THE REVISED WHO CLASSIFICATION OF LYMPHOMAS – WHAT HAS CHANGED FOR THE PATHOLOGIST AND CLINICIAN

Elaine S Jaffe, NCI, NIH
WHO Classification of Tumours of the Haematopoietic and Lymphoid Tissues

A new taxonomy of disease*

- Build a biomedical information network to promote disease discovery & pathogenetic insights
- Provide a framework for “Precision Medicine”
- Facilitate clinical trials
- Improve the standard of diagnosis and treatment in the community

New Insights Since 2008

• Rapid progress in understanding of molecular pathogenesis
  – NGS studies, Nanostring
  – Allow high throughput investigation of paraffin embedded samples

• Large scale clinical studies led to new insights into clinical behavior
  – Interest in more targeted therapy

• IARC authorized a “Revised 4th WHO classification”
Clinical Advisory Committee
Integral Part of the Process since the 2001 Edition

• Classification should be useful to both pathologists and clinicians
• Classification should be suitable for daily practice and clinical trials
• Has remained an integral part of the process
Summaries of revisions
Swerdlow et al. (Lymphoid Neoplasms)
Arber et al. (Myeloid and Acute Leukemia)
Blood May 19, 2016

Bluebook published
September 2017

WHO Press
World Health Organization
bookorders@who.int
www.who.int/bookorders/

From the USA
Stylus Publishing
22883 Quicksilver Drive
Herndon VA 20172-05
stylusmail@presswarehouse.com
www.styluspub.com
What’s New in Early Lesions

BCL2

Cyclin D1

MBL, In situ FL and In situ MCL, Duodenal type FL

BCL2
What’s New in B-cell Lymphomas

- LPL and MZL
- Pediatric type FL
- Double Hit Lymphomas
What’s new in the Peripheral T-cell lymphomas

- AITL & other nodal TFH lymphomas
- Intestinal T-cell lymphomas
- ALCL, ALK-negative
Early Events in Neoplasia come in “two flavors”

Early lesion is phenotypically and genotypically similar to malignant counterpart (well-defined entities)
  – Monoclonal B-lymphocytosis/ CLL
  – MGUS/ Myeloma
  – Follicular lymphoma in situ (FLIS)
  – Mantle cell lymphoma in situ

Clonal lymphoid proliferations of limited malignant potential (B & T) – unrelated to specific lymphoma subtypes (not precursors)
  – Pediatric-type follicular lymphoma
  – Primary cutaneous acral CD8+ T-cell lymphoma
  – Primary cutaneous CD4+ T-cell LPD
  – Indolent T-cell LPD of the GI tract
“Early” forms of well-defined lymphoid neoplasms

- Retrospective studies reveal almost universal evidence of the “early lesion” many years prior to diagnosis
  - Best shown for MBL, MGUS
- But, progression to frank lymphoma or leukemia is not the rule
- Most patients do not progress – not necessarily a precursor
  - What are the predictive factors?
Follicular Lymphoma In Situ (FLIS)
Cong et al Blood 2002

Bcl-2
BCL2/IGH in Healthy Individuals
(Limpens et al. 1991; Roulland et al. 2006)

• BCL2/IGH is found in peripheral blood of up to 70% of normal adults over age 50
  – Numbers increase with age
  – Numbers increase with pesticide use in farmers

• BCL2/IGH + B-cells are not naïve B-cells
  – Memory B-cells, Class switched
  – Have encountered the germinal center reaction
  – Prone to intense trafficking among germinal centers

• FLIS & FL-like B-cells coexist in the same pt
• FL-like B-cells home to GC environment

• FLIS seen in 2-3% of all lymph node biopsies -- sometimes coincidental with other B-cell lymphomas

• Low level of genetic aberrations beyond BCL2R

• Low risk of progression to FL < 5%

• No therapeutic intervention required
Phenotypically and genetically similar to nodal FL (*BCL2/IGH*), but usually IgA+

Commonly present in duodenum
- other sites in distal small bowel

Superficial polypoid lesions in mucosa

Express homing receptor found on intestinal lymphocytes (*α4β7* integrin)

Lack AID activity

Local recurrences without dissemination
t(14;18)

FL-like B-cell in healthy individuals
- t(14;18)+
- SHM
- CSR
- Intense trafficking

Duodenal-type follicular lymphoma
- Intestinal localized disease
- t(14;18)+
- Low level CNAs
- CREBBP, TNFRSF14, EZH2 mutations
- Low frequency KMT2D mutations

In-situ follicular neoplasia
- Nodal localized disease
- t(14;18)+
- Low level CNAs
- CREBBP, TNFRSF14, EZH2 mutations
- ? Low frequency KMT2D mutations

Conventional follicular lymphoma
- Systemic disease
- t(14;18)+
- High level CNAs
- CREBBP, TNFSF14, EZH2, BCL2 mutations
- Frequent mutations in KMT2D

Jaffe; Blood 2018
In situ Mantle Cell Lymphoma

Positive cells on the inner zone of mantle cuff

Cyclin D1
What is the preferred terminology for these “early lesions”?

In situ follicular lymphoma/ In situ mantle cell lymphoma/ Intestinal follicular lymphoma

WHO 2017

• In situ follicular neoplasia
• In situ mantle cell neoplasia
• Duodenal-type follicular lymphoma
Differential Dx of LPL vs. MZL has been challenging for patients presenting with nodal disease.
Lymphoid Malignancies with MYD88 L265P Mutation

- Waldenstrom’s mac/ LPL: 90%
- IgM MGUS: 50%
- DLBCL, ABC: 30%
- DLBCL, testis/ CNS: 70%
- CLL: 3%
- SMZL
- NMZL
- EMZL < 10 % overall

Activates NFkB pathway
MAPK pathways
Pediatric Follicular Lymphoma

in the 2008 WHO

Rare subtype in children (1-2%)
Tonsils, nasopharynx, GI tract, testis, lymph nodes
Typically “high grade” (3A/3B)
Male >> Female
85% localized, Stage I or II

- Not clearly defined as an entity
- Nodal and extranodal forms not clearly distinguished
Nodal, Usually Head and Neck, Stage I
M >> F; 22:0
CD10+, BCL6+, BCL2-, MUM1-
Chromosomal aberrations rare

Tonsil/ Waldeyer’s ring; M=F
Co-expression of MUM1, BCL6, often CD10
Frequent IRF4 breaks

Testicular, Stage I, good prognosis
CD10+, BCL6+, BCL2-, MUM-
Occasional BCL6 breaks
Pediatric-type Nodal Follicular Lymphoma

(Louissaint et al, 2012; Liu et al 2013)

- A clonal germinal center B-cell proliferation of undetermined malignant potential
- Median age, 15-18 yrs, uncommon over age 40
- Marked male predominance (~10:1)
- Clonal CD10+ B-cells by flow; IG PCR+
- Genetic aberrations for BCL2, BCL6, IRF4 not identified
- Many patients in continuous CR following surgical excision and no further treatment – conservative approach recommended
Pediatric-type of Nodal Follicular Lymphoma

CD79a

IgD

CD10
Prominent starry sky

Follicles composed of medium sized “blastoid cells”

Not typical Grade3A/B
Pediatric-type Nodal Follicular Lymphoma
(Schmidt et al., Blood 2016; 2017; Louissaint et al, Blood 2016)

• Genome wide analysis shows recurrent mutations
  • Mutations in TNFRSF14 with frequent copy-number neutral loss of 1p36
  • Region affected in > 50% of cases
    • Not specific for Pediatric type FL- also in usual FL
  • Frequent mutations involving the MAPK pathway
    • MAP2K1 (~50%); more rarely RRAS;MAPK1
Large B-cell lymphoma with IRF4 Translocation

Translocation partners include IGH, IGL, and undetermined

Salaverria et al 2011; Liu et al. 2013

Follicular, but may have diffuse areas

Waldeyer’s Ring in 80%; Median age 12; M=F

Provisional entity in revised WHO
Atypical GCB phenotype, IRF4/MUM1+
Primary mediastinal large B-cell lymphoma
Diffuse large B-cell, NOS (most cases) [ABC/GCB]
  Primary DLBCL of the CNS
  Primary cutaneous DLBCL, leg type
    *EBV positive DLBCL, NOS [of the elderly]*
    *DLBCL associated with chronic inflammation (EBV+)*
T-cell/ histiocyte rich large B-cell lymphoma
*IRF-4 positive large B-cell lymphoma*
Lymphomatoid granulomatosis (EBV+)
Intravascular large B-cell lymphoma
ALK-positive large B-cell lymphoma
Primary effusion lymphoma (KSHV/HHV8+); *HHV8+ DLBCL*
Plasmablastic lymphoma
Burkitt lymphoma/ *Burkitt-like lymphoma with 11q aberration*
B-cell lymphoma intermediate between DLBCL and CHL (GZL)
*High grade B-cell lymphoma, w MYC, BCL2, &/or BCL6 R*
*High grade B-cell lymphoma, NOS*
MYC translocation is necessary but not specific for Burkitt Lymphoma

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>% MYC R</th>
</tr>
</thead>
<tbody>
<tr>
<td>Burkitt lymphoma</td>
<td>100%</td>
</tr>
<tr>
<td>De Novo Double-hit</td>
<td>100%</td>
</tr>
<tr>
<td>t(14;18) &amp; t (8;14)</td>
<td></td>
</tr>
<tr>
<td>Plasmablastic lymphoma</td>
<td>~ 50%</td>
</tr>
<tr>
<td>DLBCL</td>
<td>5-8%</td>
</tr>
<tr>
<td>Follicular lymphoma</td>
<td>2-8%</td>
</tr>
<tr>
<td>Transformed follicular</td>
<td>~ 5%</td>
</tr>
<tr>
<td>DLBCL, high grade</td>
<td></td>
</tr>
<tr>
<td>Lymphoblastic, TDT +</td>
<td></td>
</tr>
</tbody>
</table>
Most studies have shown a poor prognosis with R-CHOP or comparable regimens

- Savage et al, Johnson et al, BCCA, 2009
- Tomita et al, Japan, 2009
- Niitsu et al Japan, 2009
- Snuderl et al, MGH, 2010
- Barrans et al, UK, 2010
Additional questions regarding the impact of a MYC translocation in Agg B-cell Lymphomas

• Does MYC rearrangement necessarily have adverse prognostic significance?
• Does “double expression” carry the same weight as a Double Hit?
• What is impact of extra MYC signals?
• What is the influence of the translocation partner with MYC: IG vs. Other?
• Does histological subtype have relevance, DLBCL vs. B-cell, unclassified?
Isolated MYC R does not confer a worse prognosis in patients with DLBCL treated with DA-EPOCH-R

Lai et al. Leukemia & Lymphoma, 2017

OS
MYC R pos _ _ _
MYC R neg ______
Time to Prog
Does Double Expression of both MYC and BCL2 carry the same prognostic significance?

- Can IHC substitute for genetic studies?
- MYC rabbit monoclonal; Y69 clone, Epitomics

 Burkit lymphoma
MYC IHC
Double Hit vs. Double Expression in DLBCL
Johnson et al JCO 2012; 30: 3452

Double expression 4x more common than Double hit

Double expression Intermediate in outcome between DH and Neither
MYC/BCL2 co-expression confers inferior prognosis independent of MYC/BCL2 co-rearrangements

OS in Double Hit (2%)  
GCB by GEP

OS in Double Expressors (31%)  
Without a double hit
Mainly ABC tumors

Hu S et al. Blood 2013
Double Hit vs. Double Expressing aggressive B-cell lymphomas

– Key Points

• Use standard criteria for DE
  – MYC > 40%; BCL2 >50%
• Double Hit Lymphomas (BCL2 & MYC R) are GCB by GEP & IHC
• Double expressing lymphomas (BCL2 & MYC + by IHC only) are ABC by GEP & IHC
  – Usually MUM1+
• Biological basis is distinct
  – Therefore, treatment implications are distinct
You cannot compare Apples & Oranges

Double Hit
GCB

≠

Double Expressor
ABC
Does histological subtype have relevance, DLBCL vs. B-cell, unclassified in DH cases?

- Some studies have found better prognosis for DLBCL than for “high-grade, unclassified”.
  - Snuderl et al 2010; Johnson et al 2009

Kaplan Meier OS
Double Hit Lymphomas
DLBCL vs. High Grade, Unclassified (WHO 2008)
Snuderl AJSP 2010

![Kaplan Meier OS graph](image-url)
Is Histological Subtype Accurately and Reproducibly Diagnosed?

DLBCL and B-cell lymphoma, Intermediate BL/DLBCL have overlapping histological features

MYC and BCL2 R
Summary

- Classical Double Hit lymphomas with MYC and BCL2 R (& BCL6 R) have a poor prognosis
- MYC rearrangement alone may not be adverse
- The translocation partner with MYC - IG vs. Other is clinically significant
- Extra MYC signals by themselves are less relevant
- Poor prognosis of DE may relate to ABC subtype
  - Don’t lump DH and DE in same category
- Relevance of histological subtype - DLBCL vs. “Burkitt-like” is controversial
Guidelines for Dx of DLBCL

• Distinguish between GCB and ABC by available means (IHC for most labs now)
• FISH for MYC, BCL2, recommended but not required
  – Can screen by IHC
  – MYC+ GCB DLBCL more likely to have a MYC R
  – MYC R rare in DLBCL of ABC type, even if MYC + by IHC
Revisions to the WHO 2016

• High grade B-cell lymphoma, with *MYC* and *BCL2* and/or *BCL6* rearrangements
  – Classify secondarily as to morphology, DLBCL or High grade unclassifiable

• High grade B-cell lymphoma, NOS
  – Includes cases with blastoid, many are MYCR

• Diffuse large B-cell lymphoma (DLBCL), NOS
  – Classify as to GCB or ABC
Comparison of Aggressive B-cell Lymphomas: WHO 2008 to 2016
Burkitt-like lymphoma with 11q aberration, Salaverria et al. Blood 2014

Pediatric, mainly nodal
Negative for MYC R
May have MYC protein
Gains and losses at 11q
Aggressive B-cell Neoplasms in the WHO (2017)

new subtypes, entities

Primary mediastinal large B-cell lymphoma
Diffuse large B-cell, NOS (most cases) \([ABC/GCB]\)
  Primary DLBCL of the CNS
  Primary cutaneous DLBCL, leg type
    \textit{EBV positive DLBCL, NOS} [of the elderly]
    \textit{DLBCL associated with chronic inflammation (EBV+)}

T-cell/ histiocyte rich large B-cell lymphoma
\textit{IRF-4 positive large B-cell lymphoma}

Lymphomatoid granulomatosis (EBV+)

Intravascular large B-cell lymphoma

ALK-positive large B-cell lymphoma

Primary effusion lymphoma (KSHV/HHV8+); \textit{HHV8+ DLBCL}

Plasmablastic lymphoma

Burkitt lymphoma/ \textit{Burkitt-like lymphoma with 11q aberration}

B-cell lymphoma intermediate between DLBCL and CHL (GZL)
\textit{High grade B-cell lymphoma, w MYC, BCL2, &/or BCL6 R}
\textit{High grade B-cell lymphoma, NOS}
What’s new in the Peripheral T-cell lymphomas

- AITL & other nodal TFH lymphomas
- Intestinal T-cell lymphomas
- ALCL, ALK-negative
Gene expression profiling and mutation analysis has helped to clarify the interrelationship among nodal T-cell lymphomas of TFH origin.
Gene expression profiling allowed reclassification of 14% of PTCL, NOS as AITL

Gene expression signatures of PTCL; Iqbal et al. *Blood* 2014
Genomic Findings in AITL and TFH derived lymphomas

- 20-45% in *IDH2*, *DNMT3A* and *TET2* in AITL
  - *Genes involved in pathogenesis of gliomas, AML*
- *TET2* mutations also seen in other PTCL of TFH origin (up to 60%)
- *RHOA* mutations in 60% of AITL and some PTCL, NOS, all with *TET2*
- *IDH2* most specific for AITL

Nodal Peripheral T-cell Lymphomas (2008)

- PTCL, NOS
- T-zone variant
- Follicular variant
- Lymphoepithelioid cell variant
- Angioimmunoblastic T-cell lymphoma
Enteropathy Associated T-cell Lymphoma, Types I & II are distinct

EATL I
Usually αβ
Celiac
disease
N European

EATL II
Usually γδ
Epitheliotropic
Asian, Hispanic

γδ
Monomorphic epitheliotropic intestinal T-cell lymphoma (EATL II)

- Medium sized cells with clear cytoplasm
- CD56 +, CD8+, CD4-
- Usually γδ+
- MAT kinase +
- SETD2 mutations (> 90%)
- STAT5B (> 60%)
JAK/STAT Pathway is an attractive target for therapy of Cytotoxic T-cell Lymphomas and Leukemias
Anaplastic Large Cell Lymphomas
overlapping clinical and biological features

- ALCL, ALK-positive
- ALCL, ALK-negative
- Primary cutaneous anaplastic large cell lymphoma & Lymphomatoid papulosis
- Breast implant associated anaplastic large cell lymphoma
Required: Cohesive growth pattern with hallmark-like cells
Strong and uniform CD30 expression
Desirable but not essential:  EMA+, Cytotoxic +, Sinusoidal growth, Loss of “T-cell ag”
Constitutive Activation of the JAK/STAT pathway in Systemic and Cutaneous ALK-negative ALCL

Activating mutations of JAK1 or STAT3 or both (20%)

Crescenzo Cancer Cell 2015
Genetic correlates with survival in ALCL, ALK+/ ALK-

Feldman et al. Blood 2014

Subset with DUSP22 R
Comparable to ALK+

p<0.0001

DUSP22 (# 22)
ALK+ (# 32)
P63 (# 6)
ALK neg, no aberrations (#45)
Breast Implant-associated anaplastic large cell lymphoma, ALK-negative

- Seen with a variety of breast implants, both saline and silicone
- Usually years after implant
- Symptoms related to accumulation of seroma fluid in cavity surrounding the implant
- Diagnosis best made by cytology
- Cells grow within cavity and on surface of cavity lining, usually without invasion
Breast implant assoc. ALCL
A provisional entity
Biological Features

- Clonal TCR reported in most but not all cases
- JAK1 & STAT3 mutations
  - Similar to ALK+/ALK- ALCL (Blombery 2016; 2017)
- Surprisingly indolent course, despite very atypical cytological features
- Therapy varies in literature
  - Chemo, Radiation, Observation following removal
- Removal of implant is probably adequate therapy in most cases – if no invasion of capsule
With acknowledgment to the many contributors &

**Lymphoid Editors**
Steven H. Swerdlow
Elias Campo
Stefano Pileri
Nancy Lee Harris
Harald Stein
Reiner Siebert

& for the **Myeloid Neoplasms**
James Vardiman; Jurgen Thiele
Dan Arber, Rob Hasserjian, Attilio Orazi,
Michelle LeBeau