BCR-ABL1 Quantitative Testing

Indications for Ordering

Chronic myelogenous leukemia (CML)
- Diagnosis and ongoing therapeutic monitoring
  - Useful for tyrosine kinase inhibitor (TKI) treatment management strategy

Acute lymphoblastic leukemia (ALL)
- Risk stratification and treatment planning
- Minimal residual disease (MRD) assessment of Philadelphia chromosome positive (Ph+) ALL

Test Description

Reverse transcription polymerase chain reaction (RT-PCR)
- Performed on peripheral blood or bone marrow (BM)
- RNA is isolated and reverse transcribed to cDNA
  - *BCR-ABL1* fusions are quantitated by real-time PCR amplification
  - Primers detect fusions with major (p210) or minor (p190) *BCR-ABL1* breakpoints
  - Test reference gene is *ABL1*
  - Copy numbers of *BCR-ABL1* fusion transcripts are expressed as ratio of *BCR-ABL1*/*ABL1*

Tests to Consider

Typical testing strategy

CML

At diagnosis:
- BM cytogenetic studies and quantitative measurement of *BCR-ABL1* transcript levels are recommended before treatment initiation

Monitoring response to TKI therapy in CML:
- Quantitative RT-PCR
  - Every 3 months when treatment response is evident
  - Every 3 months for 3 years, and every 3-6 months thereafter after complete cytogenetic response has been achieved
  - More frequent monitoring may be required in individuals with rising *BCR-ABL1* transcripts

BCR-ABL1 kinase domain mutation analysis (using next generation sequencing)
- If inadequate initial response to TKI therapy
- Rising level of *BCR-ABL1* transcripts
- Disease progression to accelerated or blast phase

ALL

At diagnosis:
- Presence of recurrent genetic abnormalities should be evaluated using karyotyping and/or FISH assays

MRD assessment:
- Flow cytometry and quantitative PCR testing (BM preferred)
- Upon completion of initial induction and additional time points, as required

Primary tests

BCR-ABL1, Qualitative with Reflex to *BCR-ABL1* Quantitative 2005010
- Recommended when submitting initial diagnostic specimen for CML or Ph+ ALL (no previous *BCR-ABL1* testing)
- If qualitative test is positive, the appropriate corresponding quantitative test is performed

BCR-ABL1, Major (p210), Quantitative 2005017
- Aids in diagnosis and monitoring of individuals with CML or Ph+ ALL who have e13a2 or e14a2 transcripts (p210)

BCR-ABL1, Minor (p190), Quantitative 2005016
- Aids in diagnosis and monitoring of individuals with Ph+ ALL who have e1a2 transcripts (p190)

Related tests

BCR-ABL1 Mutation Analysis for Tyrosine Kinase Inhibitor Resistance by Next Generation Sequencing 2008420
- Order only for patients with an established diagnosis of a *BCR-ABL1* positive leukemia
- Use to determine if a mutation is present that would interfere with response to TKI therapy in Ph+ ALL or CML
- Detects all common mutations, including T315I
- Higher sensitivity than traditional Sanger sequencing techniques
- Offers coverage of SH2, SH3, and kinase domains

Myeloid Malignancies Mutation Panel by Next Generation Sequencing 2011117
- Assesses for gene mutations, including substitutions and insertions and deletions that may have diagnostic, prognostic, and/or therapeutic significance

Acute Lymphocytic Leukemia (ALL) Panel by FISH, Pediatric 2002719
- Recommended FISH panel for children with newly diagnosed ALL

Acute Lymphocytic Leukemia (ALL) Panel by FISH, Adult 2002647

- Recommended FISH panel for adults with newly diagnosed ALL

Chromosome FISH, Interphase 2002298

- Specific FISH probe for t(9;22); *BCR-ABL1* must be requested

Disease Overview

CML

Treatment issues – goal of TKI therapy is to achieve a complete cytogenetic response within 12 months of initiation of therapy with goal of eventual major molecular response

- A subset of individuals will eventually achieve a complete molecular response (undetectable *BCR-ABL1* transcripts using a test with 4.5 log sensitivity)

Prognostic issues – a 3-log decrease in the level of *BCR-ABL1* fusion transcripts (major molecular response) within 18 months of beginning TKI therapy is an indicator of favorable outcome

- Monitoring for recurrence using quantitative measures is crucial for detecting early relapse

ALL

Incidence – 1.6/100,000

- 80% of childhood acute leukemias
  - 2-4% are Ph+
- 20% of adult leukemias
  - 25% are Ph+

Genetics

Gene – *BCR-ABL1*

Mutations

- Four regions
  - Adenosine triphosphate binding-loop (P-loop) mutations
  - Drug-binding sites
  - Catalytic domain
  - Activation loop
- >130 mutations

Test Interpretation

***BCR-ABL1*, Major (p210), Quantitative**

Analytical sensitivity – 1:125,000 normal cells

Results

- Positive – *BCR-ABL1* fusion transcripts (p210) detected
  - *BCR-ABL1/ABL1* quantitative ratio is provided (normalized copy number)
  - Results also reported in terms of *BCR-ABL1* international scale (IS)
- Weakly positive – *BCR-ABL1* fusion transcripts detected below the limit of quantitation
  - *BCR-ABL1* to *ABL1* ratio cannot be calculated
  - IS result <0.0069%
- Not detected – no *BCR-ABL1* fusion transcripts detected
  - Does not exclude *BCR-ABL1* fusion transcripts (p210) below the test limit of detection
  - Does not exclude *BCR-ABL1* fusion transcripts not detected by this test (p190 or p230)

Limitations

Does not detect p190 or p230 form

***BCR-ABL1*, Minor (p190), Quantitative**

Analytical sensitivity – 1:125,000 normal cells

Results

- Positive – *BCR-ABL1* fusion transcripts (p190) detected
  - *BCR-ABL1/ABL1* quantitative ratio is provided (normalized copy number)
- Weakly positive – *BCR-ABL1* fusion transcripts detected, but below the limit of quantitation
  - *BCR-ABL1* to *ABL1* ratio cannot be calculated
- Not detected – no *BCR-ABL1* fusion transcripts detected
  - Does not exclude *BCR-ABL1* fusion transcripts (p190) below the test limit of detection
  - Does not exclude *BCR-ABL1* fusion transcripts that are not detected by this test (p210 or p230)

Limitations

Does not detect p210 or p230