Neurovascular Diseases and Stroke Gene Panel

Genes Tested:

ABCC6, ACTA2, ACVRL1, ADA2, ATP1A2, ATP7A, ATR, BRAF, CACNA1A, CBS, CCM2, CENPJ, CEP152, CEP63, CHD4, CLDN14, CNOT3, COL3A1, COL4A1, COL4A2, COLGALT1, EFNB2, ENG, EPHA4, EPHB4, FBN1, G6PC, GDF2, GLA, GUCY1A3, HBB, HRAS, HTRA1, JAG1, KRAS, KRIT1, MAP2K1, MYH11, MYLK, NF1, NHLRC2, NIN, NOTCH2, NOTCH3, NRAS, OTC, P2RY1, P2RY12, PCNT, PDCD10, PMM2, POLG, PRRT2, PTPN11, RAF1, RASA1, RBBP8, RNF213, SAMHD1, SCN1A, SCN5A, SETD5, SLC19A2, SLC2A10, SMAD2, SMAD3, SMAD4, SMARCAL1, SOS1, SUOX, TGFB2, TGFB3, TGFBR1, TGFBR2, TREX1, TSC1, TSC2, TTC19, WFS1, YY1AP1

Description:

Pediatric ischemic stroke affects up to 1.6 in 100,000 children per year (Mallik et al. 2014) and may be caused by one of a number of different genetic etiologies. Clinical manifestations of cerebrovascular disorders may include cerebral embolism, thrombosis, seizures, hemorrhage, aneurysm, cerebral arteriopathies, or venous anomalies. Risk factors may be structural or hematologic and include hemoglobinopathies, coagulopathies, congenital heart disease, hereditary hemorrhagic telangiectasia, familial hemiplegic migraine, moyamoya, and vasculopathies (Doig et al. 2020, Kirkham 2003, McCrea et al. 2018). Identifying a genetic etiology may assist in medical management and recurrence risk estimates. The Neurovascular Diseases and Stroke Gene Panel utilizes exome sequencing technology to analyze 80 genes involved with pediatric neurovascular diseases and stroke.

Indications:

- Moyamoya
- Childhood arterial ischemic stroke
- Pediatric hemorrhagic stroke
- Cerebral arteriopathies: aneurysm, stenosis, dissection, vasospasm
- Familial hemiplegic migraine
- Hereditary hemorrhagic telangiectasia
- Head, neck and spine vascular or veno-lymphatic malformation
- Carotid or vertebral artery aneurysm
- Brain structural vascular abnormalities
    - Arterial tortuosity syndrome
    - Cerebral cavernous malformation
    - Arteriovenous malformation
    - Arteriovenous fistula
    - Cerebral proliferative angiopathy
    - Sinus pericranii
- Intracranial aneurysm
- Cerebral venous thrombosis

What Is Reported?

Variants that will be discussed in detail in the report:

- Pathogenic/likely pathogenic variants: Variants that are known to be pathogenic or for which the laboratory has sufficient evidence suggesting pathogenicity.

Variants that will be listed in the report:

- Variants of uncertain clinical significance

What is not reported?

- Variants in genes not included in the predefined gene list
- Variants where there is currently no evidence of association with the disease and that are identified in healthy individuals (benign or likely benign variants)

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changing the outcome together

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Note: Neurovascular Diseases and Stroke Panel cases with negative or uncertain findings can be reflexed to Whole Exome Sequencing (WES). A separate test order and a signed consent form is required for all WES testing. In addition, including biological parental samples is strongly encouraged to assist with the analysis of WES and to increase test yield. Reflex to WES orders can either be placed simultaneously or separately. Separate reflex to WES orders are subject to review prior to the initiation of testing. Please see our website at www.cincinnatichildrens.org/exome to obtain a WES test requisition and consent form.

Genetic Conditions CommonlyAssociated with Pediatric Neurovascular Diseases and Stroke

Gene	Inheritance	Condition
ABCC6	AR, AD	Generalized arterial calcification of infancy type 2 (AR); pseudoxanthoma elasticum (AD, AR)
ACTA2	AD	Familial thoracic aortic aneurysm type 6; multisystemic smooth muscle dysfunction syndrome; moyamoya disease 5
ACVRL1	AD	Hereditary hemorrhagic telangiectasia type 2
ADA2	AR	Sneddon syndrome; vasculitis, autoinflammation, immunodeficiency, and hematologic defects syndrome
ATP1A2	AD, AR	Alternating hemiplegia of childhood type 1 (AD); familial migraine (AD); fetal akinesia, respiratory insufficiency, microcephaly, polymicrogyria, and dysmorphic facies (AR)
ATP7A	XLR	Menkes disease; occipital horn syndrome
ATR	AR	Seckel syndrome type 1
BRAF	AD	Cardiofaciocutaneous syndrome; LEOPARD syndrome type 3; Noonan syndrome type 7
CACNA1A	AD	Familial hemiplegic migraine
CBS	AR	Homocystinuria
CCM2	AD	Cerebral cavernous malformations type 2
CENPJ	AR	Seckel syndrome type 4
CEP152	AR	Seckel syndrome type 5
CEP63	AR	Seckel syndrome type 6
CHD4	AD	Sifrim-Hitz-Weiss syndrome
CLDN14	AD	Vein of Galen malformation
CNOT3	AD	Moyamoya disease
COL3A1	AD	Ehlers-Danlos syndrome, vascular type
COL4A1	AD	Small-vessel brain disease
COL4A2	AD	Small-vessel brain disease type 2
COLGALT1	AR	Brain small vessel disease type 3
EFNB2	AD	Vein of Galen malformation
ENG	AD	Hereditary hemorrhagic telangiectasia type 1
EPHA4	AD	Vein of Galen malformation
EPHB4	AD	Capillary malformation-arteriovenous malformation type 2
FBN1	AD	Marfan syndrome
G6PC	AR	Glycogen storage disease Ia
GDF2	AD	Hereditary hemorrhagic telangiectasia type 5
GLA	XL	Fabry disease
GUCY1A3	AR	Moyamoya type 6 with achalasia
HBB	AR	Sickle cell anemia
HRAS	AD	Costello syndrome

Genetic Conditions Commonly Associated with Pediatric Neurovascular Diseases and Stroke (cont.)

Gene	Inheritance	Condition
HTRA1	AR, AD	Cerebral arteriopathy with subcortical infarcts and leukoencephalopathy (CARASIL); Cerebral arteriopathy with subcortical infarcts and leukoencephalopathy type 2 (CADASIL2)
JAG1	AD	Alagille syndrome 1
KRAS	AD	Cardiofaciocutaneous syndrome type 2; Noonan syndrome type 3; RAS-associated autoimmune leukoproliferative disorder
KRIT1	AD	Cerebral cavernous malformations type 1
MAP2K1	AD	Cardiofaciocutaneous syndrome type 3
MYH11	AD	Familial thoracic aortic aneurysm type 4
MYLK	AD	Familial thoracic aortic aneurysm type 7
NF1	AD	Neurofibromatosis type 1
NHLRC2	AR	Fibrosis, neurodegeneration and cerebral angiomatosis (FINCA) syndrome
NIN	AR	Seckel syndrome type 7
NOTCH2	AD	Hajdu-Cheney syndrome; Alagille syndrome type 2
NOTCH3	AD	Cerebral arteriopathy with subcortical infarcts and leukoencephalopathy type 1
NRAS	AD	Noonan syndrome type 6
OTC	XL	Ornithine transcarbamylase deficiency
P2RY1	AD	Ischemic stroke (IS) susceptibility
P2RY12	AD	Ischemic stroke (IS) susceptibility
PCNT	AR	Microcephalic osteodysplastic primordial dwarfism type 2
PDCD10	AD	Cerebral cavernous malformations type 3
PMM2	AR	Congenital disorder of glycosylation type Ia
POLG	AD, AR	Progressive external ophthalmoplegia (AD, AR); Mitochondrial recessive ataxia syndrome (AR); Mitochondrial DNA depletion syndrome 4A (Alpers type) (AR); Mitochondrial DNA depletion syndrome 4B (MNGIE type) (AR)
PRRT2	AD	Familial infantile convulsions with paroxysmal choreoathetosis; Episodic kinesigenic dyskinesia type 1; Benign familial infantile seizures type 2
PTPN11	AD	Noonan syndrome type 1; LEOPARD syndrome type 1; Metachondromatosis
RAF1	AD	Noonan syndrome type 5; Dilated cardiomyopathy, 1NN
RASA1	AD	Capillary malformation-arteriovenous malformation type 1
RBBP8	AR	Seckel syndrome 2
RNF213	AD, AR	Susceptibility to Moyamoya disease type 2
SAMHD1	AR	Aicardi-Goutieres syndrome 5
SCN1A	AD	Familial hemiplegic migraine type 3
SCN5A	AD	Brugada syndrome 1, Heart block, Long QT syndrome 3, Familial atrial fibrillation type 10, Dilated cardiomyopathy
SETD5	AD	Intellectual developmental disorder type 23
SLC19A2	AR	Thiamine-responsive megaloblastic anemia syndrome
SLC2A10	AR	Arterial tortuosity syndrome
SMAD2	AD	Loeys-Dietz syndrome 6
SMAD3	AD	Loeys-Dietz syndrome 3
SMAD4	AD	Juvenile polyposis/hereditary hemorrhagic telangiectasia syndrome

Genetic Conditions Commonly Associated with Pediatric Neurovascular Diseases and Stroke (cont.)

Gene	Inheritance	Condition
SMARCAL1	AR	Schimke immunoosseous dysplasia
SOS1	AD	Noonan syndrome type 4
SUOX	AR	Sulfite oxidase deficiency
TGFB2	AD	Loeys-Dietz syndrome type 4
TGFB3	AD	Loeys-Dietz syndrome type 5; Arrhythmogenic right ventricular dysplasia 1
TGFBR1	AD	Loeys-Dietz syndrome type 1
TGFBR2	AD	Loeys-Dietz syndrome type 2
TREX1	AD	Retinal vasculopathy with cerebral leukoencephalopathy and systemic manifestations; Chilblain lupus
TSC1	AD	Tuberous sclerosis type 1
TSC2	AD	Tuberous sclerosis type 2
TTC19	AR	Nuclear mitochondrial complex III deficiency type 2
WFS1	AR	Wolfram syndrome type 1
YY1AP1	AR	Grange syndrome

AD – autosomal dominant, AR – autosomal recessive, XLR – X-linked recessive, XL – X-linked

Methodology:

Neurovascular Diseases and Stroke Gene Panel uses Human Comprehensive Exome kit from Twist Bioscience to capture the exonic regions of genes from the genomic DNA extracted from the patient. Targeted regions are sequenced using an Illumina sequencing system with paired-end reads. Sequence reads are aligned to the human reference genome (build UCSC hg19). Variants within exons and flanking sequences are identified and evaluated by a validated in-house developed bioinformatics analysis pipeline that includes the usage of GATK and Fabric Genomic Analysis platform. Data quality is assessed to confirm it has a minimum coverage of 20X for >95% of targets of interest.

Technical Limitations:

- Pathogenic variants may be present in a portion of the genes not covered by this test or in regions with suboptimal data due to homologous issue, polynucleotides, or nucleotide repeats, and therefore may not be identified. Thus, the absence of identified pathogenic variants does not exclude the possibility of a genetic etiology for the patient's symptoms.
- Certain types of mutations are not detected. Only single base pair changes or small insertions or deletions of DNA are detected. Large deletions, duplications, or rearrangements, mitochondrial genome mutations, repeat expansions, low level mosaicism and many epigenetic defects may not be detected by this test.
- *HBB* analysis by NGS has limitations: gross deletions/duplications will not be detected; deep intronic, pathogenic variants in untranslated regions might not be detected due to lack of coverage and/or homology issues. However, separate comprehensive hemoglobin gene testing by Sanger sequencing and MLPA are available in our lab for patients with suspected sickle cell disease and one copy of HbS detected.

Regions of Homology

These gene regions with homology may generate suboptimal data with potential false negative results.

GENE	TRANSCRIPT	EXON	CHROM	EXON_START	EXON_END
ABCC6	NM_001171	1	16	16317255	16317291
ABCC6	NM_001079528	2	16	16315424	16315688
ABCC6	NM_001171	2	16	16315505	16315688

Regions of Homology (cont.)

GENE	TRANSCRIPT	EXON	CHROM	EXON_START	EXON_END
ABCC6	NM_001171	3	16	16313678	16313804
ABCC6	NM_001171	4	16	16313410	16313539
ABCC6	NM_001171	5	16	16308180	16308306
ABCC6	NM_001171	6	16	16306041	16306103
ABCC6	NM_001171	7	16	16302584	16302716
ABCC6	NM_001171	8	16	16297266	16297470
NOTCH2	NM_024408	2	1	120572528	120572610
NOTCH2	NM_024408	3	1	120547951	120548211
NOTCH2	NM_024408	4	1	120539619	120539955
YY1AP1	NM_001198903	9	1	155631097	155631214

Low coverage (<20X) regions

GENE	REGION	LENGTH	CHROM	EXON_START	EXON_END
ATP7A	NM_000052:c.121-8_134	22 bps	X	77243730	77243751
NF1	NM_000267:c.3198-3	1 bps	17	29559088	29559088
NRAS	NM_002524:c.112-8_115	12 bps	1	115256596	115256607
OTC	NM_000531:c.205_216+8	20 bps	X	38226671	38226690
OTC	NM_000531:c.299-8_326	36 bps	X	38240587	38240622
OTC	NM_000531:c.1006-7_1006-6	2 bps	X	38280269	38280270
SOS1	NM_005633:c.2168-8	1 bps	2	39239497	39239497
TGFBR1	NM_004612:c.97+2_97+8	7 bps	9	101867586	101867592

Please note: *These regions represent the low coverage (<20X) regions identified during our test validation. For specific patient cases, these regions may vary.*

Turn-Around Time:

56 days (8 weeks)

Specimen:

At least 3 mls whole blood in a lavender top (EDTA) tube or saliva in an Oragene saliva kit . Please call the lab at 513-636-4474 for a free saliva collection kit. Label the tube with the patient's name, birth date, and date of collection. Alternatively, 10 mcg of DNA extracted by a CLIA certified lab may be submitted.

CPT Codes:

Neurovascular Diseases and Stroke Gene Panel:
81404, 81405, 81479(x2)

Shipping Instructions:

Please enclose **test requisition** with sample. **All information must be completed before sample can be processed.**

Place samples in styrofoam mailer and ship at room temperature by overnight Federal Express to arrive Monday through Saturday.

Ship to:

Genetics and Genomics Diagnostic Laboratory
3333 Burnet Avenue NRB 1042
Cincinnati, OH 45229
513-636-4474

References:

Doig et al. 2020. Monogenetic Stroke Syndromes in Children and Young Adults. AJR Am J Roentgenol. 215(3):695-705.

Kirkham, F. 2003. Is there a genetic basis for pediatric stroke? Curr Opin Pediatr. 15(6):547-58.

Mallik et al. 2014. Childhood arterial ischaemic stroke incidence, presenting features, and risk factors: a prospective population-based study. Lancet Neurol. 13(1):35-43.

McCrea et al. 2018. Genetic and Environmental Associations With Pediatric Cerebral Arteriopathy. Stroke. 50(2):257-265.