Clinical Genetics 1989: 36: 333–334

Impact of medical genetics concerning phenylketonuria: accomplishments, status and practical future possibilities

FLEMMING GÜTTLER
The John F. Kennedy Institute, Glostrup, Denmark

Millions of newborn infants are screened for phenylketonuria (PKU) to prevent the inherited metabolic error by dietary treatment. For some PKU patients a relaxed low-phenylalanine diet will be lifelong. PKU-women must return to a strict low-phenylalanine diet before conception and during pregnancy to insure the delivery of a normal child. Two mutations that account for 60% of the PKU genes in Denmark are established and oligonucleotide probing enables carrier detection and genetic counselling. Primary hepatocytes can be successfully cultured and transformed with phenylalanine hydroxylase cDNA and somatic gene therapy of PKU may be a future possibility.

Key words: dietary treatment; gene therapy: mutations; PKU screening.

Phenylketonuria (PKU) is caused by an autosomal recessive genetic deficiency of hepatic phenylalanine hydroxylase (PAH) with an average incidence of 1 in 10 000.

Today millions of newborn infants all over the world are screened for phenylketonuria (PKU) and many thousand early-treated children and young persons are happy that they are bright and completely normal. Research workers from many disciplines have cooperated to elucidate the biochemical and genetic background of PKU and to prevent this inherited metabolic error by dietary treatment before clinical symptoms appear.

PKU was first discovered and described in 1934 by the Norwegian physician and biochemist Asbjørn Følling, and during the past five decades the disease has been regarded as a model of an inherited disorder of amino acid metabolism. Untreated, the disease causes neuropsychiatric symptoms including intellectual deterioration, seizures, severe hyperactivity, self-injury and self-mutilation. Subtle reversible changes in cerebral function, including impaired neuropsychological and behavioural functions, may occur in young adults with PKU in whom the dietary treatment has been discontinued. So, for some PKU patients a relaxed low-phenylalanine diet will be lifelong. The neuropsychiatric prognosis in the long term is unknown. Children born to PKU-women will suffer from mental retardation and microcephaly unless the women return to the rigours of a strict low-phenylalanine diet before conception and during pregnancy.

The human PAH gene spans over 95 Kbp, comprising 13 exons and 8 polymorphic binding sites for restriction enzymes. Restriction fragment length polymorphisms (RFLPs) detected by means of a cDNA probe of PAH enabled mutant genes in Danish PKU-families to be traced for the first time. Twelve distinct RFLP haplotypes

were observed among Danish PKU-families, and 90% of the PKU alleles are confined to four haplotypes (nos. 1–4). Different combinations of the mutant RFLP haplotypes contribute to clinical diversity of PKU, and patients with mutant haplotype combinations 2/2, 2/3, and 3/3 will follow a severe course of the disease. The mutations associated with haplotypes 2 and 3 have been established, and these mutations account for 60% of the PKU genes in Denmark. Specific oligonucleotide probes for mutant haplotype 2 and 3 alleles showed that these mutations are also present in England, Scotland, Switzerland and Italy.

Specific amplification of a 245 bp region containing exon 12 and the two mutation sites can be achieved by direct analysis of DNA extracted from dried blood spots and amplified by the polymerase chain reaction (PCR) followed by hybridization with ^{32}P-labelled mutant specific oligonucleotide probes. This enables carrier detection and genetic counselling before the first PKU-child has been born, as well as determination of the severity of the disease in hyperphenylalaninemic neonates.

Human PAH activity is expressed in mouse hepatoma cells after retrovirus-mediated gene transfer of the human PAH cDNA. Primary hepatocytes can be successfully cultured and transformed with PAH cDNA using retroviral vectors, which might be useful to introduce a functional PAH gene into liver cells, and hence for somatic gene therapy of PKU.

Address:

Dr. Flemming Güttler
John F. Kennedy Institute
Gl. Landevej 7
2600 Glostrup
Denmark