Overview

Useful For

Prenatal testing for a known familial pathogenic mutation in the *F9* gene in a fetus who is at risk for inheriting this mutation

Genetics Test Information

Hemophilia B is an X-linked disorder caused by mutations in the *F9* gene. This test is intended to prenatally detect a previously confirmed familial mutation (missense, nonsense, splice site variants, and small intragenic deletions/insertions) in an at-risk fetus. This mutation should be confirmed and documented in an affected family member and/or confirmed in the mother of the fetus via molecular testing. **Documentation of the specific familial mutation must be provided with the specimen in order to perform this test.** Testing will be cancelled if this documentation is not submitted. This test is not validated to detect large deletions or duplications that are a cause of approximately 3% of cases of hemophilia B.

Reflex Tests

Test ID	Reporting Name	Available Separately	Always Performed
MATCC	Maternal Cell Contamination, B	Yes	No
CULFB	Fibroblast Culture for Genetic Test	Yes	No
CULAF	Amniotic Fluid Culture/Genetic Test	Yes	No

Testing Algorithm

If amniotic fluid (nonconfluent cultured cells) is received, amniotic fluid culture/genetic test will be added and charged separately. If chorionic villus specimen (nonconfluent cultured cells) is received, fibroblast culture for genetic test will be added and charged separately. For any prenatal specimen that is received, maternal cell contamination studies will be added. A maternal peripheral blood sample is required to perform this test.

The following algorithms are available in Special Instructions:

-Hemophilia Carrier Testing Algorithm

-Hemophilia Testing Algorithm

Special Instructions

- Hemophilia B Patient Information
- Informed Consent for Genetic Testing
- Hemophilia Carrier Testing Algorithm
- Hemophilia Testing Algorithm
- Informed Consent for Genetic Testing (Spanish)

Method Name

Polymerase Chain Reaction (PCR)/Fluorescent DNA Sequencing

NY State Available

Yes

Specimen

Specimen Type

Varies

Additional Testing Requirements

Due to the complexity of prenatal testing, consultation with the laboratory is required for all prenatal testing. Prenatal specimens can be sent Monday through Thursday and **must be received by 5 p.m. CST on Friday** in order to be processed appropriately. All prenatal specimens must be accompanied by a maternal blood specimen. Order MATCC / Maternal Cell Contamination, Molecular Analysis on the maternal specimen.

Shipping Instructions

Advise Express Mail or equivalent if not on courier service

Necessary Information

Hemophilia B 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

Results will be reported and also telephoned or faxed, if requested.

A. For the purposes of maternal cell contamination studies (MCC), submit the following specimen type from the mother in addition to 1 of the 3 accepted fetal specimen types:

Specimen Type: Peripheral blood

Container/Tube:

Preferred: Yellow top (ACD solution B)

Acceptable: Lavender top (EDTA) or light blue top (sodium citrate)

Specimen Volume: 6 mL

Collection Instructions:

1. Invert several times to mix blood.

2. Send specimen in original tube.

Specimen Stability Information: Ambient (preferred)/Refrigerated

B. For the purposes of prenatal testing of the fetus, submit only 1 of the following specimens:

Specimen Type: Amniotic fluid

Container/Tube: Amniotic fluid container

Specimen Volume: 5-10 mL

Collection Instructions:

1. Optimal timing for specimen collection is during 14 to 18 weeks of gestation, but specimens collected at other weeks of gestation are also accepted.

2. Discard the first 2 mL of amniotic fluid. If the culture will be performed in conjunction with chromosome analysis and alpha-fetoprotein, a total of approximately 25 mL to 30 mL will be needed for the combined studies.

Specimen Stability Information: Ambient (preferred) <24 hours/Refrigerated

Specimen Type: Chorionic villi

Container/Tube: 15-mL tube containing 15 mL of transport media

Specimen Volume: 20-30 mg

Collection Instructions:

1. Collect specimen by the transabdominal or transcervical method.

2. Transfer the chorionic villi specimen to a Petri dish containing transport medium (T095).

3. Using a stereomicroscope and sterile forceps, assess the quality and quantity of the villi and remove any blood clots and maternal decidua.

Specimen Stability Information: Refrigerated (preferred) <24 hours/Ambient

Specimen Type: Confluent cultured cells

Container/Tube: T-25 flask

Specimen Volume: 2 Flasks approximately 90% confluent

Collection Instructions: Submit confluent cultured cells from another laboratory

Specimen Stability Information: Ambient (preferred) <24 hours/Refrigerated

Additional Information:

1. Place the tubes in a Styrofoam container (T329).

2. Fill remaining space with packing material.

3. Unavoidably, about 1% to 2% of mailed-in specimens are not viable.

4. Bloody specimens are undesirable.

5. If the specimen does not grow in culture, you will be notified within 7 days of receipt.

6. There will be no culture charge.

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

Amniotic fluid: 10 mL
Chorionic Villi: 5 mg

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 and Interpretive

Clinical Information

Hemophilia B, factor IX deficiency, is an X-linked recessive bleeding disorder with an incidence of about 1 per 30,000 live male births. It occurs as a result of mutations in the factor IX *(F9)* gene. As many as one-third of hemophiliacs have no affected family members, which reflects a high mutation rate in the *F9* gene (ie, de novo mutations). Hemophilia B affects males; however, all male offspring from an affected male will be normal. Although all female offspring of affected males will be obligatory carriers, they rarely have symptomatic bleeding. In contrast, female offspring of female carriers of hemophilia B have a 50% chance of being carriers themselves, and each male offspring has a 50% chance of being affected.

Based on factor IX activity, hemophilia B is classified as severe (factor IX activity <1%), moderate (factor IX activity 1%-5%), or mild (factor IX activity >5%-40%). In males, a low factor IX activity level establishes the diagnosis of hemophilia B. However, the wide range of normal factor IX activity precludes an accurate assessment of carrier status in females, thus making molecular testing essential in assessment of carrier status.

Inhibitors to factor IX activity are estimated to occur in 5% to 8% of hemophilia B patients, much less than that of hemophilia A. Inhibitor risk correlates with genotype and typically occurs in patients with either partial or total deletions of the *F9* gene or in certain nonsense mutations that result in no circulating factor IX:antigen. More recently, it has been observed that a subset of patients with such mutations may be at risk of experiencing anaphylactic reactions to the factor IX replacement therapy.

Reference Values

An interpretive report will be issued that will include specimen information, assay information, background information, and conclusions based on the test results (ie, information about the mutation).

Interpretation

An interpretive report will be provided.

Cautions

Special Coagulation Clinic/Laboratory and Medical Genetics consultations are available for DNA diagnosis cases and may be especially helpful in complex cases or in situations where the diagnosis is atypical or uncertain.

Clinical Reference

1. Yoshitake S, Schach BG, Foster DC, et al: Complete nucleotide sequence of the gene for human factor IX (antihemophilic factor B). Biochemistry 1985;24(14):3736-3750

2. Giannelli F, Green PM, Sommer SS, et al: Haemophilia B: database of point mutations and short additions and deletions-eighth edition. Nucleic Acids Res 1998;26(1):265-268

3. Ketterling RP, Bottema CD, Phillips JA 3rd, Sommer SS: Evidence that descendants of three founders constitute about 25% of hemophilia B in the United States. Genomics 1991;10(4):1093-1096

Performance

Method Description

Direct mutation analysis of leukocyte genomic DNA performed by PCR amplification of a single region of the *F9* gene, followed by fluorescent DNA sequencing analysis utilizing an Applied Biosystems Inc. (ABI) 3730*xl* DNA Analyzer.(Costa JM, Ernault P, Vidaud D, et al: Fast and efficient mutation detection method using multiplex PCR and cycle sequencing-application to haemophilia B. Thromb Haemost 2000;83[2]:244-247; Kaiser RJ, MacKellar SL, Vinayak RS, et al: Specific-primer-directed DNA sequencing using automated fluorescence detection. Nucleic Acids Res 1989;17[15]:6087-6102)

PDF Report

No

Day(s) and Time(s) Test Performed

Monday through Friday

Analytic Time

28 days

Maximum Laboratory Time

35 days

Specimen Retention Time

Extracted DNA indefinitely, patient must opt-out.

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

81403

LOINC® Information

Test ID	Test Order Name	Order LOINC Value
F9KMP	F9 Gene Known Mutation, AF or CVS	91681-7

Result ID	Test Result Name	Result LOINC Value
39837	F9 Known Mut Reason for Referral	42349-1
37887	F9 Known Mutation Method	85069-3
37888	F9 Known Mutation, AF or CVS	50397-9
37889	F9 Known Mutation Specimen Type	31208-2
37890	F9 Known Mutation Interpretation	69047-9
37891	F9 Known Mutation Reviewed By	18771-6