IGHV and TP53 Sequencing: Clinical Utility in Chronic Lymphocytic Leukemia (CLL)

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Disclosures
• None

Today’s Outline
• Understand the use of prognostic markers in CLL patients
• Highlight the importance of molecular analysis for IGHV and TP53 sequencing in CLL patients
• Understand how IGHV analysis provides prognostic information in CLL and helps inform clinicians on treatment decisions
• Recognize that TP53 mutations by sequencing studies are associated with poor outcomes and more likely to be resistant to standard chemotherapy/chemoimmunotherapy regimens
**Brief Overview of CLL**

- CLL is a neoplasm of small mature B-cells
  - Most common leukemia diagnosed in adults
  - Median age at diagnosis is 70 y.o.
    - ~11% are < 55 y.o. and ~2% are < 45 y.o.
  - 5 year survival rate: 84.2%
- Clinical course
  - Varies from indolent and slowly progressive to patients having a more progressive course that requires aggressive treatment
- Prognostic markers
  - Clinical staging systems, serum markers, immunophenotypic markers, and genetic anomalies have been used in predicting outcomes
- Therapies for progressive disease in CLL are expanding with improvements in overall survival

**CLL Prognostic Markers: Flow-Based**

- CD38 and ZAP70: When positive, both are associated with poor outcomes. But both have not stood up as independent markers.
- Discordance with IGHV mutation status occurs in patients with either stable disease as well as with progressive disease.

**Conclusion:**

- CD38 and ZAP70: **Cannot** be used as a replacement for IGHV mutation status in determining prognosis and in subsequent treatment decisions
- CD49d: When positive, it is associated with an unfavorable prognosis and shown to be the strongest flow cytometry-based predictor of overall survival and treatment-free survival.
CLL Prognostic Markers: FISH

- FISH is not diagnostic of CLL as the following anomalies can be found in other B-cell lymphoproliferative disorders
  - 13q-: Associated with good prognosis
  - Trisomy 12: Associated with intermediate prognosis
  - 11q-: Associated with poor prognosis
  - 17p- (p53): Associated with very poor prognosis;
    - Incidence at diagnosis 5-10%;
    - Incidence following Fludarabine treatment 30-40%

CLL Prognostic Markers: IGHV

- Unmutated IGHV
  - Defined as <2% difference from the germline VH gene sequence identity
  - Poorer prognosis: Mean overall survival of 95 months
  - 40% of CLL cases are unmutated IGHV
- Mutated IGHV HV-M
  - Defined as ≥2% difference from the germline VH gene sequence identity
  - Good prognosis: Mean overall survival of 293 months
- IGHV status is now being used to help with treatment decisions and identifies patients who may benefit from BTK inhibitors, such as Ibrutinib
CLL Prognostic Markers: TP53

- TP53 point mutations are related, but separate from FISH for 17p-
  - Incidence at diagnosis: 5-10%; incidence s/p Fludarabine treatment: 30-40%
- del(17p) and/or TP53 mutations are associated with poor outcomes and may be relatively resistant to standard chemotherapy / chemoimmunotherapy regimens
- Patients who have 17p- and/or TP53 mutation fare better when treated with small molecule inhibitors of BTK, phosphatidylinositol 3-kinase, or BCL2
- Thus, assessment of both 17p- by FISH and TP53 by sequencing has prognostic value and may help guide therapeutic decisions in routine practice

Typical Application of “Risk” in a CLL Patient

**Risk Assessment:**
- FISH (+12, 11q-, 13q-, 17p-)
- IGHV mutation status

**Lower Risk:**
- Mutated IGHV, 13q-, trisomy 12, and/or diploid

**Treatment:**
- Chemoimmunotherapy

**Intolerance or Disease Progression**

**Higher Risk:**
- Unmutated IGHV, 17p- or mutated TP53, and/or 11q-

**Treatment:**
- BTK Inhibitor

**Intolerance or Disease Progression**

**Treatment:**
- PI3Kδ inhibitor

**Intolerance or Disease Progression**

**Treatment:**
- BCL2 inhibitor
International Prognostic Index (CLL-IPI): Study

<table>
<thead>
<tr>
<th>Study Design</th>
<th>Data</th>
</tr>
</thead>
<tbody>
<tr>
<td>Analysis</td>
<td>3472 treatment naïve patients from 5 countries</td>
</tr>
<tr>
<td>External Validation</td>
<td>838 patients from Mayo Clinic</td>
</tr>
<tr>
<td>Simple Composite Score: 5 Parameters</td>
<td>17p- FISH, IGHV, Serum B2M, Clinical Stage, Age</td>
</tr>
<tr>
<td>4 Risk Groups</td>
<td>Low, intermediate, high, very high</td>
</tr>
<tr>
<td>Clinical Application</td>
<td>Improved clinical staging; useful for testing novel therapeutics in high or very high risk groups</td>
</tr>
</tbody>
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IGHV and TP53 Recommended by IWCLL

<table>
<thead>
<tr>
<th>Diagnostic Lab Tests</th>
<th>Recommendation</th>
</tr>
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<tbody>
<tr>
<td>At Diagnosis</td>
<td></td>
</tr>
<tr>
<td>CBC and Diff</td>
<td>Always</td>
</tr>
<tr>
<td>Blood flow study</td>
<td>Always</td>
</tr>
<tr>
<td>Before Treatment</td>
<td></td>
</tr>
<tr>
<td>Bone marrow study</td>
<td>When clinically indicated</td>
</tr>
<tr>
<td>Serum chemistries, immunoglobulins, DAT</td>
<td>Always</td>
</tr>
<tr>
<td>CLL FISH for 13q-, 11q-, 17p-, +12</td>
<td>Always</td>
</tr>
<tr>
<td>Chromosome karyotype</td>
<td>Not generally indicated</td>
</tr>
<tr>
<td><strong>TP53 mutation</strong></td>
<td><strong>Always</strong></td>
</tr>
<tr>
<td><strong>IGHV mutation status</strong></td>
<td><strong>Always</strong></td>
</tr>
<tr>
<td>Serum B2-microglobulin</td>
<td>Desirable</td>
</tr>
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</table>
**CLL-IPI Summary**

<table>
<thead>
<tr>
<th>Variable</th>
<th>Adverse Factor</th>
<th>Grading</th>
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</thead>
<tbody>
<tr>
<td>TP53 (17p-)</td>
<td>Deleted and/or mutated</td>
<td>4</td>
</tr>
<tr>
<td>IGHV</td>
<td>Unmutated</td>
<td>2</td>
</tr>
<tr>
<td>Serum B2M (mg/L)</td>
<td>&gt; 3.5</td>
<td>2</td>
</tr>
<tr>
<td>Clinical stage</td>
<td>Binet B/C or Rai I-IV</td>
<td>1</td>
</tr>
<tr>
<td>Age</td>
<td>&gt; 65</td>
<td>1</td>
</tr>
<tr>
<td>Prognostic score</td>
<td></td>
<td>0 – 10</td>
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**CLL-IPI and Overall Survival**

Mayo cohort [n = 838]
### Simplistic Application of CLL-IPI

<table>
<thead>
<tr>
<th>Risk Group</th>
<th>General Treatment Direction?</th>
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<tbody>
<tr>
<td>Low</td>
<td>“Do not touch”</td>
</tr>
<tr>
<td>Intermediate</td>
<td>“Do not treat”</td>
</tr>
<tr>
<td></td>
<td>(Except symptomatic)</td>
</tr>
<tr>
<td>High</td>
<td>“Treat”</td>
</tr>
<tr>
<td></td>
<td>(Except asymptomatic)</td>
</tr>
<tr>
<td>Very High</td>
<td>“Consider treatment in experimental protocol or with non-cytotoxic drugs if possible”</td>
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### Summary

- CLL knowledge has rapidly grown with molecular technologies
- It is critical that the right tests get ordered in patients with CLL
- The CLL-IPI is a relatively simple yet elegant way to stratify patients into distinct prognostic groups that can drive particular therapies
- CLL FISH testing for prognosis is well-established and understood
- IGHV mutation analysis provides critical prognostic information in CLL and also can help inform clinicians on treatment decisions
- Mutations identified by TP53 sequencing are associated with poor outcomes and may be resistant to standard chemotherapy and chemoimmunotherapy regimens
Thank You!