MAYO CLINIC LABORATORIES

Test Definition: PHEGP

Phenylalanine Disorders Gene Panel

Overview

Useful For

Follow up for abnormal biochemical results suggestive of a phenylalanine disorder

Establishing a molecular diagnosis for patients with phenylalanine disorders

Identifying variants within genes known to be associated with phenylalanine disorders, allowing for predictive testing of at-risk family members

Genetics Test Information

This test utilizes next generation sequencing to detect single nucleotide and copy number variants in 10 genes associated with phenylalanine disorders: *DDC*, *DNAJC12*, *GCH1*, *PAH*, *PCBD1*, *PTS*, *QDPR*, *SLC18A2*, *SPR*, *TH*. See Method Description for additional details.

Identification of a pathogenic variant may assist with diagnosis, prognosis, clinical management, familial screening, and genetic counseling for phenylalanine disorders.

Additional first tier testing may be considered/recommended. For more information see Advisory Information.

Special Instructions

- Molecular Genetics: Biochemical Disorders Patient Information
- Informed Consent for Genetic Testing
- Blood Spot Collection Card-Spanish Instructions
- Blood Spot Collection Card-Chinese Instructions
- Informed Consent for Genetic Testing (Spanish)
- Blood Spot Collection Instructions
- Targeted Genes and Methodology Details for Phenylalanine Disorders Gene Panel

Reflex Tests

Test Id	Reporting Name	Available Separately	Always Performed
FIBR	Fibroblast Culture	Yes	No
CRYOB	Cryopreserve for Biochem Studies	No	No

Method Name

Custom Sequence Capture and Targeted Next-Generation Sequencing followed by Polymerase Chain Reaction (PCR) and Sanger Sequencing

NY State Available

Yes

Specimen

Specimen Type

Varies

Ordering Guidance

The recommended first-tier test for disorders of phenylalanine metabolism is quantitative plasma amino acids (AAQP / Amino Acids, Quantitative, Plasma), as well as neurotransmitters in cerebrospinal fluid and pterin metabolite analysis in blood and urine.

Shipping Instructions

Specimen preferred to arrive within 96 hours of collection.

Specimen Required

Patient Preparation: A previous bone marrow transplant from an allogenic donor will interfere with testing. Call 800-533-1710 for instructions for testing patients who have received a bone marrow transplant.
Specimen Type: Whole blood
Container/Tube:
Preferred: Lavender top (EDTA) or yellow top (ACD)
Acceptable: Any anticoagulant
Specimen Volume: 3 mL
Collection Instructions:
1. Invert several times to mix blood.
2. Send specimen in original tube.
Specimen Stability Information: Ambient (preferred)/Refrigerated

Forms

1. **New York Clients-Informed consent is required.** Document on the request form or electronic order that a copy is on file. The following documents are available in Special Instructions:
-Informed Consent for Genetic Testing (T576)
-Informed Consent for Genetic Testing (Spanish) (T826)
2. Molecular Genetics: Biochemical Disorders Patient Information (T527) in Special Instructions

Reject Due To

All specimens will be evaluated at Mayo Clinic Laboratories for test suitability.

Specimen Minimum Volume

See Specimen Required

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Varies	Varies (preferred)		

Clinical & Interpretive

Clinical Information

Hyperphenylalaninemia is a heterogeneous disorder of phenylalanine catabolism caused by a deficiency of any one of 6 enzymes involved in the conversion of phenylalanine to tyrosine.
Phenylketonuria (PKU) is the most frequent inherited disorder of amino acid metabolism (about 1:10,000-1:15,000) and was the first successfully treated inborn error of metabolism and included in newborn screening programs worldwide. It

is inherited in an autosomal recessive manner and is caused by a defect in the enzyme phenylalanine hydroxylase (PAH), which converts the essential amino acid phenylalanine to tyrosine. Deficiency of PAH results in decreased levels of tyrosine and an accumulation of phenylalanine in blood and tissues. Untreated, PKU leads to severe brain damage with intellectual impairment, behavior abnormalities, seizures, and spasticity. The level of enzyme activity differentiates classic PKU (PAH activity <1%) from other milder forms; however, all are characterized by increased levels of phenylalanine (hyperphenylalaninemia). Treatment includes the early introduction of a diet low in phenylalanine.
Approximately 2% of patients with hyperphenylalaninemia have a deficiency of tetrahydrobiopterin (BH4), which causes a secondary deficit of the neurotransmitters dopamine and serotonin. There are 4 autosomal recessive disorders associated with BH4 deficiency plus hyperphenylalaninemia; guanosine triphosphate cyclohydrolase deficiency (GCH1), 6-pyruvoyl tetrahydropterin synthase deficiency (PTS), dihydropteridine reductase deficiency (QDPR), and pterin-4 alpha carbinolamine dehydratase (PCD) deficiency (PCBD1). This group of disorders, with the exception of PCD, is characterized by progressive dystonia, truncal hypotonia, extremity hypertonia, seizures, and mental retardation though milder presentations exist. PCD has no symptoms other than transient alterations in tone. Treatment may include administration of BH4, L-dopa (and carbidopa) 5-hydroxytryptophan supplements, and a low phenylalanine diet.
Recently, variants in *DNAJC12*, which encodes a heat-shock protein that interacts with the phenylalanine, tyrosine, and tryptophan hydroxylases to help catalyze the conversion of the substrates to their respective products, has been shown to cause hyperphenylalaninemia, progressive neurodegeneration, and dystonia. Treatment may include early administration of BH4 and/or neurotransmitter precursors.
Related additional disorders of neurotransmitter metabolism include:
-Aromatic l-amino acid decarboxylase (AADC) deficiency, caused by variants in *DDC*, is an autosomal recessive inborn error in neurotransmitter metabolism that leads to combined serotonin and catecholamine deficiency.
-Patients with dopa-responsive dystonia due to variants in *SPR* causing sepiapterin reductase deficiency have progressive psychomotor retardation and dystonia.
-Variants in tyrosine hydroxylase (TH) prevent the conversion of L-tyrosine to L-dopa resulting in Segawa syndrome.
-Variants in *SLC18A2*, a vesicular transporter of dopamine, cause infantile parkinsonism-dystonia-2 (PKDYS2)

Reference Values

An interpretive report will be provided.

Interpretation

All detected alterations are evaluated according to American College of Medical Genetics and Genomics (ACMG) recommendations.(1) Variants are classified based on known, predicted, or possible pathogenicity and reported with interpretive comments detailing their potential or known significance.

Cautions

Clinical Correlations:
Test results should be interpreted in context of clinical findings, family history, and other laboratory data. Misinterpretation of results may occur if the information provided is inaccurate or incomplete.
If testing was performed because of a clinically significant family history, it is often useful to first test an affected family member. Detection of a reportable variant in an affected family member would allow for more informative testing of at risk individuals.
To discuss the availability of further testing options, for assistance in general test selection, or for assistance in the interpretation of these results, Mayo Clinic Laboratory genetic counselors can be contacted at 800-533-1710.
Technical Limitations:
Next-generation sequencing may not detect all types of genomic variants. In rare cases, false-negative or false-positive results may occur. The depth of coverage may be variable for some target regions, but assay performance below the

minimum acceptable criteria or for failed regions will be noted. Given these limitations, negative results do not rule out the diagnosis of a genetic disorder. If specific clinical disorders are suspected, evaluation by alternative methods can be considered.

If the patient has had an allogeneic hematopoietic stem cell transplant or a recent heterologous blood transfusion, these results may be inaccurate due to the presence of donor DNA. Call Mayo Clinic Laboratories for instructions for testing patients who have received a bone marrow transplant.

There may be regions of genes that cannot be effectively amplified for sequencing or deletion and duplication analysis as a result of technical limitations of the assay, including regions of homology, high guanine-cytosine (GC) content, and repetitive sequences. Confirmation of select reportable variants will be performed by alternate methodologies based on internal laboratory criteria.

This assay will not reliably detect insertions/deletions (indels) of 40 or more base pairs (bp), including Alu insertions, long interspersed nuclear elements (LINES), and short interspersed nuclear elements (SINES). The bioinformatics software pipeline is verified to detect 95% of deletions up to 75 bp and insertions up to 47 bp.

Additionally, low level mosaic variants may not be detected.

This test is not designed to differentiate between somatic and germline variants. If there is a possibility that any detected variant is somatic, additional testing may be necessary to clarify the significance of results.

Reclassification of Variants-Policy:

At this time, it is not standard practice for the laboratory to systematically review previously classified variants on a regular basis. The laboratory encourages health care providers to contact the laboratory at any time to learn how the status of a particular variant may have changed over time.

Variant Evaluation:

Evaluation and categorization of variants is performed using published American College of Medical Genetics and Genomics (ACMG) and Association for Molecular Pathology (AMP) recommendations as a guideline. Other gene specific guidelines may also be considered. Variants are classified based on known, predicted, or possible pathogenicity and reported with interpretive comments detailing their potential or known significance. Variants classified as benign or likely benign are not reported.

Multiple in silico evaluation tools may be used to assist in the interpretation of these results. The accuracy of predictions made by in silico evaluation tools is highly dependent upon the data available for a given gene, and periodic updates to these tools may cause predictions to change over time. Results from in silico evaluation tools should be interpreted with caution and professional clinical judgment. Intronic and synonymous sequence variants not predicted to impact splicing or otherwise contribute to disease are not reported.

Clinical Reference

1. Richards S, Aziz N, Bale S, et al: Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. Genet Med. 2015 May;17(5):405-424

2. Burgard P, Luo X, Levy HL, Hoffmann GF: Phenylketonuria. In: Sarafoglou K, Hoffmann GF, Roth KS, eds. Pediatric Endocrinology and Inborn Errors of Metabolism. 2nd ed. McGraw-Hill Education; 2017:251-258

3. Blau N, Thony B: Hyperphenylalanemias: Disorders of tetrahydrobiopterin metabolism. In: Sarafoglou K, Hoffmann GF, Roth KS, eds. Pediatric Endocrinology and Inborn Errors of Metabolism. 2nd ed. McGraw-Hill Education; 2017:259-266

4. Anikster Y, Haack TB, Vilboux T, et al: Biallelic mutations in DNAJC12 cause hyperphenylalaninemia, dystonia, and intellectual disability. Am J Hum Genet. 2017;100(2):257-266. doi: 10.1016/j.ajhg.2017.01.002

5. OMIM. Johns Hopkins University; Accessed January 2, 2020. Available at https://omim.org/

Performance

Method Description

Next-generation sequencing (NGS) and/or Sanger sequencing is performed to test for the presence of variants in coding regions and intron/exon boundaries of the genes analyzed. NGS and/or a polymerase chain reaction (PCR)-based quantitative method is performed to test for the presence of deletions and duplications in the genes analyzed.
There may be regions of genes that cannot be effectively amplified for sequencing or deletion and duplication analysis as a result of technical limitations of the assay, including regions of homology, high guanine-cytosine (GC) content, and repetitive sequences. Confirmation of select reportable variants may be performed by alternate methodologies based on internal laboratory criteria.
PCR-based methods and/or Sanger sequencing is used to confirm variants detected by NGS when appropriate.(Unpublished Mayo method)
Genes analyzed: *DDC*, *DNAJC12*, *GCH1*, *PAH*, *PCBD1*, *PTS*, *QDPR*, *SLC18A2*, *SPR*, *TH*

PDF Report

No

Specimen Retention Time

Whole Blood: 2 weeks (if available); Extracted DNA: 3 months

Performing Laboratory Location

Rochester

Fees & Codes

Test Classification

This test was developed, and its performance characteristics determined by Mayo Clinic in a manner consistent with CLIA requirements. This test has not been cleared or approved by the US Food and Drug Administration.

CPT Code Information

81405
81406 x 2
81479

LOINC® Information

Test ID	Test Order Name	Order LOINC Value
PHEGP	Phenylalanine Disorders Gene Panel	In Process

Result ID	Test Result Name	Result LOINC Value
608788	Test Description	62364-5
608789	Specimen	31208-2

608790	Source	31208-2
608791	Result Summary	50397-9
608792	Result	82939-0
608793	Interpretation	69047-9
608794	Resources	In Process
608795	Additional Information	48767-8
608796	Method	85069-3
608797	Genes Analyzed	48018-6
608798	Disclaimer	62364-5
608799	Released By	18771-6