Lymphoma (Aggressive) Panel by FISH

Indications for Ordering

- Aid in diagnosis of aggressive large B-cell lymphoma with intermediate features between Burkitt lymphoma and diffuse large B-cell lymphoma (DLBCL)
- Confirmation of suspected double hit lymphoma

Test Description

Fluorescence in situ hybridization (FISH)
- FISH probes include
  - MYC
  - IGH
  - BCL2
  - BCL6

Tests to Consider

Typical Testing Strategy

- Lymph node biopsy with morphologic and immunohistochemical evaluation
- Leukemia/lymphoma phenotyping by flow cytometry
- In aggressive B-cell lymphomas with a high proliferation index and/or with unusual morphologic, phenotypic, or clinical features
  - Tests for MYC, BCL2, and BCL6

Panel Tests

Aggressive B-Cell Lymphoma Reflex Panel by FISH, Tissue 3001495
- Formalin-fixed, paraffin-embedded (FFPE) tissue specimens
- If MYC (8q24) Gene Rearrangement by FISH is positive, then IGH-BCL2 Fusion, t(14;18) by FISH will be added
- If IGH-BCL2 Fusion, t(14;18) by FISH is negative, then BCL6 (3q27) Gene Rearrangement by FISH will be added

Lymphoma (Aggressive) Panel by FISH 2002650
- Bone marrow (BM) or whole blood specimens; other specimens may be acceptable
  - FFPE and frozen specimens unacceptable

Individual Tests

- FFPE tissue specimens
  - MYC (8q24) Gene Rearrangement by FISH 3001300
    - Detects all MYC rearrangements, including t(8;14), t(2;8), and t(8;22)
  - IGH-MYC t(8;14) by FISH 3001299
  - IGH-BCL2 Fusion, t(14;18) by FISH 3001298
  - BCL6 (3q27) Gene Rearrangement by FISH 3001311

Related Tests

Leukemia/Lymphoma Phenotyping by Flow Cytometry 2008003
- Aids in diagnosis of hematopoietic neoplasms

Chromosome FISH, Interphase 2002298
- Specific probes must be requested
  - MYC break apart, BCL2, BCL6
- Fresh tissue specimens only

Chromosome Analysis, Bone Marrow 2002292
- Diagnosis, prognosis, and monitoring of lymphoma in BM

Chromosome Analysis, Solid Tumor 2002296
- May identify additional, useful cytogenetic abnormalities in tissues that are not targeted by FISH assays

Disease Overview

Prognostic Issues

- B-cell lymphomas with two recurrent chromosomal breakpoint aberrations are referred to as double hit lymphomas and are classified as high-grade B-cell lymphomas with MYC and BCL2 and/or BCL6 (WHO 2016)
  - Usually involve MYC oncogene in association with BCL2; less often with BCL6
- Lymphomas with three translocations (usually MYC/BCL2/BCL6) are referred to as triple hit lymphomas
  - Rare
- Important to identify these lymphomas in diagnostic evaluation of morphologically aggressive lymphomas
  - They are highly resistant to standard chemotherapy
    - Poor outcome independent of regimen intensity or inclusion of rituximab
  - Individuals have shortened survival compared with those having Burkitt lymphoma or international prognostic index (IPI)-matched DLBCL
### Genetics

#### Breakpoints Used to Identify Double Hit or Triple Hit Lymphomas

<table>
<thead>
<tr>
<th>Oncogene</th>
<th>Break apart MYC</th>
<th>BCL2</th>
<th>BCL6</th>
</tr>
</thead>
<tbody>
<tr>
<td>Locus</td>
<td>8q24</td>
<td>18q21</td>
<td>3q27</td>
</tr>
<tr>
<td>Biology</td>
<td>Accelerator of cell proliferation</td>
<td>Apoptosis inhibitor</td>
<td>Transcription modifier</td>
</tr>
<tr>
<td>Cytogenetics</td>
<td>Any MYC translocation</td>
<td>BCL2/IGH – t(14;18)(q32;q21)</td>
<td>BCL6 most commonly has a non-IG translocation partner – BCL6 (3q27)</td>
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<tr>
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<td></td>
<td>Uncommon partner – BCL6/IGH [t(3;14)(q27;q32)]</td>
</tr>
<tr>
<td>Specific lymphomas associated with translocation</td>
<td>Burkitt lymphoma, DLBCL</td>
<td>Follicular lymphoma, DLBCL</td>
<td>Follicular lymphoma, DLBCL</td>
</tr>
<tr>
<td></td>
<td>Aggressive B-cell lymphoma not otherwise specified (NOS)</td>
<td>High grade lymphomas (rare)</td>
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#### Test Interpretation

**Results**
- Abnormal – t(14;18)(q32;q21) (IGH/BCL2 translocation) or other rearrangements involving BCL6 and/or MYC detected
  - Presence of two or more translocations is associated with poor prognosis in mature B-cell lymphomas
  - Single rearrangements can provide diagnostic and/or prognostic information in the appropriate context
- Normal – t(14;18)(q32;q21) (IGH/BCL2 translocation) or other rearrangements involving BCL6 or MYC not detected

**Limitations**
- Interpretation of results requires correlation with morphology and immunophenotype
- MYC and/or BCL2 overexpression can be due to other mechanisms not detected by this test
- Chromosome alterations outside probe region are not detected