PRECIPIO EXPECT MORE
203-787-1717
203-901-1289

MOLECULAR

Patient: John A. Doe
DOB/Gender: 10/10/44 (74 yrs) - Male
Patient ID/MRN: 123456
Date Collected: 04/09/2019

Case#/Status: X19-00633 - Final

Report Category:
Detected

Provider: Jane Smith, M.D.
Hematology Oncology Associates
Tel: 800-123-4567
Fax: 800-765-4321

DIAGNOSIS:

Peripheral blood:
HemeScreen™ results reveal:
- Negative for JAK2 V617F point mutation
- Negative for JAK2 exon 12 mutations
- Negative for MPL W515L/K point mutations
- The specimen tested positive for calreticulin (CALR) mutation.
  CALR Percentage: 44.3
  CALR Mutation(s): c. 1099_1150del (52bp deletion)

INTERPRETATION

(HemeScreen): The V617F mutation of the JAK2 (Janus kinase 2) gene has been described in polycythemia vera (PV), essential thrombocythemia (ET) and primary myelofibrosis (PMF) cases.[1-2] Identification of the V617F JAK2 point mutation in myeloproliferative neoplasms (MPN) is indicated in diagnosis, classification and monitoring.

Mutations in the JAK2 exon 12 (Janus kinase 2) gene are rare in ET or PMF, and their occurrence in PV is almost always associated with the absence of JAK2V617F and the presence of a subnormal serum erythropoietin level.[3] The identification of the JAK2 exon 12 mutations in myeloproliferative neoplasms (MPN) may be useful to assist diagnosis, classification and monitoring.

MPL (W515L/K) mutations of the juxtamembrane region of the thrombopoietin receptor MPL (myeloproliferative leukemia virus oncogene homology) have been described in JAK2 V617F-negative primary myelofibrosis (PMF) and essential thrombocythemia (ET).[4] The identification of W515 L/K point mutations in myeloproliferative neoplasms (MPN) may be useful to assist diagnosis, classification and monitoring.

REFERENCES:
1. Baxter et al. The Lancet 2005: 1054 - 1061
2. Levine et al. Cancer Cell 2005: 387-397
3. Pardanani et al Leukemia 21: 2007; Pietra et al Blood 111: 2008
4. Pancrazzi et al. JMD 2008: 435 - 441

(CALR): Calreticulin (CALR) is endoplasmic reticulum protein that binds calcium and plays a role in signaling and protein expression. It is also found in the nucleus and believed to play a role in transcription regulation. Somatic insertions or deletions in exon 9 of CALR gene are detected in 67% of JAK2/MPL negative essential thrombocythemia (ET) and 88% of JAK2/MPL negative primary myelofibrosis (PMF) patients. CALR mutations are not detected in polycythemia vera (PV) patients. CALR mutations appear to be mutually exclusive of JAK2 and MPL mutations. It has been reported that patients with mutated CALR have a lower risk of thrombosis and longer overall survival than patients with JAK2 mutation.

REFERENCES:
Nangalia J, et al. Somatic CALR mutations in myeloproliferative neoplasms with nonmutated JAK2,
N Engl J Med. 2013 Dec 19;369(25):2391-405.
Klampfl T, et al. Somatic mutations of calreticulin in myeloproliferative neoplasms.
N Engl J Med. 2013 Dec 19;36 (25):2379-90.

METHOD:
(Hemescreen): Melting curve analysis in combination with real-time PCR is a natural extension of continuously monitored PCR within each cycle. During high resolution DNA melting analysis (HRM or HRMA), melting curves are produced using dyes that fluoresce in the presence of double-stranded DNA (dsDNA). Using specialized instruments designed to monitor fluorescence during heating; as the temperature increases, the fluorescence decreases, producing a characteristic melting profile.

This assay can detect mutations with a minimum sensitivity of 5~10% depending on the wild type background in the specimen. Although molecular testing is highly accurate rarely false-positive and false-negative diagnostic errors may occur.

(CALR): The CALR mutation analysis is performed by both fragment analysis and Sanger sequencing methods. Patient nucleic acid is extracted; mutation hot spots in exon 9 are amplified. The products are sequenced in both directions and point mutations as well as SNPs are identified. Fragment length analysis is performed to further determine very low levels of heterozygous insertions/deletions, which may be missed by sequencing. All mutations, including heterozygous indels, will be reported. This assay has a sensitivity of 10-15% for detecting point mutations and 5% for detecting heterozygous insertion/deletions in the wild-type background. Various factors including quantity, quality and sample age can affect assay performance.

DISCLAIMER:
The adequacy of staining is verified by the appropriate LSI controls. The reagents used for these assays are for research use only (RUO). Their performance characteristics have been initiated by Precipio, Inc., New Haven, CT. They have not been reviewed by the FDA. The FDA has deemed that such approval is unwarranted for clinical use. These assays should be viewed as experimental and/or research use only.

CLINICAL DATA

ICD-10: Secondary polycythemia

Received CBC, reported on 04/09/2019: WBC 6.2; RBC 5.68; HGB 18.2; HCT 53.6; MCV 94.0; MCH 32.1; MCHC 34.0; RDW 12.3%; PLT 147; MPV 7.1; LYM 29.8%; MON 4.8%; NEU NP; EOS NP; BAS NP (NP = not provided)

Electronically Signed By: Frank Bauer, MD (04/11/19 13:00)

Patient: John A. Doe
Case #: X19-00633
Received Information: 1 lavender-top tube
Received: 04/09/19 11:04
Reported: 04/12/19 13:00