Overview

Useful For
Diagnostic workup of patients with high probability of BCR-ABL1-positive hematopoietic neoplasms, predominantly chronic myeloid/myelogenous leukemia and acute lymphoblastic leukemia

Highlights
Following a positive BCR/ABL1 diagnostic RT-PCR result, a reflex test will be performed to provide a quantitative measurement of BCR/ABL1 mRNA transcript (either p190 or p210 types). Current National Comprehensive Cancer Network (NCCN) guidelines for chronic myeloid leukemia (CML), for example, indicate that the quantitative p210 mRNA transcript level be obtained at diagnosis. The reflex test establishes the initial patient diagnostic baseline level to assess response to therapy in follow-up samples.

Reflex Tests

<table>
<thead>
<tr>
<th>Test ID</th>
<th>Reporting Name</th>
<th>Available Separately</th>
<th>Always Performed</th>
</tr>
</thead>
<tbody>
<tr>
<td>B190R</td>
<td>BCR/ABL1, p190, Quant, Reflex</td>
<td>No, (Bill Only); For Separate Testing: (Order BA190)</td>
<td>No</td>
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<tr>
<td>B210R</td>
<td>BCR/ABL1, p210, Quant, Reflex</td>
<td>No, (Bill Only); For Separate Testing: (Order BCRAB)</td>
<td>No</td>
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</tbody>
</table>

Testing Algorithm
When a positive common p210 or p190 BCR/ABL1 result is identified by the qualitative assay, a reflex test will then be performed at an additional charge to determine the quantitative transcript level of BCR/ABL1 mRNA. A positive common p210 or p190 result will specifically trigger either quantitative p210 (B210R) or p190 (B190R) testing to provide a normalized percentage of transcript level. For the p210 target, the value is additionally defined using the International Scale (IS) convention. The results are released in an integrated report and provide a baseline quantitative transcript to monitor treatment response. If the initial qualitative testing is negative, or an alternate rare form of BCR/ABL1 is detected, then no reflex testing will be pursued and the initial results will be reported.

The following documents are available in Special Instructions:

- Myeloproliferative Neoplasm: A Diagnostic Approach to Bone Marrow Evaluation
- Myeloproliferative Neoplasm: A Diagnostic Approach to Peripheral Blood Evaluation

Special Instructions
- Myeloproliferative Neoplasm: A Diagnostic Approach to Peripheral Blood Evaluation
- Myeloproliferative Neoplasm: A Diagnostic Approach to Bone Marrow Evaluation
- Hematopathology Patient Information
- BCR/ABL1 Ordering Guide for Blood and Bone Marrow

Method Name
Reverse Transcription-Polymerase Chain Reaction (RT-PCR) Multiplex PCR
NY State Available
Yes

Specimen

Specimen Type
Varies

Advisory Information
For information on which test to order for various scenarios, see BCR/ABL1 Ordering Guide for Blood and Bone Marrow in Special Instructions.

Shipping Instructions
Specimen must arrive within 72 hours of collection. Collect and package specimen as close to shipping time as possible.

Necessary Information
The following information is required:

1. Pertinent clinical history including if the patient has a diagnosis of chronic myeloid/myelogenous leukemia or other BCR/ABL1 positive neoplasm

2. Date of collection

3. Specimen source (blood or bone marrow)

Specimen Required
Submit only 1 of the following specimens:

Specimen Type: Whole blood

Container/Tube:

Preferred: Lavender top (EDTA)

Acceptable: Yellow top (ACD)

Specimen Volume: 10 mL

Collection Instructions:

1. Invert several times to mix blood.

2. Send specimen in original tube.

3. Label specimen as blood.

Specimen Type: Bone marrow

Container/Tube:
Test Definition: BCRFX
BCR/ABL1 Reflex, Qual/Quant

Preferred: Lavender top (EDTA)
Acceptable: Yellow top (ACD)

Specimen Volume: 3 mL

Collection Instructions:
1. Invert several times to mix bone marrow.
2. Send specimen in original tube.
3. Label specimen as bone marrow.

Forms
1. Hematopathology Patient Information (T676) in Special Instructions
2. If not ordering electronically, complete, print, and send a Hematopathology/Cytogenetics Test Request (T726) with the specimen.

Specimen Minimum Volume
Blood: 4 mL
Bone marrow: 1 mL

Reject Due To
- Gross hemolysis: Reject
- Other: Moderately to severely clotted

Specimen Stability Information

<table>
<thead>
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<th>Specimen Type</th>
<th>Temperature</th>
<th>Time</th>
<th>Special Container</th>
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<tbody>
<tr>
<td>Varies</td>
<td>Refrigerated (preferred)</td>
<td>72 hours</td>
<td>PURPLE OR PINK TOP/EDTA</td>
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<tr>
<td></td>
<td>Ambient</td>
<td>72 hours</td>
<td>PURPLE OR PINK TOP/EDTA</td>
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Clinical and Interpretive

Clinical Information
The t(9;22)/BCR-ABL1 abnormality is associated with chronic myelogenous leukemia (CML) and "Philadelphia positive" acute lymphoblastic leukemia of B-cell lineage (Ph+ ALL). Very rarely, this abnormality has also been identified in cases of acute myeloid leukemia and T-lymphoblastic leukemia/lymphoma. The fusion gene on the derivative chromosome 22q11 produces a chimeric BCR-ABL1 mRNA transcript and corresponding translated oncoprotein. Despite substantial breakpoint heterogeneity at the DNA level, a consistent set of BCR-ABL1 mRNA transcripts are produced that can be readily and sensitively detected by reverse transcription PCR (RT-PCR) technique. In CML, breakpoints in BCR result in either exons 13 or 14 (e13, e14) joined to exon 2 of ABL1 (a2). The corresponding e13-a2 or e14-a2 BCR-ABL1 mRNAs produce a 210 kD protein (p210). Rare cases of CML are
Test Definition: BCRFX
BCR/ABL1 Reflex, Qual/Quant

characterized by an e19-a2 type mRNA with a corresponding p230 protein. In Ph+ ALL, the majority of cases harbor an e1-a2 BCR-ABL1 mRNA transcript, producing a p190 protein. However, chimeric mRNA type is not invariably associated with disease type, as noted by the presence of p210-positive Ph ALL and very rare cases of p190-positive CML. Therefore, positive results from a screening (diagnostic) assay for BCR-ABL1 mRNA need to be correlated with clinical and pathologic findings.

In addition to the main transcript variants described above, rare occurrences of both CML and Ph+ ALL can have alternative break-fusion events resulting in unusual BCR-ABL1 transcript types. Examples include e6-a2 and BCR exon fusions to ABL1 exon a3 (eg, e13-a3, e14-a3, or e1-a3). In addition to detecting common BCR-ABL1 mRNA transcripts, this assay also can identify these rarer BCR-ABL1 transcript variants and is therefore a comprehensive screen for both usual and uncommon BCR-ABL1 gene fusions in hematopoietic malignancies. Given the nature of genetic events in tumors however, this assay will not identify extremely rare and unexpected BCR-ABL1 events involving other exons (eg, case report level) and is therefore not absolutely specific, but is predicted to detect greater than 99.5% of BCR-ABL1 events. Therefore, it is recommended that for diagnosis, RT-PCR plus a second method (eg, BCR-ABL1 FISH or cytogenetics) should be used. However, this RT-PCR assay is invaluable at diagnosis for identifying the precise BCR-ABL1 mRNA type (eg, for future quantitative assay disease monitoring), which complementary methods cannot.

This assay is intended as a qualitative method, providing information on the presence (and specific mRNA type) or absence of the BCR-ABL1 mRNA. Results from this test can be used to determine the correct subsequent assay for monitoring of transcript levels following therapy (eg, BCRAB / BCR/ABL, p210, mRNA Detection, Reverse Transcription-PCR (RT-PCR), Quantitative, Monitoring Chronic Myeloid Leukemia (CML), Varies; BA190 / BCR/ABL, p190, mRNA Detection, Reverse Transcription-PCR (RT-PCR), Quantitative, Monitoring Assay, Varies). Because the assay is analytically sensitive, it compensates for situations such as partially degraded RNA quality, or low cell number but it is not intended for quantitative or monitoring purposes.

Reference Values
An interpretive report will be provided.

Interpretation
An interpretive report will be provided.

When positive, the test identifies which specific mRNA fusion variant is present to guide selection of an appropriate monitoring assay. If common p210 or p190 fusion variant detected, quantitative reflex will be performed.

-Common fusion variants detected: e13-a2 or e14-a2 (p210), e1-a2 (p190), and e6-a2 (p205*)

-Rare fusion variants detected: e13-a3 (p210), e14-a3 (p210), e1-a3 (p190), e19-a2 (p230)

-Potential rare fusions detected: e12-a3, e19-a3

*This is formerly observed as the e6-a2 (p185) fusion form

Cautions
No significant cautionary statements

Clinical Reference
Performance

Method Description
Total RNA is extracted from the patient's blood or bone marrow at the time of diagnosis and mRNA is reverse transcribed into cDNA. The cDNA is then subjected to PCR using 4 separate multiplex reactions. A qualitative result, which will include the relative ratio of target translocation mRNA to control GUSB gene mRNA, will be given by LightCycler 96 software. As this method employs a quantitative PCR platform, the results can be used to evaluate the relative expression levels of the translocation mRNA relative to control mRNA, thus providing an improved measure of RNA quality in the assay. Reporting of results will be qualitative; either BCR-ABL1 mRNA positive/detected (with transcript type) or negative/not detected.(Instrument manual: LightCycler 96 Real-Time PCR System. Roche Applied Science Indianapolis, IN. 2013)

PDF Report
No

Day(s) and Time(s) Test Performed
Monday through Friday

Analytic Time
7 days

Maximum Laboratory Time
10 days

Specimen Retention Time
RNA 3 months

Performing Laboratory Location
Rochester

Fees and Codes

Fees
- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their Regional Manager. For assistance, contact Customer Service.

Test Classification
This test has been cleared, approved or is exempt by the U.S. Food and Drug Administration and is used per manufacturer's instructions. Performance characteristics were verified by Mayo Clinic in a manner consistent with CLIA requirements.

CPT Code Information
81208
81206
81207
## LOINC® Information

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<td>BCRFX</td>
<td>BCR/ABL1 Reflex, Qual/Quant</td>
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<table>
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<tr>
<td>48389</td>
<td>BCR/ABL1 Reflex Result</td>
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<td>Interpretation</td>
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