CBFB-MYH11 inv(16) Fusions in Acute Myeloid Leukemia

Indications for Ordering

- Detection of *CBFB-MYH11* fusion transcripts resulting from inv(16)/t(16;16) in patients with acute myeloid leukemia (AML)
  - Necessary for diagnostic subclassification
  - Monitoring response to therapy
  - Detection of minimal residual disease

Test Description

- Reverse transcription polymerase chain reaction (RT-PCR)
- *CBFB-MYH11* fusions are quantitated by real-time PCR amplification
- Primers detect fusions with *CBFB* exon 5 and *MYH11* exons 7, 8, and 12
- Test reference gene is *ABL1*
- Copy numbers of *CBFB-MYH11* fusion transcripts are expressed as the ratio of *CBFB-MYH11*:*ABL1*
- Performed on peripheral blood or bone marrow (BM) specimen

Tests to Consider

Testing Strategy

At diagnosis

- BM cytogenetic studies and FISH are recommended to detect inv(16)/t(16;16)
- Measurement of *CBFB-MYH11* fusion transcripts at diagnosis by RT-PCR may also be helpful

Monitoring response to treatment

- Quantitative RT-PCR
  - Every 3 months when treatment response is evident
  - After complete cytogenetic response has been achieved
    - Every 3 months for 3 years, and every 3-6 months thereafter
  - More frequent monitoring may be required in individuals with increasing level of *CBFB-MYH11* transcripts

Primary test

CBFB-MYH11 inv(16) Detection, Quantitative 2011114

- Use for detection of *CBFB-MYH11* in AML

Related tests

Acute Myeloid Leukemia Panel by FISH 2011132

- Diagnosis, prognosis, and monitoring of AML
- Includes

Probe Target	Gene(s)/Unique Sequence
t(15;17)(q24;q21)	*PML-RARA*
t(8;21)(q22;q22)	*RUNX1T1-RUNX1* (*ETO-AML1*)
inv(16)(p13.3q22)	*CBFB*
11q23	*KMT2A (MLL)*
inv(3) or t(3;3)	*RPN1-MECOM (EVI1)*
del(5)(q31)	*EGR1*
del(7)(q31)/-7	D7S486

Chromosome Analysis, Bone Marrow 2002292

- Diagnosis, prognosis, and monitoring of AML

Chromosome Analysis, Bone Marrow with Reflex to Genomic Microarray 2007130

- Diagnosis, prognosis, and monitoring of AML
- If chromosome analysis is “normal” or “no growth,” then genomic microarray testing will be added

Cytogenomic SNP Microarray – Oncology 2006325

- Preferred test for fresh specimens at time of diagnosis for detecting prognostically important genomic abnormalities in leukemias/lymphomas and solid tumors involving
  - Loss/gain of DNA
  - Loss of heterozygosity (LOH)
  - Monitor disease progression and response to therapy

Chromosome FISH, Interphase 2002298

- Specific FISH probe for inv(16); *CBFB-MYH11* must be requested

Myeloid Malignancies Mutation Panel by Next Generation Sequencing 2011117

- Assess for single gene mutations, including substitutions and insertions and deletions that may have diagnostic, prognostic, and/or therapeutic significance

KIT Mutations in AML by Fragment Analysis and Sequencing 2002437

- Prognostication in core-binding factor-related (CBF) AML

Myeloperoxidase Stain 0049030

Lysozyme (Muramidase) by Immunohistochemistry 2003990

Myeloperoxidase (MPO) by Immunohistochemistry 2004014

Eosinophil Panel by FISH 2002378

CBC with Platelet Count 0040002

Disease Overview

Incidence

- Infants – 1.5/100,000
- Children 5-9 years – 0.4/100,000
- Adults <50 years – 1.0/100,000
- Adults >75 years – 25/100,000
- *CBFB-MYH11* AML – 5-8% of de novo cases of AML

Features

- ~50% of *CBFB-MYH11* AML cases belong to subtype FAB-M4 eos
- Translocation creates fusion between *CBFB* and *MYH11* genes
- Most cases demonstrate 1 of 3 types of breakpoints involving exons 33, 29, and 28 of *MYH11* and exon 5 of *CBFB*
- AML with inv(16); *CBFB-MYH11*
  - Has overall favorable prognosis
  - May be difficult to detect by classic cytogenetics
  - FISH or PCR may be required for detection
- *KIT* gene testing should also be performed
  - Presence of mutation in *KIT* gene is associated with a worse outcome

Genetics

Genes – *CBFB*, *MYH11*

De novo mutations

Chromosome 16 inversion results in fusion of *CBFB* and *MYH11* genes

- Type A - *CBFB* exon 5/*MYH11* exon 12 (88%)
- Type D - *CBFB* exon 5/*MYH11* exon 8 (5%)
- Type E - *CBFB* exon 5/*MYH11* exon 7 (5%)

Test Interpretation

Results

- Detected – *CBFB-MYH11* fusion transcripts detected
  - Diagnostic for AML regardless of the percentage of blasts in BM
- Not detected – *CBFB-MYH11* fusion transcripts not detected
  - Does not exclude the presence of *CBFB-MYH11* transcripts below detection limit of test
- Weakly positive – nonquantifiable

Limitations

- Limit of detection (LOD) for types A, D is one copy
- LOD for *CBFB-MYH11* type E is 10 copies
- Presence of fusion product <10 copies may not be detected
- BM specimens preferred for maximum sensitivity
- Poor RNA yield will lead to false negatives