Hereditary Hemolytic Anemia Cascade

Hereditary hemolytic anemia (HHA) is characterized by premature red blood cell (RBC) destruction and anemia due to intrinsic RBC defects, and encompasses a diverse group of heterogeneous disorders. Genetic testing is indicated when initial test results do not explain clinical presentation or mode of inheritance.

Disease Overview

Clinical presentation is highly variable and may include anemia, fatigue, gallstones, hyperbilirubinemia/jaundice, pallor, scleral icterus, and splenomegaly. Laboratory findings include decreased hemoglobin concentration, hematocrit, and RBC count. Blood smear abnormalities such as spherocytes, acanthocytes, schistocytes, bite cells, stomatocytes, polychromasia, and target cells may be present.

Associated Disorders

RBC Membrane Disorders

Disorders characterized by weakened RBC membrane resulting in irregular shape, increased fragility, and hemolysis:

- Hereditary spherocytosis (HS): *ANK1*, *EPB42*, *SLC4A1*, *SPTA1*, and *SPTB*
  - The most common RBC membrane disorder and is characterized by spherically shaped RBCs with decreased deformability.
  - Approximately 75% of HS is autosomal dominant and 25% is autosomal recessive or de novo.
- Hereditary elliptocytosis and hereditary pyropoikilocytosis (HE/HPP) are related disorders with a wide spectrum of clinical phenotypes:
  - HE is characterized by elliptically shaped RBCs.
  - Marked anisopoikilocytosis with elliptocytes, microspherocytes, and bizarrely shaped RBCs are characteristically seen in HPP.
  - HE is autosomal dominant and HPP is autosomal recessive; variants in *SPTA1*, *SPTB*, and, less commonly, *EPB41* are causative.
  - Individuals with HE are generally asymptomatic but can have mild compensated hemolytic anemia.
  - HPP patients usually present with moderate to severe anemia.
- Dehydrated hereditary stomatocytosis (xerocytosis) is characterized by decreased intracellular potassium content, loss of cell water, increased cytoplasmic viscosity, and increased mean cell hemoglobin concentration:
  - Inheritance is autosomal dominant and the most commonly involved gene is *PIEZO1*.
  - Splenectomy should be avoided in patients with some forms of hereditary stomatocytosis as it may predispose the patient to life-threatening thrombotic events.

RBC Enzymopathies

More than 20 recognized disorders caused by deficiencies of enzymes involved with glycolysis, hexose monophosphate shunt, glutathione metabolism, and nucleotide metabolism.

Tests to Consider

Hereditary Hemolytic Anemia Cascade 3000894

Method: High Performance Liquid Chromatography (HPLC)/Electrophoresis/RBC Solubility/Polymerase Chain Reaction (PCR)/Fluorescence Resonance Energy Transfer/Sequencing, Spectrophotometry, Visual Identification, Quantitative Enzymatic, Quantitative Flow Cytometry, Cytochemical Stain, Multiplex Ligation-Dependent Probe Amplification

Optimal test to evaluate individuals with hereditary hemolytic anemia or unexplained long-standing hemolytic anemia

Cascade testing:

Initial testing includes peripheral blood smear evaluation, osmotic fragility testing, unstable hemoglobin evaluation, hemoglobinopathy testing using high-performance liquid chromatograqphy (HPLC), capillary electrophoresis, and red blood cell solubility. In addition to these tests, qualitative flow cytometry (RBC band 3 protein reduction in hereditary spherocytosis) and quantitative enzymatic testing (pyruvate kinase and glucose-6-phosphate dehydrogenase) are performed.

A hematopathologist on the faculty of the University of Utah School of Medicine personally directs and interprets each stage of testing through to completion.

Molecular reflex testing for hemoglobinopathies/thalassemia, G6PD deficiency, and pyruvate kinase deficiency (PKD) will be performed to confirm as needed.

Hereditary hemolytic anemia next generation sequencing (NGS) gene panel will be performed in complex cases of transfusion-dependent hemolytic anemia.

A comprehensive report is provided.

Submit a recent CBC along with transfusion history to inform reflex cascade testing and aid in result interpretation.

See Test Description for cascade components. Tests listed may also be ordered individually.

Hereditary hemolytic anemia testing strategy:

See ARUP Consult's Hemolytic Anemias Testing Algorithm.

- Common forms:
  - G6PD deficiency (*G6PD*)
  - Pyruvate kinase deficiency (*PKLR*)
  - Pyrimidine 5'-nucleotidase (*NT5C3A*)
- Associated findings:
  - Usually normocytic normochromic hemolytic anemia with no specific abnormalities of RBC morphology
  - Severity of hemolysis variable and may be a result of an external stressor (eg, infection, administration of drugs, or ingestion of some foods)
  - Nonhematological manifestations may include:
    - Myopathy
    - Neurological dysfunction
    - Intellectual disability

Hemoglobinopathies

Quantitative defect in biosynthesis of one type of hemoglobin (Hb) chain or a structurally abnormal Hb:

- Alpha or beta thalassemia results from a quantitative defect in the synthesis of either the alpha- or beta-globin chain
  - The unpaired subunits precipitate, bind to the RBC membrane, and lead to hemolysis
- Structural Hb variants result from a structurally abnormal Hb that may polymerize, precipitate, or crystalize within the RBC, leading to membrane changes and hemolysis

Prevalence

HHA disorders: 1/500-1,100

- HS: 1/2,000 northern Europeans
- HE/HPP: 1/2,000-4,000 worldwide
- G6PD deficiency: 400 million worldwide
  - Varies by ethnicity: 7/10 Kurdish Jewish males, 1/6-10 African American males, 1/7-9 Arabic males, 1/6-16 Southeast Asian males
- PKD: 1/20,000 Europeans

Inheritance

Dependent on gene: autosomal recessive, autosomal dominant, or X-linked

Test Description

Cascade Components (Always Performed)

- Blood Smear with Interpretation 3001947
- Osmotic Fragility, Erythrocyte 2002257
- Hemoglobin, Unstable 0049020
- Hemoglobin Evaluation with Reflex to Electrophoresis and/or RBC Solubility 0050610
- RBC Band 3 Protein Reduction in Hereditary Spherocytosis 2008460
- Glucose-6-Phosphate Dehydrogenase 0080135
- Pyruvate Kinase 0080290

Reflexive Cascade Components (Performed as Needed)

- Alpha Globin (HBA1 and HBA2) Deletion/Duplication 2011622
- Beta Globin (HBB) Gene Sequencing 0050578
- Glucose-6-Phosphate Dehydrogenase Deficiency (G6PD) Sequencing 2007163
- Hereditary Hemolytic Anemia Panel Sequencing 2012052
  - See Genes Tested table for genes included in the hereditary hemolytic anemia sequencing panel.

Clinical Sensitivity

Variable and dependent on phenotype/condition

Limitations

- A negative result does not exclude a heritable form of hemolytic anemia.
- This testing is not suitable for acquired causes of hemolytic anemia.
- See individual components for limitations, analytical sensitivity/specificity, interpretation considerations, and reference intervals.

Additional Resources

Bolton-Maggs PHB, Langer JC, Iolascon A, et al. Guidelines for the diagnosis and management of hereditary spherocytosis--2011 update. Br J Haematol. 2012;156(1):37-49. PubMed

Gallagher PG. Abnormalities of the erythrocyte membrane. Pediatr Clin North Am. 2013;60(6):1349-1362. PubMed

Koralkova P, van Solinge WW, van Wijk R. Rare hereditary red blood cell enzymopathies associated with hemolytic anemia - pathophysiology, clinical aspects, and laboratory diagnosis. Int J Lab Hematol. 2014;36(3):388-397. PubMed

Related Information

Hemolytic Anemias
Hemolytic Anemias Testing Algorithm
Hereditary Hemolytic Anemia Panel, Sequencing

Related Tests

Alpha Globin (HBA1 and HBA2) Sequencing and Deletion/Duplication (Extended TAT as of 1/11/21-no referral available) 2011708

Method: Polymerase Chain Reaction/Sequencing./Multiplex Ligation-dependent Probe Amplification.

Beta Globin (HBB) Sequencing and Deletion/Duplication (Temporary Referral as of 12/07/20) 2010117

Method: Polymerase Chain Reaction/Sequencing/Multiplex Ligation-dependent Probe Amplification

Familial Mutation, Targeted Sequencing 2001961

Method: Polymerase Chain Reaction/Sequencing

Glucose-6-Phosphate Dehydrogenase (G6PD) 2 Mutations 0051684

Method: Polymerase Chain Reaction/TaqMAN

Hemoglobin Evaluation Reflexive Cascade 2005792

Method: High Performance Liquid Chromatography/Electrophoresis/RBC Solubility/Polymerase Chain Reaction/Fluorescence Resonance Energy Transfer/Sequencing

Pyruvate Kinase Deficiency (PKLR) Sequencing 3002059

Method: Polymerase Chain Reaction/Sequencing

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