CADASIL Genetic Testing

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Introduction

CADASIL (Cerebral Autosomal Dominant Arteriopathy with Subcortical Infarcts and Leukoencephalopathy) genetic testing is addressed by this guideline.

Procedures addressed

The inclusion of any procedure code in this table does not imply that the code is under management or requires prior authorization. Refer to the specific Health Plan's procedure code list for management requirements.

Procedures addressed by this guideline	Procedure codes
NOTCH3 Deletion/Duplication Analysis	81479
NOTCH3 Known Familial Mutation Analysis	81403
NOTCH3 Targeted Sequencing	81406

Criteria

Introduction

Requests for CADASIL (Cerebral Autosomal Dominant Arteriopathy with Subcortical Infarcts and Leukoencephalopathy) genetic testing are reviewed using these criteria.

Known Familial Mutation Testing

- Genetic Counseling:
  - Pre and post-test genetic counseling by an appropriate provider (as deemed by the Health Plan policy), AND
- Previous Genetic Testing:
  - No previous genetic testing for NOTCH3 mutations that would detect the familial mutation, AND
- Predictive Testing:
  - Member has a first-degree relative (i.e. parent, sibling, child) with an identified NOTCH3 gene mutation, and
  - Member is at least 18 years of age, OR

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- Diagnostic Testing for Symptomatic Individuals:
  - Member has a first-degree relative (i.e. parent, sibling, child) with an identified NOTCH3 gene mutation, and
  - High index of suspicion for CADASIL diagnosis based on clinical findings, AND
- Rendering laboratory is a qualified provider of service per the Health Plan policy.

NOTCH3 Targeted Sequencing

- Genetic Counseling:
  - Pre and post-test genetic counseling by an appropriate provider (as deemed by the Health Plan policy), AND
- Previous Genetic Testing:
  - No previous genetic sequencing for NOTCH3 mutations, AND
- Diagnostic Testing for Symptomatic Individuals:
  - High index of suspicion for CADASIL diagnosis based on clinical findings, AND
- Rendering laboratory is a qualified provider of service per the Health Plan policy.

NOTCH3 Deletion/Duplication Analysis

- Member meets the above criteria for NOTCH3 targeted sequencing, AND
- NOTCH3 targeted sequencing performed and detected no mutations, AND
- No previous NOTCH3 deletion/duplication analysis, AND
- Rendering laboratory is a qualified provider of service per the Health Plan policy.

What is CADASIL?

Definition

CADASIL (Cerebral Autosomal Dominant Arteriopathy with Subcortical Infarcts and Leukoencephalopathy) is an adult-onset form of cerebrovascular disease. There are no generally accepted clinical diagnostic criteria for CADASIL and symptoms vary among affected individuals.

Prevalence

CADASIL is a rare disease.[1-3] The exact prevalence is unknown. CADASIL is probably still underdiagnosed. The minimum prevalence is estimated to be between 2-5 per 100,000 based on multiple small and national registries.[1,3] More recent "reports suggest

that the prevalence of NOTCH3 cysteine-altering pathogenic variants is substantially higher, and may be as high as 1 in 300 worldwide."[2] A founder effect has been reported for Finnish individuals and individuals in the Marche region of Italy.[1]

CADASIL is the most prevalent inherited cause of cerebral small-vessel disease.[4]

Symptoms

Typical signs and symptoms include[1,3,5]

- transient ischemic attacks and ischemic stroke, occurs at a mean age of 47 years (age range 20-70 years), in most cases without conventional vascular risk factors
- cognitive disturbance, primarily affecting executive function, may start as early as age 35 years
- psychiatric or behavioral abnormalities
- migraine with aura, occurs with a mean age of onset of 30 years (age range 6-48 years), and

Less common symptoms include:

- recurrent seizures with onset in middle age, usually secondary to stroke
- acute encephalopathy, with a mean age of onset of 42 years

Cause

CADASIL is caused by mutations in the NOTCH3 gene.

To date, NOTCH3 is the only gene in which mutations are known to cause CADASIL.[1] NOTCH3 has 33 exons. CADASIL pathogenic variants occur in exons 2–24, which encode the 34 epidermal growth factor repeats (EGFR).[1,6] The majority of pathogenic variants occur in exons 2-6.[3] NOTCH3 encodes a transmembrane receptor that is primarily expressed in vascular smooth-muscle cells, preferentially in small arteries.[1] "In CADASIL, the extracellular domain of the Notch3 receptor accumulates within blood vessels. Accumulation takes place at the cytoplasmic membrane of VSMCs [vascular smooth muscle cells] and pericytes in close vicinity to the granular osmiophilic deposits (GOM) that characterize the disease. NOTCH3 recruits other proteins into the extracellular deposits, among them vitronectin and tissue inhibitor of metalloproteinase-3 (TIMP3), which may be relevant for disease pathogenesis."[3] There is a hypothesis that structural abnormalities in the vascular smooth-muscle protein NOTCH3 trigger arterial degeneration, vascular protein accumulation, and cerebrovascular failure.[4]

No clear genotype-phenotype correlations exist for individuals with CADASIL.[7,8] Some studies describe phenotype-genotype correlations. "There is reasonably strong evidence that pathogenic variants in the first six epidermal growth factor-like repeat domains (EGFR 1 to 6) of the Notch3 protein are associated with an earlier age of stroke onset, a more severe phenotype, and lower survival compared with pathogenic variants in EGFR 7 to 34."[3] However, there can be significant intrafamilial variability with the age of onset, disease severity, and disease progression. The genotype cannot

be used to predict the phenotype.[1,4] NOTCH3 cysteine-altering pathogenic variants are associated with a broad phenotypic spectrum which includes classic CADASIL, mild small vessel disease, and non-penetrance.[3]

Inheritance

CADASIL is an autosomal dominant disorder.

Autosomal dominant inheritance

In autosomal dominant inheritance, individuals have 2 copies of the gene and only one mutation is required to cause disease. When a parent has a mutation, each offspring has a 50% risk of inheriting the mutation. Males and females are equally likely to be affected.

Diagnosis

Brain Magnetic Resonance Imaging (MRI) findings include T2-signal-abnormalities in the white matter of the temporal pole and T2-signal-abnormalities in the external capsule and corpus callosum.[1,3]

CADASIL is suspected in an individual with the clinical signs and MRI findings. A positive family history for stroke or dementia is also indicative of disease in symptomatic individuals. However, a negative family history should not exclude the diagnosis, as de novo mutations have been reported, and affected family members are frequently misdiagnosed.[1,7]

Sequencing of all NOTCH3 exons encoding EGF–like domains fails to identify a mutation in up to 4% of individuals with CADASIL. Therefore, skin biopsy with histopathologic evaluation for characteristic GOM deposits is appropriate for individuals with a high index of clinical suspicion for CADASIL and negative genetic testing.[2,3]

For a firm diagnosis of CADASIL, at least one of the following is required:

- Documentation of a typical NOTCH3 mutation by genetic analysis.[1,3,7]
    - NOTCH3 mutation detection may reach >95% in individuals with strong clinical suspicion of CADASIL[1].
- Documentation of characteristic GOM deposits within small blood vessels by skin biopsy.[1,3,7]

Management

A correct diagnosis of CADASIL is important because the clinical course of disease is different from individuals with other types of cerebral small-vessel disease and proven therapies for stroke have not been validated in individuals with CADASIL.[7] However, no specific disease-modifying treatments for CADASIL exist. Management and treatment of individuals is generally symptomatic and supportive.[1,3,5,7,9]

Patients with CADASIL should avoid anticoagulants, angiography, and smoking to avoid disease-related complications, so clinical utility is represented.[1,7] Because of the risk for cerebral hemorrhage, use of antiplatelets rather than anticoagulants is considered for prevention of ischemic attacks. Evidence against the use of intravenous tissue plasminogen activator (IV tPA) has been suggested due to the possibility of hemorrhage; however, this is not conclusive.[10] Statins are used for treatment of hypercholesterolemia and antihypertensive drugs are used for hypertension and hypertension treatment may have an additional benefit.[3] Management of neurologic events (migraines, depression, psychiatric manifestations) by a neurologist or neuropsychiatrist can be beneficial; pregnancy and postpartum periods are potential risk factors.[1] The American Heart Association issued a scientific statement summarizing the current recommendations for the diagnosis and management of CADASIL.[11]

Survival

"In a retrospective analysis of 411 patients with CADASIL, the median age at death was 65 years in men and 71 years in women."[2]

Test information

Introduction

Testing for CADASIL may include genetic testing (known familial mutation analysis, sequence analysis, or deletion/duplication analysis) and/or skin biopsy.

Skin biopsy

A pathognomonic characteristic of CADASIL is the finding of characteristic GOM within the vascular media and increased NOTCH3 staining of the arterial wall, which can be evaluated in a skin biopsy.[1] Specificity of skin biopsy findings is high, as the characteristic deposits have not been documented in any other disorder. Sensitivity has been reported to range from 45%-100%. Sensitivity and specificity can be maximized to >90% by immunostaining for NOTCH3 protein.[7] When interpreted by an experienced (neuro) pathologist, combined analysis by electron microscopy and immunohistochemistry usually allows for a conclusive CADASIL diagnosis.

Known Familial Mutation (KFM) Testing

Known familial mutations analysis is performed when a causative mutation has been identified in a close biological relative of the individual requesting testing. Analysis for known familial mutations typically includes only the single mutation. However, if available, a targeted mutation panel that includes the familial mutation may be performed.

Sequence analysis

To date, all mutations in NOTCH3 causing CADASIL have been in exons 2-24, including intron-exon boundaries.[1] In the United States, laboratories offering CADASIL testing appear to perform, at minimum, next-generation sequencing (NGS) of exons 2-24 at the time of this review.

Deletion and Duplication Analysis

Analysis for deletions and duplications can be performed using a variety of technical platforms including exon array, Multiplex ligation-dependent probe amplification (MLPA), and NGS data analysis. These assays detect gains and losses too large to be identified through standard sequence analysis, often single or multiple exons or whole genes.

Large deletions and duplications in the NOTCH3 gene have not been reported.[3] Molecular testing approaches can include deletion/duplication analysis if sequencing analysis of NOTCH3 is unrevealing.[1]

Guidelines and evidence

Introduction

This section includes relevant guidelines and evidence pertaining to CADASIL testing. No evidence-based U.S. testing guidelines have been identified.

American Heart Association

The American Heart Association (AHA, 2023) published a scientific statement with information on the management of inherited cerebral small vessel disease (CSVD), including CADASIL.[11] They stated the following regarding genetic testing for CADASIL.

- "Several approaches have been developed to help clinicians prioritize gene testing. For at least two decades, it has been recognized that anterior temporal polar WMH [white matter hyperintensities] on brain MRI is a marker of CADASIL with good sensitivity and specificity. The Pescini scale ranges from 0 to 25 (>14 points suspicious of CADASIL) and uses clinical features like stroke or transient ischemic attack onset before 50 years of age."
- "Strong consideration should be given to genetic counseling to allow discussion of the ramifications of obtaining genetic test results on the individual patient and their family." The authors provided additional considerations of pre-test genetic counseling including addressing possible discrimination, the possible "negative psychological consequences", and the general consensus against predictive testing for minors.

- "Posttest counseling can help with interpretation, especially of variants of unclear significance, and navigating grief or guilt from positive, negative, or equivocal results."
- "A known pathogenic NOTCH3 mutation within a family simplifies testing because only a single mutation needs to be investigated. When the mutation is unknown or unavailable, the laboratory must undertake a more complex analysis." The genetic analysis outlined when a known familial mutation has not been identified may include targeted sequencing or full gene sequencing. If multiple inherited CVSDs are in the differential, a multigene panel may be considered.
- "Although used less frequently, there is still a role for skin biopsy to look for pathognomonic granular osmophilic material on electron microscopy that may clarify the clinical significance of a mutation of uncertain or unknown significance. The accuracy of skin biopsy is enhanced by the use of immunohistochemistry."

European Academy of Neurology

The European Academy of Neurology (EAN, 2020) consensus panel stated:[9]

- "CADASIL can only be definitively confirmed by genetic testing, revealing a NOTCH3 mutation altering the number of cysteines in one of the 34 EGFr domains of the NOTCH3 protein."
- A diagnosis of CADASIL can be established by skin biopsy with electron microscopy showing GOM, but genetic testing should be the first diagnostic line of investigation.
- "In the case of a NOTCH3 variant of unknown significance, CADASIL can be confirmed using a skin biopsy for electron microscopy and/or NOTCH3 immunostaining."
- "All or almost all variants leading to CADASIL result in a loss or gain of a cysteine in EGFr repeats. Some non-cysteine-changing variants have been reported but the consensus was that the vast majority of these variants are not pathogenic. In such cases, electron microscopy revealing GOM can be a useful diagnostic tool."
- "The diagnosis of CADASIL should be considered in any patient with unexplained symmetrical periventricular WMHs and a positive family history of migraine with aura, stroke, mood disorders or dementia."

Selected Relevant Publications

The following publications addressed CADASIL testing.

Guey et al (2021)

Guey et al (2021) stated that due to the phenotypic overlap between CADASIL and other more recently characterized hereditary cerebral small vessel diseases (e.g., CARASIL, HTRA1/CADASIL type 2, COL4A1-related small vessel disease) as well as the lack of highly specific or sensitive clinical features, a multigene panel which

includes genes associated with these related inherited conditions may be preferred when offering genetic testing to a symptomatic proband.[12]

Pescini et al (2012)

Pescini et al (2012) published a scale to help guide clinicians in selecting individuals for NOTCH3 genetic analysis due to a high probability of a CADASIL genetic diagnosis. This scale assigns weighted scores to common features of CADASIL. The authors state that their scale is accurate, demonstrating optimal sensitivity (96.7%) and specificity (74.2%). At the time of publication, results needed to be confirmed and further validated.[13]

Choi et al (2010)

A two-center cohort study found that blood pressure and hemoglobin A1c levels were associated with cerebral mini bleeds in individuals with CADASIL.[7] Therefore, controlling blood pressure and glucose levels may improve the clinical course of the disease. It is also reasonable to control for high cholesterol and high blood pressure given the high rate of ischemic stroke seen in CADASIL.[7]

Tikka et al (2009)

Evidence from a 2009 retrospective cohort study suggested that an adequate skin biopsy for analysis of GOM is a cost effective way to determine a diagnosis of CADASIL in symptomatic individuals.[14]

The authors suggest that biopsy results can be used to guide the decision for who should have genetic testing, particularly in individuals with no known familial mutation or from ethnic populations with no evidence of founder mutations.[14]

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Introduction

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