**Chronic Lymphocytic Leukemia Prognostic Markers**

### Indications for Ordering

Use to determine risk group in newly diagnosed chronic lymphocytic leukemia (CLL)
- **IGHV** mutation
- **CD38** expression

### Test Description

- **IGHV Mutation Analysis by Sequencing**
  - Polymerase chain reaction/sequencing using VH leader and JH primers
  - Identified clonal VH sequences are searched against a known database for homology
    - Percent of homology to germline sequences is reported
  - CLL clones comprising at least 50% of total B cells can be analyzed
- **Leukemia/Lymphoma Phenotyping by Flow Cytometry**
  - **CD38** expression

### Tests to Consider

**Primary tests**
- **IGHV Mutation Analysis by Sequencing 0040227**
- **Determine risk group in newly diagnosed CLL**
- **Leukemia/Lymphoma Phenotyping Evaluation by Flow Cytometry 3001780**
  - Aid in evaluation of hematopoietic neoplasms
  - Expression of **CD38** typically performed for CLL diagnosis and followup

**Related tests**
- **Cytogenomic SNP Microarray – Oncology 2006325**
  - Preferred test at time of diagnosis for detecting prognostically important genomic abnormalities in leukemias/lymphomas and solid tumors involving
    - Loss/gain of DNA
    - Loss of heterozygosity (LOH)
  - Monitor disease progression and response to therapy
- **Chromosome FISH, CLL Panel 2002295**
  - Alternate test to detect prognostically important genomic abnormalities in CLL
    - Not as sensitive as SNP microarray
    - Includes **ATM** (11q22.3), chromosome 12 centromere (Trisomy 12), (D13S319) 13q14.3, p53 (17p13.1)

### Disease Overview

#### Treatment issues
- CLL is a heterogenous disease
  - Therapy based on risk group stratification
  - Clinical course is variable
    - ~1/2 are aggressive, others are indolent
- Cytogenetic, molecular, and flow cytometry testing play an important role in prognostication of CLL

#### Significance of markers
- **IGHV**
  - **IGHV** mutation status of CLL tends to remain constant over course of disease
  - del(17p) and/or del(11q) correlate with nonmutated **IGHV** genes
  - Presence of mutation associated with more indolent course
- **CD38** expression
  - Transmembrane glycoprotein modulates intracellular signaling
  - Cases that express **CD38** often have nonmutated **IGHV** genes
  - May reflect an increased proliferative status of CLL
  - Expression levels may vary over course of disease

### Test Interpretation

**Results**

**IGHV mutation**
- Nonmutated **VH** segment
  - >98% identical to the most closely matched germline **VH** gene sequence
  - Associated with unfavorable outcome
- Mutated **VH** segment
  - <98% identical to the most closely matched germline **VH** gene sequence
  - Associated with favorable outcome
  - Borderline mutation
    - 97-97.9% identical to the most closely matched germline **VH** gene sequence
    - May have an intermediate clinical outcome (Hamblin TJ, Br J Hematology, 2008)
- Expression of **VH3-21** segment typically has an unfavorable outcome regardless of mutation status

**CD38 expression**
- Associated with unfavorable outcome
Limitations

*IGHV* mutation

- Samples that do not yield amplification product may
  - Contain too few CLL cells (<50% B cells)
  - Express *VH* genes with high numbers of mutations that may compromise clonal B-cell amplification
- Not intended to detect minimal residual disease