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 test is a third-order test and should be ordered when the patient meets the following criteria: diagnosis of lifelong and sustained erythrocytosis, JAK2 V617F is negative, serum erythropoietin levels are decreased to 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. 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

<table>
<thead>
<tr>
<th>Specimen Type</th>
<th>Temperature</th>
<th>Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Whole blood</td>
<td>Refrigerated</td>
<td>30 days</td>
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</table>
Clinical and Interpretive

Clinical Information

Erythrocytosis (ie, increased RBC mass or polycythemia) may be primary, due to an intrinsic defect of bone marrow stem cells (ie, polycythemia vera: PV), or secondary, in response to increased serum 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 these common causes of secondary erythrocytosis are excluded, a heritable cause involving hemoglobin or erythrocyte regulatory mechanisms may be suspected.

Unlike polycythemia vera, hereditary erythrocytosis is not associated with the risk of clonal evolution and should present with isolated erythrocytosis that has been present since birth. A small subset of cases is associated with pheochromocytoma and/or paraganglioma formation. It is caused by mutations in several genes and may be inherited in either an autosomal dominant or autosomal recessive manner. A family history of erythrocytosis would be expected in these cases, although it is possible for new mutations to arise in an individual.

The genes coding for hemoglobin, beta globin and alpha globin (high-oxygen-affinity hemoglobin variants), hemoglobin-stabilization proteins (2,3 bisphosphoglycerate mutase: \(BPGM\)), and the erythropoietin receptor, \(EPOR\), and oxygen-sensing pathway enzymes (hypoxia-inducible factor: \(HIF/EPAS1\), prolyl hydroxylase domain: \(PHD2/EGLN1\), and von Hippel Lindau: \(VHL\)) can result in hereditary erythrocytosis (see Table). High-oxygen-affinity hemoglobin variants and \(BPGM\) abnormalities result in a decreased p50 result, whereas those affecting \(EPOR, HIF, PHD,\) and \(VHL\) have normal p50 results. The true prevalence of hereditary erythrocytosis-causing mutations is unknown.

### Genes Associated with Hereditary Erythrocytosis

<table>
<thead>
<tr>
<th>Gene</th>
<th>Inheritance</th>
<th>Serum EPO</th>
<th>p50</th>
</tr>
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<tbody>
<tr>
<td>JAK2 V617F</td>
<td>Acquired</td>
<td>Decreased</td>
<td>Normal</td>
</tr>
<tr>
<td>JAK2 exon 12</td>
<td>Acquired</td>
<td>Decreased</td>
<td>Normal</td>
</tr>
<tr>
<td>EPOR</td>
<td>Dominant</td>
<td>Decreased to normal level</td>
<td>Normal</td>
</tr>
<tr>
<td>PHD2/EGLN1</td>
<td>Dominant</td>
<td>Normal level</td>
<td>Normal</td>
</tr>
<tr>
<td>BPGM</td>
<td>Recessive</td>
<td>Normal level</td>
<td>Decreased</td>
</tr>
<tr>
<td>Beta Globin</td>
<td>Dominant</td>
<td>Normal level to increased</td>
<td>Decreased</td>
</tr>
<tr>
<td>Alpha Globin</td>
<td>Dominant</td>
<td>Normal level to increased</td>
<td>Decreased</td>
</tr>
<tr>
<td>HIF2A/EPAS1</td>
<td>Dominant</td>
<td>Normal level to increased</td>
<td>Normal</td>
</tr>
<tr>
<td>VHL</td>
<td>Recessive</td>
<td>Markedly Increased</td>
<td>Normal</td>
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</table>

The oxygen-sensing pathway functions through an enzyme, hypoxia-inducible factor (HIF), which 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 \(EPO, VEGF,\) and \(GLUT1\). HIF-alpha is regulated by von Hippel-Lindau (VHL) protein-mediated ubiquitination and proteosomal degradation, which requires prolyl hydroxylation of HIF proline residues. The HIF-alpha subunit is encoded by the \(HIF2A\) (official name \(EPAS1\)) gene. Enzymes important in the hydroxylation of HIF-alpha are the prolyl hydroxylase
domain proteins, of which the most significant isoform is PHD2, which is encoded by the *PHD2* (official name *EGLN1*) gene. Mutations resulting in altered HIF-alpha, PHD2, and VHL proteins can lead to clinical erythrocytosis. A small subset of mutations, in *PHD2* and *HIF2A*, has also been detected in erythrocytic patients presenting with paragangliomas or pheochromocytomas.

Truncating mutations in the *EPOR* gene coding for the erythropoietin receptor can result in erythrocytosis through loss of the negative regulatory cytoplasmic SHP-1 binding domain leading to EPO hypersensitivity. All currently known mutations have been localized to exon 8, are mainly missense or small deletion and insertions resulting in stop codons, and are heterozygous. *EPOR* mutations are associated with decreased to normal EPO levels and normal p50 values (see Table).

**Reference Values**

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

An interpretive report will be provided.

**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**

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

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

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<tr>
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