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Genetic testing for central areolar choroidal dystrophy

Andi Abeshi[1,2], Francesca Fanelli[3], Tommaso Beccari[4], Munis Dundar[5], Benedetto Falsini[6] and Matteo Bertelli[2,3]

Abstract

We studied the scientific literature and disease guidelines in order to summarize the clinical utility of the genetic test for central areolar choroidal dystrophy (CACD). CACD is mostly inherited in an autosomal dominant manner. Transmission is rarely autosomal recessive. Overall prevalence is currently 1-9 per 100 000. CACD is caused by mutations in the *PRPH2* and *GUCY2D* genes. Clinical diagnosis is based on clinical findings, ophthalmological examination, fluorescein angiography, electroretinography (showing cone dystrophy) and stereo fundus photography. The genetic test is useful for confirming diagnosis, and for differential diagnosis, couple risk assessment and access to clinical trials.

[1]MAGI Balkans, Tirana, Albania
[2]MAGI'S Lab, Rovereto, Italy
[3]MAGI Euregio, Bolzano, Italy
[4]Department of Pharmaceutical Sciences, University of Perugia, Perugia, Italy
[5]Department of Medical Genetics, Erciyes University Medical School, Kayseri, Turkey
[6]Department of Ophthalmology, Catholic University of Rome, Rome, Italy

Corresponding author: M. Bertelli
E-mail: info@assomagi.org

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Central areolar choroidal dystrophy

(other synonyms: CACD, choroidal macular dystrophy, choroidal sclerosis)

General information about the disease

Central areolar choroidal dystrophy (CACD) is a genetically heterogeneous disorder, principally affecting the macula, characterized by a large central area of atrophy and loss or absence of photoreceptors, retinal pigmented epithelium and choriocapillaris (1-4), leading to progressive reduction of visual acuity, generally occurring between the ages of 30 and 60 years (4-6). The rate of disease progression is highly variable. Color vision is normal in the early stages, however strongly deteriorated later on, and there is no photophobia. CACD is classified in four stages (7). The estimated prevalence of CACDs is 1-9 per 100 000 (retrieved from orphanet.org).

The clinical diagnosis of CACD is based on clinical findings, ophthalmological examination (4), fluorescein angiography or autofluorescence, electroretinography (showing severe cone dysfunction) and optical coherence tomography. It is confirmed molecularly by detection of pathogenic variants of the causative genes.

Differential diagnosis should consider late stages of cone dystrophy, atrophic age-related macular degeneration and other adult onset peripherin/RDS-related macular dystrophies, with which CACD shares clinical characteristics such as geographic atrophy and drusen-like deposits (8-11).

Being clinically and genetically heterogeneous, the different CACD types (CACD1, CACD2, CACD3) involve variations in several causative genes that are mostly inherited in an autosomal dominant manner. CACD1 is caused by an unknown variation on 17p13. CACD2 and CACD3 are associated with variations in the peripherin/RDS gene *PRPH2* (OMIM gene: 179605; OMIM disease: 613105) (12-14). CACD was also recently linked to variations in the *GUCY2D* gene (15).

Pathogenic variants may contain small intragenic deletions/insertions, as well as splice-site, missense and nonsense variants. Exon and whole-gene duplications/deletions are not reported.

Aims of testing

- 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 about 10 accredited medical genetic laboratories in the EU, and in the GTR database, offered by 9 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 in the *PRPH2* and *GUCY2D* genes. Potentially causative variants and areas of 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 CACDs. 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 *PHPR2* and *GUCY2D* 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 variant of unknown or uncertain significance: a new variation and/or without any evident pathogenic significance or with insufficient or significant conflicting evidence to indicate it is likely benign or likely pathogenic for a given genetic disorder. 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

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

- 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;
- alterations that cannot be identified by sequencing, such as large rearrangements that cause loss (deletion) or gain (duplication) of extended gene fragments.

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

In 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.

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: variations in the *PRPH2* gene are associated with several forms of autosomal dominant retinal dystrophy, including retinitis pigmentosa (ADRP), cone dystrophy and autosomal dominant macular dystrophy (ADMD). More than 100 variations have so far been associated with forms of CACD: 8-9% are cases of ADRP and 7-23% families with ADMD; penetration is low in some cases. ADMD forms include conditions defined more on a clinical basis as CACD, where variations in the *PRPH2* gene are found in large pedigrees with autosomal dominant inheritance: a total of six variations have been described.

A variation in the *GUCY2D* gene associated with CACD was recently identified in a large Irish family (15).

Clinical specificity is: estimated at 99.99% % [Author's laboratory data] (16).

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 (17)	yes
Couple risk assessment	yes

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