Overview

Useful For

Genetic confirmation of congenital disorders of fibrinogen with the identification of an alteration in *FGA*, *FGB*, or *FGG* that is known or suspected to cause disease

Testing for close family members of an individual with a diagnosis of afibrinogenemia/hypofibrinogenemia or dysfibrinogenemia/hypodysfibrinogenemia

This test is **not intended for** prenatal diagnosis

Genetics Test Information

This test detects pathogenic alterations within the *FGA*, *FGB*, and *FGG* genes to delineate the underlying molecular defect in a patient with a laboratory diagnosis of congenital afibrinogenemia/hypofibrinogenemia or dysfibrinogenemia/hypodysfibrinogenemia.

The gene targets for this test are:

Gene name (transcript): *FGA* (GRCh37 [hg19] NM_021871)

Chromosomal location: 4q31.3

Gene name (transcript): *FGB* (GRCh37 [hg19] NM_005141)

Chromosomal location: 4q31.3

Gene name (transcript): *FGG* (GRCh37 [hg19] NM_000509)

Chromosomal location: 4q32.1

Testing Algorithm

The laboratory workup for a congenital fibrinogen disorder begins with global coagulation screening assays.

In afibrinogenemia, prothrombin time (PT), activated partial thromboplastin time (aPTT), and thrombin clotting time (TT) may be infinitely prolonged in afibrinogenemia.

In hypofibrinogenemia, TT is more sensitive than PT or aPTT for both quantitative and qualitative defects in fibrinogen.(1) Reptilase time (RT) maybe performed in addition to or instead of TT in samples known or suspected to contain heparin, which artificially prolongs TT.

PT, aPTT, and TT have poor sensitivity for mild fibrinogen deficiency or dysfunction. Further screening and identification of a mild fibrinogen deficiency or dysfibrinogenemia requires a clottable fibrinogen assay (typically Clauss-method based, eg, FIBTP / Fibrinogen, Plasma) to further test fibrinogen function as well as an immunologic (antigentic) assay (FIBAG / Fibrinogen Antigen, Plasma) to detect the quantity of fibrinogen present. Hypofibrinogenemia is indicated by a proportional decrease of functional and immunoreactive fibrinogen. Dysfibrinogenemia is indicated by a discrepancy between functional and immunoreactive fibrinogen.

Genetic testing for a congenital disorder of fibrinogen is indicated if:

-Coagulation tests indicate a quantitative or functional defect in fibrinogen

-Acquired causes of fibrinogen deficiency or dysfunction have been excluded (eg, thrombin clotting time [TT] may be prolonged by the presence of heparin, prior exposure to bovine thrombin, and high concentrations of serum proteins, as in multiple myeloma)

Special Instructions

- Informed Consent for Genetic Testing
- Informed Consent for Genetic Testing (Spanish)
- Rare Coagulation Disorder Patient Information

Method Name

Custom Sequence Capture and Targeted Next-Generation Sequencing (NGS) Followed by Polymerase Chain Reaction (PCR) and Sanger Sequencing When Appropriate

NY State Available

Yes

Specimen

Specimen Type

Varies

Advisory Information

Genetic testing for a congenital disorder of fibrinogen should only be considered if coagulation screening tests measuring thrombin clotting time (TT) with or without reptilase time (RT), clottable fibrinogen, and fibrinogen antigen labs are documented and indicate a quantitative or functional defect in fibrinogen, especially if these findings are similar between family members.

Shipping Instructions

Ambient and refrigerated specimens **must** arrive within 7 days (168 hours of draw), and frozen specimens must arrive within 14 days (336 hours of draw).

Collect and package specimens as close to shipping time as possible.

Necessary Information

Rare Coagulation Disorder Patient Information is required, see Special Instructions. Testing may proceed without the patient information, however, the information aids in providing a more thorough interpretation. Ordering providers are strongly encouraged to fill out the form and send with the specimen.

Specimen Required

Submit only 1 of the following specimens:

Specimen Type: Peripheral blood

Container/Tube:

Preferred: Lavender top (EDTA)

Acceptable: Yellow top (ACD) or green top (sodium citrate)

Specimen Volume: 3 mL

Collection Instructions:

1. Invert several times to mix blood.

2. Send specimen in original tube.

Specimen Stability: Ambient (preferred)/Refrigerated/Frozen

Specimen Type: Extracted DNA

Container/Tube: 1.5- to 2-mL tube

Specimen Volume: Entire specimen

Collection Instructions:

1. Label specimen as extracted DNA and source of specimen.

2. Provide indication of volume and concentration of the DNA.

Specimen Stability: Frozen (preferred)/Refrigerated/Ambient

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. If not ordering electronically, complete, print, and send a Coagulation Test Request (T753) with the specimen.

Specimen Minimum Volume

Blood: 1 mL
Extracted DNA: 100 mcL at 50 ng/mcL concentration

Reject Due To

Gross hemolysis	OK
Gross lipemia	OK

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Varies	Ambient (preferred)	7 days	
	Frozen	14 days	
	Refrigerated	7 days	

Clinical and Interpretive

Clinical Information

Fibrinogen forms the insoluble fibrin matrix that is a major component of the blood clots critical for stopping blood loss. Fibrinogen is made up of six polypeptide chains-one pair each of alpha, beta, and gamma chains (encoded by the *FGA*, *FGB*, and *FGG* genes respectively)-that are held together by 29 disulfide bonds.(2) The alpha, beta, and gamma fibrinogen subunits polymerize to form an insoluble fibrin matrix that is a major component of the blood clots critical for stopping blood loss. Fibrinogen also has a role in the early stages of wound repair.

Fibrinogen disorders are classified as either 1) afibrinogenemia or hypofibrinogenemia, a quantitative defect of low or absent fibrinogen plasma antigen levels, or 2) dysfibrinogenemia or hypodysfibrinogenemia, a qualitative defect in function and activity with normal or reduced antigen levels. Congenital afibrinogenemia and hypofibrinogenemia are inherited in an autosomal recessive manner. Congenital dysfibrinogenemia is, in most cases, inherited in an autosomal dominant manner but cases of recessive inheritance have also been reported.

Afibrinogenemia:

Afibrinogenemia is characterized by the complete absence of fibrinogen in circulation. Although all individuals with afibrinogenemia have unmeasurable functional fibrinogen, the severity of bleeding is highly variable, even among those with the same genetic alteration(s).(3) Abnormal bleeding may occur in the neonatal period as umbilical cord bleeding. Bleeding may occur in skin, the oral cavity, gastrointestinal tract, genitourinary tract, or central nervous system. Intracranial hemorrhage is a major cause of death in affected individuals, who are also at risk for joint bleeds and spontaneous splenic rupture. Venous and arterial thromboembolic complications and poor wound healing may also occur. Affected women have increased risk for menometrorrhagia and recurrent pregnancy loss. The prevalence of afibrinogenemia is estimated to be 1 in 1 million.

Hypofibrinogenemia:

Most individuals with hypofibrinogenemia (characterized by fibrinogen levels less than 1.5 g/L) are asymptomatic.(3) Thromboembolism may occur spontaneously or with fibrinogen substitution therapy. Affected individuals may experience abnormal bleeding after trauma or if they have a second hemostatic abnormality. Recurrent pregnancy loss and postpartum hemorrhage are reported in affected women. There is typically good correlation between fibrinogen levels and clinical severity, with levels less than 0.5 g/L associated with major bleeding.(4) Specific alterations associated with hypofibrinogenemia are strongly correlated with hepatic storage disease.(3) Acquired hypofibrinogenemia has been reported in individuals with hepatic failure or decompensation cirrhosis. Hypofibrinogenemia is also commonly associated with acute disseminated intravascular coagulation (DIC). Less common acquired causes include administration of L-asparaginase and valproic acid or other drugs that impair hepatic synthesis. These causes of acquired hypofibrinogenemia should be excluded prior to genetic testing for a fibrinogen disorder.

Dysfibrinogenemia and Hypodysfibrinogenemia:

About half of individuals with dysfibrinogenemia and hypodysfibrinogenemia are asymptomatic. However, alteration carriers carry a high risk of major bleeding and/or thromboembolic complications.(5) Patients bleed most after trauma, surgery, or postpartum. Some women have spontaneous abortions. Specific alterations associated with dysfibrinogenemia are strongly associated with thromboembolic pulmonary hypertension and amyloidosis. Causes of acquired (non-genetic) dysfibrinogenemia or defects in fibrinogen that should be excluded prior to genetic testing include cirrhosis, acute or chronic hepatitis, metastatic hepatoma, renal carcinoma, and biliary obstruction. Individuals treated with isotretinoin therapy have also been reported to develop acquired dysfibrinogenemia. Fibrinogen antibodies and inhibitors have been reported in systemic lupus erythematosus, ulcerative colitis, and

multiple myeloma. These causes of acquired dysfibrinogenemia and hypodysfibrinogenemia should be considered and excluded prior to genetic testing for a fibrinogen disorder.

Reference Values

An interpretive report will be provided

Interpretation

An interpretive report will be provided.

Evaluation and categorization of variants is performed using the most recent published American College of Medical Genetics and Genomics (ACMG) recommendations as a guideline. Variants are classified based on known, predicted, or possible pathogenicity and reported with interpretive comments detailing their potential or known significance.

Consultations with the Mayo Clinic Special Coagulation Clinic, Molecular Hematopathology Laboratory, or Thrombophilia Center are available for DNA diagnosis cases. This may be especially helpful in complex cases or in situations where the diagnosis is atypical or uncertain.

Cautions

Clinical:

Some individuals may have a mutation that is not identified by the methods performed. The absence of a mutation, therefore, does not eliminate the possibility of afibrinogenemia, hypofibrinogenemia, dysfibrinogenemia, thrombophilia, bleeding tendency, familial visceral amyloidosis, or fibrinogen storage disease. This assay does not distinguish between germline and somatic alterations, particularly with variant allele frequencies (VAF) significantly lower than 50%. 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.

Technical Limitations:

Next-generation sequencing (NGS) may not detect all types of genetic variants. Additionally, rare polymorphisms may be present that could lead to false negative or positive results. Therefore test results should be interpreted in the context of activity and antigen measurements, clinical findings, family history, and other laboratory data. If results do not match clinical findings, consider alternative methods for analyzing these genes, such as Sanger sequencing or large deletion/duplication analysis. Misinterpretation of results may occur if the information provided is inaccurate or incomplete.

If multiple alterations are identified, NGS is not able to distinguish between alterations that are found in the same allele ("in cis") and alterations found on different alleles ("in trans"). This limitation may complicate diagnosis or classification and has implications for inheritance and genetic counseling. To resolve these cases, molecular results must be correlated with clinical history, activity and antigen measurements, and family studies.

Unless reported or predicted to cause disease, alterations found deep in the intron or alterations that do not result in an amino acid substitution are not reported. These and common polymorphisms identified for this patient are available upon request.

Reclassification of Variants Policy: At this time, it is not standard practice for the laboratory to systematically review likely pathogenic variants or variants of uncertain significance that are detected and reported. 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.

Clinical Reference

1. Verhovsek M, Moffat KA, Hayward CP: Laboratory testing for fibrinogen abnormalities. Am J Hematol. 2008;83(12):928-931

2. Weisel J and Litvinov R: Mechanisms of fibrin polymerization and clinical implications. Blood. 2013;121:1712-1719

3. De Moerloose P, Casini A, Neerman-Arbez M: Congenital fibrinogen disorders: an update. Semin Thromb Hemost. 2013;39:585-595

4. De Moerloose P, Schved JF, Nugent D: Rare coagulation disorders: fibrinogen, factor VII and factor XIII. Haemophilia. 2016;22(Suppl 5):61-65

5. Casini, A, Blondon M, Lebreton A, et al: Natural history of patients with congenital dysfibrinogenemia. Blood. 2015;125:553-561

6.Casini A, Neerman-Arbez M, Ariens RA: Dysfibrinogenemia: from molecular anomalies to clinical manifestations and management. J Thromb Haemost. 2015;13(6):909-919

7. Peyvandi F: Epidemiology and treatment of congenital fibrinogen deficiency. Thromb Res. 2012;130(Suppl 2):S7-11

Performance

Method Description

Next-generation sequencing and/or Sanger sequencing are performed.

Regions of homology, high guanine-cytosine (GC)-rich content, and repetitive sequences may not provide accurate sequence. Therefore, all reported alterations detected by next-generation sequencing in these regions are confirmed by an independent reference method. However, this does not rule out the possibility of a false-negative result in these regions.

Sanger sequencing is used to confirm alterations detected by next-generation sequencing when appropriate.(Unpublished Mayo method)

PDF Report

No

Day(s) and Time(s) Test Performed

Performed weekly, Varies

Analytic Time

21 days

Maximum Laboratory Time

28 days

Specimen Retention Time

Whole Blood: 2 weeks; DNA: Indefinitely

Performing Laboratory Location

Rochester

Fees and 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 Regional Manager. 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. This test has not been cleared or approved by the U.S. Food and Drug Administration.

CPT Code Information

81479

LOINC® Information

Test ID	Test Order Name	Order LOINC Value
FIBNG	FGA/B/G Genes, Full Gene NGS	92992-7

Result ID	Test Result Name	Result LOINC Value
113084	FIBNG Result	50397-9
113078	Alterations Detected	82939-0
113077	Interpretation	69047-9
113079	Additional Information	48767-8
113080	Method	85069-3
113081	Disclaimer	62364-5
113082	Panel Gene List	48018-6
113083	Reviewed By	18771-6