

Practical guidelines for molecular testing in Leber congenital amaurosis

Disease definition : Leber congenital amaurosis (LCA) is an autosomal recessive dystrophy of the retina resulting in early-onset blindness. It is different from Leber Hereditary Optic Neuropathy (LHON), which is a mitochondrial disease characterised by subacute bilateral visual failure in young adults.

Frequency : LCA affects approximately 2-3 in 100.000 persons. It is the most common cause of inherited blindness in childhood.

Main clinical symptoms : LCA is a severe dystrophy of the retina, which becomes evident already in the first year of life with nystagmus, photophobia, diminished pupillary responses, Franceschetti's oculo-digital sign (a triad of eye poking, pressing, and rubbing), and very poor visual function with clinical blindness. Hyperopia, keratoconus, and pigmentary retinopathy usually develops later, whereas about 20 % of patients exhibit psychomotor retardation.

Inheritance : LCA is nearly always inherited in autosomal recessive way. Heterozygotes are asymptomatic, but may show abnormal electroretinography . Very rarely, LCA is transmitted in an autosomal dominant way, usually resulting from a mutation in the CRX gene then.

Clinical diagnosis : If LCA is suspected, funduscopy, determination of visual acuity and visual field, and electroretinography (ERG) should be performed. The electroretinogram (ERG) is characteristically nondetectable.

Clinical classification : No real clinical classification exists in LCA, and this retinal dystrophy is difficult to differentiate from severe early-onset retinitis pigmentosa or cone-rod dystrophy.

Molecular testing : Up to now more than 11 loci with 8 nuclear genes AIPL1, CEP290, CRB1, CRX, GUCY2D, RDH12, RPE65, and RPGRIP1 have been shown to be implicated in LCA. Additional genes, including TULP1, MERTK, LRAT and IMPDH1 are implicated in LCA-like entities (Table 1). These genes harbour more than 40 % of all LCA mutations. Molecular testing is complicated by this genetic heterogeneity and the large size of the 2 most common disease genes GUCY2D and CEP290, making testing rather expensive. Furthermore, there are no real mutation hot spots, apart from the W278X mutation in AIPL1, which occurs in 25 % of mutant AIPL1 alleles, but in few mutant LCA alleles overall.

However, a microarray-based test of more than 400 mutations in all 8 LCA disease genes (AIPL1, CRB1, CRX, GUCY2D, CEP290, RDH12, RPGRIP1 and RPE65), and 3 LCA-like genes (LRAT, TULP1, MERTK) is available. As this test detects around 30 % of mutations in LCA, it is the first-pass screening tool.

Most genes implicated in LCA are also involved in other retinal dystrophies, including retinitis pigmentosa and cone-rod dystrophy. These less severe retinal dystrophies are sometimes a consequence of specific mutations, or result when only 1 (dominant) mutation is present. Whereas 20 % of LCA patients has psychomotor retardation, also real syndromic forms of LCA, including Joubert, Meckel and Senior-Loken syndrome, may develop, especially in the case of CEP290 mutations.

References

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Databases

Retina-international database: www.retina-international.org

HUGO mutation database Initiative: www.hgvs.org

HGMD database : www.hgmd.cf.ac.uk

Table 1. Different types of LCA with the proportion of the respective gene, its size, price indication, and test advise.

Type	Specific feature	Gene	Protein	Gene contribution (%)	Number of exons (AA)	Price indication (Euro)	Test Advise
LCA	Also RP and CRD	GUCY2D (LCA1)	Retinal guanylyl cyclase 1	6-21	20 exons (1103 AA)		Test 2
	Also Joubert, Meckel and Senior-loken syndrome	CEP290 (LCA10)	Centrosomal protein Cep290	10-20	55 exons (2481 AA)	3000	No test advised
	Also RP	RPE65 (LCA2)	Retinal pigment epithelium-specific 65 kDa protein	3-16	14 exons (555 AA)	450	Test 4
		RDH12	Retinol dehydrogenase 12	4	7 exons (316 AA)	570	No test advised
	Also CRD and RP	AIPL1 (LCA4)	Aryl-hydrocarbon-interacting protein-like 1	4-8	5 exons (321 AA)		No test advised
	Also CRD and RP	CRX (LCA8)	Cone-rod homeobox protein	3	4 exons (299 AA)		No test advised
	Also CRD	RPGRIP1 (LCA6)	X-linked retinitis pigmentosa GTPase regulator-interacting protein 1	5	24 exons (1286 AA)	900	No test advised
	Also RP and pigmented paravenous chorio-retinal atrophy	CRB1 (LCA7)	Crumbs homolog 1	5-15	6 exons (674 AA)	1275	Test 3
LCA- like		LRAT	Lecithin retinol acyltransferase		3 exons (230 AA)	200	No test advised
	Mainly RP	TULP1	Tubby-like protein 1	1-2	15 exons (542 AA)		No test advised
	Mainly RP	MERTK	Mer tyrosine kinase protooncogene		19 exons (999 AA)	800	No test advised
	Also RP	IMPDH1	Imp dehydrogenase 1		17 exons (599 AA)		No test advised
All cases		AIPL1, CRB1, CRX, LRAT, GUCY2D, CEP290, RDH12, RPGRIP1, RPE65, TULP1, MERTK	Various	30	Mutations in 11 genes	600	Test 1

Figure 1. Suggested molecular testing in LCA

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