BlueCross BlueShield of Alabama

Name of Policy:
Genetic Testing of CADASIL Syndrome

Policy #: 589
Category: Laboratory

Latest Review Date: May 2018
Policy Grade: D

Background/Definitions:

As a general rule, benefits are payable under Blue Cross and Blue Shield of Alabama health plans only in cases of medical necessity and only if services or supplies are not investigational, provided the customer group contracts have such coverage.

The following Association Technology Evaluation Criteria must be met for a service/supply to be considered for coverage:

1. *The technology must have final approval from the appropriate government regulatory bodies;*
2. *The scientific evidence must permit conclusions concerning the effect of the technology on health outcomes;*
3. *The technology must improve the net health outcome;*
4. *The technology must be as beneficial as any established alternatives;*
5. *The improvement must be attainable outside the investigational setting.*

Medical Necessity means that health care services (e.g., procedures, treatments, supplies, devices, equipment, facilities or drugs) that a physician, exercising prudent clinical judgment, would provide to a patient for the purpose of preventing, evaluating, diagnosing or treating an illness, injury or disease or its symptoms, and that are:

1. *In accordance with generally accepted standards of medical practice; and*
2. *Clinically appropriate in terms of type, frequency, extent, site and duration and considered effective for the patient's illness, injury or disease; and*
3. *Not primarily for the convenience of the patient, physician or other health care provider; and*
4. *Not more costly than an alternative service or sequence of services at least as likely to produce equivalent therapeutic or diagnostic results as to the diagnosis or treatment of that patient's illness, injury or disease.*

Description of Procedure or Service:

Variants in the *NOTCH3* gene have been causally associated with CADASIL (cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy). Genetic testing is available to determine if pathogenic variants exist in the *NOTCH3* gene for patients with suspected CADASIL and their family members.

CADASIL

CADASIL is an uncommon, autosomal dominant disease. It is the most common cause of hereditary stroke and hereditary vascular dementia in adults. The CADASIL syndrome is an adult-onset, disabling systemic condition, characterized by migraine with aura, recurrent lacunar strokes, progressive cognitive impairment, and psychiatric disorders. The overall prevalence of the disease is unknown in the general population.

Diagnosis

The differential diagnosis of CADASIL includes the following conditions: (See Table 1).

Table 1. Differential Diagnosis of CADASIL

Acquired Disorders	Inherited Disorders
• Sporadic SVD with or without hypertension as the main risk factor • Multiple sclerosis • Primary angiitis of the central nervous system	• Fabry disease • Cerebral autosomal recessive arteriopathy with subcortical infarcts and leukoencephalopathy • Familial SVD caused by heterozygous variants in the *HTRA1* gene • Some forms of leukodystrophy

SVD: small vessel disease.

Since the clinical presentation of CADASIL varies, the condition may be confused with multiple sclerosis, Alzheimer dementia, and Binswanger disease. The specific clinical signs and symptoms, along with family history and brain magnetic resonance imaging (MRI) findings, are extremely important in determining the diagnosis of CADASIL. The clinical features and mode of inheritance (autosomal dominant versus autosomal recessive) help to distinguish other inherited disorders in the differential diagnosis from CADASIL.

When the differential diagnosis includes CADASIL, various diagnostic tests are available:

- Genetic testing, by direct sequencing of selected exons or of exons 2-24 of the *NOTCH3* gene. Identification of a *NOTCH3* pathogenic variant definitively establishes a diagnosis of CADASIL without the need for additional diagnostic testing (e.g. skin biopsy).
- Immunohistochemistry assay of a skin biopsy sample, using a monoclonal antibody with reactivity against the extracellular domain of the *NOTCH3* receptor. Positive immunostaining reveals the accumulation of *NOTCH3* protein in the walls of small blood vessels. Lesnick Oberstein et al in 2003 estimated sensitivity and specificity at 85% to 90% and 95% to 100%, respectively, for two observers of the test results in a population of patients and controls correlated with clinical, genetic and MRI parameters.
- Detection of granular osmiophilic material (GOM) in the same skin biopsy sample by electron microscopy. The major component of GOM is the ectodomain of the *NOTCH3* gene product. GOM accumulates directly in vascular smooth muscle cells and, when

present, is considered a hallmark of the disease. However, GOM may not be present in all biopsy samples. Sensitivity has been reported as low as 45% and 57%, but specificity is generally near or at 100%.

- Examination of brain tissue for the presence of GOM. GOM was originally described as limited to brain vessels. Examination of brain biopsy or autopsy after death was an early criterion standard for diagnosis. In some cases, peripheral staining for GOM has been absent even though positive results were seen in brain vessels.

NOTCH3 Variants

Variants in *NOTCH3* have been identified as the underlying cause of CADASIL. In almost all cases, the pathogenic variants lead to loss or gain of a cysteine residue that could lead to increased reactivity of the *NOTCH3* protein, resulting in ligand-binding and toxic effects.

The *NOTCH3* gene is found on chromosome 19p13.2-p13.1 and encodes the third discovered human homologue of the Drosophila melanogaster Type I membrane protein *NOTCH*. The *NOTCH3* protein consists of 2321 amino acids primarily expressed in vascular smooth muscle cells and plays an important role in the control of vascular transduction. It has an extracellular ligand-binding domain of 34 epidermal growth factor-like repeats, traverses the membrane once, and has an intracellular domain required for signal transduction.

Variants in the *NOTCH3* gene have been differentiated into those that are causative of the CADASIL syndrome (pathogenic variants) and those that are of uncertain significance. Pathogenic variants affect conserved cysteine residues within 34 epidermal growth factor (EGF)-like repeat domains in the extracellular portion of the *NOTCH3* protein. More than 150 pathogenic variants have been reported in at least 500 pedigrees. *NOTCH3* has 33 exons, but all CADASIL variants reported to date have occurred in exons 2-24, which encode the 34 EGF-like repeats, with strong clustering in exons 3 and 4, which encode EGFR 2-5 (>40% of variants in >70% of families occur in these exons). Some studies indicate that the clinical variability in CADASIL presentation, particularly with regard to the development of white matter hyperintensities on MRI, may be related to genetic modifiers outside the *NOTCH3* locus, but the specific role of these modifiers is not well-delineated.

The probability that CADASIL is present is an individualized assessment, depending on numerous factors such as family history, symptoms, imaging results, and other specialized testing such as skin biopsy. In 2012, Pescini et al published a study that attempted to identify clinical factors that increase the likelihood of a pathologic variant being present, with increasing likelihood with the presence of one or several factors, including migraine, migraine with aura, transient ischemic attack/stroke, psychiatric disturbance, cognitive decline, leukoencephalopathy (with greater risk for leukoencephalopathy extending to the temporal pole or external capsule), and subcortical infarcts.

Policy:

Effective for dates of service on or after May 12, 2017:

Genetic testing of *NOTCH3* to confirm the diagnosis of CADASIL meets Blue Cross and Blue Shield of Alabama's medical criteria for coverage when **one or more** of the following criteria are met:

- Patients with clinical signs, symptoms and imaging results are consistent with CADASIL, i.e., history of migraines or migraines with aura, transient ischemic attack/stroke, progressive dementia, diffuse subcortical lesion in white matter on magnetic resonance imaging, and mood disorders, with or without a family history of the condition; **or**
- The diagnosis of CADASIL is inconclusive following alternative methods of testing, including skin biopsy and magnetic resonance imaging; **or**
- Asymptomatic individuals with or without a known *NOTCH3* variant in an affected family member (first- or second-degree relative).

Genetic testing of *NOTCH3* to confirm the diagnosis of CADASIL syndrome does not meet Blue Cross and Blue Shield of Alabama's coverage criteria and is considered **investigational in all other situations**.

Effective for dates of service prior to May 12, 2017:

Genetic testing for CADASIL, cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy, meets Blue Cross and Blue Shield of Alabama's medical criteria for coverage when **one or more** of the following criteria are met:

- Patients with symptoms of CADASIL, i.e., history of migraines with aura, multiple subcortical ischemic events in the absence of hypertension and other vascular risk factors, progressive dementia, diffuse subcortical lesion in white matter on magnetic resonance imaging, and mood disorders, with or without a family history of the condition; **or**
- Pre-symptomatic individuals where there is a known mutation in an affected family member.

Genetic testing for CADASIL syndrome does not meet Blue Cross and Blue Shield of Alabama's coverage criteria and is considered investigational in all other situations.

Blue Cross and Blue Shield of Alabama does not approve or deny procedures, services, testing, or equipment for our members. Our decisions concern coverage only. The decision of whether or not to have a certain test, treatment or procedure is one made between the physician and his/her patient. Blue Cross and Blue Shield of Alabama administers benefits based on the member's contract and corporate medical policies. Physicians should always exercise their best medical judgment in providing the care they feel is most appropriate for their patients. Needed care should not be delayed or refused because of a coverage determination.

Key Points:

The most recent literature review covers the period through February 5, 2018.

Evidence reviews assess whether a medical test is clinically useful. A useful test provides information to make a clinical management decision that improves the net health outcome. That is, the balance of benefits and harms is better when the test is used to manage the condition than when another test or no test is used to manage the condition.

The first step in assessing a medical test is to formulate the clinical context and purpose of the test. The test must be technically reliable, clinically valid, and clinically useful for that purpose. Evidence reviews assess the evidence on whether a test is clinically valid and clinically useful. Technical reliability is outside the scope of these reviews, and credible information on technical reliability is available from other sources.

Testing Individuals with Suspected CADASIL Syndrome

Clinical Context and Test Purpose

The purpose of genetic testing of symptomatic individuals with suspected CADASIL syndrome are to establish the diagnosis of CADASIL without skin biopsy or other invasive testing and aid in reproductive planning, when the diagnosis cannot be made clinically.

The questions addressed in this evidence review are: In individuals with suspected CADASIL, does the use of genetic testing result in changes in management or outcome improvements, including eliminating the need for skin biopsy to confirm diagnosis of CADASIL, aid in preimplantation genetic testing to determine likelihood of an affected offspring or alter reproductive planning decisions?

The following PICOTS were used to select literature to inform this review.

Patients

The relevant population of interest includes individuals with suspected CADASIL.

Interventions

The relevant intervention of interest is genetic testing for NOTCH3 variants.

Comparators

The relevant comparator of interest is standard clinical management without genetic testing.

Outcomes

The potential beneficial outcome of primary interest would be changes in management associated with improved outcomes initiated based on confirming a genetic diagnosis of CADASIL. Reductions in skin biopsies or other invasive tests to confirm diagnosis of CADASIL are potential beneficial outcomes.

Potential harmful outcomes are those resulting from a false-positive or false-negative test results. False-positive test results can lead to inappropriate initiation of treatments or psychological harm

after receiving positive test results. False-negative test results can lead to lack of medical or neurologic treatments or surveillance.

Timing
The time frame for outcomes measures varies from short-term development of symptoms to long-term changes in disease status and outcomes.

Setting
Patients suspected of CADASIL are actively managed by neurology or psychiatry due to ischemic episodes, cognitive deficits, migraines with aura or psychiatric disturbances. Genetic testing is utilized to confirm a diagnosis of CADASIL. Referral for genetic counseling is important for explanation of genetic disease, heritability, genetic risk, test performance, and possible outcomes.

Simplifying Test Terms
There are 3 core characteristics for assessing a medical test. Whether imaging, laboratory, or other, all medical tests must be:

- Technically reliable
- Clinically valid
- Clinically useful.

Because different specialties may use different terms for the same concept, we are highlighting the core characteristics. The core characteristics also apply to different uses of tests, such as diagnosis, prognosis, and monitoring treatment.

Diagnostic tests detect presence or absence of a condition. Surveillance and treatment monitoring are essentially diagnostic tests over a time frame. Surveillance to see whether a condition develops or progresses is a type of detection. Treatment monitoring is also a type of detection because the purpose is to see if treatment is associated with the disappearance, regression, or progression of the condition.

Prognostic tests predict the risk of developing a condition in the future. Tests to predict response to therapy are also prognostic. Response to therapy is a type of condition and can be either a beneficial response or adverse response. The term predictive test is often used to refer to response to therapy. To simplify terms, we use prognostic to refer both to predicting a future condition or to predicting a response to therapy.

Technically Reliable
Assessment of technical reliability focuses on specific tests and operators and requires review of unpublished and often proprietary information. Review of specific tests, operators, and unpublished data are outside the scope of this evidence review, and alternative sources exist. This evidence review focuses on the clinical validity and clinical utility.

Clinically Valid
A test must detect the presence or absence of a condition, the risk of developing a condition in the future, or treatment response (beneficial or adverse).

Several retrospective and prospective studies have examined the association between *NOTCH3* variants and cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy (CADASIL), as shown in Table 2. Studies have been divided into two categories: Part 1, diagnostic studies, in which the patients enrolled were suspected but not confirmed to have CADASIL; and Part 2, clinical validity studies, in which the patients had already been diagnosed with the disease by some method other than genetic testing. The diagnostic studies are more likely to represent the target population in which the test would be used.

The results of the clinical validity studies demonstrate that a *NOTCH3* pathogenic variant is found in a high percentage of patients with a clinical diagnosis of CADASIL, with studies reporting a clinical sensitivity of 90% to 100%. Limited data on specificity are from testing small numbers of healthy controls, and no false positive *NOTCH3* variants have been reported in these populations. The diagnostic yield studies report a variable diagnostic yield, ranging from 10% to 54%. These lower numbers likely reflect testing in heterogeneous populations that include patients with other disorders.

Testing Strategy
Identification of a NOTCH3 pathogenic variant establishes a diagnosis of CADASIL. For individuals suspected of CADASIL:

- Perform targeted sequencing and analysis of specific NOTCH3 exons (e.g., exon 4 only, exons 2 – 6) OR
- Perform general testing of NOTCH3 exons (e.g., exons 2 – 24 or all 33 exons).
- If no NOTCH3 pathogenic variant is identified, skin biopsy is warranted for immunohistochemical staining for Notch3 protein and/or electron microscopy for granular osmiophilic material (GOM).

Table 2. Association Between *NOTCH3* With CADASIL Diagnosis: Results of Studies Supporting *NOTCH3* Genotyping Test Claims

Study	Patients Evaluated	*NOTCH3* Exons Evaluated	Results	
Part 1 diagnostic studies				
			Diagnostic Yield	**Specificity**
Mosca et al (2011)	**Patients:** 140 patients with clinical suspicion of CADASIL (Italian and Chinese) **Patient selection:** History of premature strokes; migraine with aura; vascular dementia; suggestive MRI findings; a consistent family history; or a combination of the previous criteria	Direct sequencing of exons 2-8, 10, 14, 19-20, and 22	**Patients:** 14 patients with pathogenic variants located in 10 exons. 126 patients free of pathogenic variants **Family members:** Analysis of 15 additional family members identified 11 of the same pathogenic variants	NR
Lee et al (2009)	**Patients:** 39 patients with suspected CADASIL (China); 100 healthy elderly controls ≥80 y **Patient selection:** Suggestive MRI findings and at least 1 of the following: young age at onset, cognitive decline, psychiatric disorders, or consistent family history	Direct sequencing of exons 2-23	**Patients:** 9 different SNVs identified in 21/39 patients **Family members:** No data for additional family members	100% No variants found in 100 healthy elderly controls
Markus et al (2002)	**Patients:** 83 patients with suspected CADASIL (UK) **Patient selection:** Patients were <60 y old with recurrent lacunar stroke with leukoaraiosis on neuroimaging. Migraine, psychiatric disorders, or dementia could occur but were not essential.	Direct sequencing of exons 3-4; SSCP of exons 2, 5-23	**Patients:** 15 SNVs identified in 48 families with a total of 116 symptomatic patients, 73% in exon 4, 8% in exon 3, and 6% in exons 5 and 6 **Family members:** No data for additional family members	NR
Choi et al (2011)	**Patients:** 151 consecutive Korean patients with acute ischemic stroke **Patient selection:** History of acute ischemic stroke, neurologic exam, cranial computed tomography, or MRI	Bidirectional sequencing of exons 3, 4, 6, 11, and 18	**Patients:** 6 patients (4%) were found with the identical *NOTCH3* mutation (R544C; exon 11). Of these, all had preexisting lacunar infarction, 5 (83.3%) had grade 2-3 white-matter hyperintensity lesions, and a history of hypertension; history of stroke and dementia was higher in patients with	NR

			variants **Family members:** No data for additional family members	
Yin et al (2014)	**Patients:** 47 subjects from 34 families (Chinese) diagnosed with suspected CADASIL **Patient diagnosis/selection:** MRI abnormalities and the presence of >1 typical symptom (eg, migraine, stroke, cognitive deficits, psychiatric symptoms) or the presence of atypical symptoms with a positive family history	Testing method as per Joutel et al; exons 3 and 4 screened first; if no variants detected, remaining exons analyzed	**Patients:** 6 known familial variants were identified in 8 families and 2 novel pathogenic variants were identified in 2 families (exons 3 and 4), and 1 VUS was identified in 1 family (exon 2). Overall *NOTCH3* mutation prevalence: 29.4%.	NR
Abramycheva et al (2015)	**Patients:** 30 unrelated patients with suspected CADASIL	Direct sequencing of exons 2-23 via PCR	**Patients:** 16 SNVs were identified in 18 unrelated patients, 12 of which had been previously described and 4 were novel (*C194G*, *V252M*, *C338F*, and *C484G)*	NR
Maksemous et al (2016)	Patients: 44 patients with suspected clinical diagnosis of CADASIL previously screened for standard sequencing exons (3 and 4) and/or (2,11, 18 and 19) by Sanger sequencing and classified as being negative for known pathogenic variants	Custom NGS panel	Patients: 6 typical CADASIL pathogenic variants were identified in 7 patients out of 44 total patients.	NR

Part 2 Clinical Validity Studies

			Sensitivity	**Specificity**
Study	**Patients Evaluated**	***NOTCH*3 Exons Evaluated**	**Results**	
Peters et al (2005)	**Patients:** 125 unrelated patients diagnosed with CADASIL **Patient diagnosis/selection:** Skin biopsy-proven CADASIL patients	Bidirectional sequencing of all exons	**Sensitivity:** 96% **Patients:** 54 distinct variants in 120 (96.0%) of the 125 patients. In 5 patients (4.0%), no variant was identified. **Family members:** No data for additional family patients	NR
Tikka et al (2009)	**Patients:** 131 patients from 28 families diagnosed with CADASIL (Finnish, Swedish, French) **Patient diagnosis/selection:** EM	Direct sequencing of exons 2-24	**Sensitivity:** 100% **Patients:** 131 CADASIL patients were mutation-positive **Family members:** No data for additional family patients	100% No pathogenic variants were found in the 26

	examination of skin biopsy was performed; 26 asymptomatic controls from CADASIL families		• No pathogenic variant reporting per family or per unrelated individual	negative controls
Dotti et al (2005)	**Patients:** 28 unrelated, consecutively diagnosed patients with CADASIL (Italian) **Patient diagnosis/selection:** Patients were diagnosed via clinical and MRI criteria	DHPLC, followed by confirmatory sequencing of identified pathogenic variants	**Sensitivity:** 100% **Patients:** All 28 patients had pathogenic variants	NR
Joutel et al (1997)	**Patients:** 50 unrelated patients with a clinical suspicion of CADASIL and 100 healthy controls **Patient diagnosis/selection:** History of recurrent strokes, migraine with aura, vascular dementia, or a combination; brain MRI with suggestive findings; and a consistent familial history	SSCP or heteroduplex analysis of all exons, followed by confirmatory sequencing of identified variants	**Sensitivity:** 90% **Patients:** 45/50 CADASIL patients had variants	100% No variants were found in 100 healthy controls

DHPLC: denaturing high-performance liquid chromatography; EM: electron microscope; MRI: magnetic resonance imaging; SSCP: single-stranded conformational polymorphism; VUS: variant of uncertain significance.

Section Summary: Clinical Validity

The clinical sensitivity of genetic testing is high given that *NOTCH*3 is the only gene for which pathogenic variants are known to cause CADASIL. In clinical situations where diagnosis of CADASIL cannot be confirmed by other methods (clinical presentation, magnetic resonance imaging [MRI] findings, skin biopsy), identification of a pathogenic variant in *NOTCH*3 establishes a diagnosis of CADASIL.

Clinically Useful

A test is clinically useful if the use of the results informs management decisions that improve the net health outcome of care. The net health outcome can be improved if patients receive correct therapy, or more effective therapy, or avoid unnecessary therapy, or avoid unnecessary testing.

Direct Evidence

Direct evidence of clinical utility is provided by studies that have compared health outcomes for patients managed with and without the test. Because these are intervention studies, the preferred evidence would be from randomized controlled trials.

The clinical specificity of genetic testing for CADASIL is high, and false-positive results have not been reported in studies of clinical validity. Therefore, a positive genetic test in a patient with clinical signs and symptoms of CADASIL is sufficient to confirm the diagnosis with a high degree of certainty. The clinical sensitivity is also relatively high, in the range of 90% to 100%

for patients with a clinical diagnosis of CADASIL. This indicates that a negative test reduces the likelihood that CADASIL is present. However, since false negative tests do occur, a negative test is less definitive in ruling out CADASIL. Whether a negative test is sufficient to rule out CADASIL depends on the pretest likelihood that CADASIL is present.

Pescini et al (2012) published a study that attempted to identify clinical factors that increase the likelihood of a pathologic variant being present and therefore might be helpful in selecting patients for testing. The authors first performed a systematic review to determine the frequency with which clinical and radiologic factors were associated with a positive genetic test. Evidence was identified from 15 clinical series of patients with CADASIL. Table 3 summarizes the pooled frequency of clinical and radiologic features.

Table 3: Clinical and Radiologic Features in Patients with *NOTCH3* Variants

Features	Number with *NOTCH3* Variant	Percent with *NOTCH3* Variant, %	Points
Clinical			
• Migraine	239/463	52	1
• Migraine with aura	65/85	76	3
• Transient ischemic attack/stroke	380/526	72	1 (2 if <50 y)
• Psychiatric disturbance	106/380	28	1
• Cognitive decline	188/434	43	3
Radiologic			
• LE	277/277	100	3
• LE extended to temporal pole	174/235	74	1
• LE extended to external capsule	228/303	75	5
• Subcortical infarcts	210/254	83	2

LE: leukoencephalopathy.

Using these frequencies, a preliminary scoring system was developed and tested in 61 patients with *NOTCH3* pathogenic variants, and in 54 patients with phenotypic features of CADASIL who were *NOTCH3*-negative. With the addition of family history, and age at onset of transient ischemic attack (TIA)/stroke, a scoring system was developed, as provided in Table 3. The authors recommended that a total score of 14 be used to select patients for testing, as this score resulted in a high sensitivity (96.7%) and a moderately high specificity (74.2%).

Currently, no specific clinical treatment for CADASIL has established efficacy. Supportive care in the form of practical help, emotional support, and counseling are appropriate for affected individuals and their families. Four studies were found that addressed the efficacy of potential treatments for CADASIL.

A 2008 double-blind, placebo controlled trial by Dichgans et al, evaluated the efficacy and safety of donepezil hydrochloride (HCl) in individuals with CADASIL. The trial showed donepezil HCl had no effect on the primary cognitive end point, the V-ADAS-cog score in patients with CADASIL who had cognitive impairment.

Another study, by Huang et al (2010), assessed the efficacy and tolerance of a 24-week treatment with 250 mg/d to improve cerebral hemodynamics in CADASIL patients (n=16). Treatment with ACZ resulted in a significant increase of mean blood flow velocity (MFV) in the middle cerebral artery (MCA) compared with MFV in the MCA at rest before treatment (57.68±12.7 cm/s vs 67.12±9.4 cm/s; p=0.001). During the treatment period, none of the subjects developed new neurologic symptoms, and the original symptoms in these patients, such as headaches and dizziness, were relieved.

A third study, by Peters et al (2007), evaluated the use of HMG-CoA-reductase-inhibitors (statins) in 24 CADASIL subjects treated with atorvastatin for eight weeks. Treatment was started with 40 mg, followed by a dosage increase to 80 mg after four weeks. Transcranial Doppler sonography measuring MFV in the MCA was performed at baseline and at the end of the treatment period. There was no significant treatment effect on MFV (p=0.5) or cerebral vasoreactivity, as assessed by hypercapnia (p=0.5) and intravenous L-arginine (p=0.4) in the overall cohort. However, an inverse correlation was found between vasoreactivity at baseline and changes of both CO2- and L-arginine-induced vasomotor response (both $p<0.05$). Short-term treatment with atorvastatin resulted in no significant improvement of hemodynamic parameters in the overall cohort of CADASIL subjects.

De Maria et al (2014) reported the results of a randomized, double-blinded trial of sapropterin compared with placebo for adults with CADASIL. Sapropterin is a synthetic analog of tetrahydrobiopterin, which is an essential cofactor in nitric oxide synthesis in endothelial cells. Given nitric oxide's role in cerebrovascular function, the authors hypothesized that sapropterin supplementation would improve cerebral endothelium-dependent vasodilation in CADASIL patients. Endothelial dysfunction was assessed using the reactive hyperemia peripheral arterial tonometry (RH-PAT) response, which has been shown to be impaired in patients with CADASIL syndrome. Peripheral arterial tonometry (PAT) is a noninvasive, quantitative test that measures changes in digital pulse volume during reactive hyperemia (RH) and evaluates the endothelial function of resistance arteries and nitric oxide-mediated changes in microvascular response. The study randomized 61 subjects from 38 families, 32 to sapropterin and 29 to placebo. In intention-to-treat analysis, there was no significant difference in change in RH-PAT response (mean difference in RH-PAT change, 0.19: 95% confidence interval, -0.18 to 0.56). Both groups demonstrated improvements in RH-PAT values over the course of the study, but after results were adjusted for age, sex, and clinical characteristics, the improvement was not associated with treatment.

Chain of Evidence
Indirect evidence on clinical utility rests on clinical validity. If the evidence is insufficient to demonstrate test performance, no inferences can be made about clinical utility.

Genetic testing of individuals with suspected CADASIL may have clinical utility by:

- Establishing a diagnosis of CADASIL in an individual with signs and symptoms of the disease, particularly when other disorders are being considered, without the need for a skin biopsy.

- Informing the reproductive decision-making process in preimplantation testing, prenatal (in utero) testing, or altering reproductive planning decisions when a *NOTCH3* pathogenic variant is present in a parent. Preimplantation testing is addressed in medical policy #593 - Preimplantation Genetic Testing.

Section Summary: Clinically Useful
Direct evidence for the clinical utility of genetic testing of individuals with suspected CADASIL is lacking. No specific clinical treatment for CADASIL has established efficacy. However, a chain of evidence for the clinical validity of *NOTCH3* pathogenic variants in establishing diagnosis of a CADASIL leading to initiation of supportive care in the form of practical help, emotional support and counseling may provide a chain of evidence for potential clinical utility.

Targeted Familial Variant Testing in Asymptomatic Patients with Relatives who have CADASIL Syndrome
Clinical Context and Test Purpose
The purpose of targeted familial variant testing of asymptomatic individuals with family members with CADASIL is to screen at-risk individuals and predict development of disease, determine the need for surveillance and aid in reproductive planning.

The question addressed in this evidence review is: In an asymptomatic patient with relatives who have CADASIL syndrome, does use of targeted genetic testing for a known familial variant lead to improved outcomes, including changes in surveillance, preimplantation genetic testing to determine likelihood of an affected offspring or alter reproductive planning decisions?

The following PICOTS were used to select literature to inform this review.

Patients
The relevant population of interest includes asymptomatic relatives of patients with CADASIL.

Interventions
The relevant intervention of interest is targeted familial variant testing of *NOTCH3*.

Comparators
The relevant comparator of interest is standard clinical management without genetic testing.

Outcomes
The potential beneficial outcomes of primary interest would be confirming or excluding the need for surveillance or changes in reproductive decision making. A negative genetic test result would eliminate the need for surveillance to detect development of symptoms and disease. A positive genetic test result would confirm a need for active surveillance and also inform the reproductive decision process.

Potential harmful outcomes are those resulting from a false-positive or false-negative test results. False-positive test results can lead to unnecessary medical or neurological surveillance of

asymptomatic individuals. False-negative test results can lead to lack of medical or neurological surveillance.

Timing
The time frame for outcomes measures varies from short-term surveillance of asymptomatic individuals for development of signs or symptoms of CADASIL to long-term development of disease.

Setting
Asymptomatic individuals with family members with CADASIL may be referred to medical geneticist for investigation of genetic status for carrying a known familial variant. Referral for genetic counseling is important for explanation of genetic disease, heritability, genetic risk, test performance, and possible outcomes.

Technically Reliable
Assessment of technical reliability focuses on specific tests and operators and requires review of unpublished and often proprietary information. Review of specific tests, operators, and unpublished data are outside the scope of this evidence review, and alternative sources exist. This evidence review focuses on the clinical validity and clinical utility.

Clinically Valid
A test must detect the presence or absence of a condition, the risk of developing a condition in the future, or treatment response (beneficial or adverse).

See the Clinical Validity discussion in the Testing Individuals with Suspected CADASIL section.

Testing Strategy
Identification of a *NOTCH3* pathogenic variant establishes a diagnosis of CADASIL in both symptomatic and asymptomatic individuals. For testing in asymptomatic individuals with family members with CADASIL:

- When the proband's *NOTCH3* pathogenic variant is known, targeted familial variant testing to determine genetic status

The testing strategy described here is a general approach for targeted genetic testing for a known pathogenic variant previously identified in a family member (familial variant) with CADASIL.

Clinically Useful
A test is clinically useful if the use of the results informs management decisions that improve the net health outcome of care. The net health outcome can be improved if patients receive correct therapy, or more effective therapy, or avoid unnecessary therapy, or avoid unnecessary testing.

Direct Evidence

Direct evidence of clinical utility is provided by studies that have compared health outcomes for patients managed with and without the test. Because these are intervention studies, the preferred evidence would be from randomized controlled trials.

No randomized trials were identified addressing outcomes managed with CADASIL testing.

Chain of Evidence

Indirect evidence on clinical utility rests on clinical validity. If the evidence is insufficient to demonstrate test performance, no inferences can be made about clinical utility.

Genetic testing of asymptomatic individuals with family members with CADASIL may have clinical utility by:

- Confirming or excluding the need for surveillance based on the presence or absences of a known familial variant.
- Informing the reproductive decision making process in preimplantation testing, prenatal (in utero) testing or altering reproductive planning decisions when a known *NOTCH3* familial variant is present in a parent. Preimplantation testing is addressed in medical policy #593 Preimplantation Genetic Testing.

Genetic counseling is recommended to discuss the impact of positive or negative test results, followed by molecular testing if desired. At present, for an asymptomatic individual, knowledge of familial variant status will generally not lead to any management changes that can prevent or delay the onset of the disorder. Avoiding tobacco use may be one factor that delays onset of disease, but this is a general recommendation that is not altered by genetic testing. However, a negative test may preclude the need for surveillance for complications. Genetic testing may also assist reproductive decision making.

A chain of evidence can be constructed to demonstrate that identification of a NOTCH3 pathogenic variant predicts future development of CADASIL in an asymptomatic individual, eliminates the need for additional diagnostic testing, allows for earlier monitoring for development of systems, aids in reproductive planning, and helps determine the likelihood of an affected offspring.

Section Summary: Clinically Useful

Direct evidence for the clinical utility of genetic testing of asymptomatic relatives of patients with CADASIL is lacking. No specific clinical treatment for CADASIL has established efficacy. However, a chain of evidence can be developed to for potential clinical utility, particularly for reproductive decision-making process for preimplantation and/or prenatal testing.

Genetic Testing of *NOTCH3* in Asymptomatic Patients with Relatives who have CADASIL and Unknown Genetic Status

Clinical Context and Test Purpose

The purpose of genetic testing of *NOTCH3* in asymptomatic individuals with family members with CADASIL who genetic status is unknown is to screen at-risk individuals and predict development of disease, determine the need for surveillance and aid in reproductive planning.

The question addressed in this evidence review is: In an asymptomatic patient with relatives who have CADASIL and whose genetic status is unknown, does use of *NOTCH*3 genetic testing lead to improved outcomes, including changes in surveillance, preimplantation genetic testing to determine likelihood of an affected offspring or alter reproductive planning decisions?

The following PICOTS were used to select literature to inform this review.

Patients
The relevant population of interest includes asymptomatic patients with relatives who have CADASIL and whose genetic status is unknown.

Interventions
The relevant intervention of interest is genetic testing of NOTCH3 variants.

Comparators
The relevant comparator of interest is standard clinical management without genetic testing.

Outcomes
The potential beneficial outcomes of primary interest would be confirming or excluding the need for surveillance or changes in reproductive decision making. A negative genetic test result would eliminate the need for surveillance to detect development of symptoms and disease. A positive genetic test result would confirm a need for active surveillance and also inform the reproductive decision process.

Potential harmful outcomes are those resulting from a false-positive or false-negative test results. False-positive test results can lead to unnecessary medical or neurological surveillance of asymptomatic individuals. False-negative test results can lead to lack of medical or neurological surveillance.

Timing
The time frame for outcomes measures varies from short-term surveillance of asymptomatic individuals for development of signs or symptoms of CADASIL to long-term development of disease.

Setting
Asymptomatic individuals with family members with CADASIL may be referred to medical geneticist for investigation of genetic status for carrying a known familial variant. Referral for genetic counseling is important for explanation of genetic disease, heritability, genetic risk, test performance, and possible outcomes.

Technically Reliable
Assessment of technical reliability focuses on specific tests and operators and requires review of unpublished and often proprietary information. Review of specific tests, operators, and unpublished data are outside the scope of this evidence review, and alternative sources exist. This evidence review focuses on the clinical validity and clinical utility.

Clinically Valid
A test must detect the presence or absence of a condition, the risk of developing a condition in the future, or treatment response (beneficial or adverse).

See the Clinical Validity discussion in the Testing Individuals with Suspected CADASIL section.

Testing Strategy
For testing in asymptomatic individuals with family members who have CADASIL whose genetic status is unknown:

- Perform targeted sequencing and analysis of specific NOTCH3 exons (e.g., exon 4 only, exons 2- 6) OR
- Perform general testing of *NOTCH3* exons (e.g., exons 2 – 24 or all 33 exons)

The testing strategy is a general approach for genetic testing for *NOTCH3* to perform sequence analysis of multiple *NOTCH3* exons to identify pathogenic variants.

Clinically Useful
A test is clinically useful if the use of the results informs management decisions that improve the net health outcome of care. The net health outcome can be improved if patients receive correct therapy, or more effective therapy, or avoid unnecessary therapy, or avoid unnecessary testing.

Direct Evidence
Direct evidence of clinical utility is provided by studies that have compared health outcomes for patients managed with and without the test. Because these are intervention studies, the preferred evidence would be from randomized controlled trials.

No randomized trials were identified addressing outcomes managed with CADASIL testing.

Chain of Evidence
Indirect evidence on clinical utility rests on clinical validity. If the evidence is insufficient to demonstrate test performance, no inferences can be made about clinical utility.

Genetic testing of asymptomatic individuals with family members with CADASIL may have clinical utility by:

- Confirming or excluding the need for surveillance based on the presence or absence of a *NOTCH3* pathogenic variant.
- Informing the reproductive decision making process in preimplantation testing, prenatal (in utero) testing or altering reproductive planning decisions when a known *NOTCH3* pathogenic variant is present in a parent. Preimplantation testing is addressed in medical policy #593 Preimplantation Genetic Testing.

Section Summary: Clinically Useful

Similar to the case where there is a known family variant associated with CADASIL, direct evidence for the clinical utility of genetic testing of asymptomatic relatives of patients with CADASIL is lacking. However, a chain of evidence can be developed to support the clinical utility of testing, as outlined above.

Summary of Evidence

For individuals with suspected CADASIL syndrome who receive genetic testing of *NOTCH*3, the evidence includes case reports, case series and genotype-phenotype correlations evaluating the clinical validity and yield of genetic testing for *NOTCH*3. Relevant outcomes are overall survival, test accuracy and validity, measures, changes in reproductive decision making, change in disease status, and morbid events. The clinical validity studies demonstrate that a *NOTCH*3 pathogenic variant is found in a high percentage of patients with a clinical diagnosis of CADASIL, with studies reporting a clinical sensitivity of 90% to 100%. Limited data on specificity is from testing small numbers of healthy controls, and no false-positive *NOTCH*3 pathogenic variants have been reported in these populations. The diagnostic yield studies report a variable diagnostic yield, ranging from 10% to 54%. These lower numbers likely reflect testing in heterogeneous populations that include patients with other disorders. No direct evidence was identified demonstrating outcome improvements associated with genetic testing for CADASIL. However, a chain of evidence can be constructed to demonstrate that identification of a *NOTCH*3 pathogenic variant establishes the diagnosis of CADASIL without the need for a skin biopsy and reduces the need for other diagnostic tests used in the exclude other conditions in the differential diagnosis. The evidence is sufficient to determine that the technology results in a meaningful improvement in the net health outcome.

For individuals who are asymptomatic with family members with CADASIL syndrome who receive targeted genetic testing for a known *NOTCH*3 familial variant, the evidence is limited. Relevant outcomes are overall survival, test accuracy and validity, changes in reproductive decision making, change in disease status, and morbid events. For asymptomatic family members of an individual with known CADASIL, knowledge of the presence of a familial variant may lead to changes in lifestyle decisions for the affected individual (e.g., reproduction, employment). However, the impact of these lifestyle decisions on health outcomes is uncertain, and there are no interventions for asymptomatic individuals that are known to delay or prevent the onset of disease. A chain of evidence can be constructed to demonstrate that identification of a *NOTCH*3 familial variant predicts future development of CADASIL in an asymptomatic individual, eliminates the need for additional diagnostic testing, allows for earlier monitoring for development of systems, aids in reproductive planning and helps determine the likelihood of an affected offspring. The evidence is sufficient to determine that the technology results in a meaningful improvement in the net health outcome.

For individuals who are asymptomatic with family members with CADASIL syndrome whose genetic status is unknown who receive genetic testing of *NOTCH*3, the evidence is limited. Relevant outcomes are overall survival, test accuracy and validity, changes in reproductive decision making, change in disease status, and morbid events. For asymptomatic family members of an individual with known CADASIL whose genetic status is unknown, knowledge of the presence of a *NOTCH*3 pathogenic variant may lead to changes in lifestyle decisions for

the affected individual (e.g., reproduction, employment). However, the impact of these lifestyle decisions on health outcomes is uncertain, and there are no interventions for asymptomatic individuals that are known to delay or prevent the onset of disease. A chain of evidence can be constructed to demonstrate that identification of a *NOTCH*3 pathogenic variant predicts future development of CADASIL in an asymptomatic individual, eliminates the need for additional diagnostic testing, allows for earlier monitoring for development of systems, aids in reproductive planning and helps determine the likelihood of an affected offspring. The evidence is sufficient to determine that the technology results in a meaningful improvement in the net health outcome.

Practice Guidelines and Position Statements

European Federation of Neurological Societies

The European Federation of Neurological Societies' 2010 guideline on the molecular diagnosis of channelopathies, epilepsies, migraine, stroke, and dementias notes that most *NOTCH3* pathogenic variants occur within exons 3 and 4 and suggests direct sequencing of these two exons if clinical suspicion is high.

U.S. Preventive Services Task Force Recommendations

Not applicable.

Key Words:

CADASIL, *NOTCH3,* Cerebral autosomal dominant arteriopathy

Approved by Governing Bodies:

Clinical laboratories may develop and validate tests in-house and market them as a laboratory service; laboratory-developed tests (LDTs) must meet the general regulatory standards of the Clinical Laboratory Improvement Act (CLIA). Genetic testing of *NOTCH3* is available under the auspices of CLIA. Laboratories that offer LDTs must be licensed by CLIA for high-complexity testing. To date, the U.S. Food and Drug Administration has chosen not to require any regulatory review of this test.

Benefit Application:

Coverage is subject to member's specific benefits. Group specific policy will supersede this policy when applicable.

ITS: Home Policy provisions apply.

FEP: Special benefit consideration may apply. Refer to member's benefit plan. FEP does not consider investigational if FDA approved and will be reviewed for medical necessity.

Current Coding:

CPT Codes:

81406 *NOTCH3 (notch 3)* (e.g., cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy [CADASIL]), targeted sequence analysis (e.g., exons 1-23) **(Effective 1/1/2013)**

References:

1. Abramycheva N, Stepanova M, Kalashnikova L, et al. New mutations in the Notch3 gene in patients with cerebral autosomal dominant arteriopathy with subcortical infarcts and leucoencephalopathy (CADASIL). J Neurol Sci. Feb 15 2015;349(1-2):196-201.
2. Brulin P, Godfraind C, Leteurtre E, et al. Morphometric analysis of ultrastructural vascular changes in CADASIL: analysis of 50 skin biopsy specimens and pathogenic implications. Acta Neuropathol. Sep 2002; 104(3):241-248.
3. Burgunder JM, Finsterer J, Szolnoki Z, et al. EFNS guidelines on the molecular diagnosis of channelopathies, epilepsies, migraine, stroke, and dementias. Eur J Neurol. May 2010; 17(5):641-648.
4. Chabriat H, Joutel A, Dichgans M, et al. Cadasil. Lancet Neurol. Jul 2009;8(7):643-653.
5. Choi JC, Lee KH, Song SK, et al. Screening for *NOTCH3* Gene Mutations Among 151 Consecutive Korean Patients With Acute Ischemic Stroke. J Stroke Cerebrovasc Dis. Nov 29 2013; 22(5):608-614.
6. De Maria R, Campolo J, Frontali M, et al. Effects of Sapropterin on Endothelium-Dependent Vasodilation in Patients With CADASIL: A Randomized Controlled Trial. Stroke. Sep 2 2014.
7. del Rio-Espinola A, Mendioroz M, Domingues-Montanari S, et al. CADASIL management or what to do when there is little one can do. Expert Rev Neurother. Feb 2009; 9(2):197-210.
8. Dichgans M, Markus HS, Salloway S, et al. Donepezil in patients with subcortical vascular cognitive impairment: a randomised double-blind trial in CADASIL. Lancet Neurol. Apr 2008; 7(4):310-318.
9. Donahue CP, Kosik KS. Distribution pattern of Notch3 mutations suggests a gain-of-function mechanism for CADASIL. Genomics. Jan 2004; 83(1):59-65.
10. Dotti MT, Federico A, Mazzei R, et al. The spectrum of *Notch3* mutations in 28 Italian CADASIL families. J Neurol Neurosurg Psychiatry. May 2005; 76(5):736-738.
11. Hayes, Inc. Cerebral Autosomal Dominant arteriopathy with Subcortical Infarcts and Leukoencephalopathy (CADSIL). Hayes GTE Synopsis. April 16, 2008. www.hayes.com. Accessed June 2010.
12. Huang L, Yang Q, Zhang L, et al. Acetazolamide improves cerebral hemodynamics in CADASIL. J Neurol Sci. May 15 2010; 292(1-2):77-80.
13. Joutel A, Favrole P, Labauge P, et al. Skin biopsy immunostaining with a *Notch3* monoclonal antibody for CADASIL diagnosis. Lancet. Dec 15 2001; 358(9298):2049-2051.
14. Joutel A, Vahedi K, Corpechot C, et al. Strong clustering and stereotyped nature of Notch3 mutations in CADASIL patients. Lancet. Nov 22 1997; 350(9090):1511-1515.

15. Lee YC, Liu CS, Chang MH, et al. Population-specific spectrum of *NOTCH3* mutations, MRI features and founder effect of CADASIL in Chinese. J Neurol. Feb 2009; 256(2):249-255.
16. Lesnik Oberstein SA, van Duinen SG, van den Boom R, et al. Evaluation of diagnostic NOTCH3 immunostaining in CADASIL. Acta Neuropathol. Aug 2003; 106(2):107-111.
17. Lesnik Oberstein SAJ, Boon EMJ, Dichgans M. CADASIL. In: Pagon RA, Bird TD, Dolan CR, et al., eds. GeneReviews. Seattle WA: University of Washington, Seattle; 1993.
18. Maksemous N, Smith RA, Haupt LM, et al. Targeted next generation sequencing identifies novel NOTCH3 gene mutations in CADASIL diagnostics patients. Hum Genomics. Nov 24 2016; 10(1):38.
19. Malandrini A, Gaudiano C, Gambelli S, et al. Diagnostic value of ultrastructural skin biopsy studies in CADASIL. Neurology. Apr 24 2007; 68(17):1430-1432.
20. Markus HS, Martin RJ, Simpson MA, et al. Diagnostic strategies in CADASIL. Neurology. Oct 22 2002; 59(8):1134-1138.
21. Mosca L, Marazzi R, Ciccone A, et al. NOTCH3 gene mutations in subjects clinically suspected of CADASIL. J Neurol Sci. Aug 15 2011; 307(1-2):144-148.
22. Muqtadar H, Testai FD. Single gene disorders associated with stroke: a review and update on treatment options. Curr Treat Options Cardiovasc Med. Jun 2012; 14(3):288-297.
23. Oberstein SAJL, Boon EMJ, Dichgans. CADSIL. GeneReviews NCBI Bookshelf. www.ncbi.nlm.nih.gov/bookshelf/br.fcgi?book=gene&part=cadasil. Accessed June 2010
24. Opherk C, Gonik M, Duering M, et al. Genome-wide genotyping demonstrates a polygenic risk score associated with white matter hyperintensity volume in CADASIL. Stroke. Apr 2014; 45(4):968-972.
25. Pescini F, Nannucci S, Bertaccini B, et al. The Cerebral Autosomal-Dominant Arteriopathy With Subcortical Infarcts and Leukoencephalopathy (CADASIL) Scale: a screening tool to select patients for NOTCH3 gene analysis. Stroke. Nov 2012; 43(11):2871-2876.
26. Peters N, Freilinger T, Opherk C, et al. Effects of short term atorvastatin treatment on cerebral hemodynamics in CADASIL. J Neurol Sci. Sep 15 2007; 260(1-2):100-105.
27. Peters N, Opherk C, Bergmann T, et al. Spectrum of mutations in biopsy-proven CADASIL: implications for diagnostic strategies. Arch Neurol. Jul 2005; 62(7):1091-1094.
28. Rutten J, Lesnik Oberstein SAJ. CADASIL. In: Adam MP, Ardinger HH, Pagon RA, et al., eds. GeneReviews. Seattle, WA: University of Washington; 2016.
29. Tikka S, Mykkanen K, Ruchoux MM, et al. Congruence between *NOTCH3* mutations and GOM in 131 CADASIL patients. Brain. Apr 2009; 132(Pt 4):933-939.
30. Yin X, Wu D, Wan J, et al. Cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy: Phenotypic and mutational spectrum in patients from mainland China. Int J Neurosci. Sep 1 2014.

Policy History:

Medical Policy Group, June 2010 **(2)**
Medical Policy Administration Committee, June 2010
Available for comment June 18-August 2, 2010 – Added coverage for CADASIL and Key Points for CADASIL
Medical Policy Group, September 2011 **(1):** Update to Key Points and References related to CADASIL
Medical Policy Panel, September 2012
Medical Policy Panel, October 2013
Medical Policy Panel, October 2014
Medical Policy Panel, October 2015
Medical Policy Group, January 2016 **(3):** Creation of individual policy with all references related to genetic testing for CADASIL Syndrome/*NOTCH3* mutation testing removed from medical policy #136; update to Description, Key Points, Key Words, Codes (added code 81406) and References; no change to policy statement – added considered investigational for all other indications outside of criteria noted
Medical Policy Panel, April 2017
Medical Policy Group, May 2017 **(3):** 2017 Updates to Description, Key Points & References; policy statement updated to clarify language & add coverage criteria for asymptomatic primary family members with or without a known *NOTCH3* variant
Medical Policy Administration Committee, May 2017
Available for comment May 12 through June 25, 2017
Medical Policy Panel, April 2018
Medical Policy Group, May 2018 **(4):** Updates to Key Points, Coding, and References. Removed Previous Coding section. CPT codes were deleted in 2013.
