Myeloproliferative Neoplasms Panel by FISH

Indications for Ordering

- Aids in diagnosis and classification of specific myeloproliferative neoplasms (MPNs) with eosinophilia in conjunction with cytogenetic testing
  - Chronic myelogenous leukemia (CML) – *BCR-ABL1* positive
  - Myeloid and lymphoid neoplasms with eosinophilia and abnormalities of *PDGFRA*, *PDGFRB*, or *FGFR1*
- Monitor minimal residual disease (MRD) in MPNs

Test Description

- Performed on bone marrow (BM)
  - Peripheral blood may be used but not preferred
- Probes target
  - *BCR-ABL1* fusion
  - *FGFR1* translocations
  - *FIP1L1-PDGFRA* region rearrangements
  - *PDGFRB* translocations
- Each probe can be run as a panel or individually

Tests to Consider

Primary test

Myeloproliferative Disorders Panel by FISH 2002360

- Limited role in the workup of myeloproliferative neoplasms in the setting of an otherwise optimal cytogenetic study
- Aids in exclusion of cryptic *BCR-ABL1* rearrangement in CML and in the exclusion of a *PDGFRA* abnormality in cases of neoplastic eosinophilia

Related tests

Chromosome Analysis, Bone Marrow 2002292

- Diagnosis, prognosis, and monitoring of MPNs
- Does not detect insertion in *BCR-ABL* fusion gene

Chromosome Analysis, Bone Marrow with Reflex to Genomic Microarray 2007130

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

Eosinophilia Panel by FISH 2002378

- Diagnosis, prognosis, and monitoring of eosinophilic leukemias
- Probes target
  - *FGFR1* translocations
  - *FIP1L1-PDGFRA* region rearrangements
  - *PDGFRB* translocations
  - *CBFB/MYH11* translocation

Chromosome FISH, Interphase 2002298

- Specific FISH probes must be requested and include
  - *PDGFRA*
  - *PDGFRB*
  - *FGFR1*
  - *BCR-ABL1*
  - inv(16)

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

Diagnostic issues

- FISH analysis has several advantages over chromosome studies in MPN diagnosis
  - More rapid turnaround time
  - Can be performed on interphase cells
  - Can detect subtle or cryptic rearrangements
- For clinical features of specific MPNs, see Table 1

Genetics

See Table 2

Test Interpretation

Analytic sensitivity/specificity – >95%

Results

- Normal – analysis using panel probes *BCR-ABL1* , *PDGFRA*, *PDGFRB*, and *FGFR1* shows no evidence of rearrangement

- Abnormal – rearrangement detected
  - Diagnostic of a clonal hematopoietic neoplasm
  - *BCR-ABL1*
    - Prognosis – good
    - Response to tyrosine kinase inhibitors (TKIs) such as imatinib – yes
  - *PDGFRA and PDGFRB* positive neoplasms
    - Prognosis – good
    - Response to TKIs such as imatinib – yes
  - *FGFR1*-rearranged myeloid/lymphoid neoplasms
    - Prognosis – poor
    - Response to TKIs such as imatinib – currently unclear
    - Response to chemotherapy protocols developed for acute leukemias – no

Limitations

- Only detects rearrangements targeted by the probes
- The translocation partners of the *PDGFRB* gene on 5q33 and *FGFR1* gene on 8p11 have multiple translocation partners
  - These translocation partners are not identified by this test

Table 1. MPN Subtypes, Features, and Diagnostic Criteria

WHO Classification	Features	Laboratory
Chronic myelogenous leukemia, *BCR-ABL1* positive	• Many are asymptomatic, but diagnosed when abnormal CBC is reported • Symptomatic individuals present with fatigue, night sweats, weight loss, and splenomegaly	• Morphology – peripheral leukocytosis o Anemia common • Genetics – majority of individuals have *BCR-ABL1* translocation
Myeloid and lymphoid neoplasms with *PDGFRA* rearrangement	• Most frequently presents as chronic eosinophilic leukemia (CEL), but may present as acute myeloid leukemia (AML), T-cell lymphoblastic lymphoma (T-LBL), or both o Acute transformation can follow CEL presentation • Organ infiltration by eosinophils o Heart o Lungs o CNS o GI tract • Splenomegaly in majority of individuals • Pronounced male predominance	• Morphology o Peripheral blood and BM eosinophilia – markedly elevated o Typically <20% blasts in peripheral blood and BM o Increased BM mast cells common • Genetics o Absence of *BCR-ABL1* fusion gene o Most commonly associated with *FIP1L1-PDGFRA* fusion ▪ FISH or PCR is usually necessary to document this genetic alteration; cytogenetic studies are normal o Other fusion genes have rarely been identified
Myeloid and lymphoid neoplasms with *PDGFRB* rearrangement	• Presents with features of CML (usually with eosinophilia) • Splenomegaly in majority of individuals • Male predominance, but much less marked than *PDGFRA*-associated neoplasms	• Morphology o Peripheral leukocytosis o Hypercellular BM with typically <20% blasts o Increased BM mast cells common • Genetics o Most common translocation – t(5;12)(q31-33;p13) resulting in formation of *ETV6-PDGFRB*
Myeloid and lymphoid neoplasms with *FGFR1* abnormalities	• Often presents with peripheral eosinophilia in the context of lymphadenopathy and lymphoblastic leukemia/lymphoma • Slight male predominance	• Morphology o AML, ALL, CEL (usually associated with peripheral blood or BM eosinophilia) • Genetics o Presence of t(8;13)(p11;q12) or a variant translocation at the 8p11 breakpoint leading to *FGFR1* rearrangement o Secondary cytogenetic abnormalities – trisomy 21 most often observed

Table 2. Overview of Genes and Mutations for Eosinophilic Disorders Tested for by this Panel

Gene	Structure/Function	Mutations	WHO Disease Association
BCR-ABL1	• Maps to 9q24 • Chimeric constitutively active tyrosine kinase	• Majority of cases are translocation t(9;22) (q34q11.2) o Results in Philadelphia chromosome • Remaining cases have variant translocations that involve a 3rd or 4th chromosome or cryptic translocation	CML, *BCR-ABL1* positive
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

Gene	Structure/Function	Mutations	WHO Disease Association
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 translocation – t(5;12)(q31-33;p13); *ETV6-PDGFRB*	Myeloid and lymphoid neoplasms with *PDGFRB* rearrangement
FGFR1	• Maps to 8p11 • Cell surface tyrosine kinase • Translocations result in constitutive activation of *FGFR1* with the fusion of the *FGFR1* C-terminal catalytic domain with unrelated proteins	• >10 fusion partners identified • Most common translocation – t(8;13)(p11;q12); ZNF198-*FGFR1* mutation	Myeloid and lymphoid neoplasms with *FGFR1* abnormalities