Test Definition: HFET

Hereditary Hemochromatosis, HFE Variant Analysis, Varies

Overview

Useful For

Establishing or confirming the clinical diagnosis of hereditary hemochromatosis (HH) in adults

Testing of individuals with increased serum transferrin-iron saturation and ferritin

Predictive testing of individuals who have a family history of HH, in coordination with appropriate genetic counseling

This test is **not recommended** for population screening.

Genetics Test Information

This test detects the 2 common disease-causing variants in the *HFE* gene: C282Y (c.845G>A) and H63D (c.187C>G). The S65C variant will be reported only when it is observed as part of the C282Y/S65C genotype.

Special Instructions

- Molecular Genetics: Congenital Inherited Diseases Patient Information
- Informed Consent for Genetic Testing
- Hereditary Hemochromatosis Algorithm
- Informed Consent for Genetic Testing (Spanish)

Highlights

Molecular testing can be done to establish or confirm the diagnosis of hereditary hemochromatosis in individuals with clinical symptoms.

This assay will not detect all variants in the *HFE* gene that cause hereditary hemochromatosis.

Method Name

Droplet Digital Polymerase Chain Reaction (ddPCR)

NY State Available

Yes

Specimen

Specimen Type

Varies

Specimen Required

Patient Preparation: A previous bone marrow transplant from an allogeneic 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**.
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, 2 mL with skirted conical base
Acceptable: Matrix tube, 1 mL
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. Molecular Genetics: Congenital Inherited Diseases Patient Information (T521)
3. If not ordering electronically, complete, print, and send a Benign Hematology Test Request (T755) 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
Hereditary hemochromatosis (HH) is an autosomal recessive disorder of iron metabolism with a carrier frequency of approximately 1 in 10 individuals of Northern European ancestry.(1) The disease is characterized by an accelerated rate of intestinal iron absorption and progressive iron deposition in various tissues (eg, liver, skin, heart, joints). Clinical symptoms of *HFE* hemochromatosis usually appear in men between age 40 and 60 years and after menopause in women, and they might be affected by other factors such as intake of iron and other mineral supplements, vitamin C, and alcohol consumption. Iron overload can lead to hepatic cirrhosis, hepatocellular carcinoma, diabetes mellitus, arthropathy, and cardiomyopathy. Such complications may be prevented by phlebotomy, and patients may have a normal life expectancy when treated before organ damage occurs.(2) For individuals with clinical symptoms consistent with HH or biochemical evidence of iron overload, an HH diagnosis is typically based on the results of transferrin-iron saturation and serum ferritin concentration. Molecular testing can also be performed to confirm/establish the diagnosis.

The two most common variants in the *HFE* gene are C282Y and H63D. The majority of HH patients (approximately 85-90%) show homozygosity for the C282Y variant or compound heterozygosity for the C282Y and H63D variants.(1)

Individuals with carrier status (heterozygotes) generally do not develop complications of iron overload but may have abnormal serum iron results.(1) Furthermore, clinically significant iron overload can also occur in the absence of known *HFE* variants. Therefore, a negative *HFE* test does not exclude other rare variants in the *HFE* gene or in other genes and, thus, does not exclude a diagnosis of iron overload or hemochromatosis.

The most common disease-causing variant identified in the *HFE* gene is C282Y (c.845G>A in exon 4). Individuals who are homozygous for the C282Y variant account for 60% to 90% of all HH cases, however clinical penetrance is incomplete.(3) Up to 50% of individuals homozygous for C282Y develop iron overload (elevated serum iron indices) and 10% to 33% (mainly men) develop hemochromatosis-related syndromes or end-organ damage symptoms.(2) Currently no test can predict whether an individual who is homozygous for C282Y will develop clinical symptoms. Additionally, 3% to 8% of individuals affected with HH are heterozygous for this variant. These frequencies show variability among different populations, with the highest frequency observed in individuals of Northern European ancestry.

The H63D (c.187C>G in exon 2) variant is also associated with HH; however, the presence of a single H63D variant is unlikely to be of clinical significance in the absence of other disease-causing variants. Additionally, homozygosity for H63D is insufficient to cause clinically significant iron overload in the absence of other modifying risk factors. Individuals who are compound heterozygous for C282Y/H63D have higher penetrance compared to those who are H63D homozygous and have been associated with increased hepatic iron concentrations. Approximately 0.5% to 2% of individuals with this genotype will develop clinical evidence of iron overload.(2) While individuals with this genotype may have increased iron indices, most will not develop clinical disease without comorbid factors (steatosis, diabetes, or excess alcohol consumption).(4)

The clinical significance of a third *HFE* variant, S65C (c.193A>T in exon 2), appears to be minimal. This rare variant displays a very low penetrance and is generally not associated with iron overload. Individuals who are heterozygous for S65C with either the wildtype or H63D allele do not seem to be at an increased risk for HH. Compound heterozygosity for C282Y and S65C may confer a low risk for mild HH.(1) Therefore, the C282Y/S65C genotype is reported when observed.

For more information see Hereditary Hemochromatosis Algorithm

Reference Values

An interpretative report will be provided.

Interpretation

The interpretive report includes an overview of the findings as well as the associated clinical significance.

Cautions

This assay only tests for the C282Y, H63D and S65C (reported as a part of the C282Y/S65C genotype) variants and will not detect all variants in the *HFE* gene that may be associated with hereditary hemochromatosis. Therefore, the absence of a detectable C282Y, H63D, or S65C variant does not rule out the possibility that an individual is a carrier of or affected with this disease.

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

Rare variants (ie, polymorphisms) exist that could lead to false-negative or false-positive results. If results obtained do not match the clinical findings, additional testing should be considered.

In rare cases, DNA variants of unknown significance may be identified.

Because of concerns of the overall penetrance of *HFE* variants, *HFE* genetic testing is not recommended for population screening.

Clinical Reference

1. Barton JC, Edwards CQ. *HFE* Hemochromatosis. In: Adam MP, Ardinger HH, Pagon RA, et al, eds. GeneReviews [Internet]. University of Washington, Seattle; 2000. Updated April 11, 2024. Accessed March 24, 2025. Available at www.ncbi.nlm.nih.gov/books/NBK1440/
2. Kowdley KV, Brown KE, Ahn J, Sundaram V. ACG Clinical Guideline: Hereditary Hemochromatosis. Am J Gastroenterol. 2019;114(8):1202-1218
3. Porto G, Brissot P, Swinkels DW, Zoller H, et al. EMQN best practice guidelines for the molecular genetic diagnosis of hereditary hemochromatosis (HH). Eur J Hum Genet. 2016;24(4):479-495
4. Hollerer I, Bachmann A, Muckenthaler MU. Pathophysiological consequences and benefits of HFE mutations: 20 years of research. Haematologica. 2017;102(5):809-817

Performance

Method Description

Droplet digital polymerase chain reaction (ddPCR) is used to test for the following three variants in the *HFE* gene: C282Y, H63D, and S65C. Because of the minimal effect on iron metabolism associated with the S65C variant, it is only reported when it is found with the C282Y variant (ie, if the patient has the C282Y/S65C genotype).(Unpublished Mayo method)

PDF Report

No

Day(s) Performed

Varies

Report Available

6 to 7 days

Specimen Retention Time

Whole blood/Salvia: 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.

CPT Code Information

81256-*HFE (*hemochromatosis*)* (eg, hereditary hemochromatosis) gene analysis, common variants (C282Y and H63D)

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
HFET	Hereditary Hemochromatosis HFE Test	34519-9

Result ID	Test Result Name	Result LOINC® Value
614667	Result Summary	50397-9
614668	Result	82939-0
614669	Interpretation	69047-9
614670	Specimen	31208-2
614791	Source	31208-2
614792	Method	85069-3
614793	Released By	18771-6