First Report of a Molecular Prenatal Diagnosis in a Tunisian Family with Lysinuric Protein Intolerance

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Abstract Lysinuric protein intolerance (LPI, MIM# 222700) is an inherited aminoaciduria caused by defective transport of cationic amino acids (CAAs; arginine, lysine, ornithine) at the basolateral membrane of epithelial cells in the intestine and kidney. We report the first prenatal diagnosis by direct mutational analysis of LPI performed in a Tunisian family. An amniotic fluid sample was carried out at 16 weeks of gestation in a 32-year-old Tunisian woman who consulted for prenatal diagnosis. The 1471 delTTCT mutation at homozygous state was identified indicating that the fetus was affected by LPI. The identification of this specific mutation provides a tool, which can be easily applied in Tunisia for molecular diagnosis, genetic counseling, and prenatal diagnosis of LPI.

Keywords Direct mutational analysis · Lysinuric protein intolerance · Prenatal diagnosis

Lysinuric protein intolerance (LPI, MIM# 222700) is a rare autosomal recessive aminoaciduria caused by defective cationic amino acid (CAA; arginine, lysine, ornithine) transport at the basolateral membrane of epithelial cells in the intestine and kidney. Major symptoms include vomiting,

diarrhea, failure to thrive, hepatosplenomegaly, bone marrow abnormalities, osteoporosis, episodes of coma, mental retardation, lung involvement (mainly as alveolar proteinosis), altered immune response, and chronic renal disease (Simell 2001). LPI has an autosomal recessive mode of inheritance and was first described in Finnish patients in 1965 (Perheentupa and Visakorpi 1965), with high prevalence of 1/60,000. It also occurs in Southern Italy and Japan. Sporadic cases have been described worldwide in 22 additional countries (Climbalistiene et al. 2007; Sperandeo et al. 2008). Five Tunisian patients (four families) were diagnosed with LPI, four of which have been previously described (Esseghir et al. 2006). LPI causes a severe clinical phenotype. Dietary regime and supplementation with citrulline may not be sufficient to avoid lethal complications. Prenatal diagnosis by linkage analysis of LPI is now available as reported by (Sperandeo et al. 1999). However, prenatal diagnosis by direct mutational analysis of LPI has not been previously reported in the literature. We report the first prenatal diagnosis by direct mutational analysis of LPI performed in a Tunisian Family.

A 32-year old woman (gravid 2, para 2) consulted for a prenatal diagnosis because her first child, an 11-year-old girl from consanguineous parents, is affected by LPI. The diagnosis of index case was made at the second year of life, on the occasion of Macrophage Activation Syndrome. Early signs of protein intolerance (vomiting, diarrhea, and selective protein aversion), poor growth and a pathological fracture at 18 months oriented the biochemical investigations revealing hyperammonemia and cationic amino aciduria with high urinary excretion of orotic acid, which confirmed the diagnosis of LPI (Table 1). Mutational analysis permitted the identification of the 1471 delTTCT mutation at homozygous state by DHPLC assay (Transgenomic) and was confirmed by direct sequencing (Applied Biosystem). For the prenatal diagnosis of the third pregnancy, an amniotic fluid sample was taken at 16 weeks of gestation. The 1471 delTTCT mutation at homozygous state was identified, indicating that the fetus was affected by LPI (Fig. 1).

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Table 1 Biochemical data of the affected LPI girl

Parameters	Concentrations	
Plasma amonemia	73 $\mu\text{mol/L}$ (10–60)	
Urinary orotic acid	103.5 mmol/mol creatinine (0.35–3.5)	
Amino acids	<i>Urine</i> ($\mu\text{mol}/\text{mmol}$ of creatinine)	<i>Plasma</i> ($\mu\text{mol/L}$)
Ornithine	3.058 (0–80)	33 (29–105)
Lysine	2.517 (0–850)	41 (119–203)
Arginine	1.548 (0–70)	28 (28–101)

Fig. 1 The sequence electropherogram of the sense strand in exon 9: an arrow indicates the position's deletion. (a) The normal sequence from a control. (b) The heterozygous sequence from a patient's parent. (c) The homozygote delTTCT present in patient

The 1471 delTTCT mutation is one of 51 different SLCA7A-specific mutations that have been identified in 142 patients with LPI (Font-Llitjos et al. 2009). Subsequently, the same mutation was identified at homozygous state in five Tunisian patients with LPI (Essegghir et al. 2009). Previously, it had been found at homozygous state in two others Tunisian siblings (Sperandeo et al. 2000).

In conclusion, the 1471 delTTCT mutation, which has been identified in a total of seven Tunisian unrelated patients with LPI, seems to be the common mutation for this disorder in Tunisia. Considering these results, Tunisian patients suspected of LPI can first be screened for this specific mutation, allowing a cost-effective and rapid diagnosis of LPI in Tunisia.

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