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Genetic testing for inherited eye misalignment

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Abstract

We studied the scientific literature and disease guidelines in order to summarize the clinical utility of genetic testing for Inherited eye misalignment (IEM). Forms of IEM associated with variations in the *SALL4*, *CHN1*, *TUBB3* and *KIF21A* genes have autosomal dominant inheritance, whereas those associated with variations in the *ROBO3*, *PHOX2A*, *HOXA1* and *HOXB1* genes have autosomal recessive inheritance. The prevalence of MS is currently unknown. Diagnosis is based on clinical findings, family history, visual acuity testing and fundus examination. The genetic test is useful for confirming diagnosis, and for differential diagnosis, couple risk assessment and access to clinical trials.

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Inherited eye misalignment

(other synonyms: eye misalignment, lazy eye)

General information about the disease

Inherited eye misalignments (acronym IEM) are inherited disorders of the eyes characterized by amblyopia and loss of binocular vision. While the causes of IEM are largely unknown, twin studies and family studies demonstrate a substantial genetic contribution (1). The age of onset distribution of IEM is bimodal, with approximately 22% diagnosed before the age of 12 months and approximately 43% detected between 2 and 3 years of age. IEM can be observed in disorders associated with mitochondrial cytopathies, such as chronic progressive external ophthalmoplegia and Kearns-Sayre syndrome (2), or disorders associated with cranial nerve misrouting, such as Duane syndrome, Moebius syndrome and congenital fibrosis of the extraocular muscles (3).

IEM has unknown prevalence whereas Duane syndrome accounts for 1-5% of all cases of strabismus (4).

Diagnosis of IEM is based on clinical findings, family history, visual acuity and fundus examination. It is confirmed by detection of pathogenic variants in known causative genes.

Differential diagnosis should consider other disorders presenting with strabismus, such as common strabismus, esotropia, exotropia, dissociated vertical deviation, microstrabismus, monofixation syndrome and sixth nerve palsy.

IEM has different patterns of inheritance. Variations in the *CHN1* (OMIM gene: 118423; OMIM disease: 604356), *KIF21A* (OMIM gene: 608283; OMIM disease: 135700), *SALL4* (OMIM gene: 607343; OMIM disease: 607323), and *TUBB3* (OMIM gene: 602661; OMIM disease: 600638) genes have autosomal dominant inheritance. Variations in the *HOXA1* (OMIM gene: 142955; OMIM disease: 601536), *HOXB1* (OMIM gene: 142968; OMIM disease: 614744), *PHOX2A* (OMIM gene: 602753; OMIM disease: 602078), *ROBO3* (OMIM gene: 608630; OMIM disease: 607313), genes have autosomal recessive inheritance.

Pathogenic variants may contain sequence variations (splicing, missense, nonsense, small deletions/insertions); exon or whole-gene duplications/deletions are typically not reported.

Aims of the test

- To determine the gene defect responsible for the pathology
- To confirm clinical diagnosis of the disease
- To determine carrier status for the disease.

Test characteristics

Expert centers/Published guidelines

The test is listed in the Orphanet database and is offered by 13 accredited medical genetic laboratories in the EU, and in the GTR database, offered by 13 accredited medical genetic laboratories in the US.

The guidelines for clinical use of the test are described in "Genetics home reference" (ghr.nlm.nih.gov).

Test strategy

A multi-gene NGS panel is used for the detection of nucleotide variations in coding exons and flanking introns known genes. Potentially causative variants and region with low coverage are Sanger-sequenced. Sanger sequencing is also used for family segregation studies.

The test identifies variations in known causative genes in patients suspected to have MS. To perform molecular diagnosis, a single sample of biological material is normally sufficient. This may be 1 ml blood in a sterile tube with 0.5 ml K3EDTA or 1 ml saliva in a sterile tube with 0.5 ml ethanol 95%. Sampling rarely has to be repeated. Gene-disease associations and the interpretation of genetic variants are rapidly developing fields. It is therefore possible that the genes mentioned in this note may change as new scientific data is acquired. It is also possible that genetic variants today defined as of "unknown or uncertain significance" may acquire clinical importance.

Genetic test results

Positive

Identification of pathogenic variants in the *CHN1, HOXA1, HOXB1, KIF21A, PHOX2A, ROBO3, SALL4, TUBB3* genes confirms the clinical diagnosis and is an indication for family studies.

A pathogenic variant is known to be causative for a given genetic disorder based on previous reports or predicted to be causative based on the loss of protein function or expected significant damage to protein or protein/protein interactions. In this way it is possible to obtain a molecular diagnosis in new/other subjects, establish the risk of recurrence in family members and plan preventive and/or therapeutic measures.

Inconclusive

Detection of a new variation, not described in the literature and/or without any evident pathogenic significance, cannot indicate any clear genotype-phenotype correlation. In these cases, it is advisable to extend testing to the patient's relatives in order to assess variant segregation and clarify its contribution. In some cases it could be necessary to perform further examinations/tests or to do a clinical reassessment of pathological signs.

Negative result

The absence of variations in the genomic regions investigated does not exclude a clinical diagnosis but suggests the possibility of:

- alterations that cannot be identified by sequencing, such as large rearrangements that cause loss (deletion) or gain (duplication) of extended gene fragments;
- sequence variations in gene regions not investigated by this test, such as regulatory regions (5' and 3' UTR) and deep intronic regions;
- variations in other genes not investigated by the present test.

Unexpected

Unexpected results may come out from the test, for example information regarding consanguinity; absence of family correlation or the possibility of developing genetically based diseases.

Risk for progeny

For genes with autosomal dominant transmission, the probability that a carrier transmits the disease variant to his/her children is 50% in any pregnancy independently of the sex of the conceived.

Autosomal recessive transmission needs that both healthy carrier parents transmit their disease variant to his/her children. In this case, the probability of having an affected boy or girl is therefore 25%.

Limits of the test

The test is limited by current scientific knowledge regarding the genes and disease.

Analytical sensitivity (proportion of positive tests when the genotype is truly present) and analytical specificity (proportion of negative tests when the genotype is not present)

NGS: Analytical sensitivity: >99% (with a minimum coverage of 10X); Analytical specificity: 99.99%.

SANGER: Analytical sensitivity: >99.99%, Analytical specificity: 99.99%.

Clinical sensitivity (proportion of positive tests if the disease is present) and clinical specificity (proportion of negative tests if the disease is not present)

Clinical sensitivity: data not available

Clinical specificity: is estimated at about 99% [Author's laboratory data] (5).

Prescription appropriateness

The genetic test is appropriate when:

a) the patient meets the diagnostic criteria for the disease;

b) the genetic test has diagnostic sensitivity greater than or equal to other published tests.

Clinical utility

Clinical management	Utility
Confirmation of clinical diagnosis	yes
Differential diagnosis	yes
Access to clinical trial (6)	yes
Couple risk assessment	yes

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