Update on New WHO Classification of B-cell Lymphomas

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Objectives

- Updates of B-cell NHL in WHO 2017
- Understand the paradigm used for lymphoma classification
  - Preneoplastic conditions
  - In situ lesions
  - Genetics in diagnosis, prognosis, therapy
  - Evolution of knowledge base
Outline

- CLL/SLL and MBL
- \textit{in situ} follicular neoplasia
- FL which are BCL2 negative
- Pediatric-type FL
- MYD88 mutations in LPL and other B-NHL
- MZL/Splenic MZL
- Hairy cell leukemia (BRAF and MAP2K)
- Mantle cell lymphoma (indolent, cyclin D1 negative, blastoid)
  
  - Aggressive B-cell lymphoma
  - Large B-cell lymphoma with IRF4 rearrangement
  - High grade B-cell lymphomas
Cellular origin of B-NHLs

Ig gene V(D)J recombination

Somatic Mutation Antigen Selection Isotype switch

Pro B → Pre B → Naive B → Germinal Center

Precursor B-LL

MCL

MM

Plasma cell

LPL

MZBCL

Memory

NLPHEL

FCL BL DLBCL

Classical HL

CLL
Common Chromosomal Deletions in CLL and Prognosis

- Deletion 13q
  - 14-40% of CLLs (indolent)
- Deletion 11q
  - 10-32% of CLLs (intermediate)
- Deletion 17p
  - 3-27% of CLLs (most aggressive)

*(NEJM, 2000, 343, 1910-1916)*

![Graph showing survival rates with different chromosomal deletions and abnormalities](image-url)
The genetic landscape of chronic lymphocytic leukemia

Genes with significant mutation frequencies CLL

Association between recurrent gene mutations and genetic features in CLL

Figure 1. Probability of Survival from the Date of Diagnosis among the Patients in the Five Genetic Categories. The median survival times for the groups with 17p deletion, 11q deletion, 12q trisomy, normal karyotype, and 13q deletion as the sole abnormality were 32, 79, 114, 111, and 133 months, respectively. Twenty-five patients with various other chromosomal abnormalities are not included in the analysis.
Monoclonal B cell lymphocytosis/MBL

- Neoplastic peripheral blood lymphocyte count < 5 x 10⁹/L
- Similar immunophenotype as CLL
- No extramedullary tissue involvement

- MBL is detected in 3% of adults
- Incidence increases with age
  - 0.3% of adults age 18 to 40 years
  - > 5% of individuals over age 60 years
  - 13.5% - 18% of first-degree relatives of CLL patients
Monoclonal B cell lymphocytosis prognosis (WHO 2016)

- Precedes virtually all cases of CLL
- Distinguish low-count MBL ($<0.5 \times 10^9/L$) from high count MBL
  - A higher monoclonal B-cell count
  - Trisomy 12 or del17p

Proliferation centers in CLL/SLL

• Cyclin D1+ in 20-30% of CLL/SLL without t(11;14) or *CCND1* copy number alterations (AJCP 138:132,2012)
• MYC+ in proliferation centers without MYC rearrangements of extra copies (Br J Haematol 2015)
• PI3K P85+, EZH2+
• Proliferative fraction in proliferation centers may have prognostic value
Follicular Lymphoma

- Most common low-grade B-cell lymphoma in adults in the U.S. and Europe
- Indolent clinical course - disseminated
- Characterized by $t(14;18)(q32;q21)$
In situ follicular neoplasia (ISFN)

- Follicular lymphoma in situ
- Partial or total colonization of GC by clonal B-cells with BCL2 translocation in otherwise reactive LN
- Observed in 2% of randomly selective LNs
- Need to distinguish from partial involvement by FL which has greater chance of developing FL/DLBCL

Bcl-2 break apart probe: Positive for t(14;18)
In situ follicular neoplasia (ISFN)

- Not apparent on histologic sections
- Affected follicles are similar in size and shape to reactive follicles
- Positive for t(14;18) but do not have genetic aberrations typically seen in FL
- Risk of subsequent FL development is low (<5%)
- May represent very early step in lymphomagenesis

In situ follicular neoplasia
t(14;18)+

EZH2
TNFRSF14
KMT2D
CREBBP

Follicular lymphoma

Haematologica 98: 1571, 2013
BCL2 negative FL

Subtypes of FL that are BCL2 negative
- Primary cutaneous FL, primary testicular FL
- Pediatric-type FL
- A subset of nodal FL grade 3B, diffuse inguinal

- Increased copies of chromosome 18/BCL2 (15%)
  - t(BCL6)
Other types of FL recognized

- Diffuse FL including a distinctive subset with 1p36 deletions
  Large, localized, with small follicular structures
- Duodenal-type FL (indolent localized, overlapping features with ISFN, MALT)
- Testicular FL (more frequent in children, BCL2 translocation negative, grade 3A)

Katzenbergerger, T et al., Blood, 2009
Bacon CM et al., Am J Surg Pathol 2007
Pediatric follicular lymphoma

- Pediatric FL: a provisional entity in WHO 2008
- Pediatric-type FL is a definite entity in WHO 2016
- Very rare, <3% of pediatric NHL
- Localized disease
- Most are diagnosed in stage I, thus curable
Pediatric-type follicular lymphoma

- 30-50% express BCL2 (mechanism of expression unknown)
- CD10+, BCL6+
- Brisk proliferation index
- t(14;18) is negative
- Different molecular pathogenesis from adult FL
Two types of follicular lymphomas in children and adults

**Pediatric-type**
- Low stage, extranodal
- Curable
- Grade 3
- CD10+ BCL6+ MUM1+
- BCL2 variable
- *BCL2/IGH* negative
- MAP2K1 (49%)
- TNFRSF14 (54%)
- IRF8 (15%)
- Low genome complexity

**Usual-type**
- Stage II-III, nodal
- Incurable
- Grade 1-2
- CD10+ BCL6+ MUM1-
- BCL2 variable
- BCL2/IGH positive

Schmidt J et al., Blood 2016
Since pediatric FL can occur in adults it is now called pediatric-type FL

- Be cautious in adults to misdiagnose a more aggressive grade 3 FL of usual type
- Requires presence of expansile large proliferative follicles that are composed of blastoid cells
Large B-cell lymphoma with *IRF4* rearrangement

- New provisional entity in WHO 2016
- In children and young adults
- Waldeyer ring and or cervical LN
- Low stage
- Follicular, follicular and diffuse or diffuse
- Grade 3B
- BCL6+, MUM1+, BCL2+/- and CD10 +/-
- *IRF4* GR+, *BCL6* GR+, *BCL2*GR-
Enhanced genetic understanding of small B-cell lymphomas (WHO 2017)
## Genetics of small B-cell lymphoma

### CLL/SLL

<table>
<thead>
<tr>
<th>Gene</th>
<th>Overall frequency (%)</th>
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<tbody>
<tr>
<td>NOTCH1</td>
<td>12.2</td>
</tr>
<tr>
<td>MYD88</td>
<td>2.9</td>
</tr>
<tr>
<td>XPO1</td>
<td>2.4</td>
</tr>
<tr>
<td>KLHL6</td>
<td>1.8</td>
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### MZL

<table>
<thead>
<tr>
<th>Gene</th>
<th>Overall frequency (%)</th>
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<tbody>
<tr>
<td>NOTCH2</td>
<td>25</td>
</tr>
<tr>
<td>KLF2</td>
<td>25</td>
</tr>
<tr>
<td>PTPRD</td>
<td>20</td>
</tr>
<tr>
<td>A20</td>
<td>35-50 MALTs</td>
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### FCL

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<th>Gene</th>
<th>Overall frequency (%)</th>
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<tr>
<td>XPO1</td>
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<tr>
<td>KLHL6</td>
<td>1.8</td>
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<tr>
<td>CXCR4</td>
<td>30</td>
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<tr>
<td>MYD88</td>
<td>90</td>
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### MCL

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<th>Gene</th>
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<tr>
<td>NOTCH1</td>
<td>10-15</td>
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<tr>
<td>KMT2D</td>
<td>87</td>
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<td>CREBBP</td>
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<td>EP300</td>
<td>9</td>
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<tr>
<td>EZH2</td>
<td>9</td>
</tr>
<tr>
<td>MEF2B</td>
<td>13</td>
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<tr>
<td>BRAF</td>
<td>100</td>
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### FCL

<table>
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<tr>
<th>Gene</th>
<th>Overall frequency (%)</th>
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</thead>
<tbody>
<tr>
<td>NOTCH2</td>
<td>25</td>
</tr>
<tr>
<td>KLF2</td>
<td>25</td>
</tr>
<tr>
<td>MAP2K</td>
<td>50% HCLv</td>
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</table>
MYD88 mutations in lymphoplasmacytic lymphoma

• Indolent B-cell LPD with overlapping features with MZL
• Encompasses Waldenstrom’s Macroglobulinemia

• Activating mutations in MYD88 (90%) define nodal LPL
• MYD88 mutation leads to TLR signaling
• CXCR4 mutations in 30%
• Combination of MYD88 and CXCR4 associated with response to ibrutinib
• MYD88 mutated patients have best response

https://www.bing.com/images/search?q=MyD88+Signaling+Pathway&FORM=RESTAB
**MYD88 mutations in other B-cell lymphomas**

- IGM monoclonal gammopathy but not in MM
- Not present in IGG/IGA MGUS
- CLL, MZL and SMZL
- Subtypes of extranodal large B-cell lymphoma
- ABC subtype of DLBCL, NOS
**MYD88 mutations in large B-cell lymphoma**

<table>
<thead>
<tr>
<th>Subtypes</th>
<th>Genetic alterations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Primary CNS</td>
<td>MYD88 (30%)</td>
</tr>
<tr>
<td></td>
<td>CD79B (30%)</td>
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<tr>
<td>Primary testicular DLBCL</td>
<td>MYD88 (68%)</td>
</tr>
<tr>
<td></td>
<td>CD79B (19%)</td>
</tr>
<tr>
<td>Primary cutaneous DLBCL, leg type</td>
<td>MYD88 (60%)</td>
</tr>
<tr>
<td></td>
<td>CD79B (20%)</td>
</tr>
<tr>
<td>ABC-DLBCL, NOS</td>
<td>MYD88 (30%)</td>
</tr>
<tr>
<td></td>
<td>CD79B (18%)</td>
</tr>
<tr>
<td></td>
<td>A20 (57%)</td>
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</tbody>
</table>
NOTCH2 and KLF2 in SMZL

- NOTCH2 is recurrently mutated in SMZL
- KLF2 is recurrently mutated in SMZL
- NOTCH2 mutations confer an adverse prognosis

Piva R et al., Leukemia 2014
BRAF MUTATION IN HAIRY CELL LEUKEMIA

BRAF V600E is strongly associated with classic HCL

MAPK2K (MEK) is mutated in HCLv and IGHV4-34+ HCL
Mantle cell lymphoma

- 3-10% of B-cell lymphoma
- Median age of 60 years
- Extranodal involvement >2 sites in 30-50% (Lymphomatoid polyposis, CNS, Waldeyers ring)
- PB involvement in 20-70%
- Aggressive clinical course
Molecular pathogenesis of MCL and variants

- Arises from naïve pre-GC B cells
- Lacks somatic mutations in *IGHV* genes
- t(11;14)(q13;q32)
Mantle cell lymphoma WHO 2017

Broader biologic spectrum than previously recognized (indolent behavior in some)

- In situ mantle cell neoplasia

- MCL, leukemia non-nodal type (SOX11 neg with mutated IGH)
In situ mantle cell neoplasia

- Analogous to *in situ* follicular neoplasia
- Uncommon and often incidental finding
- May represent early step in MCL
- Indolent but maybe disseminated
- Most patients do not develop overt MCL

*Haematologica 2012;97(2):270-278*
MCL, leukemia non-nodal type

- Biologically different subset relative to classic MCL
- Identified by gene expression profile studies
- BM +/- splenic involvement
- Mutated IGHV
- SOX11 negative
- Monoclonal asymptomatic lymphocytosis, cyclin D1 positive

Ondrejka SL et al., Haematologica 2011,96: 1121-7
Cyclin D1 negative MCL

- 10% of MCL are cyclin D1 negative
- Diverse mechanisms of pathogenesis
- *CyclinD2* or *CyclinD3* rearrangements
- Mutations of *CyclinD1* gene
SOX11 in Cyclin D1 negative MCL

- Neural transcription factor
- Highly sensitive for MCL but not specific
- SOX11+ in Burkitt lymphoma (25%) and hairy cell leukemia (50%)
- Histology and immunophenotype should be otherwise typical
- Other D type cyclins involved
  - IG/CyclinD2 translocations (>75%)

Mozoz et al 2009; Chen et al, 2010
Take home points

• Recognize pre-neoplastic or early presentation of B-cell lymphomas (MBL in CLL, ISFN in FL, ISMCL)
• MCLs exhibit broader biologic spectrum than previously recognized (indolent forms of MCL, cyclin D1 negative MCL, SOX11 negative MCL)
• Pediatric-type FL (spectrum of disease, can occur in adults, have unique genetic features)
• Pediatric population can have usual-type FL
• Primary FL (duodenal, testicular) have distinct biology
Take home points

• BRAF V600E mutation in virtually all HCL
• MYD88 mutations in vast majority of LPL but only a small percentage in other small B-LPD
• NOTCH1 mutations in CLL/SLL, MCL: prognosis?
• NOTCH2 and KLF2 mutations in a subset of SMZL: diagnosis, prognosis and potentially therapeutic