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MUTATION IN BRIEF

A 3-Base Pair In-Frame Deletion in Exon 8 (delGlu272/273) of the Ornithine Transcarbamylase Gene in Late-Onset Hyperammonemic Coma

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INTRODUCTION

Ornithine transcarbamylase (OTC, MIM 311250) is a mitochondrial enzyme of ureagenesis that catalyses the synthesis of citrulline from ornithine and carbamyl phosphate. OTC deficiency is the most common cause of urea cycle disorder in humans and has a partially dominant X-linked inheritance (Brusilow and Horwich, 1989). The OTC gene maps to chromosome Xp21.1 and contains 10 exons encoding a 354-amino acid protein (Hata et al., 1988). Most patients diagnosed are males with acute neonatal hyperammonemic coma and die shortly after birth with unmeasurable OTC activity in the liver. It is probable that an equal or even larger number of males with late-onset milder disease exists, but a proportion may not be diagnosed (Tuchman et al., 1989). Here, we report on a 3-base pair (bp) in-frame deletion of OTC exon 8 (delGlu272/273) in a male patient with late-onset hyperammonemic coma.

PATIENT AND METHODS

The patient is a boy who had feeding difficulties during the first months of life and developed a severe hyperammonemic coma at 10 months of age. His liver OTC activity averaged 5% of controls. At 14 years of age, his blood ammonia is normal, and he is doing well on a low-protein diet (30 g/day). The results of his mother's and sister's protein-loading tests were negative, but several maternal relatives of both sexes have unexplained mental retardation.

DNA was extracted from polymorphonuclear leukocytes and OTC exons were amplified as described (Gilbert-Dussardier et al., 1995). Polymerase chain reaction (PCR) amplification of exon 8 was as follows: denaturation at 94°C for 40 sec, annealing at 57°C for 40 sec, and extension at 72°C for 40 sec, for 30 cycles.

RESULTS AND DISCUSSION

Gel analysis of amplified exon 8 detected a low-molecular-weight band indicative of a small deletion in the proband. Sequence analysis on an automatic DNA sequencer Applied Biosystems 373A revealed one instead of two GAG at codons 272–273 (delGlu272/273). The mother and the sister of the proband were heterozygous for the delGlu272/273 mutation, and this deletion was not detected in 100 X chromosomes from unrelated controls (Fig. 1). This in-frame deletion is likely to be deleterious but does not abolish completely OTC enzymatic activity, resulting in a milder clinical phenotype. It is interesting to note that the GAG deletion occurred in an AG-rich domain, a feature that might have promoted intramolecular recombinations between repeated sequences.

The severity of the mutation is determined in part by the location of the residue on the tertiary structure of the enzyme and by the type of substitution. At the genomic DNA level, it is difficult to correlate exon locations with severity of the phenotype. Nevertheless, it is worth noting that mutations in exons 1 and 5–6 of the OTC gene have been reported in patients with neonatal-onset OTC deficiency, while mutations in exons 2–4 and 9 were observed in both neonatal- and in late-onset OTC deficiency. Exon 8 mutations frequently re-

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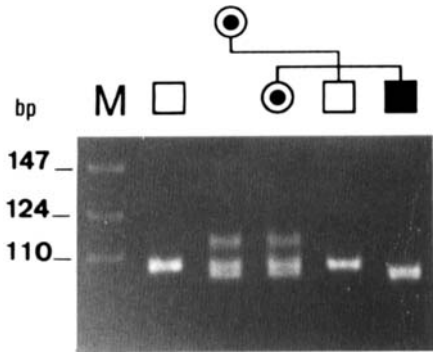

FIGURE 1. **GFC analysis of exon 8 in a patient with the del-Glu272/273 genotype. Exon 8 of the *OTC* gene was amplified by PCR using the following primers: A, 5′-GAC ACT TGG ATA AGC ATG GGA T-3′; B, 5′-CCT GAG AGA GCA TCA ATT TG-3′ with an annealing temperature of 57°C. Amplification products were electrophoresed on a 6% polyacrylamide gel. M, DNA molecular-weight-marker VIII (Boehringer). Control displays a normal 104-bp band. A 101-bp band appears in the affected boy, his mother, and sister. Note the presence of heteroduplex bands in heterozygous females.**

TABLE 1. **Genotype–Phenotype Correlations in Exon 8 of the Ornithine Transcarbamylase Gene**

Codon	Nucleotide change	Codon change	Onset
264	**ACT→GCT**	**Thr→Ala**	**Late**
268	**ATG→ACG**	**Met→Thr**	**Late**
272–273	**ΔGAG**	**Deletion of Glu**	**Late**
277	**CGG→TGG**	**Arg→Trp**	**Late**
277	**CGG→CAG**	**Arg→Gln**	**Late**

sulted in late-onset OTC deficiency (Table 1). The observation of a Glu272/273 deletion in a case of late-onset OTC deficiency adds to the view that exon 8 encompasses mutations that are mildly deleterious for OTC activity, which is not exclusive of more deleterious mutations in this exon (Gilbert-Dussardier et al., 1994, 1995; Oppliger-Leibundgut et al., 1995; Tuchman and Plante, 1995).

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