

Overview

Useful For

The definitive evaluation of an individual with *JAK2*-negative erythrocytosis associated with lifelong sustained increased RBC mass, elevated RBC count, hemoglobin, or hematocrit

Genetics Test Information

This is a third-order test and should be ordered when the patient meets the following criteria: diagnosis of erythrocytosis, serum erythropoietin levels are normal, and p50 values are normal.

Testing Algorithm

This evaluation is recommended for patients presenting with lifelong erythrocytosis, usually with a positive family history of similar symptoms. Polycythemia vera should be excluded prior to testing as it is much more common than hereditary erythrocytosis and can be present even in young patients. A *JAK2* V617F or *JAK2* exon 12 mutation should not be present. Additionally, p50 testing should be performed and a normal result confirmed before ordering this test. Serum Epo levels are typically normal (inappropriately so for the level of hemoglobin). For a complete evaluation including p50 testing, hemoglobin electrophoresis testing, and hereditary erythrocytosis mutation analysis in an algorithmic fashion, order REVP / Erythrocytosis Evaluation.

Method Name

Only orderable as part of a profile. For more information see HEMP / Hereditary Erythrocytosis Mutations.

Polymerase Chain Reaction (PCR) Amplification/Sanger Sequence Analysis

NY State Available

Yes

Specimen

Specimen Type

Whole blood

Additional Testing Requirements

This test does not provide a serum erythropoietin (EPO) level. If EPO testing is desired, see EPO / Erythropoietin (EPO), Serum.

Specimen Required

Only orderable as part of a profile. For more information see HEMP / Hereditary Erythrocytosis Mutations.

Specimen Minimum Volume

0.5 mL

Specimen Stability Information

Specimen Type	Temperature	Time
Whole blood	Refrigerated	30 days

Clinical and Interpretive

Clinical Information

Erythrocytosis (increased RBC mass or polycythemia) may be primary, due to an intrinsic defect of bone marrow stem cells (polycythemia vera: PV), or secondary, in response to increased erythropoietin (EPO) levels. Secondary erythrocytosis is associated with a number of disorders including chronic lung disease, chronic increase in carbon monoxide (due to smoking), cyanotic heart disease, high-altitude living, renal cysts and tumors, hepatoma, and other EPO-secreting tumors. When extrinsic causes of erythrocytosis are excluded, a heritable cause intrinsic to the RBC or erythrocyte regulatory mechanisms may be suspected.

Mutations in genes coding for hemoglobin (high-oxygen-affinity hemoglobin variants), hemoglobin-stabilization proteins (2,3 bisphosphoglycerate deficiency), the erythropoietin receptor and oxygen-sensing pathway enzymes (hypoxia-inducible factor, prolyl hydroxylase domain, and von Hippel Lindau) can result in erythrocytosis (see Table).

Erythrocytosis Testing			
Gene	Inheritance	Serum EPO	p50
<i>JAK2V617F</i>	Acquired	Decreased	Normal
<i>JAK2</i> exon 12	Acquired	Decreased	Normal
<i>EPO</i>	Dominant	Decreased to Normal	Normal
<i>PHD2</i>	Dominant	Normal	Normal
<i>BPGM</i>	Dominant	Normal	Decreased
Beta Globin	Dominant	Increased to Normal	Decreased
Alpha Globin	Dominant	Increased to Normal	Decreased
<i>HIF2A</i>	Dominant	Increased to Normal	Normal
<i>VHL</i>	Recessive	Increased	Normal

The oxygen sensing pathway functions through an enzyme, hypoxia-inducible factor (HIF) that regulates RBC mass. A heterodimer protein comprised of alpha and beta subunits, HIF functions as a marker of depleted oxygen concentration. When present, oxygen becomes a substrate mediating HIF-alpha subunit degradation. In the absence of oxygen, degradation does not take place and the alpha protein component is available to dimerize with a HIF-beta subunit. The heterodimer then induces transcription of many hypoxia response genes including *EPO*. HIF-alpha is regulated by von Hippel-Lindau (vHL) protein-mediated ubiquitination and proteosomal degradation, which requires prolyl hydroxylation of the serine and histidine residues. Enzymes important in the hydroxylation of HIF are the prolyl hydroxylase domain proteins, which have 3 isoforms-PHD1, PHD2, and PHD3. The most significant isoform associated with erythrocytosis is PHD2. PHD enzymes are oxygen dependent and have an iron-containing active site. Ascorbic acid enhances, but is not essential for, the activity of PHD. Therefore, activity can be modulated by low iron and ascorbic acid levels as well as by low oxygen. Clinically significant *PHD2* (official designation *EGLN1* [egl nine homolog 1]) mutations are heterozygous and have been found in exons 1 through 4. These mutations result in amino acid substitutions and are associated with inappropriately normal EPO levels.

Reference Values

Only orderable as part of a profile. For more information see HEMP / Hereditary Erythrocytosis Mutations.

Interpretation

An interpretive report will be provided as a part of the HEMP / Hereditary Erythrocytosis Mutations, and will include specimen information, assay information, and whether the specimen was positive for any mutations in the gene. If positive, the mutation will be correlated with clinical significance, if known.

Cautions

Polycythemia vera and acquired causes of erythrocytosis should be excluded before ordering this evaluation. The p50 value should be normal.

This test is not intended for prenatal diagnosis.

This test will not detect somatic or gonadal mosaicism.

Certain sequence alterations have no clinical manifestations and, in essence, are clinically benign. Correlation with all relevant clinical information is necessary to provide appropriate patient care.

Performance

Method Description

DNA is extracted from whole peripheral blood and amplified in 7 separate PCR reactions to cover *EPOR* exon 8, *HIF2A* exons 9 and 12, and *PHD2* exons 1 through 5. PCR products are then sequenced by the Sanger sequencing method and analyzed with sequencing software. Patient sequence results are compared with the genomic reference sequences and the single nucleotide polymorphisms known to occur in the genes. If a mutation is detected, the messenger RNA reference sequence will be used to determine the amino acid number and resulting amino acid change if there is one. (Percy MJ, McMullin MF, Roques AW, et al: Erythrocytosis due to a mutation in the erythropoietin receptor gene. *Br J Haematol* 1998;100:407-410; Martini M, Teofili L, Cenci T, et al: A novel heterozygous HIF2a[M535I] mutation reinforces the role of oxygen sensing pathway disturbances in the pathogenesis of familial erythrocytosis. *Haematologica* 2008;93[7]:1068-1071; Percy MJ, Zhao Q, Flores A, et al: A family with erythrocytosis establishes a role for prolyl hydroxylase domain protein 2 in oxygen homeostasis. *PNAS* 2006;103[3]:654-659)

PDF Report

No

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-Unlisted molecular pathology procedure

LOINC® Information

Test ID	Test Order Name	Order LOINC Value
PHD2	PHD2 Gene, Mutation Analysis, B	In Process

Result ID	Test Result Name	Result LOINC Value
34646	PHD2 Gene Sequencing Result	82939-0