MAYO CLINIC LABORATORIES

Test Definition: CALX

CALR Mutation Analysis, Myeloproliferative Neoplasm (MPN), Reflex, Varies

Overview

Useful For

Aiding in the distinction between a reactive cytosis and a chronic myeloproliferative disorder

Evaluating mutations in *CALR* in an algorithmic process for the MPNR / Myeloproliferative Neoplasm, *JAK2* V617F with Reflex to *CALR* and *MPL*, Varies

Method Name

Only orderable as a reflex. For more information see MPNR / Myeloproliferative Neoplasm, *JAK2* V617F with Reflex to *CALR* and *MPL*, Varies.

Polymerase Chain Reaction (PCR) and Fragment Analysis

NY State Available

Yes

Specimen

Specimen Type

Varies

Specimen Required

Only orderable as a reflex. For more information see MPNR / Myeloproliferative Neoplasm, *JAK2* V617F with Reflex to *CALR* and *MPL*, Varies.

Submit only 1 of the following specimens:

Specimen Type: Whole Blood
Container/Tube: Lavender top (EDTA) or yellow top (ACD solution B)
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. Label specimen as blood.
Specimen Stability Information: Ambient (preferred)7 days/Refrigerate 7 days

Specimen Type: Bone marrow
Container/Tube: Lavender top (EDTA) or yellow top (ACD solution B)
Specimen Volume: 2 mL

Collection Instructions:
1. Invert several times to mix bone marrow.
2. Send bone marrow specimen in original tube. **Do not** aliquot.
3. Label specimen as bone marrow.
Specimen Stability Information: Ambient (preferred)7 days/Refrigerate 7 days

Specimen Type: Extracted DNA from blood or bone marrow
Container/Tube: 1.5- to 2-mL tube
Specimen Volume: Entire specimen
Collection Instructions:
1. Label specimen as extracted DNA from blood or bone marrow.
2. Provide volume and concentration of the DNA on the label.
Specimen Stability Information: Frozen (preferred)/Refrigerate/Ambient

Specimen Minimum Volume
Whole blood/Bone marrow: 1 mL

Reject Due To

Gross hemolysis	Reject
Paraffin shavings	Reject
Slides	Reject
Moderately to severely clotted	Reject
Paraffin embedded bone marrow aspirate clot or biopsy blocks	Reject

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Varies	Varies	7 days	

Clinical & Interpretive

Clinical Information
The *JAK2* (Janus kinase 2) gene codes for a tyrosine kinase (JAK2) associated with the cytoplasmic portion of a variety of transmembrane cytokine and growth factor receptors important for signal transduction in hematopoietic cells. Signaling

via JAK2 activation causes phosphorylation of downstream signal transducers and activators of transcription (STAT) proteins (eg, STAT5) ultimately leading to cell growth and differentiation. *BCR*::*ABL1*-negative myeloproliferative neoplasms (MPN) frequently harbor an acquired single nucleotide mutation in *JAK2* characterized as c.G1849T; p.Val617Phe (V617F). The *JAK2* V617F is present in 95% to 98% of polycythemia vera cases and 50% to 60% of primary myelofibrosis (PMF) and essential thrombocythemia (ET) cases. It has also been described infrequently in other myeloid neoplasms, including chronic myelomonocytic leukemia and myelodysplastic syndrome. Detection of the *JAK2* V617F is useful to help establish the diagnosis of MPN. However, a negative *JAK2* V617F result does not indicate the absence of MPN. Other important molecular markers in *BCR*::*ABL1*-negative MPN include *CALR* exon 9 mutation (20%-30% of PMF and ET) and *MPL* exon 10 mutation (5%-10% of PMF and 3%-5% of ET). Mutations in *JAK2*, *CALR*, and *MPL* are essentially mutually exclusive. A *CALR* mutation is associated with decreased risk of thrombosis in both ET and PMF and confers a favorable clinical outcome in PMF patients. A triple negative (*JAK2* V617F, *CALR*, and *MPL*-negative) genotype is considered a high-risk molecular signature in PMF.

Reference Values

Only orderable as a reflex. For more information see MPNR / Myeloproliferative Neoplasm, *JAK2* V617F with Reflex to *CALR* and *MPL*, Varies.

An interpretive report will be provided.

Interpretation

An interpretation will be provided under the MPNR / Myeloproliferative Neoplasm, *JAK2* V617F with Reflex to *CALR* and *MPL*, Varies.

Cautions

A positive result is not specific for a particular subtype of myeloproliferative neoplasm and clinicopathologic correlation is necessary in all cases.

A negative result does not exclude the presence of a myeloproliferative neoplasm or other neoplastic process.

In rare cases, a mutation other than the V617F may be present in an area that interferes with primer or probe binding and cause a false-negative result.

Clinical Reference

1. Klampfl T, Gisslinger H, Harutyunyan AS, et al. Somatic mutation of calreticulin in myeloproliferative neoplasms. N Engl J Med. 2013;369(25):2379-2390
2. Nangalia J, Massie CE, Baxter EJ, et al. Somatic CALR mutation in myeloproliferative neoplasms with nonmutated JAK2. N Engl J Med. 2013;369(25):2391-2405
3. Rumi E, Pietra D, Ferretti V, et al. JAK2 or CALR mutation status defines subtypes of essential thrombocythemia with substantially different clinical course and outcomes. Blood. 2014;123(10):1544-1551
4. Rotunno G, Mannarelli C, Guglielmelli P, et al. Impact of calreticulin mutations on clinical and hematological phenotype and outcome in essential thrombocythemia. Blood. 2014;123(10):1552-1555
5. Tefferi A, Lasho TL, Finke CM, et al. CALR vs JAK2 vs MPL-mutated or triple-negative myelofibrosis: clinical, cytogenetic and molecular comparisons. Leukemia. 2014;28(7):1472-1477
6. Greenfield G, McMullin MF, Mils K. Molecular pathogenesis of the myeloproliferative neoplasms. J Hematol Oncol. 2021;14(1):103

Performance

Method Description
Polymerase chain reaction (PCR) amplification of *CALR* exon 9 is performed on DNA isolated from the patient sample. The PCR product is then run on an ABI Genetic Analyzer for fragment analysis to detect insertions and deletions. An unmutated *CALR* will show an amplicon at 266 base pairs (bp), a mutated *CALR* with insertion will show an amplicon greater than 266 bp, and a mutated *CALR* with deletion will show an amplicon smaller than 266 bp. This assay has an analytical sensitivity of approximately 6% (ie, 6 mutation-containing cells in 100 total cells) in most mutation types, except for the rare type of 1-bp deletion, which has a sensitivity of approximately 20%.(Unpublished Mayo method)

PDF Report
No

Day(s) Performed
Monday through Friday

Report Available
7 to 10 days

Specimen Retention Time
Whole blood/Bone marrow: 2 weeks; 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
81219-*CALR* (calreticulin) (eg, myeloproliferative disorders), gene analysis, common variants in exon 9

LOINC® Information

Test Definition: CALX

CALR Mutation Analysis, Myeloproliferative Neoplasm (MPN), Reflex, Varies

Test ID	Test Order Name	Order LOINC® Value
CALX	CALR, Gene Mutation, Exon 9, Reflex	77174-1

Result ID	Test Result Name	Result LOINC® Value
36998	Final Diagnosis	22637-3