1. Dietary Treatment of Amino Acids Inborn Errors of Metabolism

A MacDonald, A Daly, A Chakrapani

Introduction

Dietary treatment is essential for many amino acid disorders, particularly phenylketonuria (PKU), homocystinuria, maple syrup urine disease (MSUD) and tyrosinaemia type I, II and III. Diet may be the sole form of therapy or used in combination with other treatments e.g. betaine in homocystinuria or NTBC in tyrosinaemia type I. Although simple in principle, diet therapy is complex for patients and carers, difficult to administer, and highly restrictive. It is important to acknowledge that diet therapy can significantly impact on family lifestyle, demands self-discipline, good organisational skills, increases parental anxiety, exacerbates family conflict and may cause embarrassment and even bullying in school age children. Severe dietary restriction is usually less acceptable to patients diagnosed after infancy.

The goals of dietary management in amino acid disorders are four-fold:

1. Prevention of excessive accumulation of substrate amino acids by strict control of natural protein intake. This is in combination with the administration of an appropriate protein substitute.
2. Achievement of normal growth and nutritional status.
4. Provision of a diet that is palatable, flexible and compatible with a modern day lifestyle.

Although the principles of diet therapy are similar for all these amino acid disorders, their prevalence, presentation, symptoms, and complications are quite different.

PKU

PKU is usually caused by a deficiency of the hepatic enzyme, phenylalanine hydroxylase (phenylalanine 4-mono-oxygenase, EC 1.14.16.1). This is a mixed function oxidase which catalyses the hydroxylation of phenylalanine...
to tyrosine, the rate limiting step in phenylalanine catabolism. Deficiency of this enzyme leads to an accumulation of phenylalanine, resulting in hyperphenylalaninaemia and abnormalities in the metabolism of many compounds derived from aromatic amino acids. Phenylalanine hydroxylase deficiency is heterogeneous with a continuum of metabolic phenotypes ranging from classical PKU, characterised by blood phenylalanine of 20 times the normal rate to mild hyperphenylalaninaemia with blood phenylalanine levels 3-5 times higher than normal. There are over 400 mutations with good genotype and phenotype correlation. Overall, the prevalence amongst Caucasians is approximately 1 in 10,000, corresponding to a carrier frequency of about 1 in 50. In Asian populations, PKU is rare and prevalence figures range from approximately 1:16,500 in China to 1:120,000 in Japan.

Untreated PKU leads to mental retardation, hyperactive behaviour with autistic features, and seizures. If dietary treatment is started within the first 3 weeks of life, irreversible mental retardation is prevented. However, even when patients with PKU treated continuously and carefully, following neonatal diagnosis, mildly depressed IQ is common in treated PKU, and depressive mood, anxiety, and social isolation have been reported. Even so, most early treated children who have started diet by 4 weeks of age fall within the broad normal range of general ability and there is strong evidence to indicate that outcome is closely related to the quality of early blood phenylalanine control.

A low phenylalanine diet is recommended for life. At diagnosis, dietary treatment is widely advocated when blood phenylalanine concentrations are consistently over 600 µmol/l. There is less agreement about the use of dietary treatment when presenting phenylalanine concentrations persist between 360-600 µmol/l. Although dietary treatment has been the main stay of therapy since the mid 1950's, the enzyme phenylalanine ammonia lyase may be used as an alternative therapy in the future. It converts phenylalanine to a nontoxic derivative and in a PKU rat model has been shown to lower plasma and tissue phenylalanine more effectively than diet.

**MSUD**

MSUD is caused by a deficiency in activity of the branched chain α-keto acid dehydrogenase (BCKD) complex. This metabolic block results in the accumulation of the branch chain amino acids leucine, isoleucine, and valine and the corresponding branched chain keto-acids. It was described in 1954 and dietary treatment was first used in 1959. It is named after the sweet, malt, caramel like odour produced by elevated concentrations of 2-oxo-3-methyl-N-valeric acid. There are four forms, which differ in the age of onset, biochemical findings, and responsiveness to thiamin, a cofactor for the BCKD complex. The genetic heterogeneity is explained by the various mutations that occur in the E1 alpha, E1 beta, E2, and E3 loci of the BCKD complex. Treatment involves both long term dietary management and aggressive intervention during acute metabolic decompensation. At any age, an emergency regimen must be adhered to during intercurrent infections.

Classic MSUD has a neonatal onset, with poor feeding, irritability, lethargy and encephalopathy and is the most common and severe form. Any delay in diagnosis or treatment may result in permanent neurological damage and early death. Toxic metabolites may need to be removed by haemodialysis or haemofiltration. Diagnosis before 10 days is imperative. An intermediate form presents at any age, infancy to adulthood, with failure to thrive, neurological features, and ketoacidosis. An intermittent form manifests episodic ataxia and ketoacidosis, often associated with increased protein consumption or intercurrent illness. Children are normal between attacks but there is still a chance of permanent neurological damage from acute episodes. The fourth type is a thiamin responsive form.

The concentrations of branch chain amino acids, particularly leucine are greatly increased in the plasma and urine. The presence of alloisoleucine is diagnostic of MSUD. The worldwide frequency is only approximately 1 in 185,000, but is common among the Mennonites of North America where the incidence is 1 in 176. It is found in all racial types. Long-term outcome is variable but the average intellectual ability is below normal. Approximately one third of classic MSUD patients have IQ scores greater than 90 and a further one third have IQ scores between 70 and 90.

**Homocystinuria**

Cystathione β-synthase (CBS) deficiency is the most frequently encountered cause of homocystinuria (HCU). Homocysteine, methionine and other sulphur containing metabolites accumulate in the body or are excreted in the urine. Plasma cystine is usually low. It was first described in 1962. The worldwide incidence of HCU is approximately 1 in 335,000 but varies from 1:65,000 (Ireland) to 1:900,000 (Japan). Two clinical forms of CBS deficiency have been described on their basis to respond to pyridoxine treatment (pyridoxine responsive and non pyridoxine responsive HCU). There is considerable genetic heterogeneity.

The most frequent complications of the disease are divided into four areas: 1) dislocation of the optic lens,
myopia and glaucoma; 2) osteoporosis, scoliosis, thinning and lengthening of the long bones; 3) learning difficulties, developmental delay affecting approximately 60% of patients, psychiatric problems, EEG abnormalities and epilepsy; and 4) thromboembolism affecting large and small arteries and veins are the most common clinical features.\textsuperscript{15} Thrombosis is a frequent cause of death. Patients vary widely in the extent to which they manifest these abnormalities. Accumulation of homocysteine probably plays an important role in the development of many of these complications.

In non-pyridoxine responsive patients, early diagnosis together with a life-long low methionine diet can be highly successful in preventing complications. Strategies for treatment of CBS deficiency include: 1) pyridoxine and folic acid supplementation in pyridoxine responsive HCU; 2) reducing the methionine substrate load and supplementing the diet with cysteine; and 3) betaine supplementation as a homocysteine lowering agent. Betaine may be effective in vitamin B6 nonresponsive patients in whom dietary management is unsatisfactory but patient compliance may be poor.\textsuperscript{16} It acts as a methyl donor for the remethylation of homocysteine to methionine. Its use is often associated with an increase in plasma methionine, but not always.\textsuperscript{17}

**Tyrosinaemia**

**Type I:** caused by deficiency of fumarylacetoacetate hydroxylase, the last enzyme in tyrosine degradation. The fumarylacetoacetase gene is located at 15q 23-25. The condition is clinically heterogeneous, more than 30 different mutations have been identified and it presents either as an acute or chronic form. Plasma tyrosine is elevated in most patients and alpha-fetoprotein may reach high concentrations. Symptoms are variable and include acute liver failure, cirrhosis, hepatocellular carcinoma, renal Fanconi syndrome, glomerulosclerosis, and neurological crisis resembling acute intermittent porphyria. Vitamin D resistant rickets develops due to severely impaired renal function. Elevated levels of succinylacetone in plasma or urine are diagnostic for this function.

NTBC (2-[2-nitro-4-trifluoro-methylbenzoyl]-1, 3-cyclohexanedione), a potent inhibitor of 4-hydroxyphenylpyruvate dioxygenase (4HPPD) prevents tyrosine degradation and production of succinylacetone. Since it was first used in the treatment of this disorder in 1991, patients have not developed acute hepatic or neurological crisis, but current data do not allow conclusions on the long-term risk of hepatocellular in NTBC treated patients.\textsuperscript{18}

A low tyrosine diet was first used in 1964. Most patients show only a partial response to dietary restriction of tyrosine and phenylalanine and now diet is used in combination with NTBC treatment.

**Type II:** caused by L-tyrosine aminotransferase deficiency, it was first described in 1983. It causes corneal lesions and keratitis and blisterous lesions on the soles and palms but the onset of symptoms may vary. Furthermore, approximately 50% of patients suffer from neurological complications including fine co-ordination and language deficits, microcephaly, self-mutilation and severe developmental delay. Plasma tyrosine concentrations are elevated but is successfully treated with a low tyrosine diet. There is no consensus on optimal blood levels of tyrosine or what age diet should be started.

**Type III:** a rare disorder caused by deficiency of 4HPPD, the second enzyme in the catabolic pathway of tyrosine. Plasma tyrosine is highly elevated. Ataxia, convulsions, and a cerebral atrophy have been reported. It may be asymptomatic or is associated with neurological symptoms. All patients reported so far have normal liver and renal function and none have skin or eye abnormalities. It is not clear how beneficial dietary tyrosine restriction is but it is thought it may be important, particularly in infancy.\textsuperscript{19}

**Dietary Management**

The principles of dietary management for all these amino acid disorders are similar. There are five key elements to dietary management.

1. Restriction of substrate amino acids to maintain blood phenylalanine concentrations within desirable reference ranges. High protein foods such a meat, fish, eggs and cheese are not permitted in the diet.
2. Daily allocation of dietary substrate amino acids from measured quantities of moderate protein containing foods to provide minimum requirements. These are given in the form of an exchange system, whereby one food can be exchanged or substituted for another of equivalent content.
4. Maintenance of a normal energy intake by encouraging liberal use of foods naturally low in protein and specially manufactured low protein foods such as bread, pasta and biscuits. These are called ‘free’ foods.
5. Provision of all vitamins and minerals to meet dietary requirements. These can either be given together in the protein substitute or as separate modules.
Restrictions of Substrate Amino Acids

The tolerance of substrate amino acids is variable and dependent on:
- Severity of disorder.
- Target plasma amino acids range.
- Compliance with protein substitute.
- Energy intake.
- Age and weight of the child.

Requirements per kg body weight for amino acids are highest in early infancy and decreases with increasing age. Total daily requirements change very little after initial stabilisation of diet. Acosta et al reported in PKU that to maintain blood phenylalanine concentrations between 60-324 µmol/l in infants, mean phenylalanine requirements were 0-3 months: 55 mg/kg/day; 4-6 months: 36 mg/kg/day; 7-9 months: 31 mg/kg/day and 9-12 months: 27 mg/kg/day. Suggested leucine requirements in MSUD are 100-120 mg/kg/day in 2-3 month old infants, reducing to 40-50 mg/kg/day in 1-year-old children. In tyrosinaemia type I, natural protein requirements varies from a peak of 1.8-2.4 g/kg/day at 5 months of age to 1 g/kg/day in later infancy.21 Average daily tolerance of substrate amino acids is given in Table 1.

The substrate amino acid is given in the form of a daily allowance via a food exchange system. This does not take into account the small quantities of protein obtained from the very low protein foods allowed without restriction. Foods such as meat, fish, eggs, cheese, milk, nuts, ordinary bread, biscuits, cakes and chocolates are avoided because these are too high in natural protein. The exchange foods are made up from moderate protein foods like potatoes, peas, sweetcorn, rice and breakfast cereals. Examples of exchange systems from the different amino acid disorders are given in Table 2. Ideally amino acid exchanges should be spread evenly throughout the day so that a load of dietary substrate amino acids is not given at any one time.

Table 1  Daily tolerance of substrate amino acids

<table>
<thead>
<tr>
<th>Amino acid disorder</th>
<th>Amino acid tolerance</th>
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<tbody>
<tr>
<td>PKU</td>
<td>200-400 mg/daily phenylalanine</td>
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<tr>
<td>MSUD</td>
<td>400-600 mg/daily leucine</td>
</tr>
<tr>
<td>Homocystinuria</td>
<td>160-900 mg/daily methionine</td>
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<tr>
<td>Tyrosinaemia Type I</td>
<td>1 g/kg/day of protein in late infancy</td>
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</tbody>
</table>

NB. This is the amount of amino acid from exchange foods. It does not take into consideration the small, additional quantities consumed from low protein free foods.

Table 2  Examples of amino acid/protein exchange systems for amino acid disorders

<table>
<thead>
<tr>
<th>Amino acid disorder</th>
<th>Exchange system</th>
<th>Examples of exchanges</th>
</tr>
</thead>
<tbody>
<tr>
<td>PKU</td>
<td>50 mg phenylalanine</td>
<td>30 ml cow's milk</td>
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<tr>
<td></td>
<td></td>
<td>80 g potato</td>
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<tr>
<td></td>
<td></td>
<td>45 g chips</td>
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<tr>
<td></td>
<td></td>
<td>20 g baked beans</td>
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<tr>
<td></td>
<td></td>
<td>25 g peas</td>
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<tr>
<td></td>
<td></td>
<td>45 g boiled rice</td>
</tr>
<tr>
<td>MSUD</td>
<td>50 mg leucine</td>
<td>15 ml cow's milk</td>
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<tr>
<td></td>
<td></td>
<td>60 g potato</td>
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<tr>
<td></td>
<td></td>
<td>35 g chips</td>
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<tr>
<td></td>
<td></td>
<td>15 g baked beans</td>
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<tr>
<td></td>
<td></td>
<td>15 g peas</td>
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<tr>
<td></td>
<td></td>
<td>25 g boiled rice</td>
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<tr>
<td>Homocystinuria</td>
<td>20 mg methionine</td>
<td>20 ml cow's milk</td>
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<tr>
<td></td>
<td></td>
<td>85 g potato</td>
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<tr>
<td></td>
<td></td>
<td>35 g chips</td>
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<tr>
<td></td>
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<td>35 g baked beans</td>
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<tr>
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<td></td>
<td>45 g peas</td>
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<tr>
<td></td>
<td></td>
<td>40 g boiled rice</td>
</tr>
<tr>
<td>Tyrosinaemia Type I</td>
<td>1 g protein</td>
<td>30 ml cow's milk</td>
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<tr>
<td></td>
<td></td>
<td>55 g potato</td>
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<td>15 g peas</td>
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<td></td>
<td></td>
<td>45 g boiled rice</td>
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Protein Substitute

These are essential in the treatment of amino acids disorders for several reasons:
- Provide sufficient amino acids for normal growth.
- Helps suppress plasma precursor amino acids concentrations.
- May supply a source of energy.
- May also supply a source of vitamins and minerals.

In the UK, protein substitutes are supplied in generous quantities as L-amino acids may be inefficiently utilised. Guidelines for the total protein requirements per kg body weight (i.e. protein from amino acid exchanges and protein substitute) are given in Table 3. They should be given evenly during the day. The effect of timing of protein substitute has been extensively studied; and it is better to give protein substitute in small frequent doses, three to four times daily spread evenly throughout the day.22 Theoretically it is better given with some of the substrate amino acid allowance. Added carbohydrate to the protein substitute may increase net protein synthesis.23 There is evidence that infrequent administration of large doses of protein substitute increases nitrogen excretion as well as oxidative utilisation of amino acids; so this practice is not advocated.

The protein substitutes are available in a variety of different presentations and the range of novel presentations is increasing. They include: L-amino acids with added carbohydrate, +/- fat, vitamins and minerals designed to be administered as a drink or gel; powdered protein substitutes which contain L-amino acids only; modular protein substitutes (presented in 3 different formats i.e. tablets, powder, and bar); and amino acid tablets. The latter two have been developed for PKU only.

Administration of Protein Substitute

Protein substitutes can be taken as a drink or paste. The traditional method of administering protein substitute is in the form of a drink. However, when dissolved in water it is bitter tasting and produces a hyperosmolar solution. If diluted with less water than recommended by the manufacturers, an additional drink of water should be taken at the same time.

Alternatively, protein substitute can be given as a paste or gel. A small amount of water, or concentrated fruit juice is added to each dose of protein substitute to make a thick paste. An additional drink of water should be given with each dose to dilute this hyperosmolar mixture. Protein substitutes are now being developed in a gel format for all amino acid disorders. Giving protein substitute as a paste appears an acceptable method of protein substitute delivery to young children.

Other Considerations

**PKU:** The inability to convert phenylalanine into tyrosine transforms tyrosine from a non-essential to an essential amino acid. Consequently, patients are dependent on a dietary source of tyrosine and so UK protein substitutes are supplemented with tyrosine to supply tyrosine requirements.

**Homocystinuria:** Cystine is usually deficient due to the metabolic block, so an additional source is needed. The UK methionine free protein substitutes are supplemented with cystine.

**MSUD:** Sometimes, additional, small amounts of valine and isoleucine must be given as the tolerance to leucine is lower than valine and isoleucine.

Low Protein Free Foods

These are essential and encouraged liberally in amino acid disorders. Their benefits include:
- Provide a good source of calories.
- Ensure normal growth.
- Enhance protein synthesis.
- Minimise catabolism. Long periods of fasting should be avoided – particularly in conditions such as MSUD.

<table>
<thead>
<tr>
<th>Age (year)</th>
<th>Total protein (g/kg/body weight)</th>
</tr>
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<tbody>
<tr>
<td>0-2</td>
<td>3.0</td>
</tr>
<tr>
<td>3-5</td>
<td>2.5</td>
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<tr>
<td>6-10</td>
<td>2.0</td>
</tr>
<tr>
<td>11-14</td>
<td>1.5</td>
</tr>
<tr>
<td>&gt;14</td>
<td>1.0</td>
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</tbody>
</table>
There are a number of foods naturally low in protein which are given freely in the UK low amino acid diets.

- **Fruits and vegetables**: many fruits and vegetables.
- **Fats**: butter, margarine, lard and vegetable oils.
- **Sugars and starches**: cornflour, custard powder, sago, tapioca, sugar, glucose, jam, honey, marmalade, golden syrup, treacle and sweets <0.3 g protein/100 g.
- **Miscellaneous**: Vegetarian jelly, agar-agar, salt, pepper, herbs, spices and vinegar. Tomato and brown sauce. Baking power, bicarbonate of soda and cream of tartar. Food essences and colouring.
- **Drinks**: Aspartame-free squash, lemonade, Coca-Cola and fruit juice. Tea, coffee, tonic water, soda water and mineral water.
- **Low protein special foods**: a selection of low protein breads, flour mixes, cake mixes, pizza bases, pasta, biscuits, egg replacers, milks, cheese, cheese sauses and chocolate are available in the UK. Most are available free of charge on the UK government prescription system so families do not need to pay for these. These are an important source of calories in the diet.

It is important to introduce a variety of low protein 'free' foods into the diet as early as possible to give variety and adequate energy to meet estimated average requirements. Families need simple and practical ideas on how to incorporate 'free' foods into the diet effectively. They need help in interpreting food labels so they can fully utilise all free foods on the market. Low protein recipe books, cookery workshops, cookery demonstrations can all help parents prepare 'free' suitable meals and dishes.

**Vitamins and Mineral Supplementation**

Comprehensive vitamin and mineral supplementation is added to some protein substitutes and providing adequate quantities of protein substitute are taken no additional supplementation is necessary. Other protein substitutes contain no vitamins and minerals so complete supplementation is necessary. Reports of vitamin and mineral deficiency is common. They include selenium deficiency, low ferritin concentrations, low vitamin B12 concentrations and decreased bone mineral density. Vitamin and mineral deficiency is due to four main reasons:

1. Failure of a protein substitute or vitamin and mineral supplement to contain a specific micronutrient e.g. selenium deficiency has been commonly associated with lack of added selenium to the supplement; 2) low bioavailability of micronutrients added to supplements; 3) non-compliance with the protein substitute with added vitamin and mineral supplement or separate vitamin and mineral supplement; and 4) excessive use of emergency regimen without added vitamins and minerals.

**Essential Fatty Acid Status**

The long chain polyunsaturated fatty acid status of patients on low protein diets with inherited metabolic disease has been the subject of much debate, particularly in PKU. Evidence suggests that children on low protein diets have reduced concentrations of arachidonic acid and docosahexanoic acid in plasma and membrane phospholipids compared to controls and may require supplementation. A strict low phenylalanine diet is high in linoleic acid, but low in alpha-linolenic acid, arachidonic acid (AA) and devoid of any sources of eicosapentanoic acid (EPA) and docosahexanoic acid (DHA). In amino acid disorders, the extent to which long chain polyunsaturated fatty acids (LCU's) can be synthesised from the parent fatty acids (linoleic acid and alpha-linolenic acid) is debatable and some would argue that a direct source should be provided. In the UK, new protein substitutes are being developed for PKU and other amino acid disorders with added essential fatty acids. In addition, long chain polyunsaturated fatty acid capsules have improved DHA concentrations and visual function in children with PKU.

**Illness Management**

During illness protein catabolism will greatly increase production of substrate amino acids. High plasma leucine concentrations in MSUD could cause rapid neurological deterioration. All parents should have a regularly updated emergency regimen, and should be carefully instructed on appropriate action during intercurrent infections.

The following measures are recommended:

- Reduce or stop intake of substrate amino acids.
- Two-three hourly administration day and night of high calorie carbohydrate drinks.
- Maintain protein substitute intake, particularly in MSUD. If a child cannot drink the recommended volume of high carbohydrate drinks or protein substitute, administration via a nasogastric tube should be considered.
- Regular monitoring of plasma amino acids.

An emergency regimen should only be used in the short term. Prolonged and frequent use of an emergency regimen for illness may lead to nutritional deficiencies.
Breast Feeding in Amino Acid Disorders

The successful use of breast-feeding in amino acid disorders has now been increasingly reported in the form of case studies. In 1981, it was first reported in PKU and more recently, Touati reported four infants with MSUD who were successfully breast fed. Breast feeding is based on the principle of giving a measured volume of infant protein substitute before breast feeds, so reducing stimulation and protection of breast milk, thus breast milk and substitute amino acid intake is reduced.

Plasma amino acid concentrations are used to determine how much infant formula to give. If substrate amino acid concentrations are high, more protein substitute is given so less breast milk is taken. If substrate amino acid concentrations are low, less infant protein substitute is given so more breast milk is taken. Motzfeldt has recently reported successfully breast feeding seventy-four out of eighty-three babies born with PKU since 1979. It took a mean of eight days to normalise phenylalanine concentrations. Breast feeding duration was anything from four weeks to sixteen months. The growth was within normal parameters.

Monitoring

Regular monitoring of plasma substrate amino acids is recommended for all amino acid disorders. An UK MRC Working Group published a set of guidelines that included monitoring of blood phenylalanine concentrations in PKU but target ranges are set by UK clinics for all the other disorders. Blood samples are taken at a standard time each day, preferably before the first dose of protein substitute in the morning when blood substrate amino acid concentrations are usually highest.

With the exception of homocystinuria, parents are taught how to collect heal or thumb prick blood samples at home by a specialist nurse. The parents then post the blood sample to the hospital. The dietitian then contacts the parents with the results to discuss their interpretation and instruct on any dietary changes.

Conclusions

Early diagnosis and subsequent metabolic control are important for successful outcome in all amino acid disorders. Diet therapy in amino acid disorders requires close supervision by an experienced dietitian, with the close support of a metabolic team. Parental and child understanding and their co-operation are also paramount in achieving satisfactory metabolic control. Improvements in nutritional therapy, dietary foods, monitoring, and management of acute infections have helped contribute to improved outcome in all these conditions.

References


