood analyses and food records, a restrictive diet, and periodic clinic visits (every 3 to 6 months for children under 5 years and once yearly for older children and adults if good metabolic control has been achieved) (Acosta & Yannicelli). To meet the challenges of this complex regimen, families must have a positive attitude toward treatment, strong social support, and a belief that HCU is manageable. HCU requires lifelong cooperation of the metabolic team, patient, and family for an optimal outcome.

**Treatment of Case Patient**
During the initial interview, the patient expressed concern about the diet he followed prior to diagnosis, which was primarily high in protein as he was frequently lifting weight and playing sports. It was determined from his dietary records that he had been consuming approximately 100 grams of protein per day, with 75% coming from high protein food sources. Before his visit with UNC Genetics and Metabolism, he had attempted to follow a vegetarian, low-protein dietary prescription from the previous metabolic team, but he admitted to having difficulty with adherence because he felt hungry most days. Further, the taste of Hominex-II, the medical food previously prescribed, made the formula hard to consume regularly. Since medical foods often provide additional satiety to the protein-restricted diet followed by HCU patients, finding an acceptable formula for this patient was a priority for the patient. Starting a medical formula at an older age is often challenging for older patients because of difficulties related to taste, consistency, or deviation from their normal dietary routine when compared to young adults who have consumed the formula from infancy. For HCU, failure with compliance leads to inadequate metabolic control and poor growth and development; therefore, the metabolic team must work diligently to accommodate the patient’s preferences and lifestyle when prescribing a medical food.

Since the patient’s eating habits during this first visit were difficult to ascertain, the dietitian approached the visit as if it were a new diagnosis. Medications were kept consistent, but protein tolerance was determined with a dietary challenge. A dietary prescription was created for the patient to determine his response to protein restriction through measurement of HCY levels after one month. At that point, either protein and/or medications would be adjusted to achieve optimal metabolic control and establish a consistent dietary regimen for the patient to follow. The dietary prescription provided 50 gm of food protein, with the recommendation that only one high protein meal be consumed per day, and the serving size of high protein food items
restricted to 3 oz. per day. He was also switched from Hominex-II to HCU Coolers, an alternative medical food specifically designed for HCU, in order to provide an additional source of protein that tasted better and would increase compliance. The dietitian calculated that the food protein and the MET-free protein provided by 3-4 HCU coolers per day would provide approximately 102 gm total protein (~ 1.0 g/kg body weight) and 600 mg methionine. Finally, the family was given educational brochures and booklets on HCU, and instructions on how to purchase low protein food items such as pasta, rice, breads, and pizzas to add variety to the patient’s diet. The patient was asked to return to the clinic one month later to reassess the diet and check his HCY levels. In the interim, his blood levels from his previous visit became available and revealed a plasma total HCY level greater than 150 umol/L and a MET level of 731 umol/L. The elevated levels were not wholly unexpected given that the patient had not adhered to the low protein diet and metabolic formula previously prescribed, and based upon these results, his dietary protein allowance was decreased and the amount of betaine increased between visits.

When the patient returned in October 2012, his anthropometric measurements had not changed. He continued to complain about pain in his leg, particularly that intensity of pain increased with longer periods of activity. Additionally, his parents expressed the desire to discontinue the Plavix prescription if the HCY levels were lower; however, the family was directed to speak with the patient’s vascular surgeon who had prescribed the medication. His three-day diet record showed that he was eating approximately 40 grams of food protein per day, which provided approximately 500 mg of methionine. Compliance with the metabolic formula had also improved in the interim between visits, as the patient consumed 3-4 HCU coolers daily without difficulty, providing approximately 50 grams of MET-free protein.
The patient continued to be motivated to bring down HCY levels and reported attempts to comply despite the difficult aspects of following a low protein diet. He continued to take his medications regularly, and reported an excellent appetite. The clinic education focused on keeping variety in his diet to avoid tiring of low protein foods, which he had done an excellent job with as high protein foods had been avoided, despite the preference for these items. The patient was encouraged by the metabolic team to continue to incorporate more fruits and vegetables. The possibility that his betaine medication would be increased before further protein restriction was attempted was also discussed at this visit. A dietary prescription prepared at this time provided 92 grams total protein and approximately 500 mg methionine, with 40 grams of protein coming from food. The patient’s betaine prescription had previously been 10 grams per day, and the dosage for other medications and supplements remained the same. A sample grocery list was given to the family to take shopping to increase the variety of low protein food items. A week after his second appointment, his HCY levels drawn at the second visit were 75 umol/L, decreased from 150 umol/L a month prior. As a result, his betaine was increased to 14 grams per day, which is below the upper recommended limit (20 grams/day). The plan was to assess total HCY and plasma amino acids in one month’s time after this adjustment, and the patient was to return to clinic in three months.

Discussion

This patient presented a unique learning opportunity, given the late diagnosis and classical presentation of CBS deficiency. Further, HCU serves as a good example of the nutritional management required to treat metabolic disorders, as it requires substrate restriction, use of alternative pathways, and provision of the amino acids and cofactors that become essential in reducing the offending amino acid. Despite compliance and motivation on his part, and strong support from his family, the patient experienced challenges in reducing HCY levels.
primary goals for the metabolic team were to improve adherence to the HCU diet, achieve metabolic control, and prevent future clot formation. Achievement of nutritional goals would be a reduction in HCY levels to within a therapeutic range, optimally below 20 umol/L. The challenge for the dietitian was trying to determine how much protein could be restricted without limiting intake to the point that the diet was unsatisfying and difficult to follow. When HCY levels were higher than 20 to 40 umol/L, the desired range at follow-up, the clinical team decided to first adjusting the betaine dosage before reducing protein intake to less than 40 grams per day. The fact that the patient was an active teenager was also considered when determining whether further protein restriction would be beneficial given his complaints of hunger and the limited variety of foods appropriate for him.

Early detection and adequate treatment of patients with CBS deficiency clearly results in a more positive clinical outcome than that seen in untreated patients. However, given the limited reliability of newborn screening, and the clinical problems that occur when HCU goes undetected, siblings of known HCU patients should be tested during early childhood to minimize the risk of developing the symptoms associated with HCU. Effective treatment of both vitamin B6-responsive and nonresponsive patients has significantly reduced the cardiovascular risk associated with classical HCU. However, vascular events have been reported, even in treated B6-responsive patients who maintain long-term plasma HCY concentrations below 20 umol/L (Wilcken et al.). Thus, frequent monitoring in a cardiovascular clinic is recommended for all patients with classical HCU. Similarly, patients who develop ectopic lentis should be evaluated for homocystinuria as well as Marfan Syndrome.

A monitoring schedule should be established for all patients with CBS deficiency, regardless of their B6-responsiveness (Acosta & Yannicelli). Growth parameters, laboratory analyses, and general nutrition status, and nutrient intake should be assessed as part of
monitoring. Laboratory analyses should always include plasma concentrations of MET, CYS, and free and total HCY (Acosta & Yannicelli). Deficient intakes of MET and CYS, total protein, medical food, and/or total energy can all adversely affect growth, and each should be evaluated monthly at first and then yearly once metabolic control is achieved. Biochemical goals for long-term treatment of patients with CBS include maintaining plasma HCY concentrations within or near the normal range (20 to 40 umol/L), free plasma HCY concentrations below 20 umol/L, and total HCY concentrations below 50 umol/L (Acosta & Yannicelli). In addition to routine nutrition and biochemical monitoring, a team-based approach often results in more successful clinical outcomes. The delivery of individualized dietary and medical treatment, in addition to both the physician and dietitian reinforcing one another’s respective discipline, leads to better understanding and continuity of care in the HCU setting. Patients are engaged, receptive, and reported that they felt their questions were addressed in an efficient manner, making the challenging aspects of dietary adherence more manageable. When patients are able to adhere without difficulty, their quality of life with an inborn error in metabolism can be much improved.
References


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