MAYO CLINIC LABORATORIES

Test Definition: PCGP

Porphyria Comprehensive Gene Panel, Varies

Overview

Useful For

Follow up for abnormal biochemical results suggestive of porphyria

Establishing a molecular diagnosis for patients with porphyria

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

Reflex Tests

Test Id	Reporting Name	Available Separately	Always Performed
CULFB	Fibroblast Culture for Genetic Test	Yes	No

Genetics Test Information

This test utilizes next-generation sequencing to detect single nucleotide and copy number variants in 11 genes associated with porphyria: *ALAD*, *ALAS2*, *CLPX*, *CPOX*, *FECH*, *GATA1*, *HFE*, *HMBS*, *PPOX*, *UROD*, and *UROS*. See Targeted Genes and Methodology Details for Porphyria Comprehensive Gene Panel and Method Description for additional details.

Identification of a disease-causing variant may assist with diagnosis, prognosis, clinical management, familial screening, and genetic counseling for porphyria.

The biochemical testing approach for diagnosis of patients with a suspected porphyria is most effective when done in a thoughtful manner. For recommendations for first-tier biochemical testing, the following algorithms are available:
-Porphyria (Acute) Testing Algorithm
-Porphyria (Cutaneous) Testing Algorithm

Testing Algorithm

For skin biopsy or cultured fibroblast specimens, fibroblast culture testing will be performed at an additional charge. If viable cells are not obtained, the client will be notified.

For more information see:
-Porphyria (Acute) Testing Algorithm
-Porphyria (Cutaneous) Testing Algorithm

Special Instructions

- Molecular Genetics: Biochemical Disorders Patient Information
- Informed Consent for Genetic Testing
- Porphyria (Acute) Testing Algorithm
- Porphyria (Cutaneous) Testing Algorithm
- 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 Porphyria Comprehensive Gene Panel

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

NY State Available
Yes

Specimen

Specimen Type
Varies

Ordering Guidance
Customization of this panel and single gene analysis for any gene present on this panel is available. For more information see CGPH / Custom Gene Panel, Hereditary, Next-Generation Sequencing, Varies.

Targeted testing for familial variants (also called site-specific or known mutations testing) is available for the genes on this panel. See FMTT / Familial Variant, Targeted Testing, Varies. To obtain more information about this testing option, call 800-533-1710.

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. For instructions for testing patients who have received a bone marrow transplant, call 800-533-1710.

Submit only 1 of the following specimens:

Specimen Type: Whole blood
Container/Tube: Lavender top (EDTA) or yellow top (ACD)
Specimen Volume: 3 mL
Collection Instructions:
1. Invert several times to mix blood.
2. Send whole blood specimen in original tube. **Do not aliquot**.
Specimen Stability Information: Ambient (preferred) 4 days/Refrigerated 14 days

Specimen Type: Skin biopsy
Supplies: Fibroblast Biopsy Transport Media (T115)

Container/Tube: Sterile container with any standard cell culture media (eg, minimal essential media, RPMI 1640). The solution should be supplemented with 1% penicillin and streptomycin.
Specimen Volume: 4-mm punch
Specimen Stability Information: Refrigerated (preferred)/Ambient
Additional Information: A separate culture charge will be assessed under CULFB / Fibroblast Culture for Biochemical or Molecular Testing. An additional 3 to 4 weeks is required to culture fibroblasts before genetic testing can occur.

Specimen Type: Cultured fibroblast
Container/Tube: T-25 flask
Specimen Volume: 2 Flasks
Collection Instructions: Submit confluent cultured fibroblast cells from a skin biopsy from another laboratory. Cultured cells from a prenatal specimen will not be accepted.
Specimen Stability Information: Ambient (preferred)/Refrigerated (<24 hours)
Additional Information: A separate culture charge will be assessed under CULFB / Fibroblast Culture for Biochemical or Molecular Testing. An additional 3 to 4 weeks is required to culture fibroblasts before genetic testing can occur.

Specimen Type: Blood spot
Supplies: Card-Blood Spot Collection Filter Paper (T493)
Container/Tube:
Preferred: Collection card (Whatman Protein Saver 903 Paper)
Acceptable: PerkinElmer 226 (formerly Ahlstrom 226) filter paper or blood spot collection card
Specimen Volume: 5 Blood spots
Collection Instructions:
1. An alternative blood collection option for a patient older than 1 year is a fingerstick. For detailed instructions, see How to Collect Dried Blood Spot Samples.
2. Let blood dry on the filter paper at ambient temperature in a horizontal position for a minimum of 3 hours.
3. Do not expose specimen to heat or direct sunlight.
4. Do not stack wet specimens.
5. Keep specimen dry
Specimen Stability Information: Ambient (preferred)/Refrigerated
Additional Information:
1. Due to lower concentration of DNA yielded from blood spot, it is possible that additional specimen may be required to complete testing.
2. For collection instructions, see Blood Spot Collection Instructions
3. For collection instructions in Spanish, see Blood Spot Collection Card-Spanish Instructions (T777)
4. For collection instructions in Chinese, see Blood Spot Collection Card-Chinese Instructions (T800)

Specimen Type: Saliva
Patient Preparation: Patient should not eat, drink, smoke, or chew gum 30 minutes prior to collection.
Supplies:
DNA Saliva Kit High Yield (T1007)
Saliva Swab Collection Kit (T786)
Container/Tube:
Preferred: High-yield DNA saliva kit
Acceptable: Saliva swab
Specimen Volume: 1 Tube if using T1007 or 2 swabs if using T786
Collection Instructions: Collect and send specimen per kit instructions.

Specimen Stability Information: Ambient (preferred) 30 days/Refrigerated 30 days
Additional Information: Saliva specimens are acceptable but not recommended. Due to lower quantity/quality of DNA yielded from saliva, some aspects of the test may not perform as well as DNA extracted from a whole blood sample. When applicable, specific gene regions that were unable to be interrogated will be noted in the report. Alternatively, additional specimen may be required to complete testing.

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:
-Informed Consent for Genetic Testing (T576)
-Informed Consent for Genetic Testing (Spanish) (T826)
2. Molecular Genetics: Biochemical Disorders Patient Information (T527)
3. If not ordering electronically, complete, print, and send a Biochemical Genetics Test Request (T798) with the specimen.

Specimen Minimum Volume

See Specimen Required

Reject Due To

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

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Varies	Varies		

Clinical & Interpretive

Clinical Information

The porphyrias are a group of inherited disorders resulting from enzyme defects in the heme biosynthetic pathway. Depending on the specific enzyme involved, various porphyrins and their precursors accumulate in different specimen types. The patterns of porphyrin accumulation in erythrocytes and plasma and excretion of the heme precursors in urine and feces allow for the detection and differentiation of the porphyrias.

The porphyrias are typically classified as erythropoietic or hepatic based upon the primary site of the enzyme defect. In addition, hepatic porphyrias can be further classified as acute hepatic or chronic cutaneous, based on their clinical presentation.

The primary acute hepatic porphyrias: acute intermittent porphyria (AIP), hereditary coproporphyria (HCP), and variegate porphyria (VP), are associated with neurovisceral symptoms that typically onset during puberty or later. Common symptoms include severe abdominal pain, peripheral neuropathy, and psychiatric symptoms. A broad range of medications (including barbiturates and sulfa drugs), alcohol, infection, starvation, heavy metals, and hormonal changes may precipitate crises. Photosensitivity is not associated with AIP but may be present in HCP and VP. Aminolevulinic acid dehydratase deficiency porphyria (ADP) is a rare, autosomal recessive porphyria that has a variable age at presentation.

Clinical manifestations of acute porphyria include attacks of neurologic dysfunction, commonly characterized as

abdominal pain. However, these acute attacks are variable and can include vomiting, diarrhea, constipation, urinary retention, acute episodes of neuropathic symptoms, psychiatric symptoms, seizures, respiratory paralysis, tachycardia, and hypertension. Respiratory paralysis can progress to coma and death.

HCP and VP are also associated with cutaneous manifestations, including edema, sun-induced erythema, acute painful photodermatitis, and urticaria. In some cases, patients present with isolated photosensitivity.

Acute attacks may be prevented by avoiding both endogenous and exogenous triggers. These triggers include porphyrogenic drugs, hormonal contraceptives, fasting, alcohol, tobacco, and cannabis.

Acute hepatic porphyrias are caused by autosomal dominant variants in 1 of 3 genes: *HMBS*, associated with AIP; *CPOX*, associated with HCP; and *PPOX*, associated with VP. Variants in these genes show incomplete penetrance, and patients with a confirmed deleterious variant may be asymptomatic. ADP is inherited in an autosomal recessive manner, due to two disease-causing variants in *ALAD*.

The recommended first-tier tests to screen for acute hepatic porphyria are quantitative urinary porphyrins analysis (PQNRU / Porphyrins, Quantitative, Random, Urine) and fecal porphyrins analysis (FQPPS / Porphyrins, Feces).

Cutaneous photosensitivity is associated with the chronic porphyrias: porphyria cutanea tarda (PCT) and the erythropoietic porphyrias; erythropoietic protoporphyria (EPP), X-linked protoporphyria (XLP), and congenital erythropoietic porphyria (CEP). Although genetic in nature, environmental factors may exacerbate symptoms, significantly impacting the severity and course of disease.

CEP is an erythropoietic porphyria caused by uroporphyrinogen III synthase deficiency. Symptoms typically present in early infancy with red-brown staining of diapers, severe cutaneous photosensitivity with fluid-filled bullae and vesicles. Other common symptoms may include thickening of the skin, hypo- and hyperpigmentation, hypertrichosis, cutaneous scarring, and deformities of the fingers, eyelids, lips, nose, and ears. A few milder adult-onset cases have been documented as well as cases that are secondary to myeloid malignancies (sometimes referred to as erythropoietic uroporphyria).

PCT is the most common form of porphyria and is most commonly sporadic (acquired), but approximately 25% of cases are inherited in an autosomal dominant manner. The most prominent clinical characteristics are cutaneous photosensitivity and scarring on sun-exposed surfaces. Patients experience chronic blistering lesions resulting from mild trauma to sun-exposed areas. These fluid-filled vesicles rupture easily, become crusted, and heal slowly. Secondary infections can cause areas of hypo- or hyperpigmentation or sclerodermatous changes and may result in the development of alopecia at sites of repeated skin damage. Liver disease is common in patients with PCT as evidenced by abnormal liver function tests and with 30% to 40% of patients developing cirrhosis. In addition, there is an increased risk of hepatocellular carcinoma.

Hepatoerythropoietic porphyria (HEP) is observed when an individual inherits PCT from both parents. Patients exhibit a similar clinical presentation to what is seen in CEP.

The clinical presentation of EPP and XLP is identical with onset of symptoms typically occurring in childhood. Cutaneous photosensitivity in sun-exposed areas of the skin generally worsens in the spring and summer months. Common symptoms may include itching, edema, erythema, stinging or burning sensations, and occasionally scarring of the skin in

sun-exposed areas.

Chronic porphyrias are caused by autosomal dominant disease-causing variants in *UROD* that are associated with inherited PCT or autosomal recessive variants in *UROD* that are associated with HEP. They are also due to autosomal recessive disease-causing variants in *UROS* or X-linked variants in *GATA1* that are associated with CEP, or autosomal recessive variants in *FECH* that are associated with EPP. In addition, autosomal dominant variants in *CLPX*, associated with EPP2, and X-linked variants in *ALAS2,* associated with XLP, are also causes of chronic porphyrias.

A comprehensive gene panel is a helpful tool to establish a targeted diagnosis for patients with suggestive clinical and biochemical features of porphyria.

The recommended first-tier biochemical testing for patients with a suspected porphyria is most effective when following a stepwise approach.

The following algorithms are available:
-Porphyria (Acute) Testing Algorithm
-Porphyria (Cutaneous) Testing Algorithm

Reference Values
An interpretive report will be provided.

Interpretation
All detected alterations are evaluated according to American College of Medical Genetics and Genomics 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 the 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 at least one 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 or for assistance in the interpretation of these results, contact the Mayo Clinic Laboratories genetic counselors 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; 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 a specific clinical disorder is suspected, evaluation by alternative methods can be considered.

There may be regions of genes that cannot be effectively evaluated by 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 test is validated to detect 95% of deletions up to 75 base pairs (bp) and insertions up to 47 bp. Deletions-insertions (delins) of 40 or more bp, including mobile element insertions, may be less reliably detected than smaller delins.

Deletion/Duplication Analysis:
This analysis targets single and multi-exon deletions/duplications; however, in some instances single exon resolution cannot be achieved due to isolated reduction in sequence coverage or inherent genomic complexity. Balanced structural rearrangements (such as translocations and inversions) may not be detected.

This test is not designed to detect low levels of mosaicism or 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.

Genes may be added or removed based on updated clinical relevance. For detailed information regarding gene-specific performance and technical limitations, see Method Description or contact a laboratory genetic counselor.

If the patient has had an allogeneic hematopoietic stem cell transplant or a recent heterologous blood transfusion, 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.

Reclassification of Variants:
Currently, it is not standard practice for the laboratory to systematically review previously classified variants on a regular basis. The laboratory encourages healthcare providers to contact the laboratory at any time to learn how the classification of a particular variant may have changed over time. Due to broadening genetic knowledge, it is possible that the laboratory may discover new information of relevance to the patient. Should that occur, the laboratory may issue an amended report.

Variant Evaluation:
Evaluation and categorization of variants are performed using published American College of Medical Genetics and Genomics and the Association for Molecular Pathology recommendations as a guideline.(1) 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.

Rarely, incidental or secondary findings may implicate another predisposition or presence of active disease. These findings will be carefully reviewed to determine whether they will be 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;17(5):405-424
2. Siegesmund M, van Tuyll van Serooskerken AM, Poblete-Gutierrez P, Frank J. The acute hepatic porphyrias: current status and future challenges. Best Pract Res Clin Gastroenterol. 2010;24(5):593-605
3. Tortorelli S, White A, Raymond K. Disorders of porphyrin metabolism. In: Dietzen DJ, Wong ECC, Bennett MJ, Haymond S, eds. Biochemical and Molecular Basis of Pediatric Disease. 5th ed. AACC Press; 2020:chap 15

Performance

Method Description

Next-generation sequencing (NGS) and/or Sanger sequencing are performed to test for the presence of variants in coding regions and intron/exon boundaries of the genes analyzed, as well as some other regions that have known disease-causing variants. The human genome reference GRCh37/hg19 build was used for sequence read alignment. At least 99% of the bases are covered at a read depth over 30X. Sensitivity is estimated to be over 99% for single nucleotide variants, over 94% for deletions-insertions (delins) less than 40 base pairs (bp), and over 95% for deletions up to 75 bp and insertions up to 47 bp. NGS and/or a polymerase chain reaction-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 evaluated by 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. See Targeted Genes and Methodology Details for Porphyria Comprehensive Gene Panel for details regarding the targeted genes analyzed and specific gene regions not routinely covered.(Unpublished Mayo method).

Genes analyzed: *ALAD, ALAS2, CLPX, CPOX, FECH, GATA1, HFE, HMBS, PPOX, UROD, UROS*

PDF Report

Supplemental

Day(s) Performed

Varies

Report Available

14 to 21 days

Specimen Retention Time

Whole Blood: 2 weeks (if available); Extracted DNA: 3 months; Blood spots/Saliva:1 month

Performing Laboratory Location

Mayo Clinic Laboratories - Rochester Main Campus

Fees & Codes

Fees

- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their account representative. For assistance, contact Customer Service.

Test Classification

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

CPT Code Information

81405
81406 x 2
81479
88233-Tissue culture, skin, solid tissue biopsy (if appropriate)
88240-Cryopreservation (if appropriate)
81479 (if appropriate for government payers)

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
PCGP	Porphyria Comprehensive Gene Panel	105350-3

Result ID	Test Result Name	Result LOINC® Value
608680	Test Description	62364-5
608681	Specimen	31208-2
608682	Source	31208-2
608683	Result Summary	50397-9
608684	Result	82939-0
608685	Interpretation	69047-9
608686	Resources	99622-3
608687	Additional Information	48767-8
608688	Method	85069-3
608689	Genes Analyzed	48018-6
608690	Disclaimer	62364-5
608691	Released By	18771-6