Test Definition: F822B
HA F8 Intron 22 Inversion KM, B

Overview

Useful For
First-tier molecular testing for males affected with severe hemophilia A, when a familial intron 22 inversion has been previously identified.

Determining hemophilia A carrier status for at-risk females, ie, individuals with a family history of severe hemophilia A due to F8 intron 22 inversion

Genetics Test Information
Detects the intron 22 inversion within the F8 gene. The intron 22 inversion mutation accounts for approximately 45% of mutations associated with severe hemophilia A.

Intron 22 inversion known mutation analysis can only be performed for individuals when an intron 22 inversion has already been identified in the family. If a mutation has not already been identified in the family, order F8INV / Hemophilia A F8 Gene, Intron 1 and 22 Inversion Mutation Analysis, Whole Blood.

Reflex Tests

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<th>Reporting Name</th>
<th>Available Separately</th>
<th>Always Performed</th>
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<tbody>
<tr>
<td>MATCC</td>
<td>Maternal Cell Contamination, B</td>
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Testing Algorithm
Maternal cell contamination testing will be performed for all cord blood specimens. A maternal whole 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

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

Method Name
Inverse Shifting-Polymerase Chain Reaction (IS-PCR)

NY State Available
Yes
Specimen

Specimen Type
Whole blood

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.

Container/Tube:

Preferred: Lavender top (EDTA) or yellow top (ACD)

Acceptable: Sodium citrate

Specimen Volume: 3 mL

Collection Instructions:
1. Invert several times to mix blood.
2. Send specimen in original tube.

Additional Information: Each molecular coagulation test requested must have its own tube.

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. Hemophilia A Patient Information (T712) in Special Instructions

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

Specimen Minimum Volume
1 mL

Reject Due To

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<th>Condition</th>
<th>Acceptance</th>
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<tr>
<td>Hemolysis</td>
<td>Mild OK; Gross OK</td>
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<tr>
<td>Lipemia</td>
<td>Mild OK; Gross OK</td>
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<tr>
<td>Icterus</td>
<td>NA</td>
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<tr>
<td>Other</td>
<td>Green-top (heparin) tube or DNA</td>
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Specimen Stability Information
Test Definition: F822B
HA F8 Intron 22 Inversion KM, B

<table>
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<tr>
<th>Specimen Type</th>
<th>Temperature</th>
<th>Time</th>
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<td></td>
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<tr>
<td></td>
<td>Refrigerated</td>
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**Clinical and Interpretive**

**Clinical Information**

Hemophilia A (HA) is caused by a deficiency of clotting factor VIII (FVIII). HA is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males. Males are typically affected with bleeding symptoms, whereas carrier females generally do not have bleeding symptoms but are at risk of having affected sons. Rarely, approximately 10% of carrier females have FVIII activity levels below 35% and are at risk for bleeding.

Bleeding, the most common clinical symptom in individuals with HA, correlates with FVIII activity levels. FVIII activity levels of <1% are associated with severe disease, 1% to 5% activity with moderate disease, and 5% to 40% with mild disease. In males with severe deficiency, spontaneous bleeding may occur. In individuals with mild HA, bleeding may occur only after surgery or trauma.

FVIII is encoded by the factor VIII (F8) gene. Approximately 98% of patients with a diagnosis of HA are found to have a mutation in F8 (ie, intron 1 and 22 inversions, point mutations, insertions, and deletions). The intron 22 inversion mutations account for approximately 45% of mutations associated with severe HA. These inversions are typically not identified in patients with mild or moderate HA.

Intron 22 inversion known mutation analysis is only recommended for individuals when an intron 22 inversion has already been identified in the family.

If a familial mutation has not been identified in a severely affected HA patient the F8 gene intron 1 and 22 inversion analysis (F8INV / Hemophilia A F8 Gene, Intron 1 and 22 Inversion Mutation Analysis, Whole Blood) should be ordered.

If the intron 22 inversion analysis is negative, the tested individual has not inherited the familial mutation.

It is recommended that the F8 mutation be confirmed in the affected male or obligate carrier female prior to testing at-risk individuals. Affected males are identified by FVIII activity (F8A / Coagulation Factor VIII Activity Assay, Plasma) and clinical evaluation, while obligate carrier females are identified by family history assessment. If the intron inversion assays do not detect an inversion in these individuals, additional analysis (ie, F8 sequencing) may be able to identify the familial mutation. Of note, not all females with an affected son are germline carriers of a F8 mutation, as de novo mutations in F8 do occur. Approximately 20% of mothers of isolated cases do not have an identifiable germline F8 mutation. Importantly, there is a small risk for recurrence even when the familial F8 mutation is not identified in the mother of the affected patient due to the possibility of germline mosaicism.

**Reference Values**

Not applicable

**Interpretation**

An interpretive report will be provided.

**Cautions**
Intron 22 inversion known mutation analysis is only recommended for individuals when an intron 22 inversion has already been identified in the family.

If a familial mutation has not been identified in a severely affected hemophilia A patient the F8 gene intron 1 and 22 inversion analysis (F8INV / Hemophilia A F8 Gene, Intron 1 and 22 Inversion Mutation Analysis, Whole Blood) should be ordered.

Obtaining a medical genetics or hematology (coagulation) consultation prior to ordering is advisable. Molecular genetic or hemophilia center consultation is available for all possible hemophilia A cases and is particularly indicated in complex cases or in situations in which the diagnosis is atypical or uncertain.

For evaluation of a patient with bleeding symptoms and no known personal history of a bleeding disorder consider BDIAL / Bleeding Diathesis Profile, Limited (Mayo Clinic Laboratories clients); or BDIAC / Bleeding Diathesis Profile, Comprehensive (Mayo Clinic only), or the specific factor assays.

This assay detects only F8 intron 22 inversion mutations. Thus, a negative result does not exclude the presence of other mutations in F8.

The intron 22 inversion mutation targeted by this assay is found in approximately 45% of individuals with severe hemophilia A; if an intron 22 inversion has not been already identified in the family, the assay may be uninformative.

Test results should be interpreted in the context of clinical findings, family history, and other laboratory data. Errors in our interpretation of results may occur if information given is inaccurate or incomplete.

**Clinical Reference**


**Performance**

**Method Description**

Genomic DNA from whole blood or cord blood is digested with Ksp 22 I restriction enzyme, ligated with T4 DNA ligase, and amplified by PCR with primers specific for the F8 intron 22 inversion mutations.(Rosetti LC, Radic CP, Larripa IB, De Brasi CD: Developing a new generation of tests for genotyping hemophilia-causative rearrangements involving int22h and int1h hotspots in the factor VIII gene. J Thromb Haemost 2008;6:830-836)

**PDF Report**

No

**Day(s) and Time(s) Test Performed**
Monday through Friday

Analytic Time
14 days

Maximum Laboratory Time
21 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

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