Test Definition: G6PDZ

Glucose-6-Phosphate Dehydrogenase (G6PD)
Full Gene Sequencing, Varies

Overview

Useful For

Genetic test for individuals at high risk for glucose-6-phosphate dehydrogenase (G6PD) deficiency

Aiding in the diagnosis of G6PD deficiency

Determining G6PD deficiency status in individuals with inconclusive or unexpected phenotyping results

Differentiation of heterozygotes with skewed X-inactivation from homozygotes and compound heterozygotes

Definitive diagnosis of carrier status

Evaluation of neonates with unexplained jaundice

Identifying individuals at risk of drug-induced acute hemolytic anemia related to G6PD deficiency

Reflex Tests

Test Id	Reporting Name	Available Separately	Always Performed
CULFB	Fibroblast Culture for Genetic Test	Yes	No
CULAF	Amniotic Fluid Culture/Genetic Test	Yes	No
_STR1	Comp Analysis using STR (Bill only)	No, (Bill only)	No
_STR2	Add'l comp analysis w/STR (Bill Only)	No, (Bill only)	No
MATCC	Maternal Cell Contamination, B	Yes	No

Genetics Test Information

This test is for molecular sequencing of the *G6PD* gene and does not assess glucose-6-phosphate dehydrogenase (G6PD) enzyme activity. Enzymatic testing may be suggested as follow-up to this assay. For G6PD enzyme testing order G6PD1 / Glucose 6-Phosphate Dehydrogenase Enzyme Activity, Blood.

G6PD deficiency is a common X-linked condition, estimated to affect up to 500 million people worldwide. Both male and female patients may be impacted due to how common G6PD deficiency is in the population.

Acute hemolytic anemia (AHA) can be triggered in individuals with G6PD deficiency by fava beans, several types of medications (including rasburicase, dapsone-containing combinations of antimalarial drugs, and methylene blue), and

infection. Less commonly, chronic congenital nonspherocytic hemolytic anemia (CNSHA) may occur in severe forms of G6PD deficiency.

US Food and Drug Administration labeling and Clinical Pharmacogenetics Implementation Consortium (CPIC) guidelines recommend that G6PD testing be undertaken in high-risk populations before prescribing drugs known to cause AHA. Knowing a patient's genotype is generally sufficient to avoid contraindicated drugs, but follow-up with the phenotyping (enzyme) assay may be necessary to clarify results in some cases.

This test involves full gene sequencing of all exons and exon/intron boundaries of the *G6PD* gene. A comprehensive interpretation will be provided including congenital and pharmacogenomic implications of results. Testing should be considered before prescribing medication associated with hemolysis in individuals with G6PD deficiency.

Testing Algorithm

For cord blood specimens that have an accompanying maternal blood specimen, maternal cell contamination studies will be performed at an additional charge.

The following are available:
-Glucose-6-Phosphate Dehydrogenase (G6PD) Deficiency Diagnostic Algorithm
-Glucose-6-Phosphate Dehydrogenase (G6PD) Genotyping Interpretive Algorithm

Special Instructions

- Informed Consent for Genetic Testing
- Pharmacogenomic Association Tables
- Glucose-6-Phosphate Dehydrogenase (G6PD) Genotyping Interpretive Algorithm
- Glucose-6-Phosphate Dehydrogenase (G6PD) Deficiency Diagnostic Algorithm

Method Name

Polymerase Chain Reaction (PCR) followed by DNA Sequence Analysis

NY State Available

Yes

Specimen

Specimen Type

Varies

Ordering Guidance

For initial or time-sensitive screening for glucose-6-phosphate dehydrogenase deficiency, order G6PD1 / Glucose 6-Phosphate Dehydrogenase Enzyme Activity, Blood.

Specimen Required

Patient Preparation: A previous hematopoietic stem cell transplant from an allogenic donor will interfere with testing. For information about testing patients who have received a hematopoietic stem cell 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**.
3. Whole blood collected postnatal from an umbilical cord is also acceptable. See Additional Information
Specimen Stability Information: Ambient (preferred) 4 days/Refrigerated 4 days/Frozen 4 days
Additional Information:
1. Specimens are preferred to be received within 4 days of collection. Extraction will be attempted for specimens received after 4 days, and DNA yield will be evaluated to determine if testing may proceed.
2. To ensure minimum volume and concentration of DNA are met, the requested volume must be submitted. Testing may be canceled if DNA requirements are inadequate.
3. For postnatal umbilical cord whole blood specimens, maternal cell contamination studies are recommended to ensure test results reflect that of the patient tested. A maternal blood specimen is required to complete maternal cell contamination studies. Order MATCC / Maternal Cell Contamination, Molecular Analysis, Varies on both the cord blood and maternal blood specimens under separate order numbers.

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.

Specimen Type: Extracted DNA
Container/Tube:
Preferred: Screw Cap Micro Tube, 2mL with skirted conical base
Acceptable: Matrix tube, 1mL
Collection Instructions:

1. The preferred volume is at least 100 mcL at a concentration of 75 ng/mcL.
2. Include concentration and volume on tube.

Specimen Stability Information: Frozen (preferred) 1 year/Ambient/Refrigerated

Additional Information: DNA must be extracted in a CLIA-certified laboratory or equivalent and must be extracted from a specimen type listed as acceptable for this test (including applicable anticoagulants). Our laboratory has experience with Chemagic, Puregene, Autopure, MagnaPure, and EZ1 extraction platforms and cannot guarantee that all extraction methods are compatible with this test. If testing fails, one repeat will be attempted, and if unsuccessful, the test will be reported as failed and a charge will be applied. If applicable, specific gene regions that were unable to be interrogated due to DNA quality will be noted in the report.

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. If not ordering electronically, complete, print, and send 1 of the following forms with the specimen:
-Therapeutics Test Request (T831)
-Benign Hematology Test Request Form (T755)

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

Glucose-6-phosphate dehydrogenase (G6PD) deficiency is the most common human enzymopathy, estimated to affect up to 500 million people worldwide. It is most frequently found in populations where *Plasmodium falciparum* malaria is (or was) endemic, but G6PD deficiency may be present in any population.(1)

Glucose-6-phosphate dehydrogenase deficiency primarily manifests as episodic acute hemolytic anemia (AHA), chronic non-spherocytic hemolytic anemia (CNSHA), and neonatal jaundice. These clinical manifestations can be triggered in individuals with G6PD deficiency by fava beans, several types of medications (including rasburicase, dapsone-containing combinations of antimalarial drugs, and methylene blue), and infection.(1,2)

Glucose-6-phosphate dehydrogenase converts glucose-6-phosphate to 6-phosphoglyconolactone in the first step of the

pentose phosphate pathway, this reaction also produces nicotinamide adenine dinucleotide phosphate (NADPH) from NADP(+). NADPH, through subsequent enzymatic reactions, protects erythrocytes from damage by detoxifying hydrogen peroxide and other sources of oxidative stress. (3)

Glucose-6-phosphate dehydrogenase is encoded by the gene *G6PD*, which lies on the X-chromosome. G6PD deficiency is inherited in an X-linked recessive manner; therefore, male patients are more commonly affected than female patients, but due to the high prevalence of G6PD deficiency, homozygous and compound heterozygous female patients are not uncommon. Over 200 *G6PD* variants have been discovered and are classified based on guidance from the World Health Organization (WHO). In 2022, WHO proposed updated guidance for the classification of *G6PD* variants (Table).(4) This revised guidance is based on the median residual enzyme activity and seeks to resolve problems identified with the WHO *G6PD* classification system that has been in place since 1985 (Table).(4)

Table. Updated and Legacy *G6PD* Variant WHO Classification and Associated G6PD Deficiency Phenotype

2022 WHO class	Median G6PD activity	Hemolysis	Legacy WHO class	Level of residual enzyme activity (% of normal)
A	<20%	Chronic (CNSHA)	I	<10%
B	<45%	Acute, triggered	II	<10%
			III	10%-60%
C	60-150%	No hemolysis	IV	Normal
U	Any	Uncertain clinical significance		

With the exception of those with CNSHA, individuals with G6PD deficiency are typically asymptomatic until they are challenged with an exogenous factor, such as a drug, infection, or fava beans.(1) The exogenous factor can trigger AHA in individuals with G6PD deficiency. The severity of AHA is highly variable, ranging from mild neonatal jaundice to life-threatening complications, such as kernicterus.(1) Therefore, determining the G6PD deficiency status is recommended on the US Food and Drug Administration label of several drugs either proven or suspected to cause AHA in patients with G6PD deficiency. For more information on drugs known to cause AHA in individuals with G6PD deficiency, see Pharmacogenomic Associations Tables. In addition, the Clinical Pharmacogenetics Implementation Consortium has published a guideline related to medication use in the context of *G6PD* genotype.(5)

Preemptive genotyping allows for the identification of patients at risk for an adverse reaction to drugs known to cause AHA in those with G6PD deficiency. In most cases, genotyping provides sufficient information to avoid the use of contraindicated drugs. In some cases, including heterozygous female patients, the phenotyping assay is necessary to determine if such drugs should be avoided. Skewed X-inactivation in heterozygous female patients has been reported to result in G6PD deficiency, so the phenotyping assay is necessary to determine G6PD activity level.(3)

Reference Values

An interpretive report will be provided.

Interpretation

All detected alterations will be evaluated according to the latest American College of Medical Genetics and Genomics

recommendations and the most recent World Health Organization system for classifying genetic variants of *G6PD*.(1,2) Variants will be classified based on known, predicted, or possible effect on gene pathogenicity and reported with interpretive comments detailing their potential or known significance.

Cautions

Patients who have received a non-leukocyte reduced blood transfusion within the preceding 6 weeks can have inaccurate genetic test results due to the presence of both donor and recipient DNA. For patients who have been transfused within the preceding 6 weeks, the glucose-6-phosphate dehydrogenase (G6PD) enzyme assay will also be affected, so it is **not** an appropriate alternative test. Patients who have received an allogeneic hematopoietic stem cell transplant would be expected to convert to the G6PD status of the donor; therefore, it is appropriate to perform *G6PD* sequencing on a whole blood sample. However, if the patient's transplant is not fully engrafted (chimerism is present) or if there is a relapse of an underlying hematologic malignancy, a mixture of donor and recipient genotype may be seen on genetic analysis. The enzyme assay can be run after transplantation; order G6PD1 / Glucose 6-Phosphate Dehydrogenase Enzyme Activity, Blood.

Rare variants exist that could lead to false-negative or false-positive results. Other variants in the primer binding regions can affect the testing, and ultimately, the genotype assessment made.

Test results should be interpreted in the context of clinical findings, family history, and other laboratory data. Large deletions or rearrangements are not detected by this assay.

Sometimes a genetic variant of uncertain significance may be identified. In this case, testing of appropriate family members, as well as testing enzyme activity, may be useful to determine pathogenicity of the alteration.

This test is not designed to provide specific dosing or drug selection recommendations and is to be used as an aid to clinical decision making only. Drug-label guidance should be used when dosing patients with medications regardless of the predicted phenotype.

Skewed X-inactivation in heterozygous female patients has been reported to result in G6PD deficiency. In these cases, the phenotyping (enzyme) assay is necessary to determine G6PD activity level and assign G6PD deficiency status.

Rarely, incidental or secondary findings may implicate another predisposition or presence of active disease. Incidental findings may include, but are not limited to, results related to the sex chromosomes. These findings will be carefully reviewed to determine whether they will be reported.

Clinical Reference

1.Luzzatto L, Ally M, Notaro R. Glucose-6-phosphate dehydrogenase deficiency. Blood. 2020;136(11):1225-1240. doi:10.1182/blood.2019000944
2.Luzzatto L, Seneca E. G6PD deficiency: a classic example of pharmacogenetics with on-going clinical implications. Br J Haematol. 2014;164(4):469-480
3.Cappellini MD, Fiorelli G. Glucose-6-phosphate dehydrogenase deficiency. Lancet. 2008;371(9604):64-74
4.Global Malaria Programme, Malaria Policy Advisory Group. Meeting report of the technical consultation to review the classification of glucose-6-phosphate dehydrogenase (G6PD). World Health Organization; 2022. Accessed July 3, 2023. Available at www.who.int/publications/m/item/WHO-UCN-GMP-MPAG-2022.01

5. Gammal RS, Pirohamed M, Somogyi AA, et al. Expanded clinical pharmacogenetics implementation consortium guideline for medication use in the context of *G6PD* genotype. Clin Pharmacol Ther. 2023;113(5):973-985.
6.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):105-423

Performance

Method Description
Genomic DNA is extracted from whole blood. The *G6PD* gene is amplified by polymerase chain reaction (PCR). The PCR products are then purified and sequenced in both directions using fluorescent dye-terminator chemistry. Sequencing products are separated on an automated sequencer and trace files analyzed for variations in the exons and intron/exon boundaries of all exons using variant detection software and visual inspection. Variant nomenclature is based on GenBank accession number NM_001042351.2 using human genome assembly GRCh37 (hg19).(Unpublished Mayo method)

PDF Report
No

Day(s) Performed
Varies

Report Available
3 to 7 days

Specimen Retention Time
Whole blood: 28 days (if available); Saliva: 30 days (if available), Extracted DNA: 3 months

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.

Test Definition: G6PDZ

Glucose-6-Phosphate Dehydrogenase (G6PD)
Full Gene Sequencing, Varies

CPT Code Information

81249

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
G6PDZ	G6PD Full Gene Sequencing, V	94231-8

Result ID	Test Result Name	Result LOINC® Value
618837	G6PD Phenotype	47998-0
618838	Result Details	82939-0
618839	Interpretation	69047-9
618840	Additional Information	48767-8
618841	Method	85069-3
618842	Disclaimer	62364-5
618843	Reviewed By	18771-6