CALR Mutation Analysis, Myeloproliferative Neoplasm (MPN)

Useful For

- Aiding in distinction between reactive cytosis and myeloproliferative neoplasm, especially essential thrombocythemia (ET) and primary myelofibrosis (PMF), and also possibly for disease monitoring

- In particular, detection of CALR mutation is highly informative in cases of suspected myeloproliferative neoplasm (MPN) in which JAK2 and MPL testing are negative

Clinical Information

The most frequent genetic mutation in BCR-ABL1-negative myeloproliferative neoplasm (MPN), essential thrombocythemia (ET), and primary myelofibrosis (PMF) is the JAK2V617F mutation, which is present in approximately 50% to 60% of patients. It serves as a confirmatory molecular marker of these diseases. Mutations in the MPL gene are found in an additional 5% to 10% of ET and PMF cases. It was recently discovered that somatic mutation (insertions and/or deletions) in exon 9 of the CALR gene is the second most frequent somatic mutation after JAK2 in ET and PMF patients, and it is mutually exclusive of JAK2 and MPL mutations. It has a frequency of approximately 49% to 88% in JAK2 and MPL-wild type (WT) ET and PMF, and is not found in polycythemia vera (PV) patients. Therefore, CALR mutation serves as an important diagnostic molecular marker in ET and PMF.

The CALR gene encodes for calreticulin, a multifunctional protein with a C-terminus rich in acidic amino acids and a KDEL ER-retention motif. All the pathologic CALR mutations reported to date are out-of-frame insertion and/or deletions (indel) in exon 9, generating a 1 base-pair (bp) frame shift and a mutant protein with a novel C-terminus rich in basic amino acids and loss of the KDEL ER-retention signal. The most common mutation types are 52-bp deletion (c.1092_1143del, L367fs*46) and 5-bp insertion (c.1154_1155insTTGCC, K385fs*47), and they comprise approximately 85% of CALR mutations in MPN. CALR mutations have been found in hematopoietic stem and progenitor cells in MPN patients and may activate the STAT5 signaling pathway. They are associated with decreased risk of thrombosis in both ET and PMF, and better survival in PMF compared to JAK2 mutations.
Interpretation
An interpretive report will be issued.

The results will be reported as 1 of the 3 states if DNA amplification is successful (see Cautions):
- Positive. A deletion/insertion-type mutation was detected in CALR, exon 9.
- Negative. No deletion or insertion was detected in CALR, exon 9.
- Equivocal. A small amplicon suspicious for a deletion/insertion type mutation was detected in CALR, exon 9.

Positive mutation status is highly suggestive of a myeloid neoplasm, but must be correlated with clinical and other laboratory and morphologic features for definitive diagnosis.

Negative mutation status does not exclude the presence of a myeloproliferative neoplasm or other neoplastic disorders.

Cautions
A positive result is not specific for a particular myeloproliferative neoplasm (MPN) diagnosis and clinicopathologic correlation is necessary in all cases.

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

This test is a fragment analysis assay, and only detects insertions and deletions (indels). It will not detect point mutations. However, all reported pathologic mutations in MPN described to date are insertions and/or deletions.

This test does not differentiate between out-of-frame and in-frame indels. However, in-frame indel mutations are very rare (<0.5%), and have only been reported in few healthy individuals and myeloproliferative neoplasm patients with JAK2V617F mutation or out-of-frame CALR mutation. Most of the rare in-frame indels are considered germline mutations and may represent non-pathogenic polymorphisms.

Infrequently, amplification failure can be encountered in a given sample, due to inadequate DNA, poor DNA quality, or a PCR inhibitor. In these circumstances, the assay will be reattempted and if persistently unsuccessful, the report will be issued with an "Invalid" result.

Clinical References


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. Published online before print December 23, 2013


For additional interpretation information and clinical references, please visit the following website:
http://www.mayomedicallaboratories.com/index.html