Genomic Analysis of Mature B-cell Malignancies

Update and Lessons Learned

Omar Abdel-Wahab, MD
Memorial Sloan Kettering Cancer Center
Human Oncology and Pathogenesis Program
and Leukemia Service

Disclaimer:
This presentation is being provided for informational purposes only. The data and conclusions shown are either of academic authors (from work cited herein) or the presenter’s alone and do not represent the views of Incyte Corporation
Key Advances in Genomic Analysis of Chronic Lymphocytic Leukemia (CLL)

• >265 whole exomes/genomes of CLL patients have been published to date. Lessons learned:

  1. CLL is marked by *inter-tumoral heterogeneity*: genetic differences between patients.
  2. But also, striking *intra-tumoral heterogeneity*: the presence of multiple subclones within patients.
  3. Central importance of the B-Cell Receptor in CLL pathogenesis & therapy.

• Resistance to novel therapeutics in CLL.
Genomic Landscape of CLL: Inter-tumoral Heterogeneity

Vast majority of genes mutated in CLL are mutated in <10% of patients.

Somatic mutations are rarely shared amongst individual patients.

Genomic Landscape of CLL: Intra-tumoral heterogeneity

Majority of CLL patients have multiple subclones which are genetically distinct.

Understanding how these subclones are affected by therapy and/or influence response to therapy will be critical.
Genomic Landscape of CLL: Intra-tumoral heterogeneity

Wu, C. Blood 2012
Serial genomic analysis of CLL: Convergent evolution

04/16/2008
WBC 153.4

05/12/2010
WBC 272

12/02/2011
WBC 60

<table>
<thead>
<tr>
<th>Genes</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>FAT3 A2925T</td>
<td>46%</td>
</tr>
<tr>
<td>SF3B1 R625H</td>
<td>47%</td>
</tr>
<tr>
<td>NOTCH1</td>
<td>48%</td>
</tr>
<tr>
<td>TP53 R273P</td>
<td>4%</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Genes</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>FAT3 A2925T</td>
<td>46%</td>
</tr>
<tr>
<td>SF3B1 R625H</td>
<td>47%</td>
</tr>
<tr>
<td>NOTCH1</td>
<td>48%</td>
</tr>
<tr>
<td>TP53 R273P</td>
<td>65%</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Genes</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>FAT3 A2925T</td>
<td>46%</td>
</tr>
<tr>
<td>SF3B1 R625H</td>
<td>47%</td>
</tr>
<tr>
<td>NOTCH1</td>
<td>48%</td>
</tr>
<tr>
<td>TP53 R273P</td>
<td>65%</td>
</tr>
<tr>
<td>TP53 I232T</td>
<td>06%</td>
</tr>
<tr>
<td>TP53 R175H</td>
<td>27%</td>
</tr>
</tbody>
</table>
Serial genomic analysis of CLL: Convergent evolution

07/11/2011

WBC 78.5

ATM E641fs*18 | 15%
ATM Y2009X | 45%

Del11q
+E641fs*18

Del11q
+Y2009X

12/22/2011

WBC 7

ATM E641fs*18 | 5%

Del11q
+E641fs*18

01/24/2012

WBC 6.2

ATM Y2009X | 9%

DNMT3A R598X | 4%

RUNX1T1 R149H | 2%

GS1101 + Rituximab

GS1101 + Rituximab
Genomic Landscape of CLL: Questions

• True frequency of spectrum of somatic mutations in CLL not fully resolved.
  – Combined analysis of Broad & ICGC data
  – Publication of comprehensive analysis of well-annotated and more uniform cohorts

• Role of specific mutations in response/resistance to new targeted therapeutic in CLL.

• Are any of these novel therapeutic targets?
  – MAP Kinase pathway alterations: KRAS, BRAF, NRAS, MAP2K1, PTPN11
  – Spliceosomal gene mutations: SF3B1
  – Notch activating mutations: NOTCH1, FBXW7, SPEN, NOTCH4
Central Importance of the B-cell receptor (BCR) in CLL

- Response to inhibitors of proximal BCR signaling highlight importance of BCR in driving CLL.

BCR’s in CLL patients show:
- Skewed repertoire of immunoglobulin heavy variable (IGHV) genes
- Highly stereotyped and quasi identical Ag binding sequences amongst different patients
- Cell-autonomous receptor activity

Evidence for the recognition of individual, discrete antigens or classes of structurally similar epitopes, likely selecting the leukemic clones.
Central Importance of the B-cell receptor (BCR) in CLL

Normal B cell: BCR signaling induced by external antigen

CLL cells: Cell autonomous BCR signal

Dependent on the heavy-chain complementarity determining region (HCDR3) & an internal epitope of the BCR.

LETTER

Chronic lymphocytic leukaemia is driven by antigen–independent cell–autonomous signalling

Marcus Döhren-von Mierendorfs, Isabel Schellhase, Dietrich Schneider, Thomas Wessling, Martina P. Bach, Malte Buchner, Daniel Rohman, Elena Simova, Mario Folko, Fabian Köhler, Hedda Wiedemann, Katja Zelik, Hendrik Vosslamber & Harumi Umeda
Drug resistance to targeted therapeutics in CLL

**BTKC481S mutation**: abolishes Ibrutinib binding to BTK.

**PLCG2 mutations**:
- R665W
- S707Y
- L845F

Familial inflammatory disorder marked by PLCG2 R665W germline mutation

Woyach, JA, et al. NEJM 2014
Furman, RR, et al. NEJM 2014
Drug resistance to targeted therapeutics in CLL: Questions

• How frequently do BTK/PLCG2 mutations account for ibrutinib resistance?  
  - CLL: Richter’s transformation pattern vs progressive pattern without Richter’s  
  - MCL: primary ibrutinib resistance/transient response vs resistance in relapse

• What about mutations in kinases downstream of BTK/PLCG2?

• Could ATP-competitive BTK inhibitors be developed which don’t require C481 for activity?

• Could PLCG2 activating mutations bypass the need for BTK altogether?

• What about drug combinations of small molecule inhibitors in CLL?

• What drugs to use for ibrutinib resistant CLL?

• What about resistance to inhibitors of PI3Kδ or BCL2?
Genomic techniques for serial disease monitoring

- **Diagnosis**
- **Treatment**
- **Relapse**

Plasma cell-free, PBMC, circ tumor DNA analysis

- Fusion detection by qRT-PCR
- MRD detection by qRT-PCR or NGS for IGH genes
- More comprehensive mutational analysis for clonal evolution/resistance