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, prognostic, and monitoring of eosinophilic leukemias
- Probes target
  - FGFR1 translocations
  - FIP1L1-PDGFRA region rearrangements
  - PDGFRB translocations
  - CBF/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
  o Diagnostic of a clonal hematopoietic neoplasm
  o \textit{BCR-ABL1}
    ▪ Prognosis – good
    ▪ Response to tyrosine kinase inhibitors (TKIs) such as imatinib – yes
  o \textit{PDGFR}A and \textit{PDGFRB} positive neoplasms
    ▪ Prognosis – good
    ▪ Response to TKIs such as imatinib – yes
  o \textit{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 \textit{PDGFRB} gene on 5q33 and \textit{FGFR1} gene on 8p11 have multiple translocation partners
  o These translocation partners are not identified by this test

Table 1. MPN Subtypes, Features, and Diagnostic Criteria

<table>
<thead>
<tr>
<th>WHO Classification</th>
<th>Features</th>
<th>Laboratory</th>
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</thead>
<tbody>
<tr>
<td>Chronic myelogenous leukemia, \textit{BCR-ABL1} positive</td>
<td>Many are asymptomatic, but diagnosed when abnormal CBC is reported</td>
<td>Morphology – peripheral leukocytosis</td>
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<td>Symptomatic individuals present with fatigue, night sweats, weight loss, and splenomegaly</td>
<td>Anemia common</td>
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<td>Genetics – majority of individuals have \textit{BCR-ABL1} translocation</td>
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<tr>
<td>Myeloid and lymphoid neoplasms with \textit{PDGFRA} rearrangement</td>
<td>Most frequently presents as chronic eosinophilic leukemia (CEL), but may present as acute myeloid leukemia (AML), T-cell lymphoblastic lymphoma (T-LBL), or both</td>
<td>Morphology – peripheral blood and BM eosinophilia – markedly elevated</td>
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<td>o Acute transformation can follow CEL presentation</td>
<td>Typically &lt;20% blasts in peripheral blood and BM</td>
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<td>o Organ infiltration by eosinophils</td>
<td>Increased BM mast cells common</td>
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<td></td>
<td>o Heart</td>
<td>Genetics – absence of \textit{BCR-ABL1} fusion gene</td>
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<td>o Lungs</td>
<td>Most commonly associated with \textit{FIP1L1-PDGFR}A fusion</td>
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<td></td>
<td>o CNS</td>
<td>\textit{FISH} or PCR is usually necessary to document this genetic alteration; cytogenetic studies are normal</td>
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<td></td>
<td>o GI tract</td>
<td>o Other fusion genes have rarely been identified</td>
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<td>Splenomegaly in majority of individuals</td>
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<td>Pronounced male predominance</td>
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<tr>
<td>Myeloid and lymphoid neoplasms with \textit{PDGFRB} rearrangement</td>
<td>Presents with features of CML (usually with eosinophilia)</td>
<td>Morphology – peripheral leukocytosis</td>
</tr>
<tr>
<td></td>
<td>Splenomegaly in majority of individuals</td>
<td>o Peripheral blood and BM eosinophilia – markedly elevated</td>
</tr>
<tr>
<td></td>
<td>Male predominance, but much less marked than \textit{PDGFR}A-associated neoplasms</td>
<td>Typically &lt;20% blasts in peripheral blood and BM</td>
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<td>Increased BM mast cells common</td>
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<td>Genetics – most common translocation – t(5;12)(q31-33;p13) resulting in formation of \textit{ETV6-PDGFRB}</td>
</tr>
<tr>
<td>Myeloid and lymphoid neoplasms with \textit{FGFR1} abnormalities</td>
<td>Often presents with peripheral eosinophilia in the context of lymphadenopathy and lymphoblastic leukemia/lymphoma</td>
<td>Morphology – peripheral eosinophilia – markedly elevated</td>
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<td>Slight male predominance</td>
<td>o AML, ALL, CEL (usually associated with peripheral blood or BM eosinophilia)</td>
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<td>Genetics – presence of t(8;13)(p11;q12) or a variant translocation at the 8p11 breakpoint leading to \textit{FGFR1} rearrangement</td>
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<td>o Secondary cytogenetic abnormalities – trisomy 21 most often observed</td>
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Table 2. Overview of Genes and Mutations for Eosinophilic Disorders Tested for by this Panel

<table>
<thead>
<tr>
<th>Gene</th>
<th>Structure/Function</th>
<th>Mutations</th>
<th>WHO Disease Association</th>
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</table>
| \textit{BCR-ABL1} | • Maps to 9q24  
  • Chimeric constitutively active tyrosine kinase | • Majority of cases are translocation t(9;22) (q34;q11.2)  
  o Results in Philadelphia chromosome  
  • Remaining cases have variant translocations that involve a 3\textsuperscript{rd} or 4\textsuperscript{th} chromosome or cryptic translocation | CML, \textit{BCR-ABL1} positive |
| \textit{PDGFR}A | • Maps to 4q12  
  • Cell surface tyrosine kinase receptor for members of the platelet-derived growth factor family  
  • Results in a constitutively active tyrosine kinase oncprotein | \textit{FIP1L1-PDGFR}A rearrangement is a karyotypically occult 800-kb interstitial deletion (ie, \textit{CHIC2} deletion) | Myeloid and lymphoid neoplasms with \textit{PDGFR}A rearrangement |
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</table>
| **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); 2NF198-FGFR1 mutation | Myeloid and lymphoid neoplasms with FGFR1 abnormalities |