Overview

Useful For
Monitoring response to therapy in patients with chronic myeloid leukemia who are known to have the e13/a2 or e14/a2 BCR/ABL1 fusion transcript forms.

Testing Algorithm
See BCR/ABL1 Ordering Guide for Blood and Bone Marrow in Special Instructions.

Special Instructions
- Hematopathology Patient Information
- BCR/ABL1 Ordering Guide for Blood and Bone Marrow

Method Name
Quantitative Reverse Transcription-Polymerase Chain Reaction (RT-PCR)

NY State Available
Yes

Specimen

Specimen Type
Varies

Shipping Instructions
Specimen must arrive within 72 hours of collection. Collect and package specimen as close to shipping time as possible. Specimens greater than 3 days old at the time of receipt will be considered unacceptable.

Necessary Information
The following information is required:

1. Pertinent clinical history including if the patient has a diagnosis of chronic myeloid 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:**
- **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) (T726) with the specimen.

**Specimen Minimum Volume**
- Blood: 4 mL
- Bone Marrow: 1 mL

**Reject Due To**

<table>
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<tr>
<th>Gross hemolysis</th>
<th>Reject</th>
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**Specimen Stability Information**

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<tr>
<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

Chronic myeloid leukemia (CML) is a hematopoietic stem cell neoplasm included in the broader diagnostic category of myeloproliferative neoplasms. CML is consistently associated with fusion of the breakpoint cluster region gene (BCR) at chromosome 22q11 to the Abelson gene (ABL1) at chromosome 9q23. This fusion is designated BCR/ABL1 and may be seen on routine karyotype as the Philadelphia chromosome.

Although various breakpoints within the BCR and ABL1 genes have been described, more than 95% of CMLs contain a consistent mRNA transcript in which either the BCR exon 13 (e13) or BCR exon 14 (e14) is fused to the ABL1 exon 2 (a2), yielding fusion forms e13/a2 and e14/a2, respectively. The e13/a2 and e14/a2 fusion forms produce a 210-kDa protein (p210). The p210 fusion protein is an abnormal tyrosine kinase known to be critical for the clinical and pathologic features of CML, and agents that block the tyrosine kinase activity (ie, tyrosine kinase inhibitors or TKI, such as imatinib mesylate) have been used successfully for treatment. Monitoring the level of BCR/ABL1 mRNA in CML patients during treatment is helpful for both prognosis and management of therapy. (1-3) Rising BCR/ABL1 mRNA levels following attainment of critical therapeutic milestones (see Clinical References) can be indicative of acquired resistance mutations involving the ABL1 portion of the BCR/ABL1 fusion gene.

Quantitative reverse-transcription PCR (qRT-PCR) is the most sensitive method for monitoring BCR-ABL1 levels during treatment. This test detects the BCR/ABL1 mRNA fusion forms found in CML (e13/a2 and e14/a2).

Reference Values

The presence or absence of BCR/ABL1 mRNA fusion form e13/e14-a2 producing the p210 fusion protein is identified. If positive, the quantitative level is reported as the normalized ratio of BCR/ABL1 (p210) to endogenous ABL1 mRNA with conversion to a percentage referenced to the international scale (IS), on which 0.1% BCR/ABL1:ABL1 (also represented on a log scale as Molecular Response 3, or MR3) is designated as a major molecular response (MMR) threshold.

Interpretation

An interpretive report will be provided. When BCR/ABL1 mRNA is present, quantitative results are reported on the international scale (IS), established from data originally reported in the IRIS (International Randomized Study of Interferon versus STI571) trial involving newly diagnosed chronic myeloid leukemia patients. Using the IS, a result of less than 0.1% BCR/ABL1 (p210):ABL1 is equivalent to a major molecular remission. This value is also designated on a log scale as Molecular Response 3, or MR3. For further discussion of the international scale, see Clinical References.

Cautions

This test detects only the e13/a2 and e14/a2 fusion forms, which code for the p210 protein. Other fusion forms are not detected, including those containing the BCR e1 exon, which codes for the p190 protein commonly found in acute lymphoblastic leukemia (ALL). If the patient is known to carry an e1/a2 (p190) fusion form, the test BA190 / BCR/ABL, p190, mRNA Detection, Reverse Transcription-PCR (RT-PCR), Quantitative, Monitoring Assay should be used for monitoring.

This test should not be used to screen for BCR/ABL1 fusions at the time of diagnosis; if a diagnostic screen for BCR-ABL1 transcripts is desired, the test BADX / BCR/ABL1, Qualitative, Diagnostic Assay, which is designed to detect all reported common and rare BCR-ABL1 mRNA fusion variants, should be ordered for this purpose.

The precision of this assay at low BCR/ABL1 levels is more variable, such that inter-run variation can be as high as + or - 0.5 log. Only level changes above 0.5 log should be considered clinically significant. For example, if a result is given as 0.1% BCR/ABL1:ABL1, then any result between 0.05% and 0.5% should be considered essentially
equivalent. If the results are being used to make major therapeutic decisions, significant changes during monitoring should be verified with a subsequent specimen.

In general, the results of this assay cannot be directly compared with results generated from other PCR assays, including identical assays performed in other laboratories. Monitoring should be performed using the same method and laboratory for each subsequent specimen.

The results of this assay cannot be directly compared with BCR/ABL1 results obtained using FISH technology. FISH measures DNA alleles and RT-PCR-based assays measure mRNA transcripts. Because a single fusion DNA allele can produce many mRNA transcripts, the values are not directly comparable and FISH results are not applicable to the IS or to disease monitoring.

Blood is the specimen of choice for monitoring CML patients. The majority of CML patients show similar BCR/ABL1 mRNA levels in blood and bone marrow drawn at the same time, although occasional patients may exhibit a difference in concurrent blood and marrow levels for technical or biological reasons, requiring follow-up testing to resolve.

Clinical Reference

Performance
Method Description
The assay is performed using an automated platform, GeneXpert (Cepheid). Four mL of whole blood is processed, added to an individual sample cartridge and loaded onto the GeneXpert machine. All subsequent reactions are performed within the cartridge and the results are processed and calculated by the instrument. Within the cartridge, RNA is extracted and converted to complementary DNA (cDNA).

Quantitative, reverse transcription PCR is performed with a nested PCR reaction containing primers designed to amplify cDNA from the e13/a2 and e14/a2 BCR/ABL1 fusion products. A fragment of ABL1 cDNA is also amplified as a control for RNA degradation and for normalization of BCR/ABL1 results. The ratio of BCR/ABL1 (p210) to ABL1 is calculated from the difference in the crossing thresholds of BCR/ABL1 (p210) and ABL1 products in relation to a lot-specific standard curve, referenced to the international scale (IS). Lot-to-lot variation in the cartridges is corrected using a calibration calculation to reference standard curve data to the IS provided by the manufacturer. (Unpublished Mayo method)

PDF Report
Supplemental
Day(s) and Time(s) Test Performed
Monday through Friday

**Analytic Time**
3 days

**Maximum Laboratory Time**
6 days

**Specimen Retention Time**
2 weeks

**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 was developed and its performance characteristics determined by Mayo Clinic in a manner consistent with CLIA requirements. This test has not been cleared or approved by the U.S. Food and Drug Administration.

**CPT Code Information**
81206

**LOINC® Information**

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<td>BCR/ABL1, p210, Quant, Monitor</td>
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