**BCR-ABL1** Qualitative and 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 identified by polymerase chain reaction (PCR) and capillary electrophoresis. Minor (p190) and major (p210) transcripts can be quantified by real time PCR
  - Primers detect fusions with major (p210), minor (p190), or micro (p230) BCR-ABL1 breakpoints
  - Test reference gene is ABL1
  - Copy numbers of BCR-ABL1 fusion transcripts (p190 or p210) are expressed as ratio of BCR-ABL1/ABL1

### Tests to Consider

**Typical Testing Strategy**

**CML**
- 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 (p190 or p210), 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**

- 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
  - Results 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