Eosinophilia Panel by FISH

Indications for Ordering

- Diagnose and classify specific eosinophilic myeloid neoplasms
  - Acute myeloid leukemia (AML) with inv(16) or t(16;16)
  - Myeloid neoplasms with eosinophilia and abnormalities of *PDGFRA*, *PDGFRB*, or *FGFR1*
- Provide prognostic and predictive information for acute or chronic leukemia with eosinophilia
- Monitor therapeutic response

Test Description

- Performed on cultured bone marrow (BM)
  - Peripheral blood may be used
- Multiple fluorescence in situ hybridization (FISH) probes target specific genes
  - *FGFR1* rearrangement
  - *FIP1L1-PDGFRA* region rearrangement
  - *PDGFRB* rearrangement
  - *CBFB/MYH11* rearrangement
- Probes can be run as a panel or individually

Tests to Consider

Primary test

Eosinophilia Panel by FISH 2002378

- Diagnosis, prognosis, and monitoring for newly diagnosed acute or chronic leukemia with eosinophilia

Related tests

Chromosome Analysis, Bone Marrow 2002292

- Diagnosis, prognosis, and monitoring of eosinophilic leukemia

Chromosome Analysis, Bone Marrow with Reflex to Genomic Microarray 2007130

- Diagnosis, prognosis, and monitoring of eosinophilic disorders
- 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 probes must be requested and include
  - *PDGFRA*
  - *PDGFRB*
  - *JAK2*
  - +8
  - +9
  - Monosomy 7 or 7q deletion
  - 5q deletion
  - 13q deletion
  - 20q deletion

Myeloproliferative Disorders Panel by FISH 2002360

- Detect specific recurrent genomic aberrations in suspected MPNs
  - *BCR/ABL1*
  - *PDGFRA*
  - *PDGFRB*
  - *FGFR1*

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

Disease Overview

Consensus criteria

- 2016 WHO classification of eosinophilic myeloid disorders
  - Myeloid and lymphoid neoplasms with *PDGFRA* rearrangement
  - Myeloid and lymphoid neoplasms with *PDGFRB* rearrangement
  - Myeloid and lymphoid neoplasms with *FGFR1* rearrangement
  - Chronic eosinophilic leukemia-not otherwise specified (CEL-NOS)
  - Myeloid and lymphoid neoplasms with *PCM1-JAK2* (provisional entity)

Incidence/prevalence

- *PDGFRA/B-* and *FGFR1*-related disorders are not well characterized
- inv16; t(16;16)
  - 5-8% of AMLs, predominantly in childhood

Diagnostic criteria

See Table 1

Genetics

See Table 2

Test Interpretation

Analytic sensitivity/specificity – >95%

Results

- Normal – no evidence of rearrangement
- Abnormal **–** rearrangement detected
  - Diagnostic of a clonal hematopoietic neoplasm
  - inv(16); t(16;16)
    - Prognosis – favorable in children and adults
      - Less favorable if *KIT* mutation is also present
    - Response to high dose cytarabine- and anthracycline-based chemotherapy – yes
      - Remission rate – 92%
      - 10-year survival – 55%
  - *PDGFRA and PDGFRB*
    - Prognosis – good
    - Response to tyrosine kinase inhibitors (TKIs) such as imatinib – yes
  - *FGFR1*-rearranged
    - Prognosis – poor
    - Response to TKIs such as imatinib – currently unclear
    - Response to chemotherapy protocols developed for acute leukemias – no

Limitations

- Detects only rearrangements targeted by the probes
- *PDGFRB* gene on 5q33 and *FGFR1* gene on 8p11 have multiple rearrangement partners
  - Rearrangement partners are not identified by this test

Table 1

WHO Classification	Features	Laboratory
AML with inv(16)(p13.1q22) or t(16;16)(p13.1;q22); *CBFB-MYH11*	• Presents as AML • Myeloid sarcomas may be present at initial diagnosis or relapse	• Morphology – acute myelomonocytic leukemia with increased eosinophils containing immature eosinophilic granules in the BM ○ Peripheral eosinophilia is unusual ○ Diagnosis of AML even if blasts <20% • Genetics ○ inv(16)(p13.1q22) or t(16;16)(p13.1;q22) found in most cases ▪ inv(16)(p13.1q22) is found in vast majority ▪ FISH or PCR may be necessary to document this genetic alteration ○ Secondary cytogenetic abnormalities – +22, +8, del(7q) ○ *KIT* mutations may be present
Myeloid and lymphoid neoplasms with *PDGFRA* rearrangement	• Most frequently presents as CEL, but may present as AML, T-lymphoblastic lymphoma, or both ○ Acute transformation can follow CEL presentation • Organ infiltration by eosinophils ○ Heart ○ Lungs ○ Central nervous system ○ Gastrointestinal tract • Splenomegaly in majority of patients • Pronounced male predominance	• Morphology ○ Peripheral blood and BM eosinophilia (markedly elevated) ○ Typically <20% blasts in peripheral blood and BM ○ Increased BM mast cells common • Genetics ○ Absence of *BCR-ABL1* fusion gene ○ Most commonly associated with *FIP1L1-PDGFRA* fusion ▪ FISH or PCR is usually necessary to document this genetic alteration; cytogenetic studies are normal ○ Other fusion genes have rarely been identified
Myeloid and lymphoid neoplasms with *PDGFRB* rearrangement	• Presents with features of chronic myelomonocytic leukemia (usually with eosinophilia) • Splenomegaly in majority of patients • Male predominance, but much less marked than *PDGFRA*-associated neoplasms	• Morphology ○ Peripheral leukocytosis ○ Hypercellular BM with typically <20% blasts ○ Increased BM mast cells common • Genetics ○ Most common rearrangement – t(5;12)(q31-33;p13), resulting in *ETV6-PDGFRB* fusion
Myeloid and lymphoid neoplasms with *FGFR1* rearrangement	• Often presents with peripheral eosinophilia in the context of lymphadenopathy and lymphoblastic leukemia/lymphoma • Slight male predominance	• Morphology ○ AML, acute lymphoblastic leukemia (ALL), CEL (usually associated with peripheral blood or BM eosinophilia) • Genetics ○ Presence of t(8;13)(p11;q12) or a variant rearrangement at the 8p11 breakpoint leading to FGFR1 rearrangement ○ Secondary cytogenetic abnormalities – trisomy 21 most often observed

Table 2

Gene	Structure/Function	Mutations	WHO Disease Association
CBFB-MYH11	• *CBFB* ○ 16q22 ○ Core binding transcription factor • *MYH11* ○ 16p13.1 ○ Codes for smooth muscle myosin heavy chain	• inv(16)(p13.1q22) or (t16;16)(p13.1;q22) • Inversion results in fusion of *CBFB* on 16q22 to *MYH11* on 16p13.1	AML with inv(16)(p13.1q22) or t(16;16)(p13.1;q22); previously FAB M4Eo
PDGFRA	• Maps to 4q12 • Cell surface tyrosine kinase receptor for members of the platelet-derived growth factor family • Results in a constitutively active tyrosine kinase oncoprotein	• *FIP1L1-PDGFRA* rearrangement is a karyotypically occult 800-kb interstitial deletion (ie, *CHIC2* deletion)	Myeloid and lymphoid neoplasms with *PDGFRA* rearrangement
PDGFRB	• Maps to 5q31-33 • Cell surface tyrosine kinase receptor for members of the platelet-derived growth factor family • Results in a constitutively active tyrosine kinase oncoprotein	• 20 fusion partners reported • Most common rearrangement – t(5;12)(q31-33;p13) resulting in *ETV6-PDGFRB* fusion	Myeloid and lymphoid neoplasms with *PDGFRB* rearrangement
FGFR1	• Maps to 8p11 • Cell surface tyrosine kinase • Rearrangement results in constitutive activation of *FGFR1* with the fusion of the FGFR1 C-terminal catalytic domain with unrelated proteins	• >10 fusion partners identified • Most common rearrangement – t(8;13)(p11;q12) resulting in *ZNF198-FGFR1* fusion	Myeloid and lymphoid neoplasms with *FGFR1* rearrangement