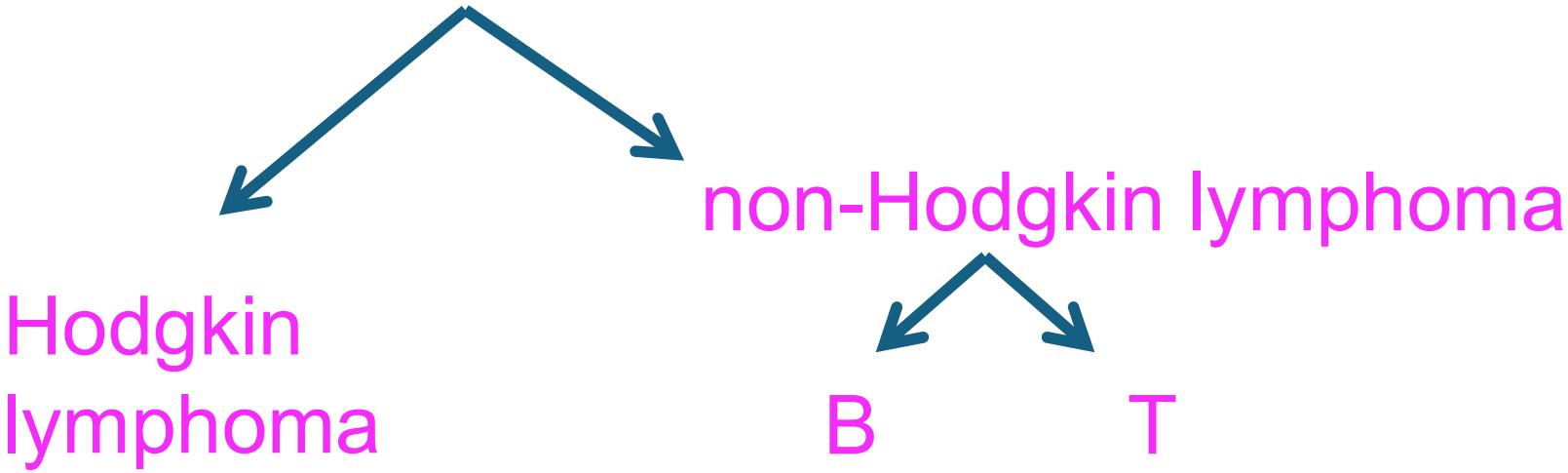
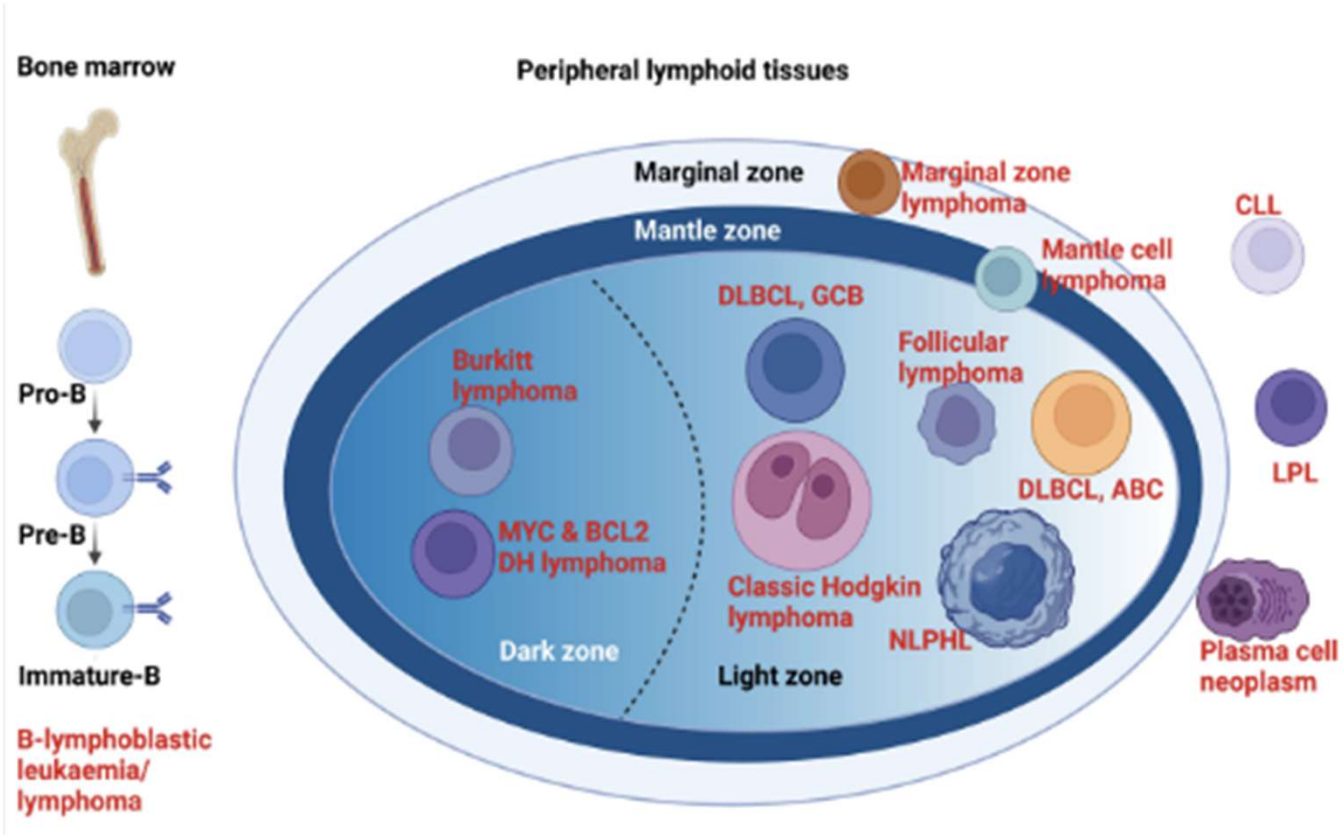
Lymphomas

P. Korkolopoulou, Professor in Pathology, NKUA
E. Lakiotaki, Ass. Professor in Pathology, NKUA
A. Sepsa, MD, PhD

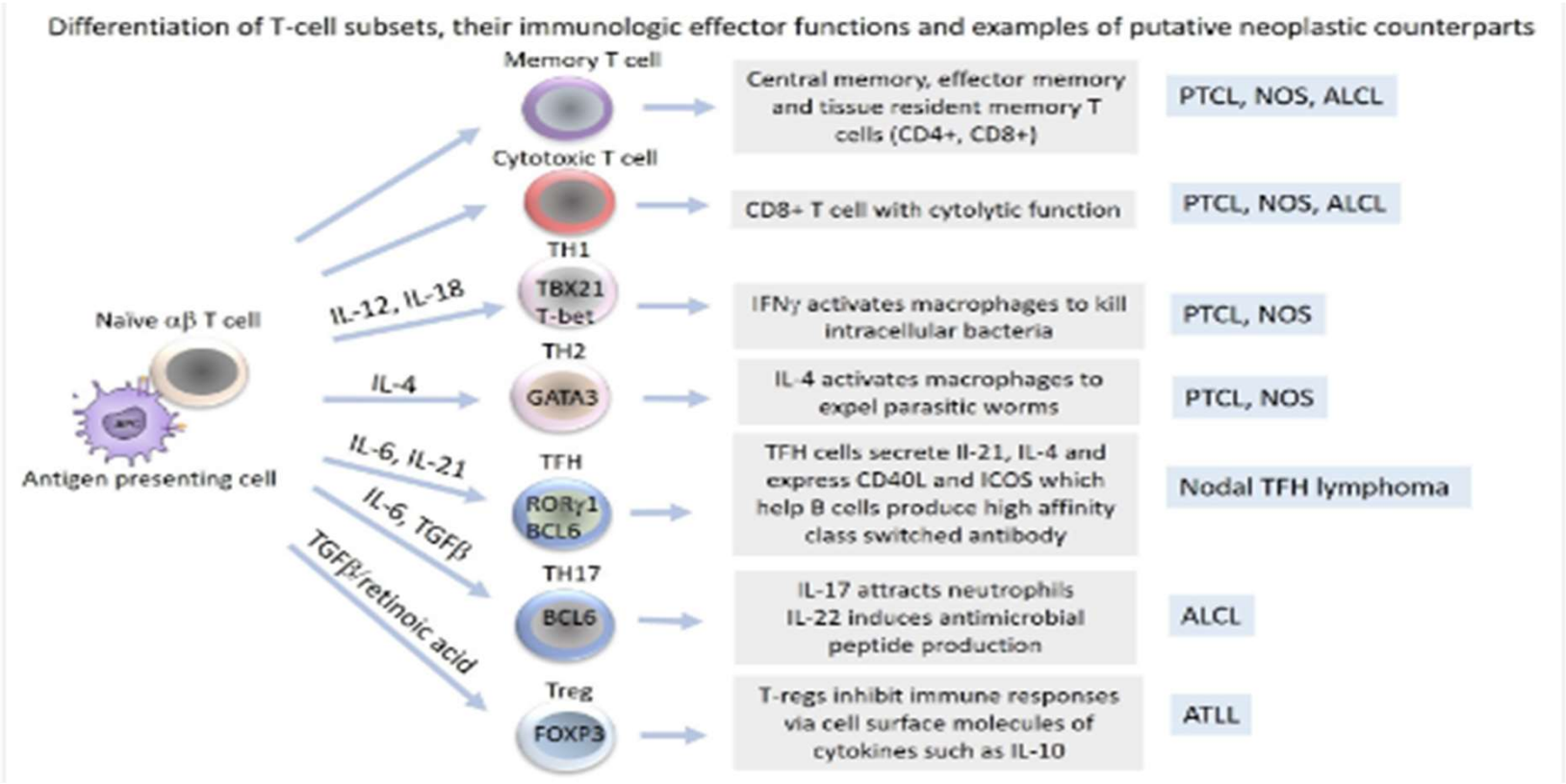
- **Lymphoma:** Malignant neoplasm of lymphoid lineage, often recapitulating differentiation stages of normal lymphoid cells





Different B-cell malignancies show features characteristic or reminiscent of B-cells at specific points during their differentiation/maturation. Please note that the image represents ontogenic relationship, but not morphological patterns of the lymphoma entities.

WHO, Haematolymphoid tumours 2022



The differentiation of effector T cell subsets. Effector T cell subsets are derived from interaction of naïve CD4+ or CD8+ T cells with antigen presenting cells through antigen-specific MHC-dependent interaction. Antigen-specific memory T cells can be found in central, effector or tissue resident subsets which can be either CD4+ or CD8+. The presence of polarizing cytokines that are present during T cell activation lead to epigenetic modifications and expression of transcription factors which contribute to the differentiation of naïve helper T cells into T helper cell subsets (TH1, TH2, TFH, TH17 and T-reg). The expression of transcription factors, cell surface molecules and cytokines are helpful in characterisation of T cell subsets. Examples of putative neoplastic counterparts of T cell subsets are listed.

WHO, Haematolymphoid tumours 2022

Main immunohistochemical markers related to lymphoid follicles

	CD20	CD10	Bcl6	Bcl2	CD21	CD23	DBA44	CD3
Follicular B-cells	+	+	+	-	-	-	-	-
Follicular dendritic cells	-	-	-	-	+	+	-	-
Follicular T-cells	-	+	+	+	-	-	-	+
Mantle B-cells	+	-	-	+	+	+	+	-
Marginal zone B-cells	+	-	-	+	+	-	-	-

Main immunohistochemical markers related to cells in the paracortical area

	CD3	CD4	CD5	CD8	CD43	S-100	CD34
T-cells	+	+	+	+	+	-	-
Interdigitating dendritic cells	-	-	-	-	-	+	-
High endothelial venules	-	-	-	-	-	-	+

HODGKIN Lymphoma

malignant cells

Definition: Reed-Sternberg (RS) and Hodgkin cells
[minority population]
+
Inflammatory background cells
[majority population]

Heterogenous neoplasm:
- Nodular lymphocyte predominant Hodgkin lymphoma
- Classic Hodgkin lymphoma

ANN-ARBOR STAGING

Stage	Disease Involvement
I	Single lymph node region (I) or one extralymphatic site (I_E)
II	Two or more lymph node regions, on the same side of the diaphragm (II) or local extralymphatic extension plus one or more lymph node regions on the same side of the diaphragm (II_E)
III	Lymph node regions on both sides of the diaphragm (III), which may be accompanied by local extralymphatic extension (III_E)
IV	Diffuse involvement of one or more extralymphatic organs or sites

Classic Hodgkin lymphoma

- **Definition: Classic Hodgkin lymphoma (CHL) is a neoplasm derived from germinal centre B-cells, characterized by a low fraction of tumour cells embedded in a reactive micro-environment rich in immune cells. The large neoplastic Hodgkin and Reed–Sternberg cells show a defective B-cell expression program.**

- **Four subtypes due to differences in**
  1)reactive micro-environment cellular composition
  2)neoplastic cells' morphology

Classic Hodgkin lymphoma

- **Epidemiology:**
  95% of HL cases

Nodular sclerosis (58,1%)
Mixed cellularity (18,9%)
Lymphocyte-rich classical Hodgkin lymphoma (5,8%)
Lymphocyte depleted (1,5%)
Not further classified (15,7%)

- **Etiology-Pathogenesis: Genetic alteration of HRS cells (activating mutations in NF-kB, JAK/STAT and MAPK/ERK signalling pathways), intense cross-talk between HRS cells and reactive micro-environment, EBV infection, particularly in immunosuppression setting (HIV+)**

-In tropical countries 100% of cases are EBV+

Age: 2 peaks – adolescents, young adults (15-35 yrs) and elderly (>60 yrs)

- **Clinical features: Lymphadenopathy-B symptoms (fever, night sweats, and weight loss) in 40% of cases**

– Primary extranodal disease very rare

Localization: cervical lymph nodes (75%), mediastinal, axillar, paraortic-
60% of patients staged I/II – in fewer cases spleen (20%) or bone marrow (5%) involvement

CHL - Morphology

RS cells +

Large size

Abundant cytoplasm

≥2 nuclei/nuclear lobes

Prominent eosinophilic nucleolus

Mononuclear cells →

Hodgkin cells

Reactive inflammatory cellular environment

neutrophils

eosinophils

lymphocytes

Plasma cells

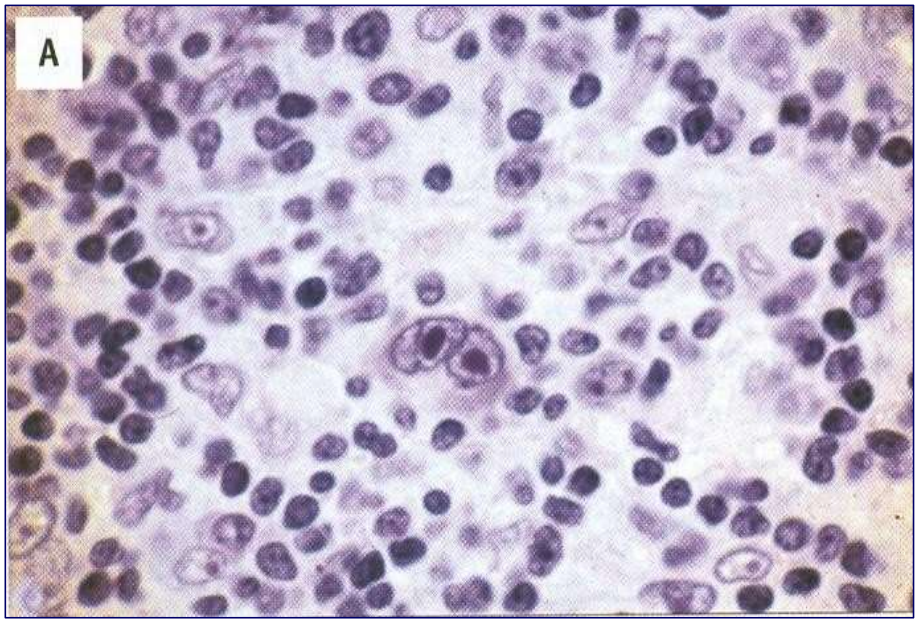
(epithelioid) histiocytes

fibroblasts

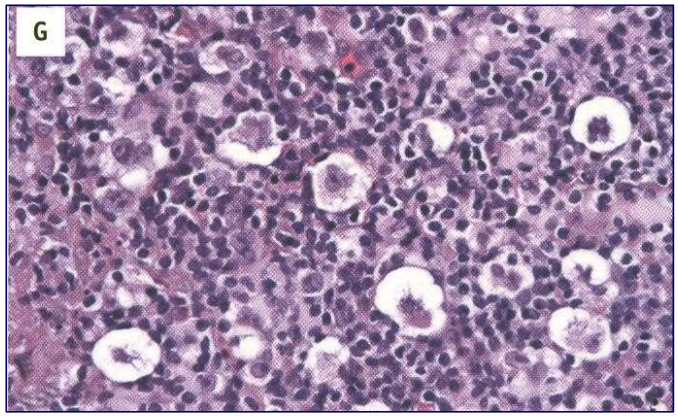
REED-STERNBERG cells – morphology forms

- **Classic RS cell: Mixed cellularity** (Nodular sclerosis-Lymphocyte depleted)
- **Lacunar: Nodular sclerosis** (Mixed cellularity-Lymphocyte-rich classical Hodgkin lymphoma)
- **Pleomorphic: Lymphocyte depleted**

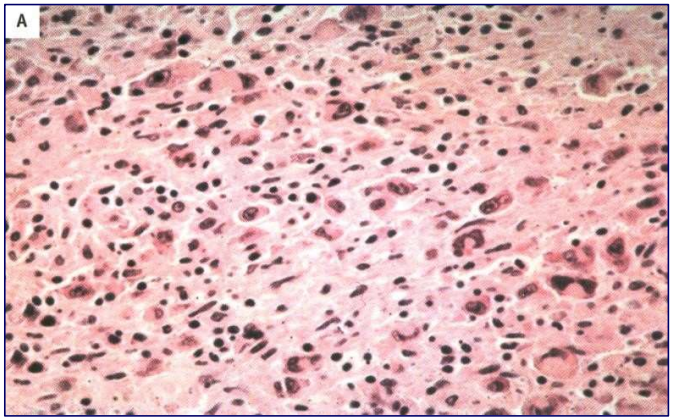
REED-STERNBERG cells – morphology forms



Classic



Lacunar



Pleomorphic

CHL – HRS Immunophenotype

CD30 + (100% of cases)

CD15 + (75-85% of cases / usually a small percentage of cells)

Membranous-
Dot-like paranuclear staining

CD20 - (+ in 30-40% of cases/ variable intensity/ small percentage of cells)

CD79α – (very rarely +)

PAX5 + (faint intensity, in comparison to B-cells)

MUM-1+

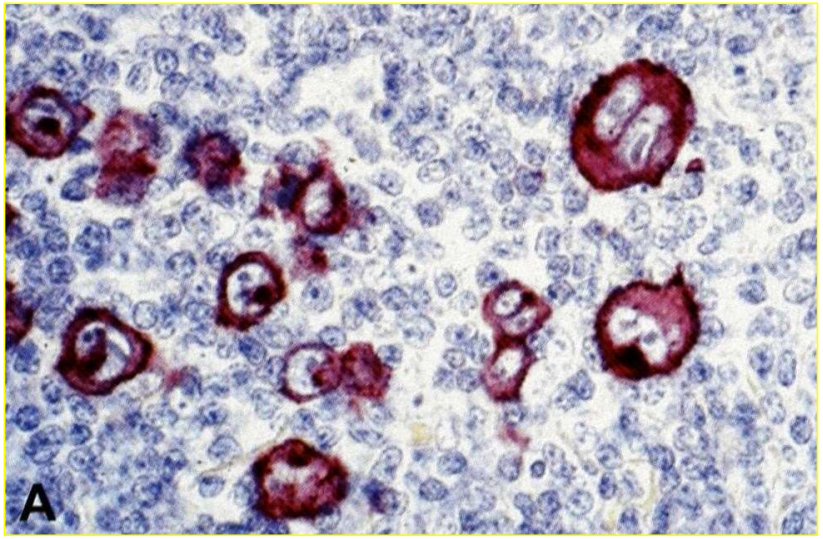
CD138-

OCT-2 or BOB-1 rarely + (10%)

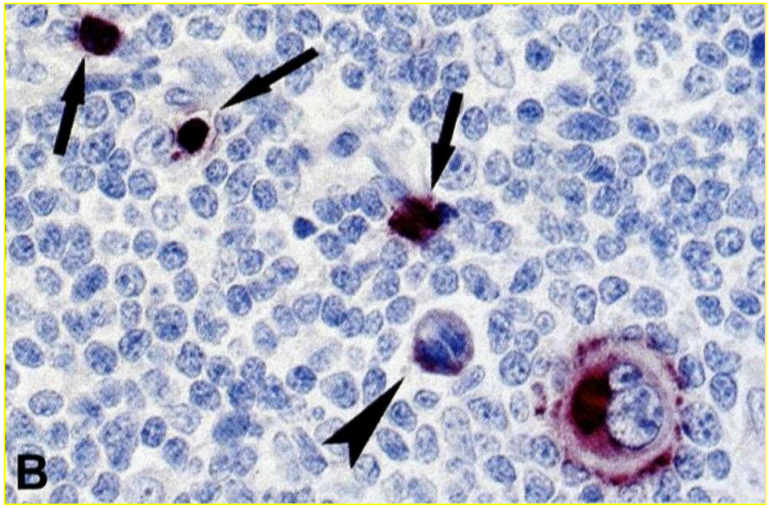
EMA, T-cell antigens – (very rarely+)

EBV + (20-75% of cases): EBNA-1/LMP-1

CHL



CD30



CD15

Comparison of immunophenotype of neoplastic cells in various Hodgkin Lymphoma types

	CHL	NLPHL
CD45	-	+
CD15	+	-
CD30	+	-
B-cell markers	- (+)	+
T-cell markers	-	-
EMA	-	+ (-)
EBV	+/-	-

CHL

Postulated normal counterpart: Germinal centre B-cell (95% of cases) with deregulated B-cell program

Genotype: Clonal rearrangements and somatic hypermutations of immunoglobulin genes

Prognosis: Curable disease in 85% of cases (radio-chemo-therapy) – therapeutic choices according to patient's stage

Nodular sclerosis CHL

- **Definition: CHL showing collagen bundles that surround at least one nodule and HRS cells "lacunar" type**
- **F > M**
- **Median age 28 yrs (bimodal age distribution)**
- **Clinical features: mediastinal mass (80%) (usually stage II) – B symptoms (40%) –**

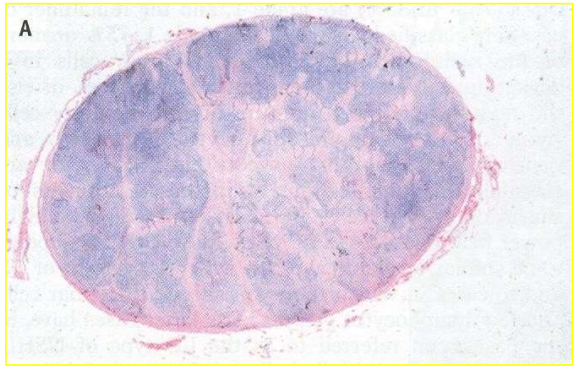
  Involvement: spleen/lung 8-10% of cases- bone marrow 3% - liver 2%
- **Prognosis: stage-dependent**
- **EBV+ (10-40% of cases)**

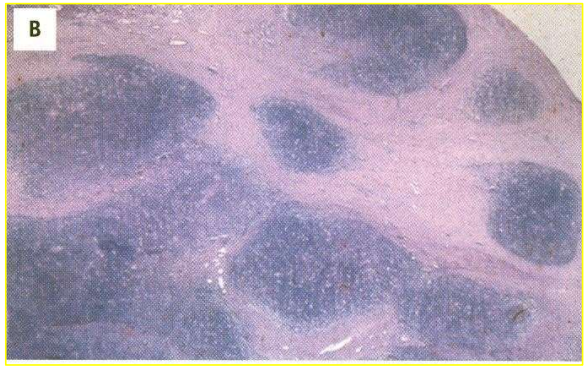
Nodular sclerosis CHL -Morphology

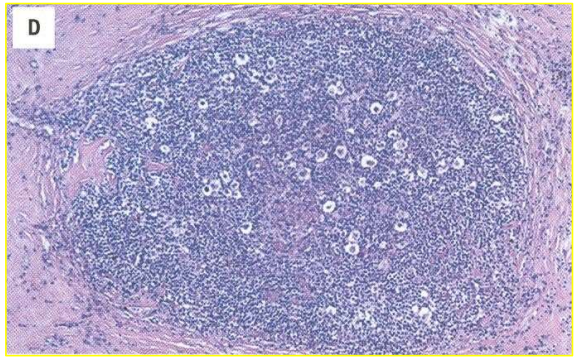
- Nodules surrounded by fascicles of hypocellular fibrous tissue
- Nodal capsular thickening
- “Lacunar” HRS cells with small nuclei and nucleoli, with retraction artefact that creates a hollow space
- Eosinophils, eosinophilic abscesses
- Histiocytes, necrotic abscesses
- “Lacunar” cell sheets «syncytial» variant

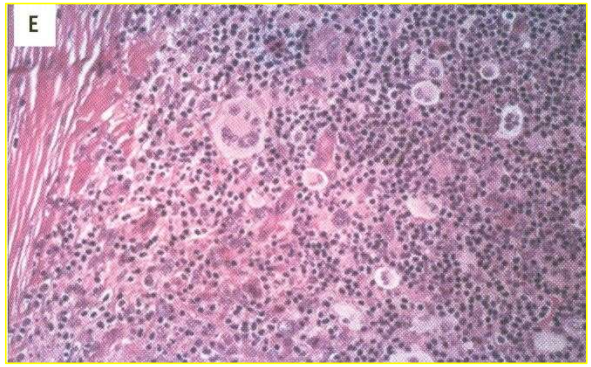
Extent of eosinophil presence and HRS cell percentage prognostic factors

Nodular sclerosis CHL







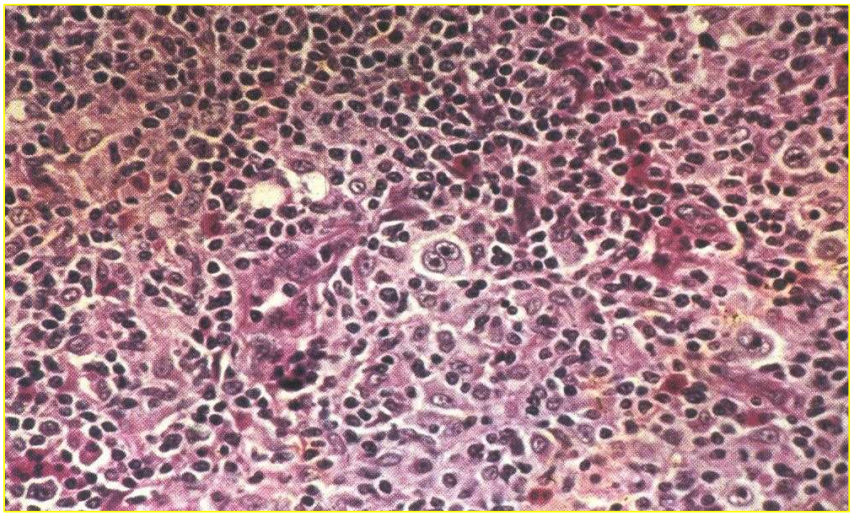


Mixed cellularity CHL

- **Definition:** CHL with HRS cells in reactive inflammatory micro-environment without nodules or sclerosis
- M > F
- Median age: 37 yrs
- **Clinical features:** peripheral lymphadenopathy (often stage III/IV) – usually with B symptoms
- Often in HIV+ patients
- Prognosis: stage-dependent
- EBV+ 75% of cases

Mixed cellularity CHL - morphology

- **HRS cells may be distributed interfollicularly**
- **In some cases there may be sclerotic fascicles without nodule formation or capsular thickening**
- **Typical HRS cells**
- **↑ ↑ (epithelioid) histiocytes in EBV+ cases**



Lymphocyte-rich CHL

- **Definition: CHL with rare HRS cells in lymphocyte-rich reactive micro-environment and usually nodular pattern**
- **M > F**
- **Age: usually 50-70 yrs**
- **Clinical features: peripheral lymphadenopathy (often stage I/II) – no B symptoms**
- **Prognosis: stage-dependent**

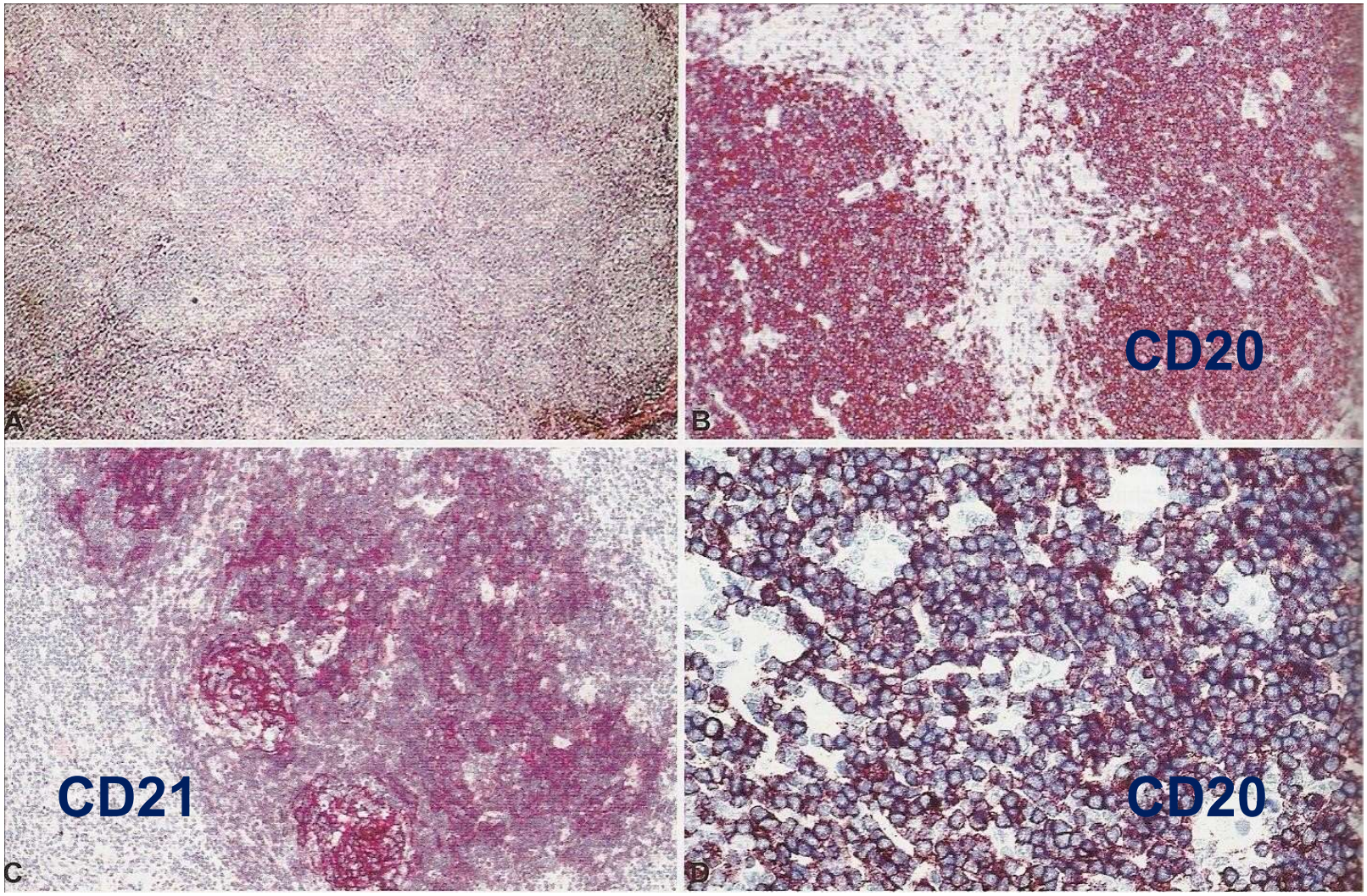
Lymphocyte-rich CHL - morphology

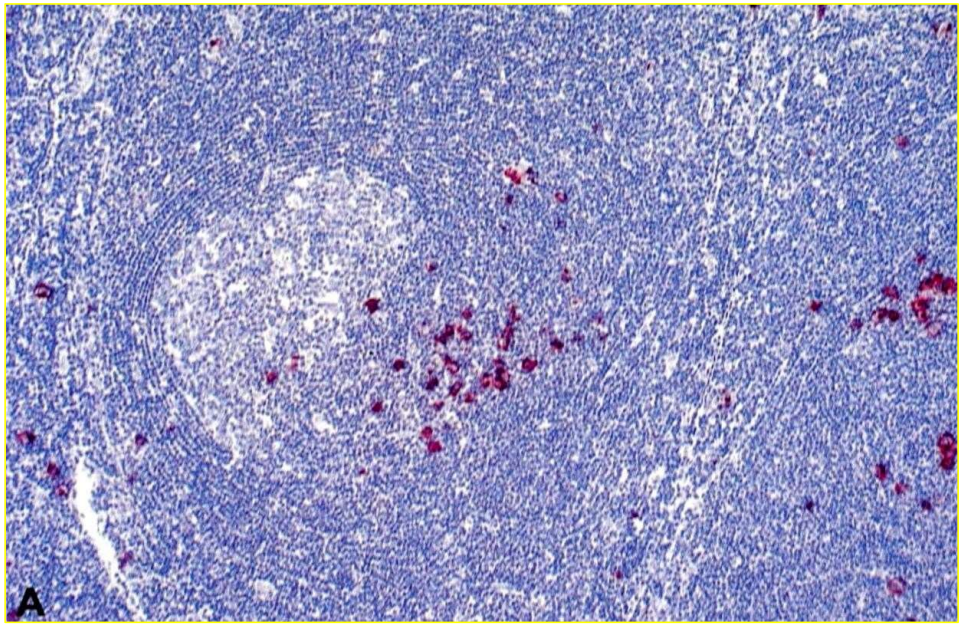
- Nodular (rarely diffuse) pattern
- Rich follicular dendritic cell network
- Cellular composition in the nodules: small B-cells (IgM+/IgD+) and rare typical HRS cells –
  no eosinophils/neutrophils
- Eccentric, atrophic germinal centres
- HRS cells into the expanded mantle zone of the follicles

** Immunophenotypic studies are crucial for differential diagnosis from NLPHL

** When diffuse growth pattern → small T-cells

Lymphocyte-rich CHL





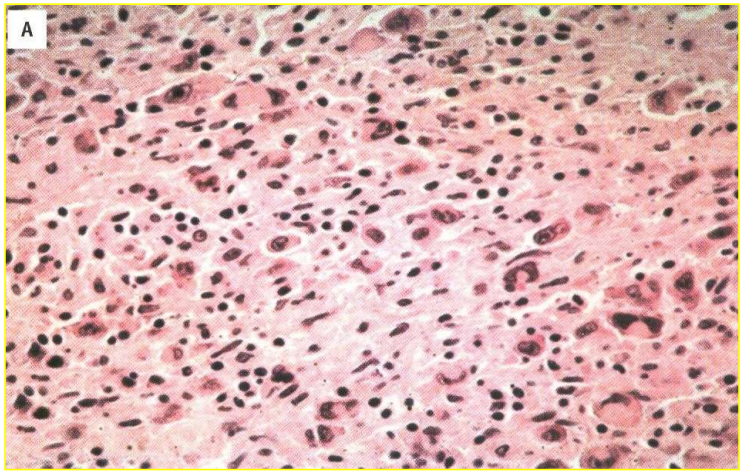
CD30

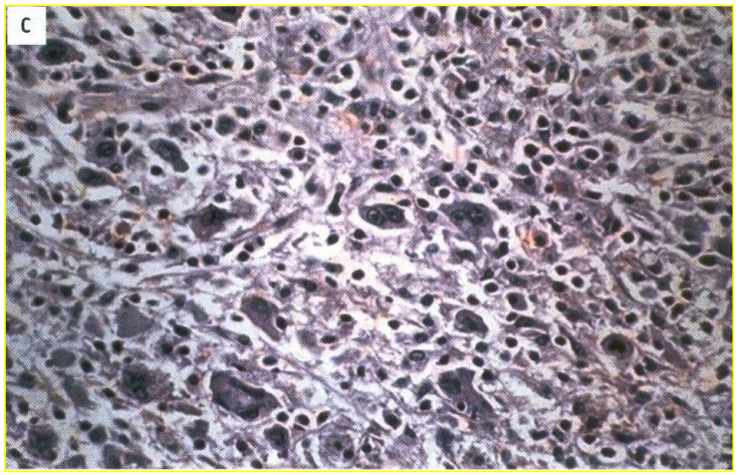
Lymphocyte depleted CHL

- **Definition: abundant HRS cells with diffuse growth pattern with or without reduction in the numbers of small-sized lymphocytes**
- **M > F**
- **Median age: 37 yrs**
- **Clinical features: peripheral-abdominal lymphadenopathy (often stage III/IV), often with B symptoms**
- **Prognosis: usually poor**
- **HIV infection → EBV+**

Lymphocyte depleted CHL - morphology

- Increased numbers of HRS cells in relation to the number of small lymphocytes
- Morphologically similar to mixed cellularity cellular composition with ↑↑ HRS cells
- Very pleomorphic neoplastic cells resembling anaplastic T-cell lymphoma
- OR relatively small number of HRS cells in cellular or hypocellular fibrous background





Nodular lymphocyte predominant Hodgkin lymphoma (NLPHL)

Definitions

- **B-cell neoplasm characterized by nodular ± diffuse proliferation of large neoplastic lymphocyte-predominant (LP) cells associated with small lymphocytes and histiocytes**
  - **Neoplastic cells are designated as LP cells**
    - a.k.a. **popcorn cells** because of their hyperlobated nuclei with vesicular chromatin
    - Formerly called lymphocytic &/or histiocytic (L&H) cells
  - **LP cells reside mostly within follicular dendritic cell meshworks**
  - **Background is composed of reactive small lymphocytes and histiocytes**
    - Inflammatory cells greatly outnumber neoplastic LP cells
    - Eosinophils and neutrophils are usually absent; plasma cells few
- **Diffuse form of NLPHL**
  - Term derived from Lukes and Butler classification
  - Most cases in this category have been reclassified as
    - T-cell/histiocyte-rich large B-cell lymphoma (THRLBCL)-like transformation of NLPHL
    - NLPHL with diffuse THRLBCL-like areas
    - Classic Hodgkin lymphoma (CHL)
  - True cases of NLPHL with diffuse pattern likely exist but are truly rare

Postulated Normal Counterpart

- **Germinal center B lymphocyte at centroblast stage of differentiation**

Associated Lesions

- **NLPHL is often associated with**
  - **Progressive transformation of germinal centers (PTGC)**
  - **Reactive follicular hyperplasia**

Diagnostic Pathology: Lymph Nodes and Extranodal Lymphomas. Medeiros, Miranda. Elsevier, 2023

Nodular lymphocyte predominant Hodgkin lymphoma

CLINICAL ISSUES

Epidemiology

- **Incidence**
  - 0.1-0.2 per 100,000 per year
  - 5-6% of all HLs
- **Age**
  - Median: 35 years
  - All age groups affected
- **Sex**
  - Male predominance
    - M:F = > 3:1

Site

- **Lymph nodes**
- Most commonly affected groups include cervical, axillary, or inguinal lymph nodes
  - Paraaortic and iliac lymph nodes less often involved
- **Liver &/or spleen involved in ~ 10% of cases**
- **Mediastinum involved in ~ 7% of cases**
- **Bone marrow rarely involved (~ 2%)**
  - Usually evidence of transformation to large B-cell lymphoma (LBCL)

Presentation

- **Peripheral lymphadenopathy**
  - Stage I or II in ~ 80% of patients
- **B symptoms uncommon (~ 10%)**
- **20% of patients present with advanced stage disease**
  - Involvement of spleen and liver
  - Abdominal lymphadenopathy
  - B symptoms often (+)

Laboratory Tests

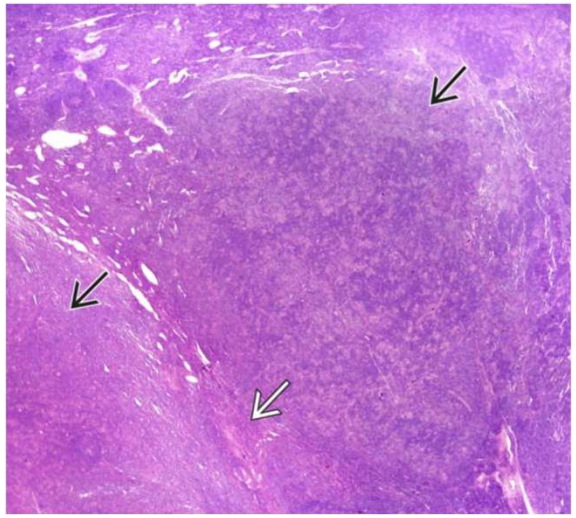
- Normal complete blood count; no leukemic phase
- Usually unremarkable chemistry panel
  - Serum LDH or β-2-microglobulin levels elevated in patients with high-stage disease

Diagnostic Pathology: Lymph Nodes and Extranodal Lymphomas. Medeiros, Miranda. Elsevier, 2023

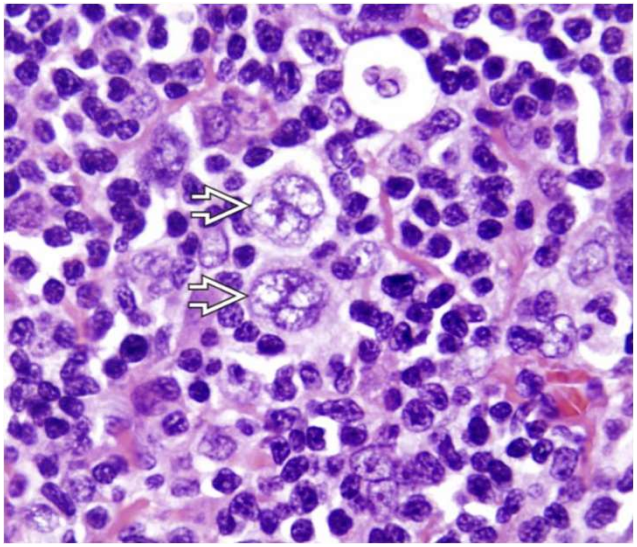
Nodular lymphocyte predominant Hodgkin lymphoma

Morphology

- **Nodular pattern pure or predominant in ~ 75% cases**
- Few LP cells in sea of small lymphocytes and histiocytes
    - Mixture imparts moth-eaten pattern
- LP cells have vesicular chromatin, small nucleoli, and thin nuclear membranes
- **No necrosis or thick, fibrous bands**



The nodal architecture is distorted by multiple expansile nodules with compressed interfollicular zones . The nodules have a moth-eaten low-power appearance.



The large neoplastic cells, known as lymphocyte-predominant (LP) cells , often have multilobated nuclear contours and resemble popcorn.

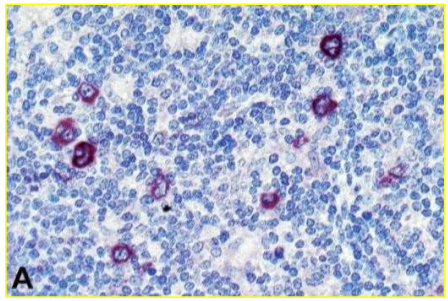
Diagnostic Pathology: Lymph Nodes and Extranodal Lymphomas. Medeiros, Miranda. Elsevier, 2023

Nodular lymphocyte predominant Hodgkin lymphoma

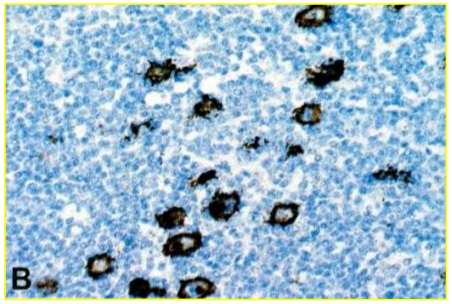
Immunophenotype-1

LP cells

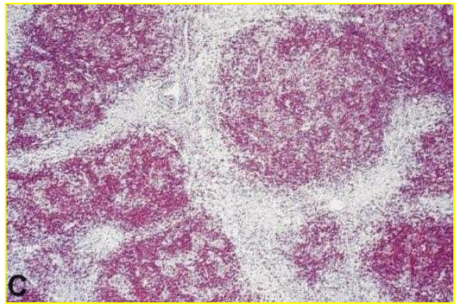
Positive markers	Negative markers
B-cell markers (CD20,CD79a,PAX-5)	CD30
BCL-6	CD15
CD45	
OCT-2	
BOB-1	
EMA	
J chain	
Heavy/light chain markers	

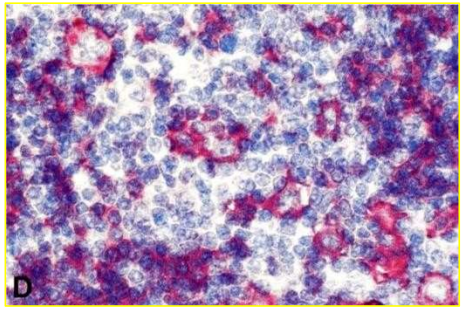


J chain



EMA





CD20

Nodular lymphocyte predominant Hodgkin lymphoma

Immunophenotype-2

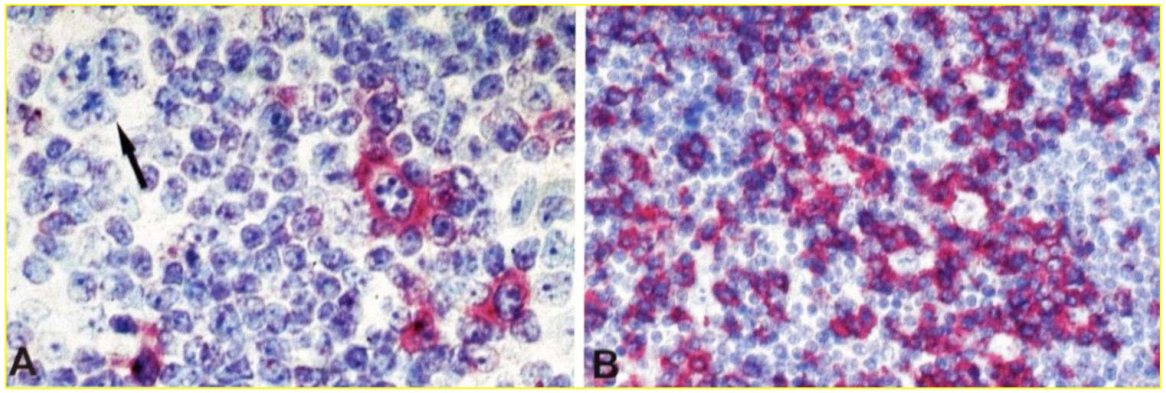
Small-sized lymphoid cells

↓ ↓

B **T (CD4+/CD57+)**

PD1, MUM-1+ ⇨ T follicular helper (TFH) cells

» » ↑ T-cells in diffuse areas



CD30 **CD57**

NLPHL with TRBCL-like areas and primary TCRBCL – Differential Diagnosis

Presence of small-sized B-cells

Presence of CD4+/CD57+ T-cells

Absence of CD8+/TIA-1+ T-cells

Nodular pattern, at least focally

} **NLPHL with TRBCL-like areas**

Nodular lymphocyte predominant Hodgkin lymphoma

Natural History - Prognosis

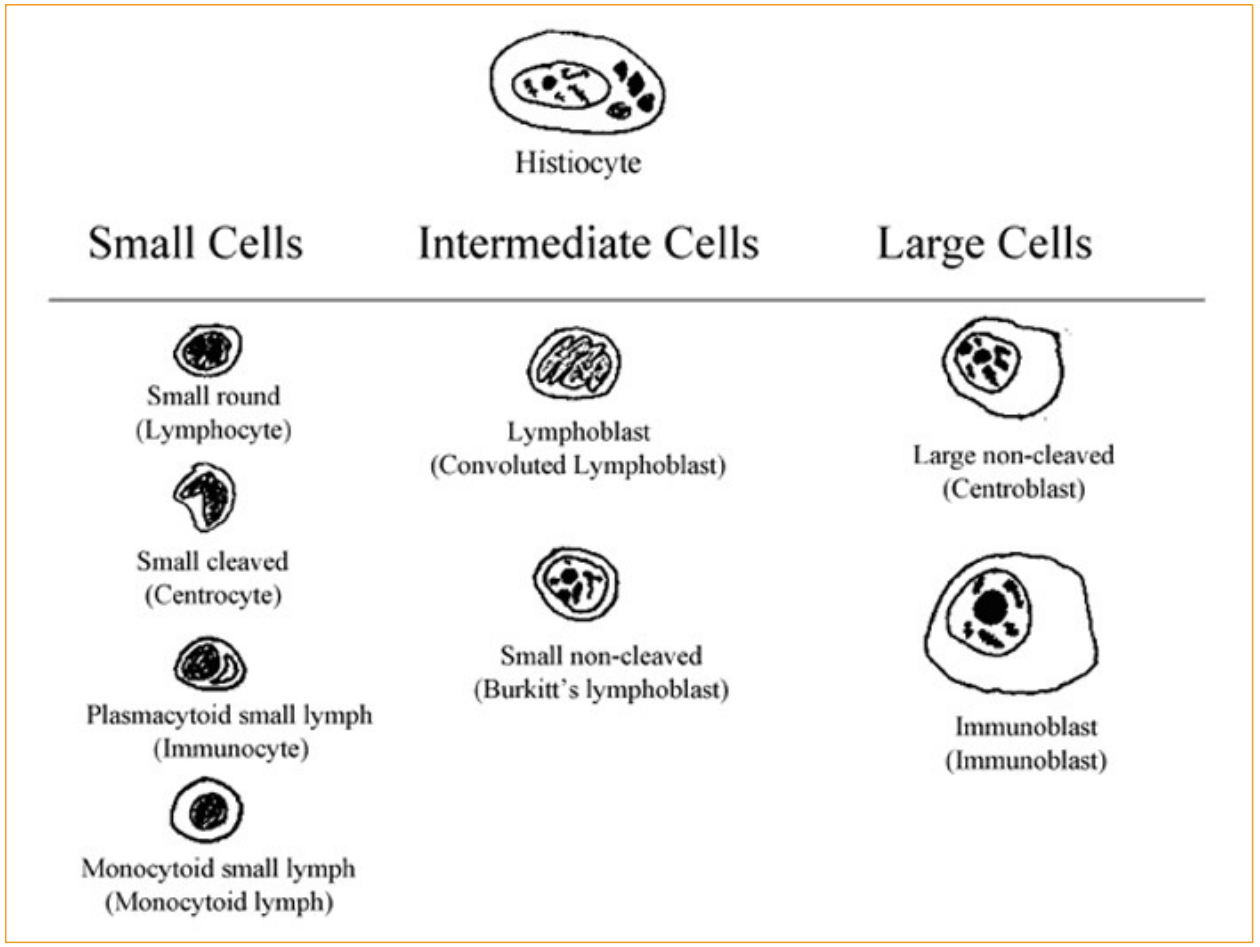
- **Clinically indolent disease with frequent relapses**
- **Relapse-free survival curves show staircase pattern**
    - **No plateau suggestive of cure**
    - Early and late (> 10 years) relapses occur
    - Risk of relapse independent of stage or therapy
- ~ 10-15% of NLPHL transform to aggressive B cell lymphoma
- With prolonged follow-up, ~ 15% of patients die
    - Deaths related to disease refractory to therapy or 2nd malignancies
    - 2nd malignancies represent ~ 4% of all deaths
        - Acute leukemia (2%)
        - Non-HL (1%)
        - Solid organ tumors (1%)

Diagnostic Pathology: Lymph Nodes and Extranodal Lymphomas. Medeiros, Miranda. Elsevier, 2023

Small B cell lymphomas

- **CHRONIC LYMPHOCYTIC LEUKEMIA/SMALL LYMPHOCYTIC LYMPHOMA**
- **MANTLE CELL LYMPHOMA**
- **FOLLICULAR LYMPHOMA**
- **MARGINAL ZONE LYMPHOMA**
- **LYMPHOPLASMACYTIC LYMPHOMA**

B CELL LYMPHOMAS – CELL SIZE



CHRONIC LYMPHOCYTIC LEUKEMIA (CLL) / SMALL LYMPHOCYTIC LYMPHOMA

- Definition: Chronic lymphocytic leukaemia/small lymphocytic lymphoma (CLL/SLL) is a B-cell lymphoma comprising monomorphic small mature B-cells that frequently co-express CD5 and CD23. A peripheral blood diagnosis of CLL requires a B-cell count >5×10^9/L, with the characteristic morphology and immunophenotype. A tissue-based diagnosis of SLL requires organ enlargement (e.g., lymphadenopathy >15 mm) and its infiltration by the above neoplastic B-cells. Although CLL and SLL represent the same disease, the name SLL is used for cases with <5×10^9/L circulating B cells and nodal, splenic, or other extramedullary involvement.
- Common involvement of lymph nodes, liver and spleen
- Extranodal involvement, such as skin, GI, CNS uncommon

CHRONIC LYMPHOCYTIC LEUKEMIA (CLL) / SMALL LYMPHOCYTIC LYMPHOMA

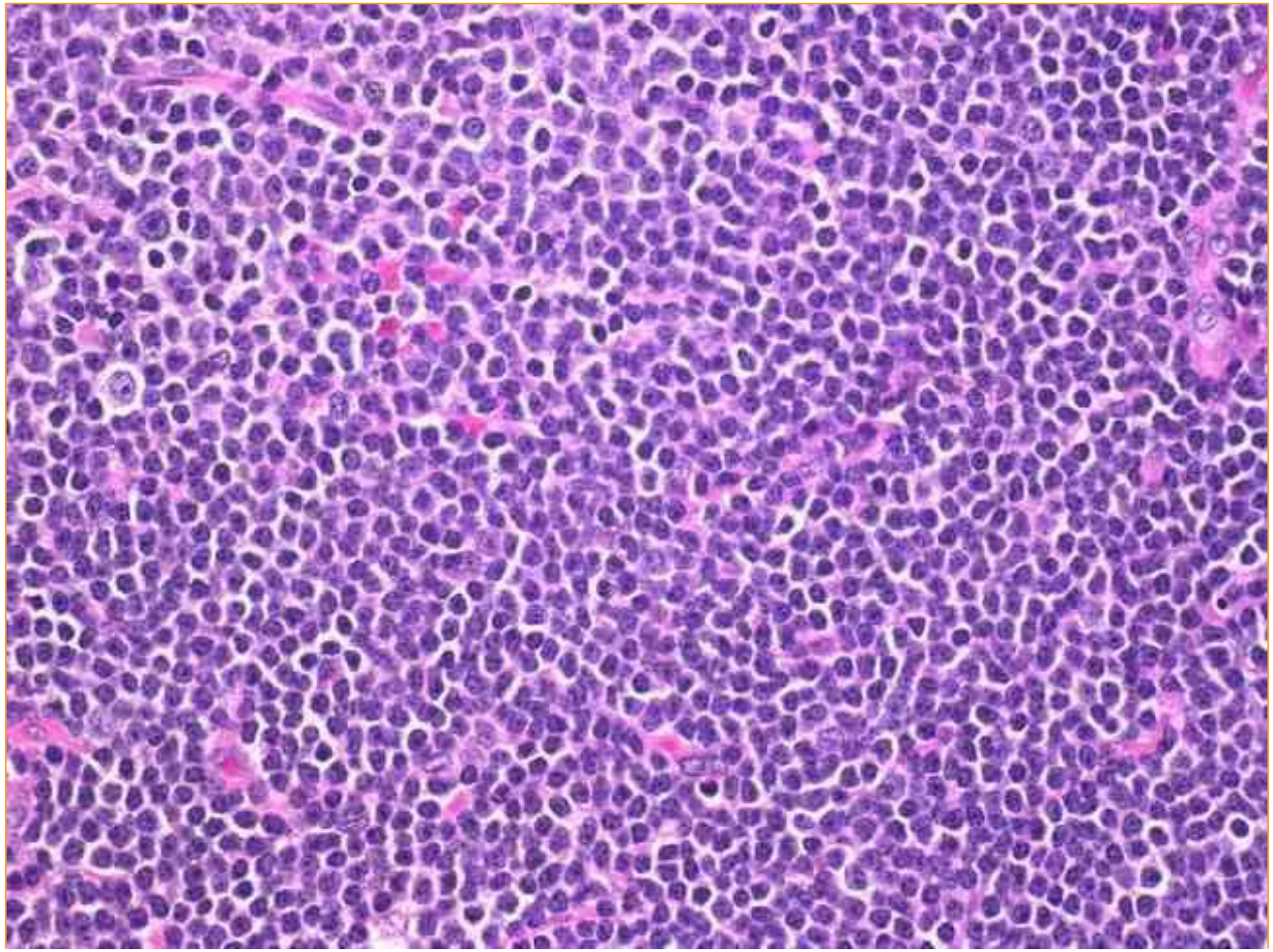
- CLL/SLL is most common in fair-skinned populations with an age-adjusted annual incidence of 4.9 per 100,000. It is much less common amongst Asian populations and in Latin Americans with the age-adjusted annual incidence of 0.1-0.5 and 0.5-1.4 per 100,000, respectively.
- M/F 1.5-2/1
- No symptoms at diagnosis at most patients
- Symptoms include autoimmune hemolytic anaemia, various infections, splenomegaly, hepatomegaly, lymphadenopathy

Morphology

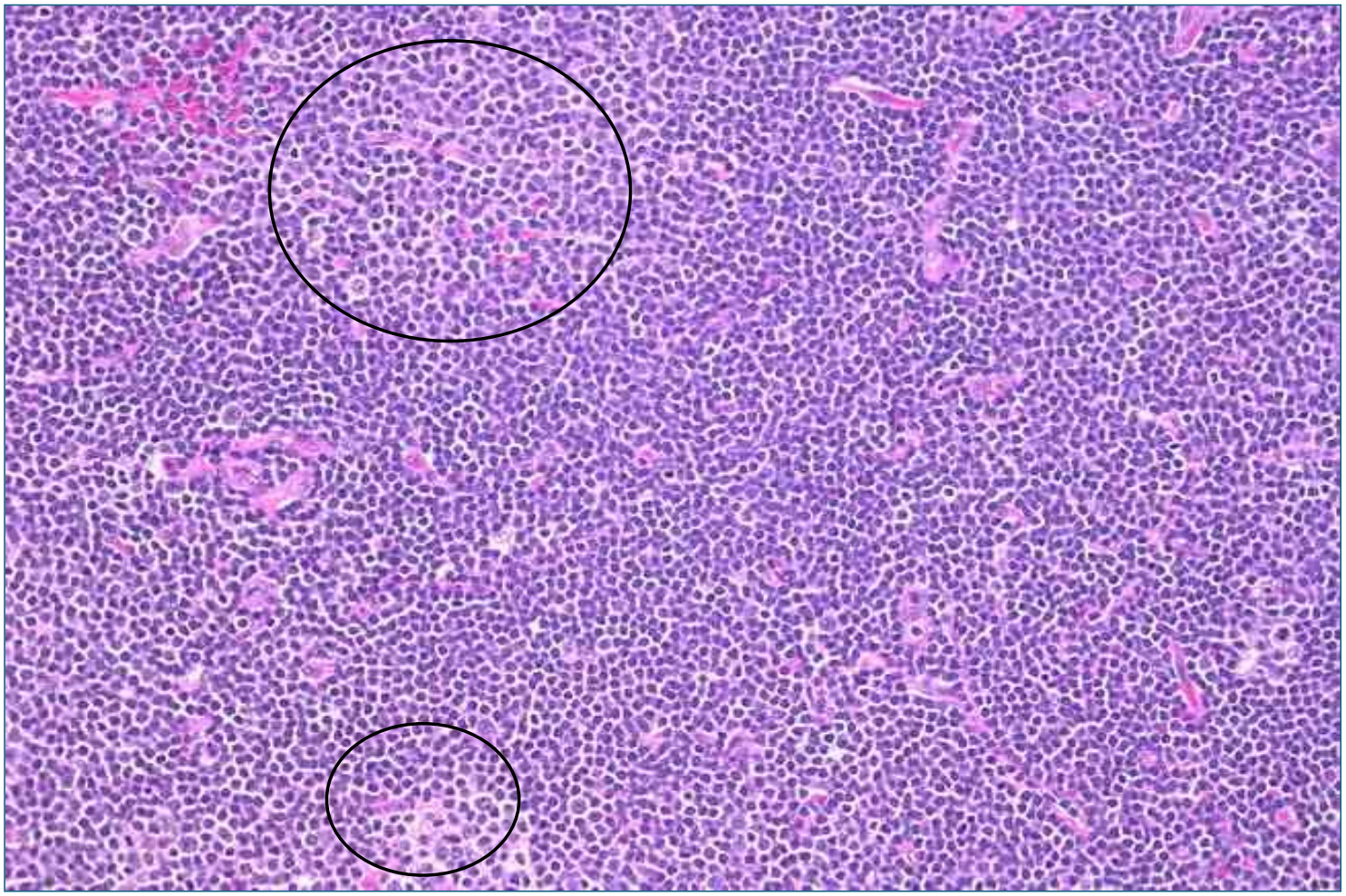
Lymph nodes-spleen

- Diffuse-vaguely nodular pattern
- pseudofollicles (*pale proliferation centers*): small/ broad (>20XPF)/absent
- Partial involvement, intrafollicular or perifollicular infiltration pattern
- Small lymphocytes
- Prolymphocytes: small-medium-sized cells with relatively clumped chromatin and small nucleoli (up to 55% of cells)
- Paraimmunoblasts: larger cells with round to oval nuclei, dispersed chromatin, central eosinophilic nucleoli, slightly basophilic cytoplasm
- Morphologic variations:
  - Cells with nuclear irregularity
  - Plasmacytoid differentiation (serum monoclonal protein)

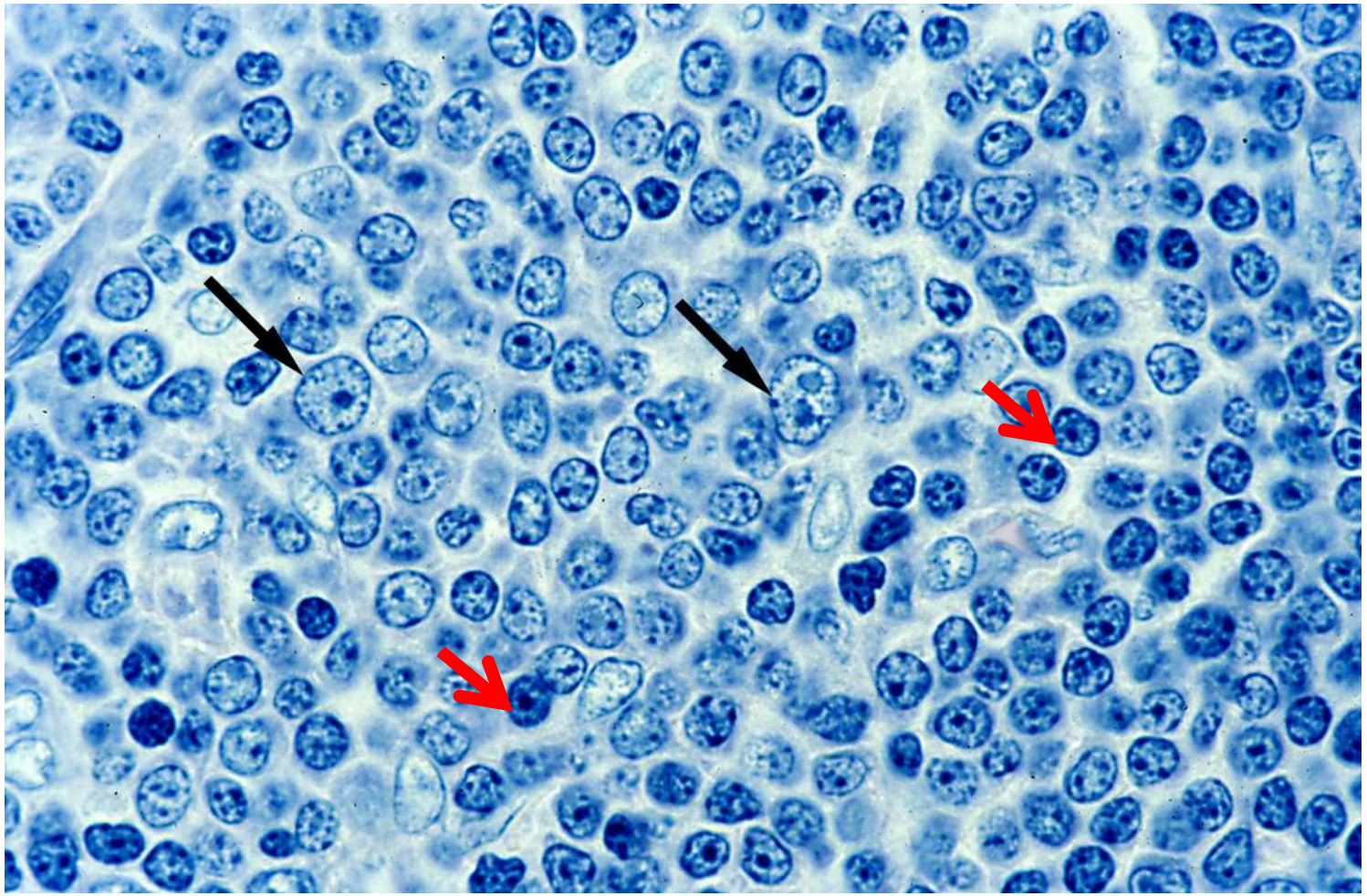
Morphology



Morphology - pseudofollicles

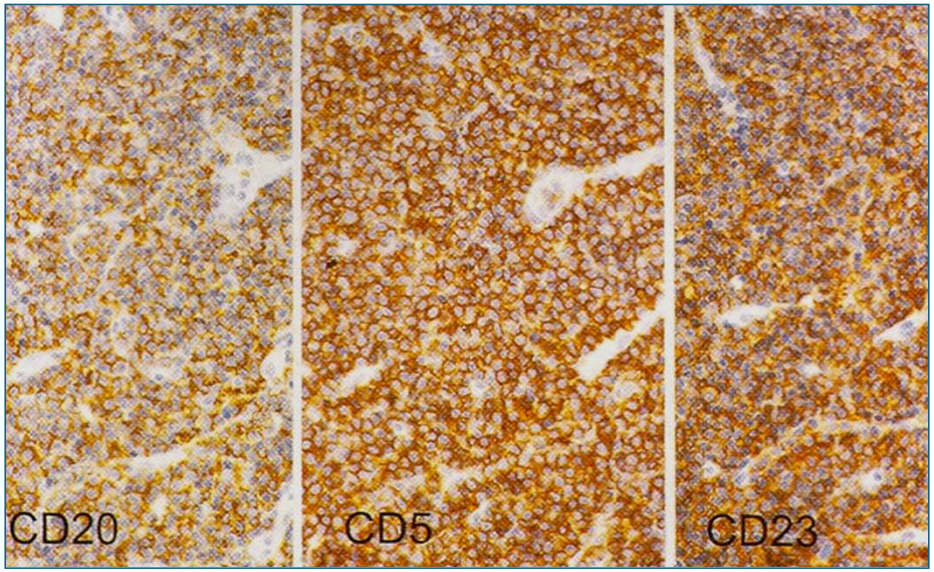


Prolymphocytes, paraimmunoblasts



Immunophenotype

✓CD20
✓CD79a
✓CD19
✓CD5
✓CD43
✓CD23
✓LEF1 (lymphoid enhancing factor-1): 100%



✓CyclinD1, MUM1 και cMyc + in proliferation centers

Genetic profile – IgHV mutation

- 40-50% unmutated cases
- 50-60% mutated cases (favourable prognosis)

	IGHV unmutated	IGHV mutated
Stage at diagnosis	Intermediate	Low
Age	Similar	
Sex	M>F	M=F
Time to cell count duplication	<12 months	>12 months
IgV mutations	No or low numbers	Numerous
ZAP-70 expression	High	Low
CD38 expression	High	Low
Chromosomal aberrations	Indicating poor prognosis	Indicating favourable prognosis

MANTLE CELL LYMPHOMA

- ✓ **Definition: Mantle cell lymphoma (MCL) is a mature B-cell neoplasm derived from the mantle zone of lymphoid follicles, and typically composed of small to medium-sized monomorphic cells expressing CD5, SOX11 and cyclin D1. It is associated with CCND family rearrangements, most commonly CCND1.**
- ✓ 3-10% of non-Hodgkin lymphomas
- ✓ Median age of occurrence is 68-69 years
- ✓ M>F (3-4:1)
- ✓ Median survival 3-5 yrs

- ✓ Usually stage III/IV at diagnosis
- ✓ Common lymph node, spleen and bone marrow involvement (+/- leukemic presentation in 25% of cases)
- ✓ Frequent extranodal involvement sites: GI (***lymphomatous polyposis***), Waldeyer ring

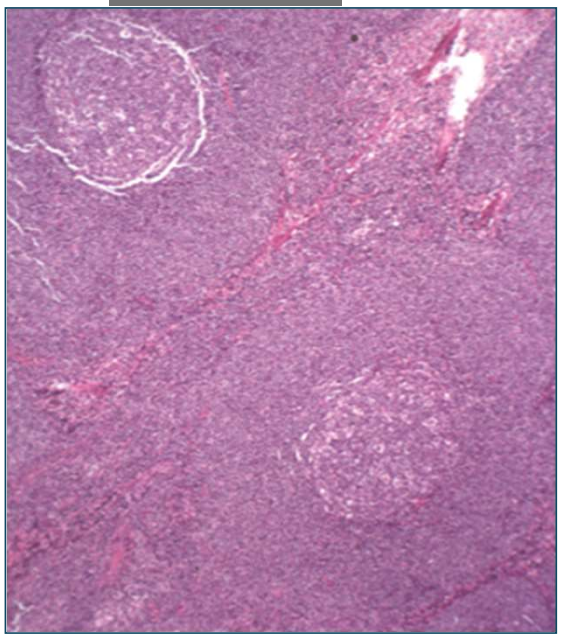
Morphology

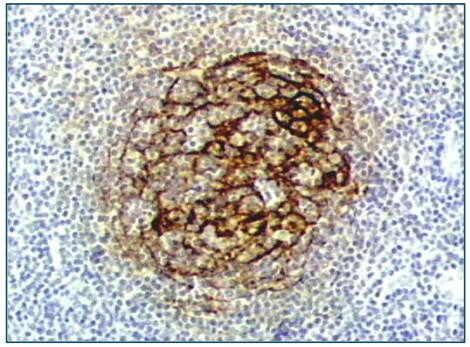
- ✓ Infiltration pattern: mantle zone, vaguely nodular, diffuse
- ✓ Monomorphic small/medium-sized cells with irregular nuclei with inconspicuous nucleoli (**classic morhology**), small amount of cytoplasm
- ✓ Hyalinized small vessels common
- ✓ Scattered epithelioid histiocytes
- ✓ Variants:
  - ✓ **blastoid** (resembling lymphoblastic lymphoma – ↑↑ mitoses- ≥ 20-30/HPF)
  - ✓ **pleomorphic** (large cells with oval to irregular nuclear contours, pale cytoplasm and prominent nucleoli)
  - ✓ **small cell** (small lymphocytes CLL-like)

Aggressive behaviour

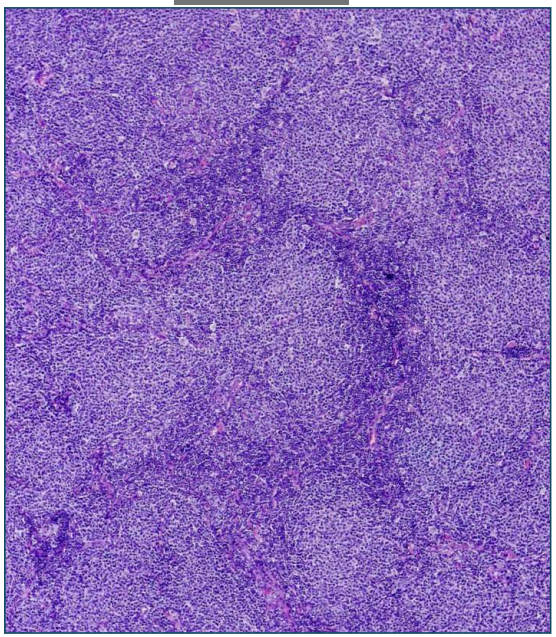
MANTLE CELL LYMPHOMA – growth patterns

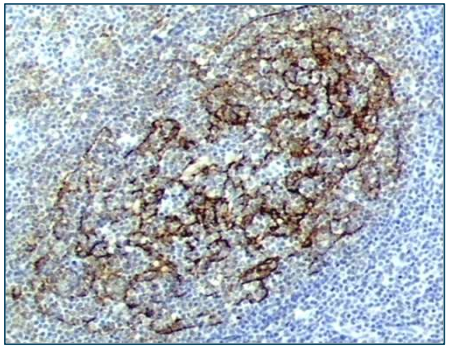
Mantle zone



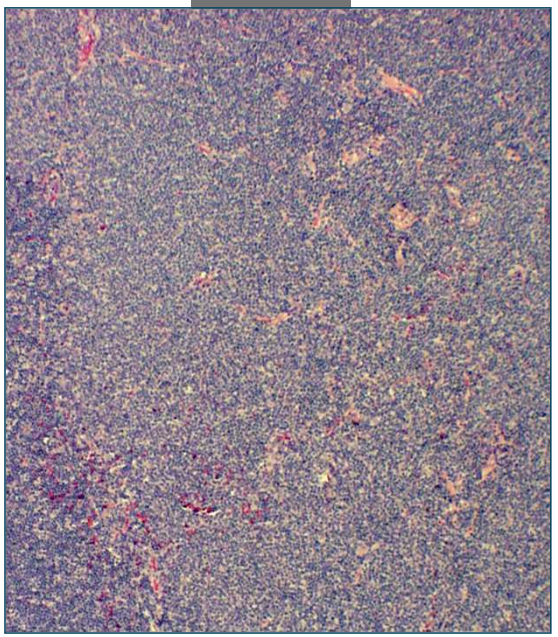


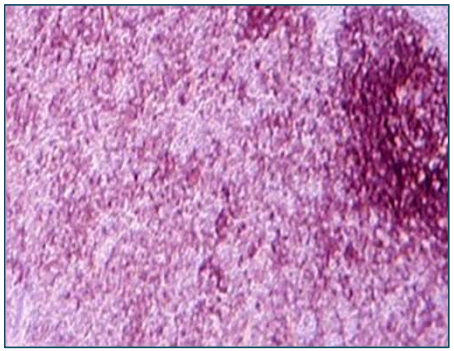
Nodular





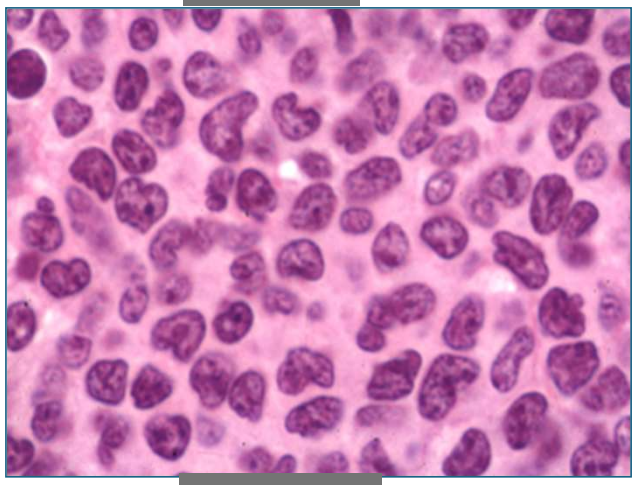
Diffuse



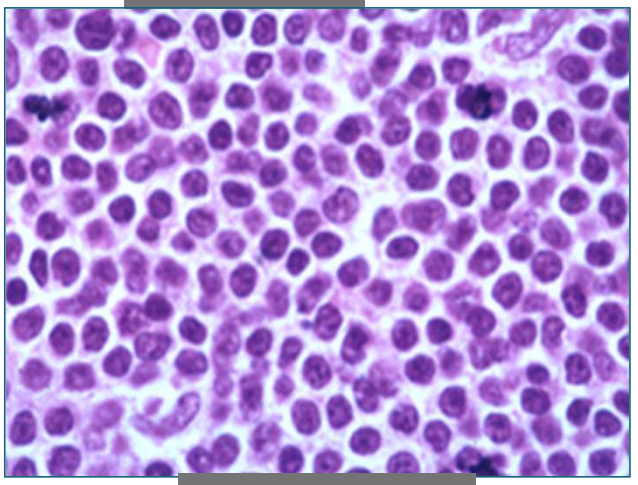


MANTLE CELL LYMPHOMA – cellular morphology

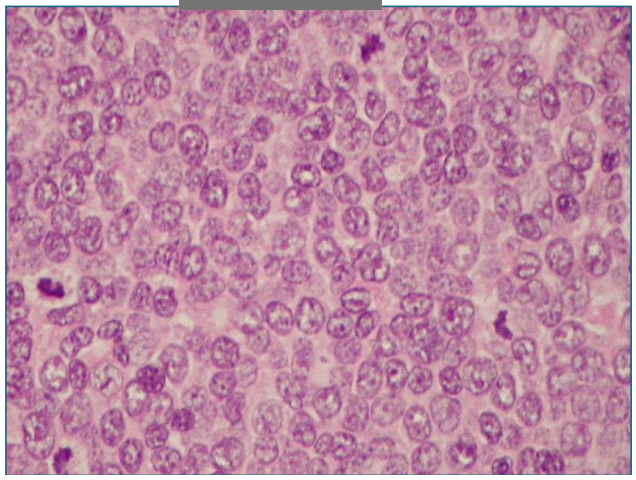
Classic



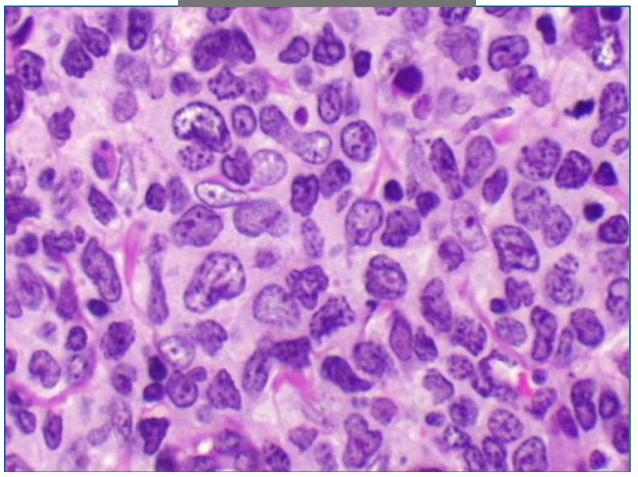
Small cell



Blastoid

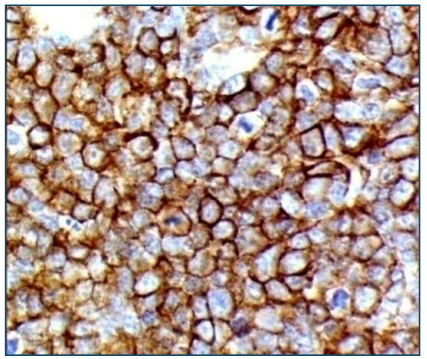


Pleomorphic

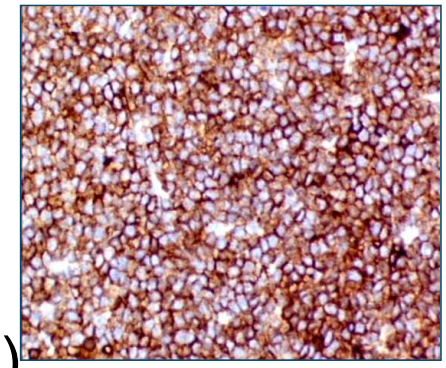


Immunophenotype

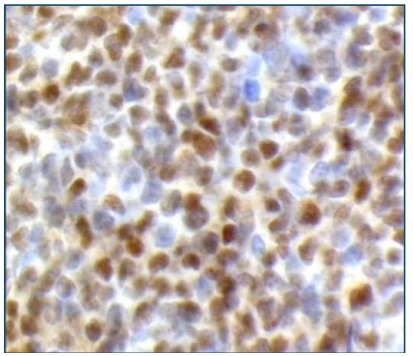
- CD20+/CD3-
- CD5, CD43+ (common)
- CD23- (common)
- Bcl-6-
- CD10- (usually)
- FMC-7+
- Bcl-2+
- IgM/IgD+ (intense expression) (λ>κ)
- cyclinD1+ (95%) **[t(11;14)]**
- SOX-11+ (85-90%)



CD20



CD5



Cyclin D1

MCL – MOLECULAR BIOLOGY

- Translocation **t(11;14) (IGH@ and cyclinD1)** as molecular hallmark (overcoming suppressor effects on G1/S cell cycle transition, favouring antiapoptotic bcl2 function)

- ***SOX11 overexpression:*** enhanced PI3K/AKT signaling

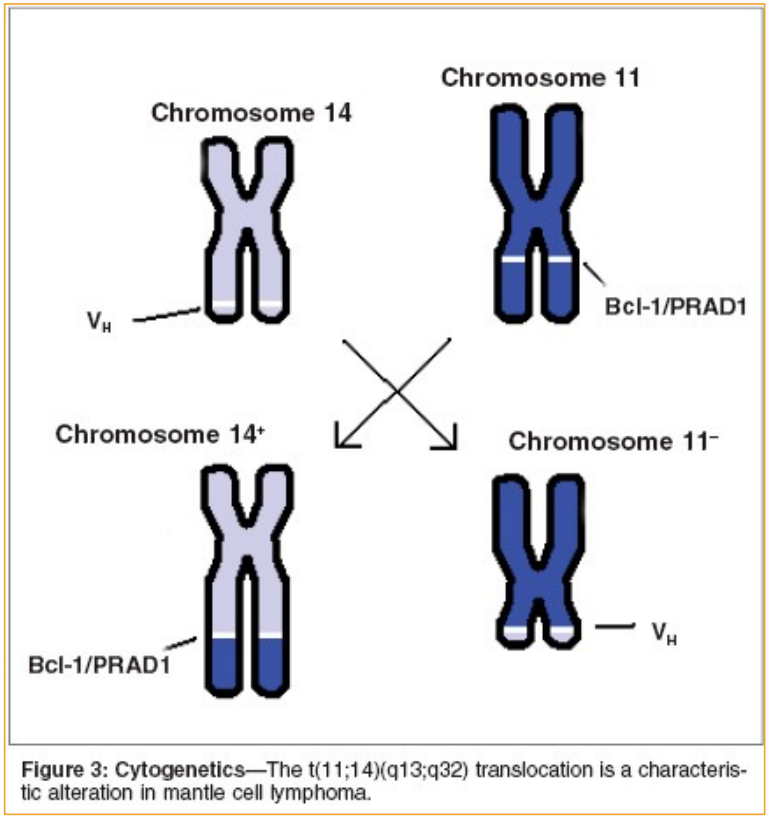


Figure 3: Cytogenetics—The t(11;14)(q13;q32) translocation is a characteristic alteration in mantle cell lymphoma.

Leukemic non-nodal mantle cell lymphoma

- Indolent clinical course, not requiring therapy for long time periods
- Clinical presentation: peripheral blood, bone marrow, often splenic involvement
- No significant lymphadenopathy
- Few abnormalities other than t(11;14)
- **SOX-11-**

FOLLICULAR LYMPHOMA

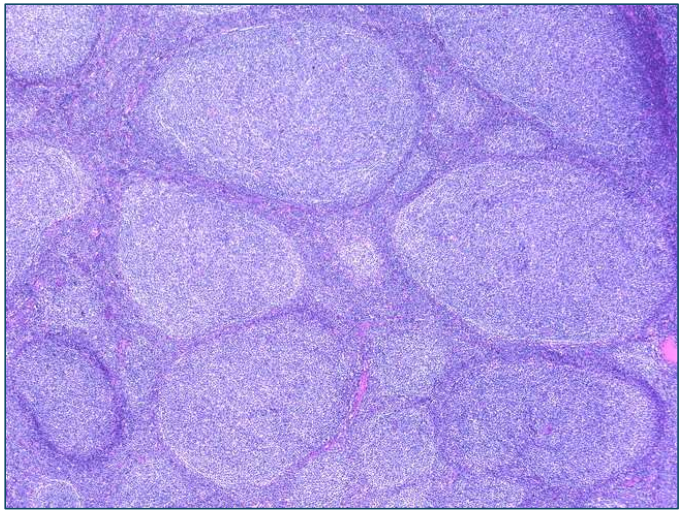
- Definition: Follicular lymphoma (FL) is a neoplasm of germinal centre (GC) B-cells with varying proportions of centrocytes (CC) and centroblasts (CB) or large transformed cells and at least a partially follicular growth pattern. In rare cases with an entirely diffuse growth pattern, the neoplastic cells should still show GC B-cell morphology and immunophenotype.

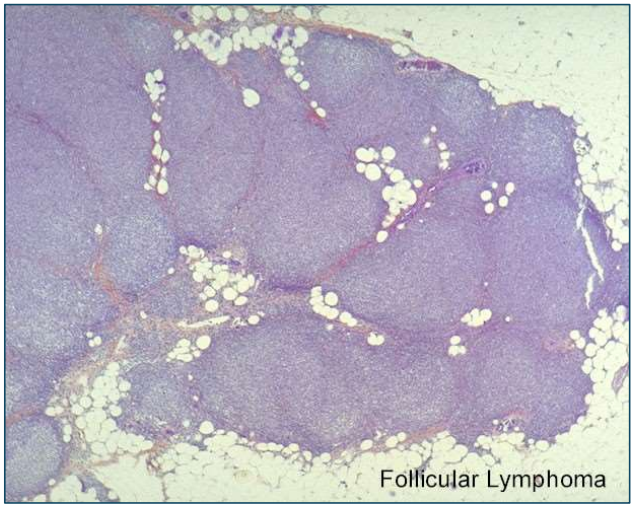
- 10-20% of all lymphomas
- Highest incidence in USA and Western Europe
- Median age: 6th decade of life
- M/F=1/1.7
- Prognosis closely related to the extend of disease at diagnosis
- Paediatric type usually in boys

Localization – Clinical Presentation

- Mainly lymph node involvement
- Common spleen, bone marrow, blood and Waldeyer ring involvement
- May extend to GI or soft tissue in high stage disease
- Common primary extranodal sites: GI (duodenum), ocular adnexa, breast, testis
- Usually widespread disease at diagnosis, with bone marrow involvement in 40-70% of cases
- Only 10-15% stage I/II at diagnosis
- Usually no symptoms

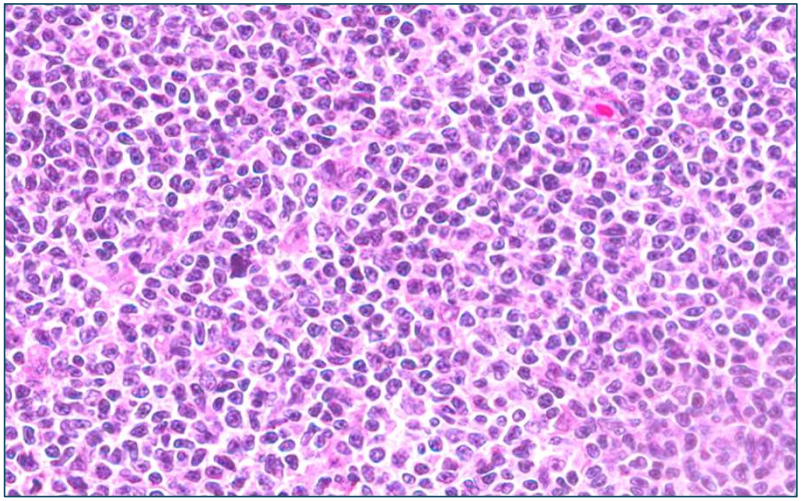
Morphology



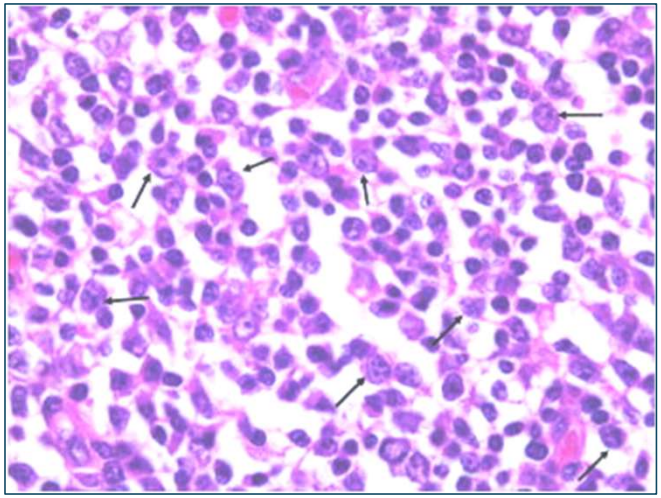


- Lymph node effacement with **at least partially follicular pattern** (CD21/CD23+ FDCs), which should be mentioned in the report
  - Follicular pattern (>75%)
  - Follicular and diffuse pattern (25-75%)
  - Focally follicular/predominantly diffuse pattern (<25%)
- Closely packed follicles with attenuated/absent mantle zones
- Extension to perinodal tissue

Cellular Morphology



Centrocytes
Small to medium-sized cells
Angulated, elongated, twisted or cleaved nuclei
Inconspicuous nucleoli
Scant cytoplasm



Centroblasts
Large round cells (X3 the size of a lymphocyte)
Round or oval nuclei
1-3 conspicuous nucleoli
Scant cytoplasm

Also cases with monocytoid morphology- plasmacytic differentiation

Grading

Table II. FL grading according to updated 2017 4th WHO classification.

Grade	Proportion of centroblasts	Percentage
Grade 1-2 [Low grade FL]	0-15 centroblasts/HPF	80-90%
Grade 3A	> 15 centroblast /HPF (centrocytes present)	10-20%
Grade 3B	> 15 centroblasts/HPF (follicles entirely composed of large centroblasts)	Rare

Fratoni et al, 2020

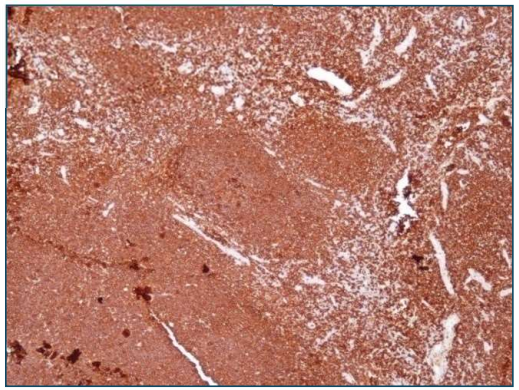
- *If grade 3 και 1/2 occur in different areas, the approximate percentages should be reported*
- **If a diffuse component is present with >15 centroblasts/HPF, an additional diagnosis of DLBCL** should be warranted

- **Limitations:** population heterogeneity, distinction between large centrocytes or small centroblasts, poor tissue handling and fixation

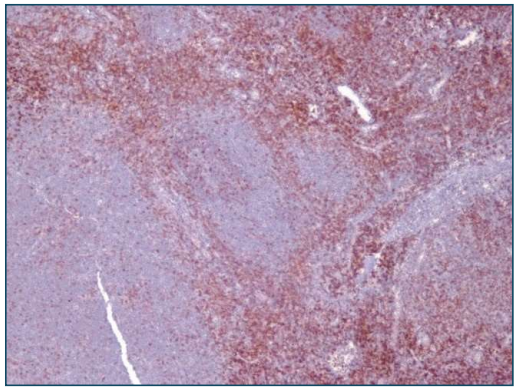
Immunophenotype

- **CD20, CD79a**, CD22, CD19
- **BCl-2 (+)**
- **BCL-6 (+)**
- **CD10 (+)**
- **LMO2 (+)**
- **HGAL (+)**
- **MEF2 (+)**
- **GCET1 (+)**
- **CD 21/CD23 meshwork** → **follicular dendritic cell**
- **Ki 67: grade 1/2 usually <20%**
  grade 3 usually> 20%

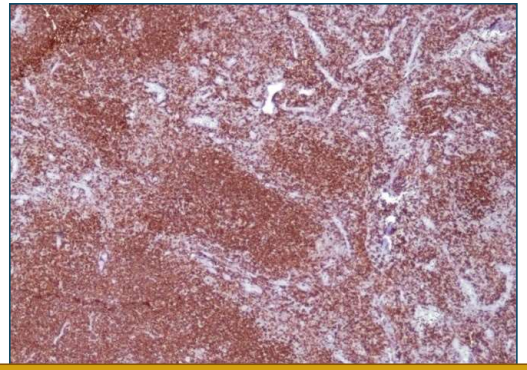
CD20



CD3



Bcl-2



Quintanilla-Fend, Jaffe, EAHP 2021

FOLLICULAR LYMPHOMA – WHO 2022 subtypes

- **Classic follicular lymphoma, cFL**
- **Follicular Large B cell Lymphoma-FLBCL (WHO 2022)/ Follicular lymphoma grade 3B (ICC2022)**
- **Follicular lymphoma with unusual features (blastoid chromatin, large cells with cleaved nuclei)**

FOLLICULAR LYMPHOMA – Molecular profile

- Follicular lymphoma with BCL-2 translocation **(BCL-2-R)**
- Follicular lymphoma with BCL-6 translocation **(BCL-6-R)**
- Follicular lymphoma bearing neither **BCL-2-R** nor **BCL-6-R**

FOLLICULAR LYMPHOMA – Molecular biology

- 85-90% of cFL cases show t(14;18) ➔ bcl-2 protein overexpression
- t(14;18) does not suffice to diagnose cFL – it may occur to healthy individuals
- BCL-6-R occurs often [especially in cases not showing t(14;18)]
- in cases not showing t(14-;18): p53 mutations important in lymphomagenesis

MARGINAL ZONE LYMPHOMA (MZL)

- Extranodal MZL (MALT type)
- Nodal MZL
- Splenic MZL

NODAL MARGINAL ZONE LYMPHOMA (NMZL)

- Nodal localization, morphology of MALT or splenic type, *no previous or concurrent occurrence of extranodal or splenic MZL*

- Epidemiology

➢6 per 1,000,000 person-years.

➢Median age 60 years

➢M=F

➢cases related to HCV infection

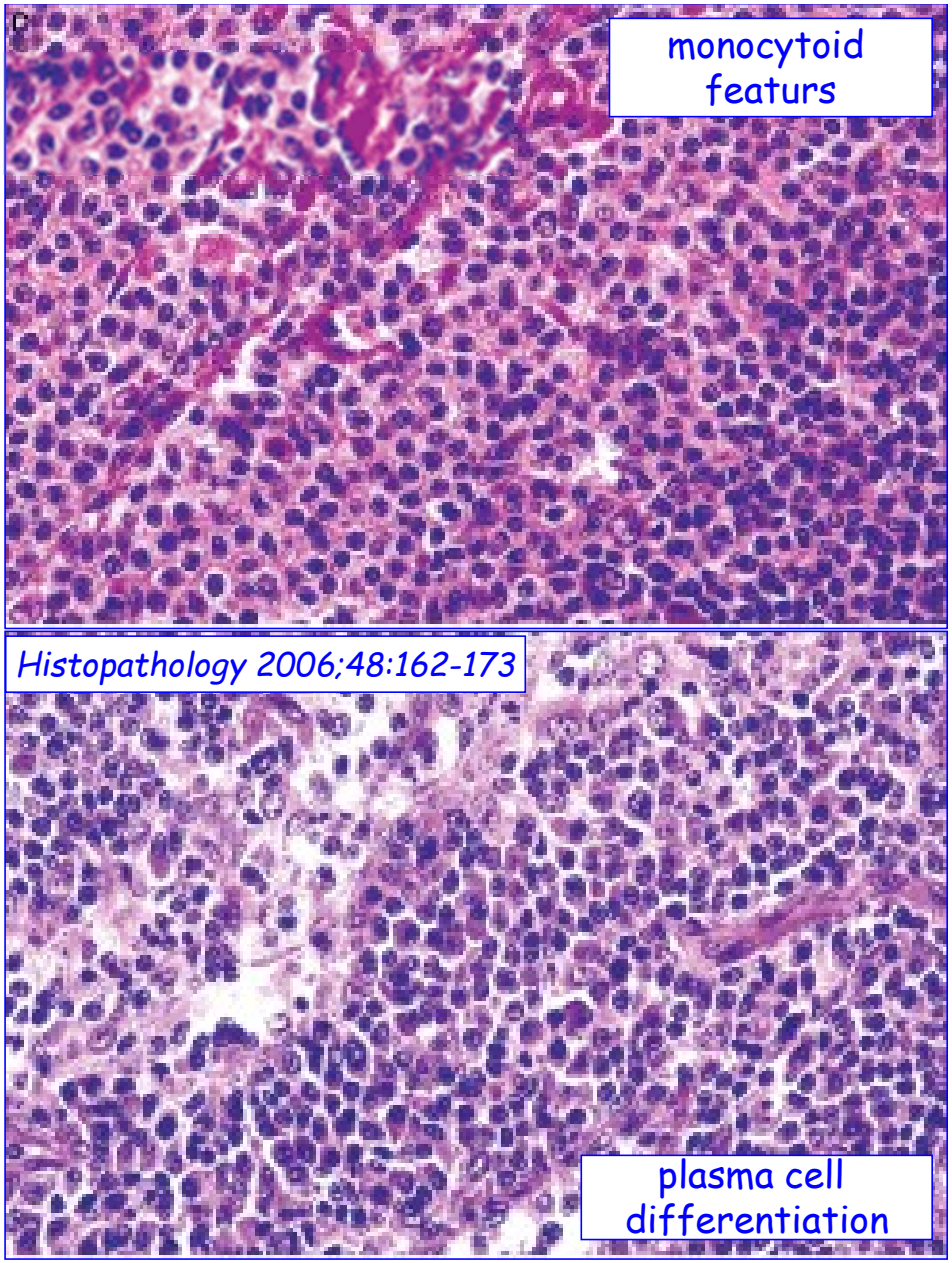
Lack of distinct morphologic or phenotypic features makes its diagnosis challenging

Morphology

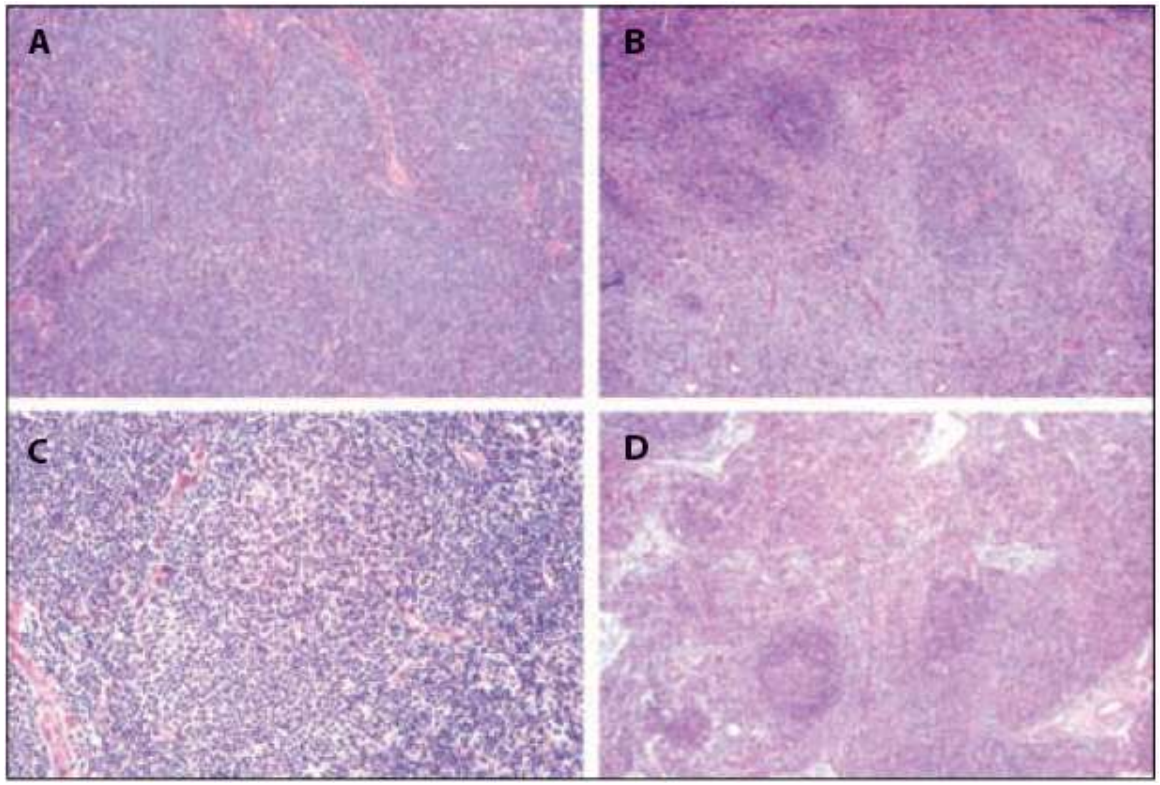
Polymorphous infiltrate consisting of:

-small sized B cells with monocytoid and/or centrocytoid morphology and/or plasma cell differentiation

-fewer large cells (immunoblasts) *(<10%)*



Morphology



A. Vaguely nodular pattern
B. Intrafollicular pattern
C. Perifollicular pattern
D. Inverse follicular pattern

EXTRANODAL MARGINAL ZONE LYMPHOMA (MALT) – General features

- Occurence and relapses in MALT, indigenous of acquired
- Tendency to remain localized for long periods of time
- Recapitulation of Peyer plaques' anatomy
- Usually in autoimmune setting (44X tendency to occur in Sjogren syndrome and 70X in Hashimoto thyroiditis)
- Prodromal lesions: H.p. gastrits, Sjogren syndrome, Hashimoto thyroiditis, HCV hepatitis, follicular bronchiolitis, Borrellia burgdorferi infection (skin), Campylobacter jejuni infection (small bowel)
- Stage I/II commonly

EXTRANODAL MARGINAL ZONE LYMPHOMA (MALT) – Epidemiology

- 7-8% of B non-Hodgkin lymphomas, 50% of primary gastric lymphomas
- Usually in adults (median age 60 years)
- F>M slight predominance *(especially for MALT lymphoma of salivary glands)*

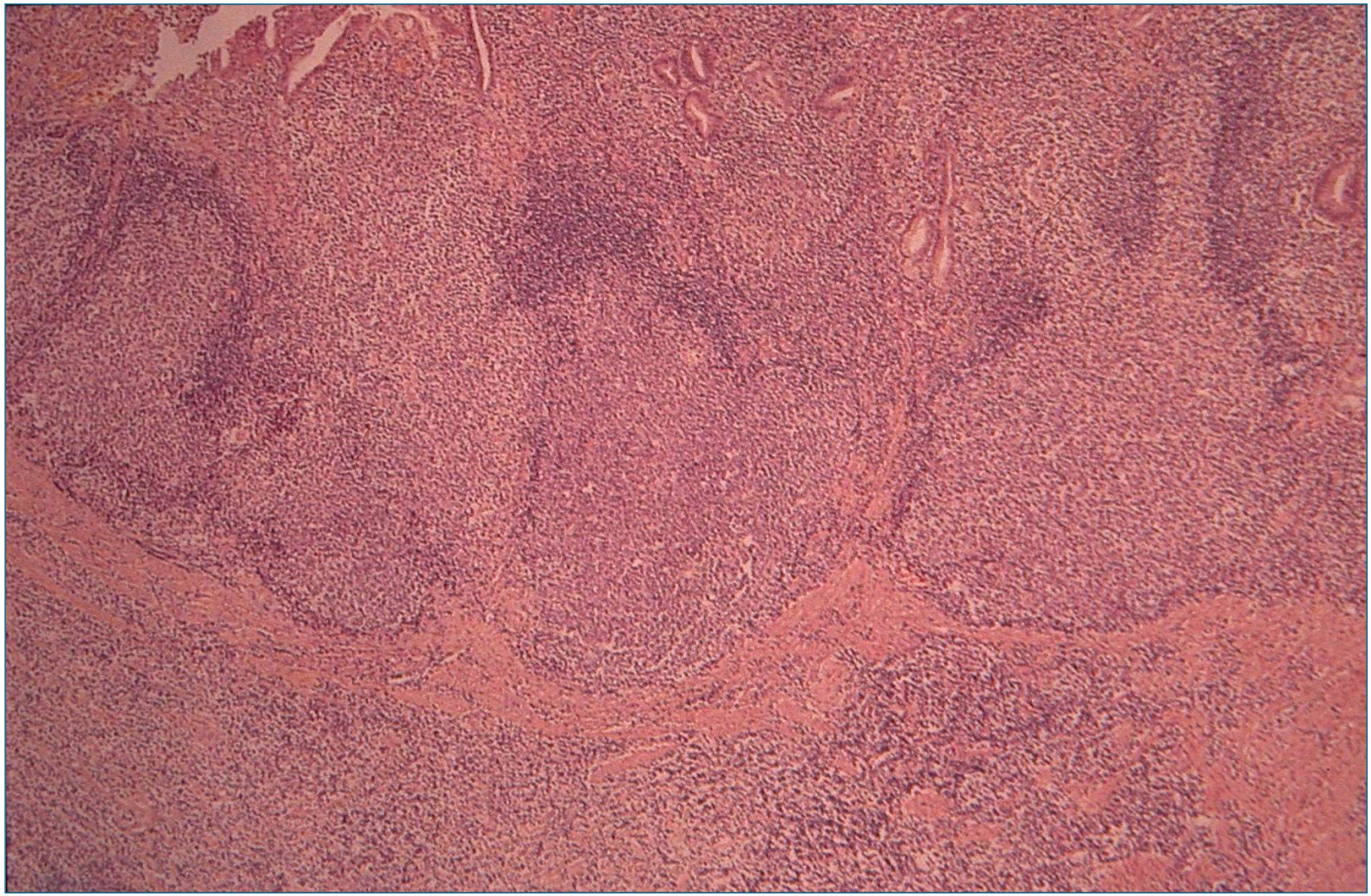
Localization

- ✓GI tract [stomach, intestine/ IPSID included)
- ✓Salival glands
- ✓Lungs, pharynx, bronchi
- ✓Eye, ocular adnexa
- ✓Thyroid
- ✓Skin
- ✓Liver
- ✓Urinary tract (urinary bladder, prostate, kidney)
- ✓Breast
- ✓Thymus

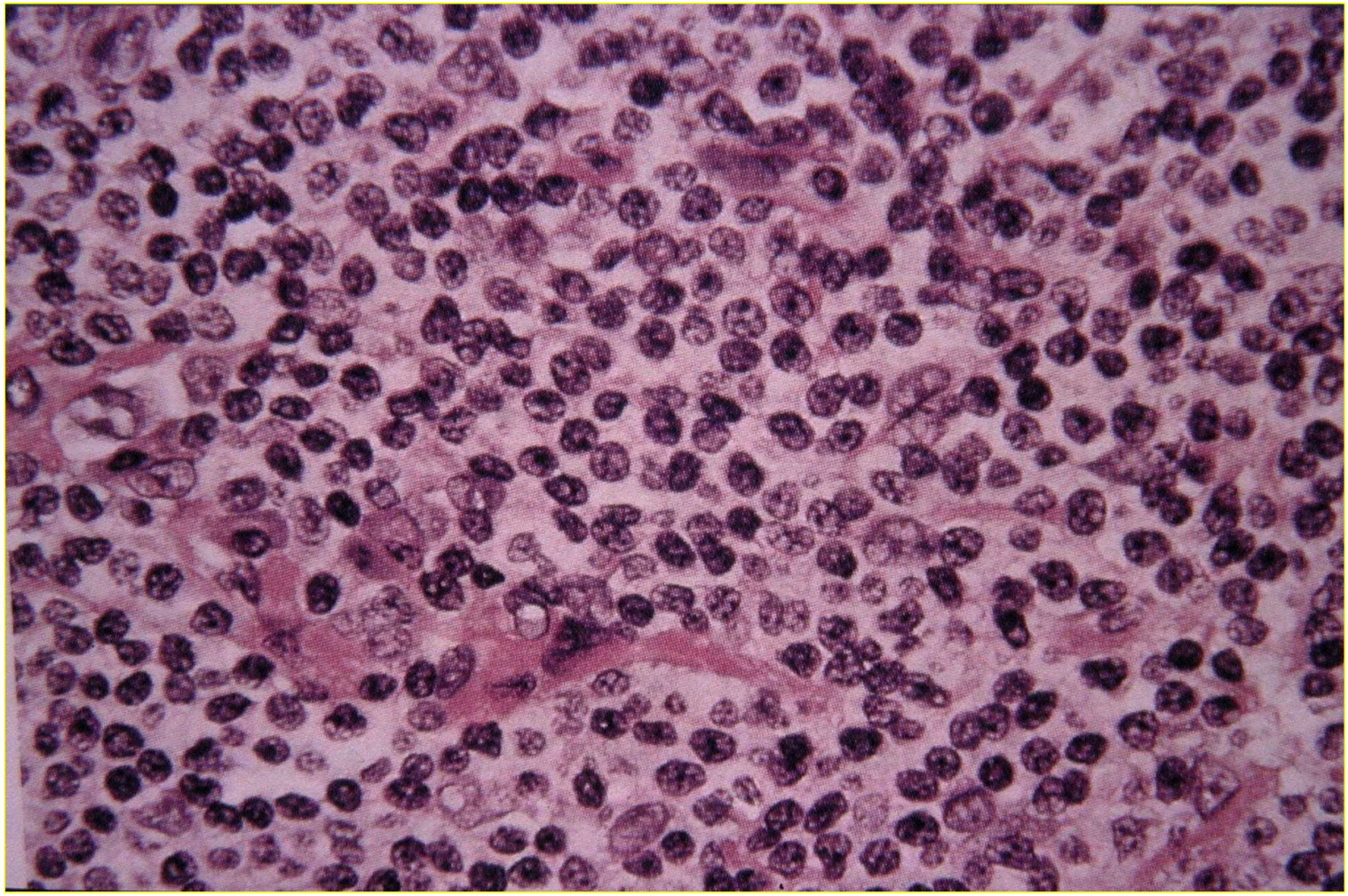
EXTRANODAL MARGINAL ZONE LYMPHOMA (MALT) – Morphology

- Polymorphic infiltrate: interstitial blastic cells (immunoblasts – centroblasts)
- Centrocytoid – monocytoid cells
- Infiltration of marginal zone and intrafollicular space
- Plasma cell differentiation (35%)
- Lymphoepithelial lesions
- Follicular colonization
- Polyclonal plasma cells

** when blastoid cells > 5-10% ↑ probability to progress to aggressive lymphoma

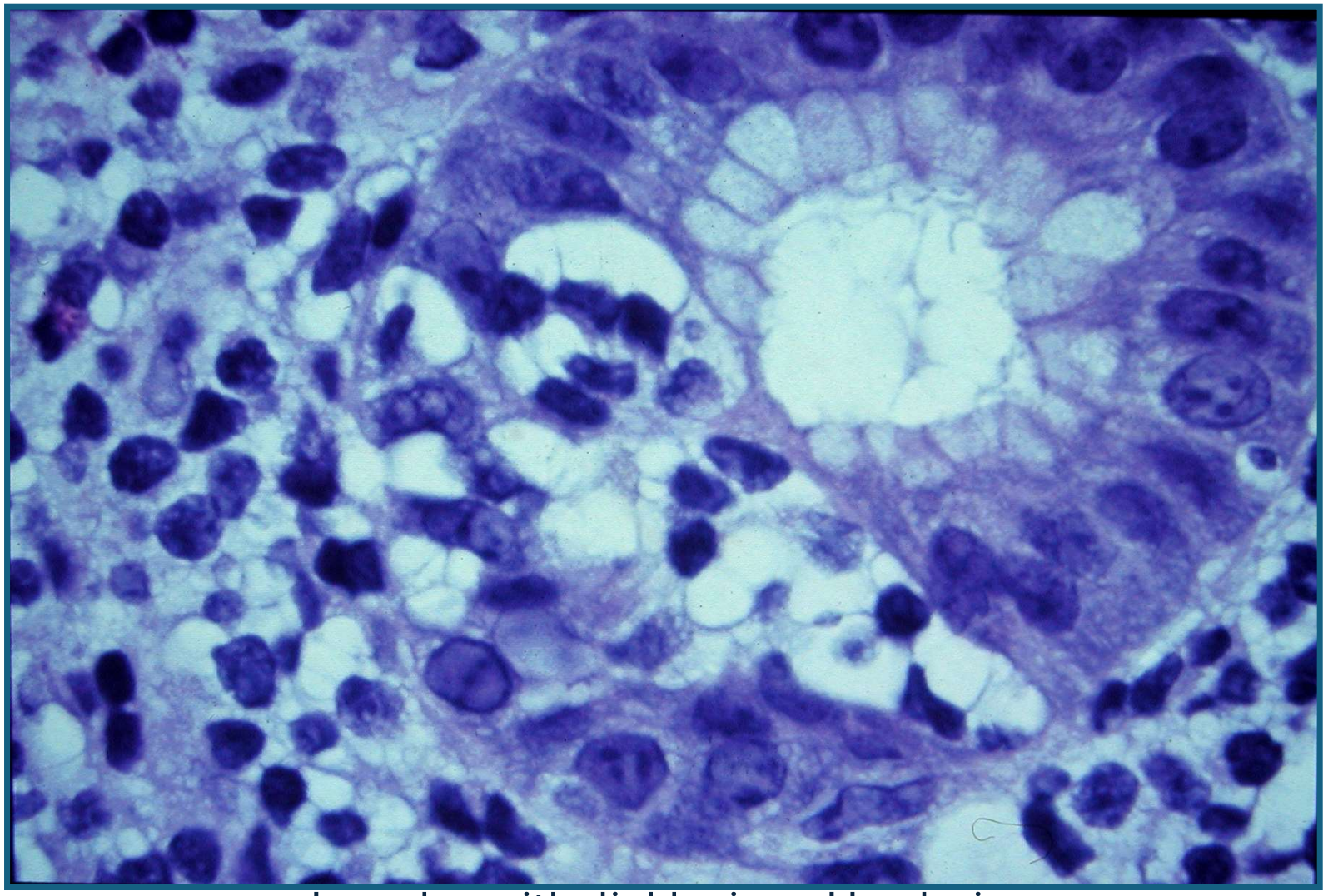


Gastric MALT lymphoma – follicular colonization



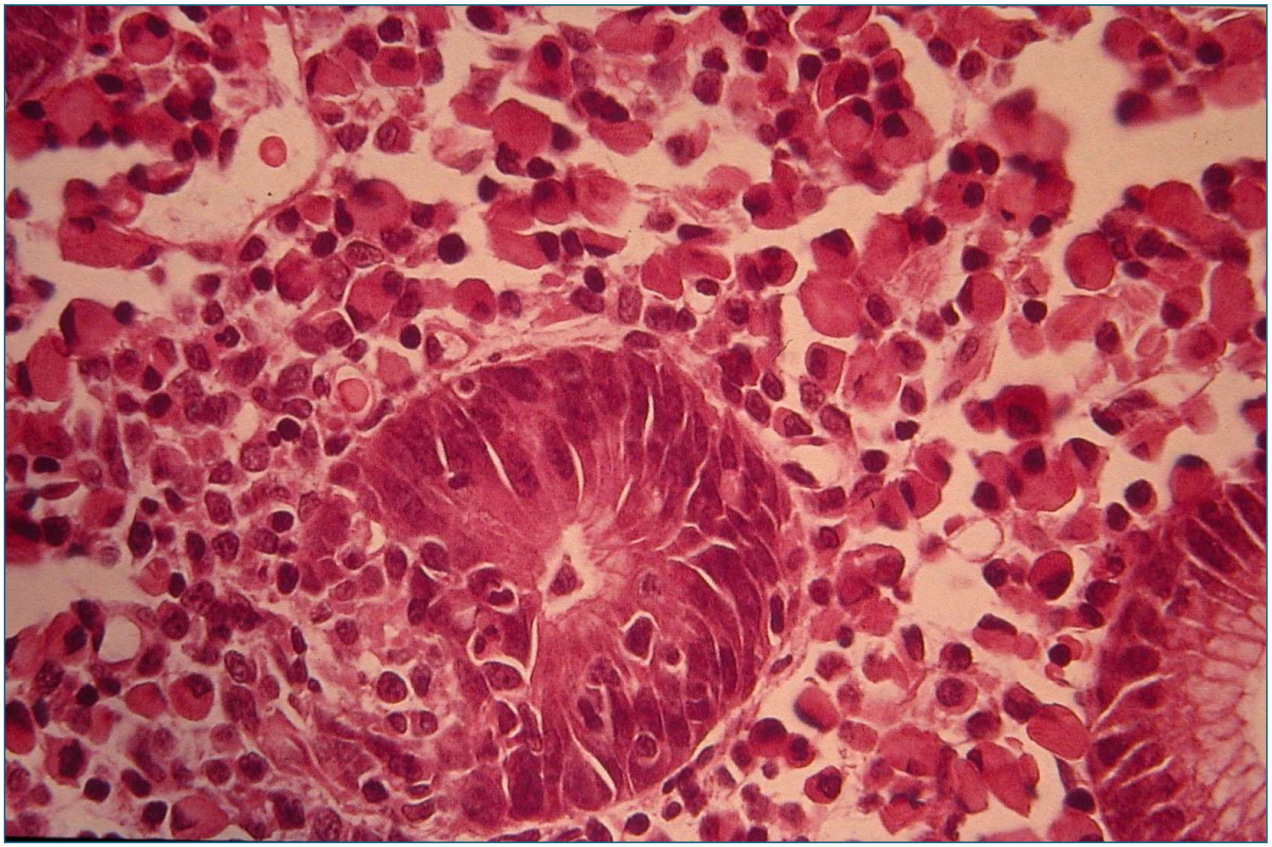
MALT lymphoma – cell morphology

MALT lymphoma



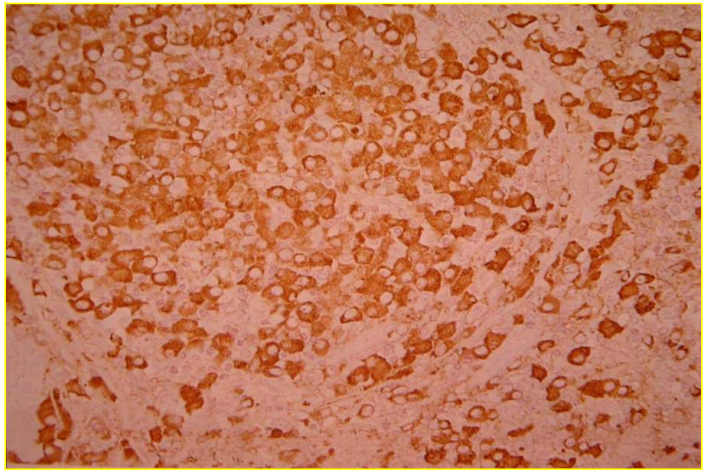
Lymphoepithelial lesion - H.pylori +

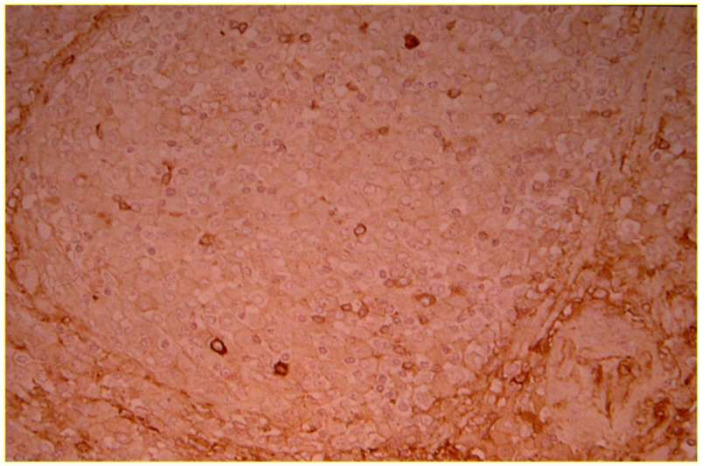
Gastric MALT lymphoma



Plasmacytic differentiation – Lymphoepithelial lesion

Gastric MALT lympoma



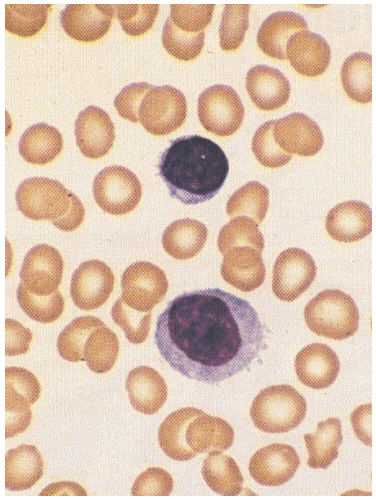


Monoclonal κ light chain

SPLENIC MARGINAL ZONE LYMPHOMA (SMZL)

- Definition: Low grade primary splenic B cell lymphoma, composed by small B cells with biphasic pattern and apart from splenic infiltration, splenic hilar infiltration and bone marrow infiltration. No peripheral lymphadenopathy

- There may be villous B cells in peripheral blood



SPLENIC MARGINAL ZONE LYMPHOMA (SMZL)

Macroscopy

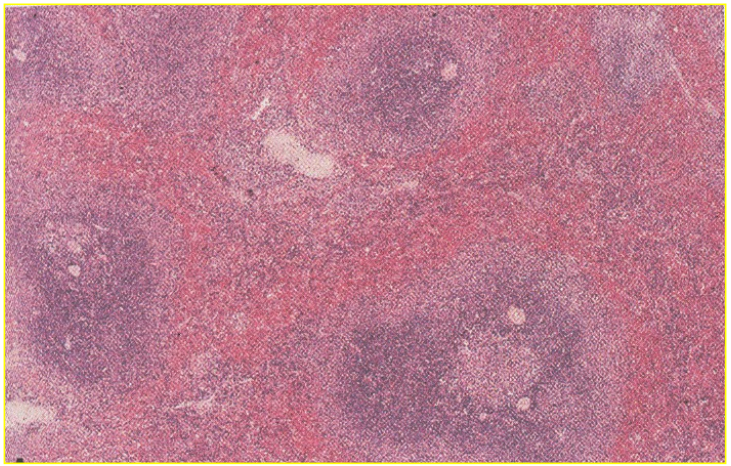
- Prominent increase in splenic weight (>1000 gr)

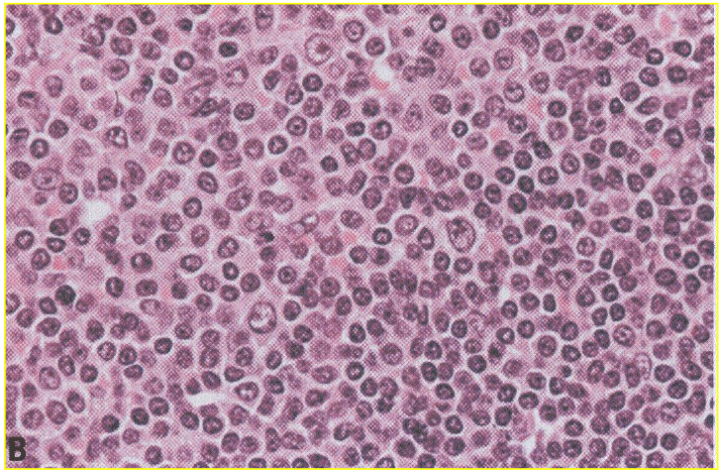
- Whitish nodules of various size (white pulp expansion)

SPLENIC MARGINAL ZONE LYMPHOMA (SMZL)

Microscopy - Spleen

- Nodules of small B cells located in the white pulp
- Biphasic cellular composition: small sized B cells located in the centre, small/medium sized B cells with abundant clear cytoplasm at the periphery, rare blastoid cells
- Variable (but always present) red pulp infiltration
- Rare findings: epithelioid histiocytes aggregates, plasma cell differentiation

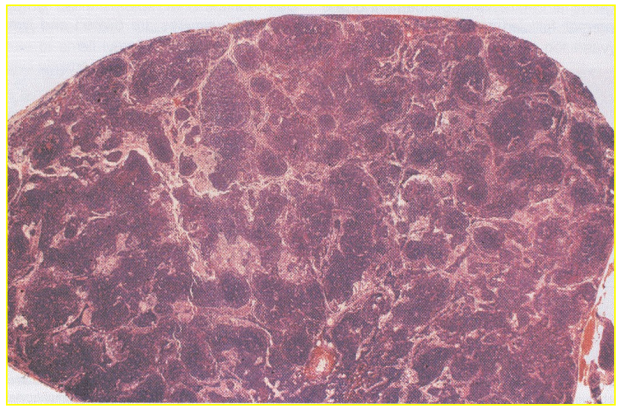




SPLENIC MARGINAL ZONE LYMPHOMA (SMZL)

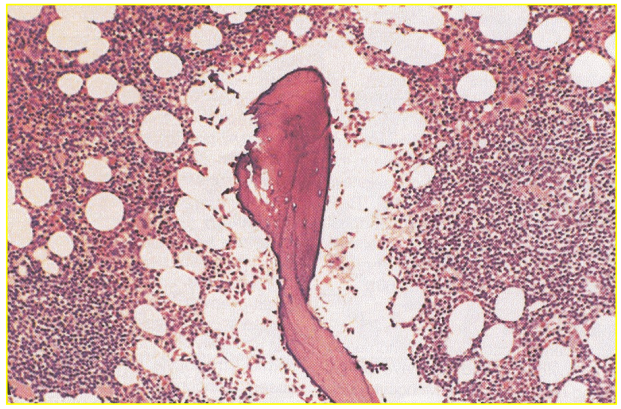
Microscopy – Hilar lymph nodes

- Nodular pattern
- Sinusoidal extension
- Follicular residues



Microscopy – Bone Marrow

- Bone marrow is infiltrated in most cases, even though this may not be readily identifiable in H&E
- Nodular, interstitial, intrasinusoidal pattern
- Germinal centres in the centre of the nodules



LYMPHOPLASMACYTIC LYMPHOMA

- B cell neoplasm consisting of small-sized B cells, plasmacytoid lymphocytes and plasma cells infiltrating the bone marrow, lymph nodes and spleen

**diagnostic criteria of other types of B cell lymphomas with plasmacytic differentiation are not met*

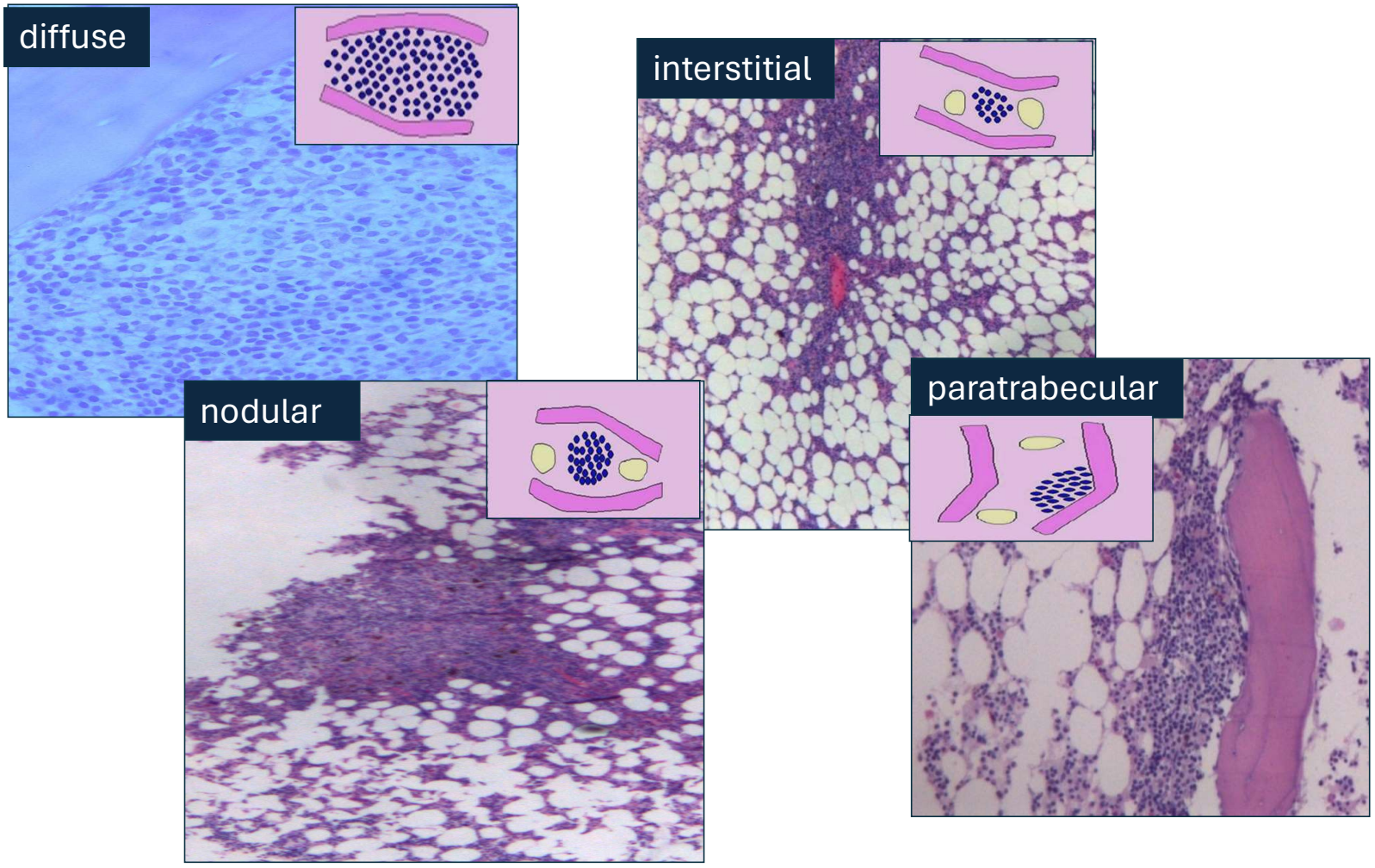
****Waldenström macroglobulinaemia**= LPL+ IgM serum paraprotein + bone marrow infiltration

- **Median survival: 5-10 yrs**

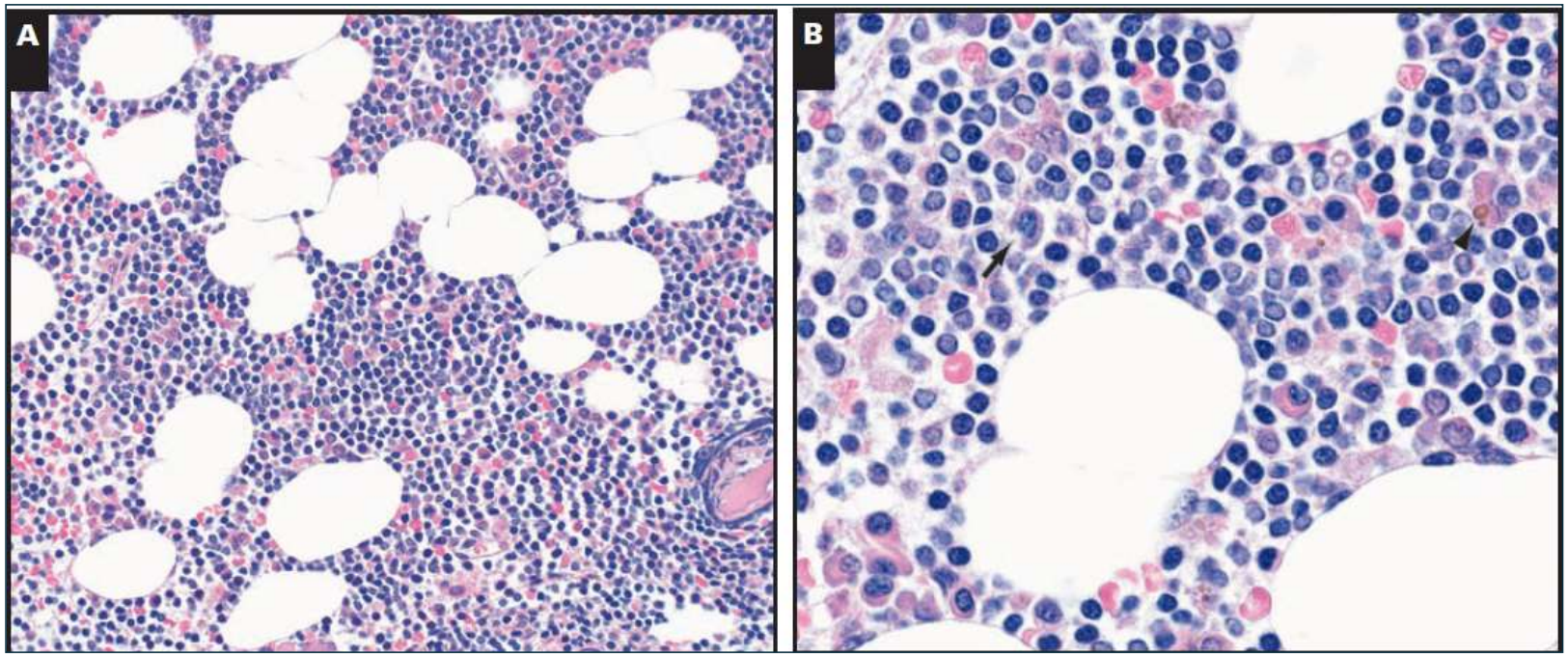
LYMPHOPLASMACYTIC LYMPHOMA

- Adult patients, median age 60 yrs
- Possibly related with HCV infection
- Bone marrow infiltration common
- Lymph node, extranodal site infiltration less common (e.g. spleen), blood
- 15-30% of WM cases: hepatosplenomegaly, lymphadenopathy
- Anaemia, weakness, fatigue
- IgM paraprotein (95% of cases)
  - Hyperviscosity (30%), coagulopathy
  - Cryoglobulin/autoimmune activity
  - Neuropathy due to paraprotein accumulation/cryoglobulinaemia

Bone Marrow Infiltration - Patterns



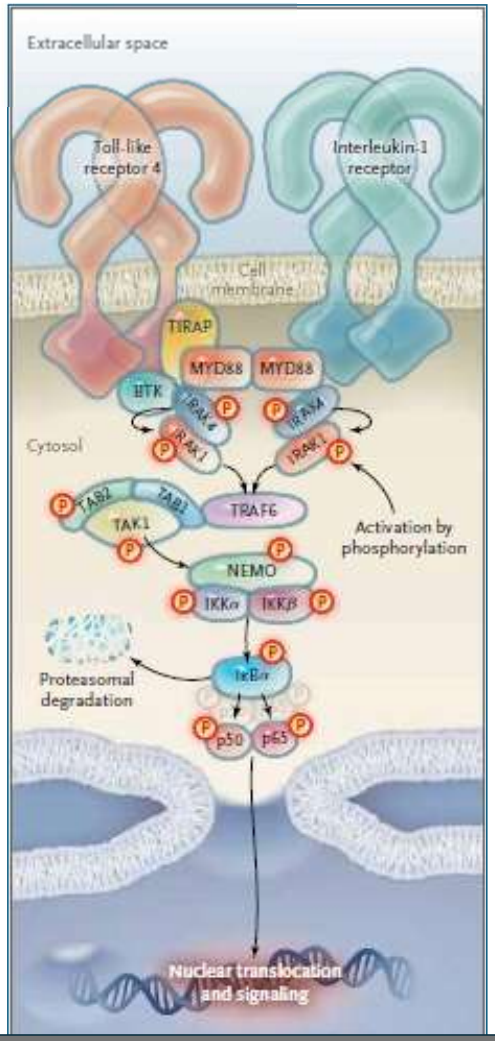
Bone Marrow Infiltration – Cellular Morphology



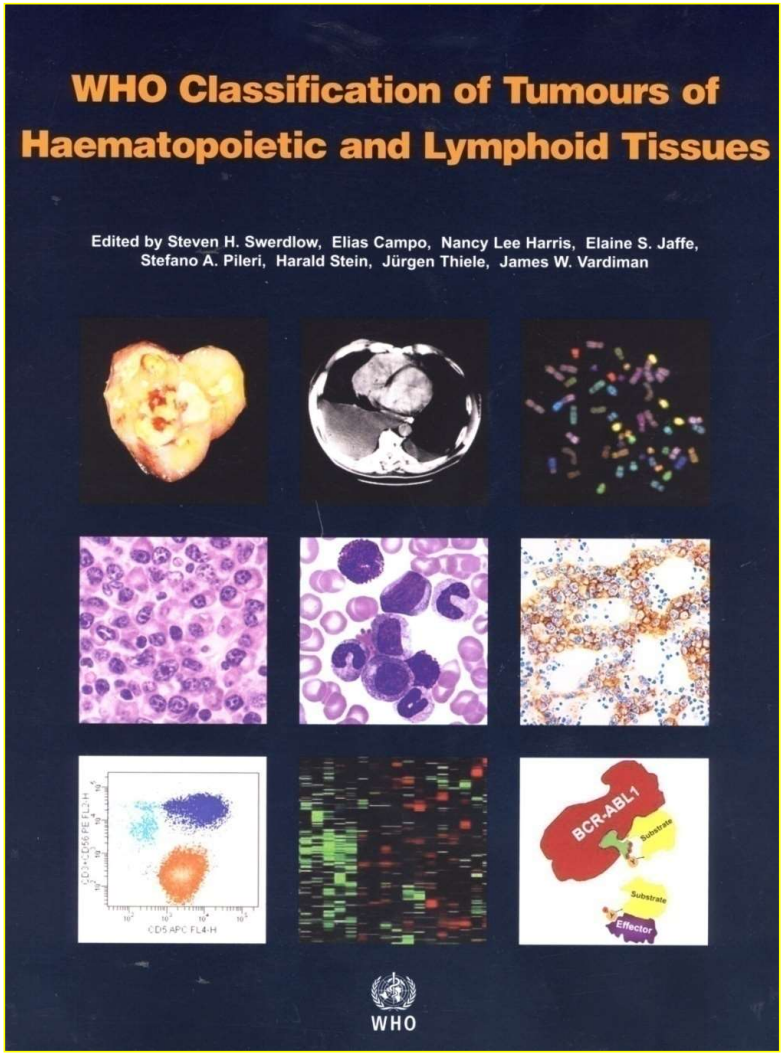
✓Neoplastic infiltrate composed of small-sized B cells, various number of plasma cells and plasmacytoid lymphocytes
✓**Increased mast cells**

Waldenström Macroglobulinaemia- Molecular Biology

- ✓ **MYD88 mutation** (T>C, L265P) in 87-91% WM
- ✓ Absent from plasma cell myeloma, low incidence in MZL (10%)
- ✓ Related with NFκB activation
- ✓ CXCR4 mutation
- ✓ MYD88 and CXCR4 activation confer poor prognosis



Xu L et al Blood 2013, Treon SP et al NEJM 2012



AGGRESSIVE B CELL LYMPHOMAS

I. Diffuse Large B Cell Lymphomas (DLBCLs)

Definition: A diffuse proliferation of medium to large size neoplastic B cells with a nuclear size at least equal to that of a histiocyte nucleus or more than twice the size of the small lymphocyte.

✓ **Heterogenous disease**

- **Clinical presentation**
- **Localization**
- **Prognosis**
- **Morphology**
- **Pathogenesis**
- **Immunophenotype / Genotype**

DLBCL not otherwise specified (NOS)

- **Epidemiology: ~30% of NHL in adults [the largest group]**
- **May arise de novo or from an underlying low grade lymphoma**
- **Cases arising in the setting of primary or acquired immunodeficiency are usually EBV positive and are classified separately**
- **Sites of involvement at presentation: Nodal (70%) – Extranodal (30%)**
- **Clinical features: -Rapidly enlarging lymph node or extranodal mass**
  -50% early stage (I-II)

-<30% bone marrow involvement with

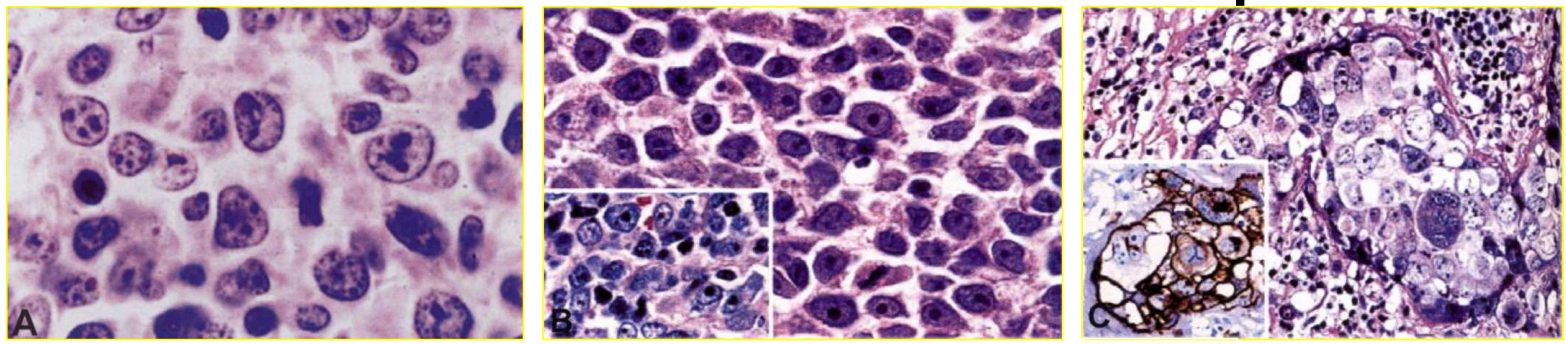
↓	↓
Large B cells	**Small B cells**
(concordant)	**(discordant)**

DLBCL NOS

- **Lymph node infiltration pattern:** **diffuse, vaguely nodular, interfollicular, sinusoidal pattern**

- **Main cytomorphologic variants**

Centroblastic **Immunoblastic** **Anaplastic**

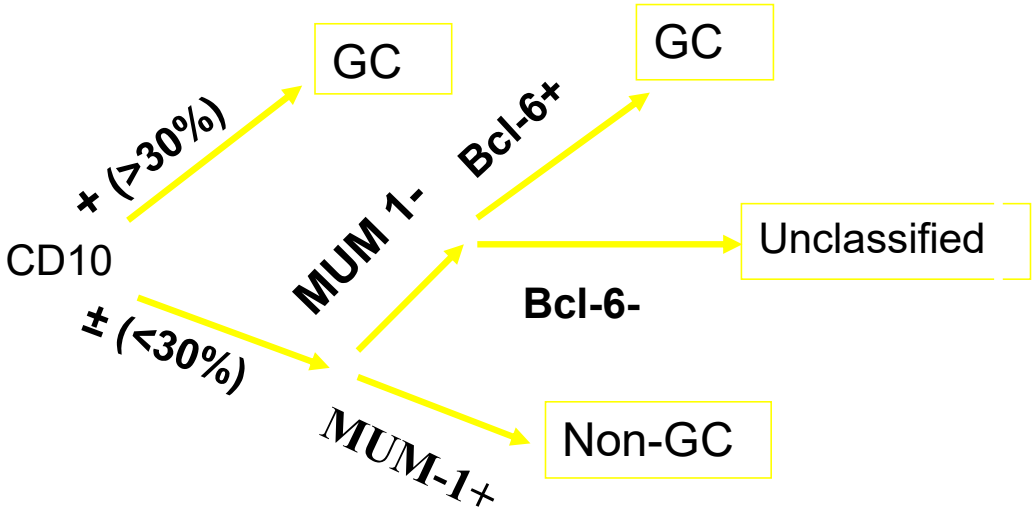


- Rarely: myxoid stroma, pseudorozettes, spindle cells, signet ring cells

DLBCL-NOS Immunophenotypic variations

Germinal centre (GC)/ non-Germinal centre (non-GC) phenotype

Hans algorithm

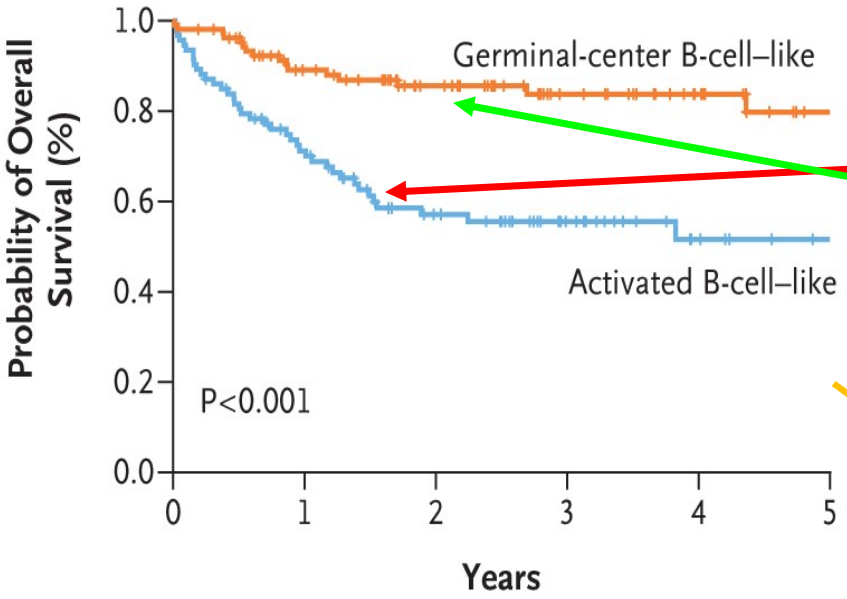


No prognostic or genetic correlation

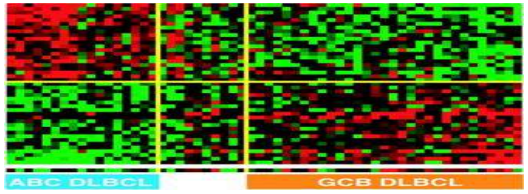
DLBCL-NOS Genotypic features

- Often cytogenetic abnormalities
- No specific diagnostic chromosomal alterations
- Clonal IgH rearrangements, somatic hypermutations, abnormal somatic hypermutations
- Translocations involving genes BCL2 (20-30%), BCL6 (~30%), MYC (10%)
- 10% of cases harbouring MYC translocations also show BCL2 and/or BCL6 translocations

DLBCL-NOS Molecular subtypes



Molacular (GEP)



Activated B-cell (ABC)

Germinal Center B-cell like (GCB)

Clinical-Prognostic Correlation

Before Mabthera era

After Mabthera

Lenz G. N. Engl. J. Med. 2008

DLBCL-NOS Molecular subtypes

- Defined by gene expression prolifing (GEP)
  - ✓ (GCB-like) germinal centre type [45-50%] (expression of genes characteristic of GCB cells)
  - ✓ (ABC-like) activated B cell type (expression of genes up-regulated during the in vitro activation of peripheral blood B cells)
  - ✓ (type 3): cases unclassified as GCB or ABC).
- Different chromosomal alterations between the 2 main sybtypes
- BCL2 rearrangement most common in GCB-like type
- The 2 main subtypes cannot safely be identified based on morphology or immunophenotype

Burkitt Lymphoma (BL)

- A highly aggressive but curable lymphoma, with very high proliferation index and an IG::MYC rearrangement.
- Presentation: Nodal/Extranodal/Leukemic
- Three epidemiological variants recognized:

  Endemic BL :

  - children of African origin in regions endemic for malaria, facial bones (50-70%), gonads, breasts, distal ileum, caecum (EBV present in all cases)

  Sporadic BL:

  - throughout the world , usually in young patients, EBV present in 20-30% but prevalence may be higher in adults; ileocecal region, ovaries and breast most commonly involved

  Immunodeficiency-associated BL:

  - (usually in HIV+ patients) EBV present in 25-40% of cases; nodal and BM involvement common
- *Advanced-stage disease with ↑↑↑ serum LDH*

Burkitt Lymphoma (BL)

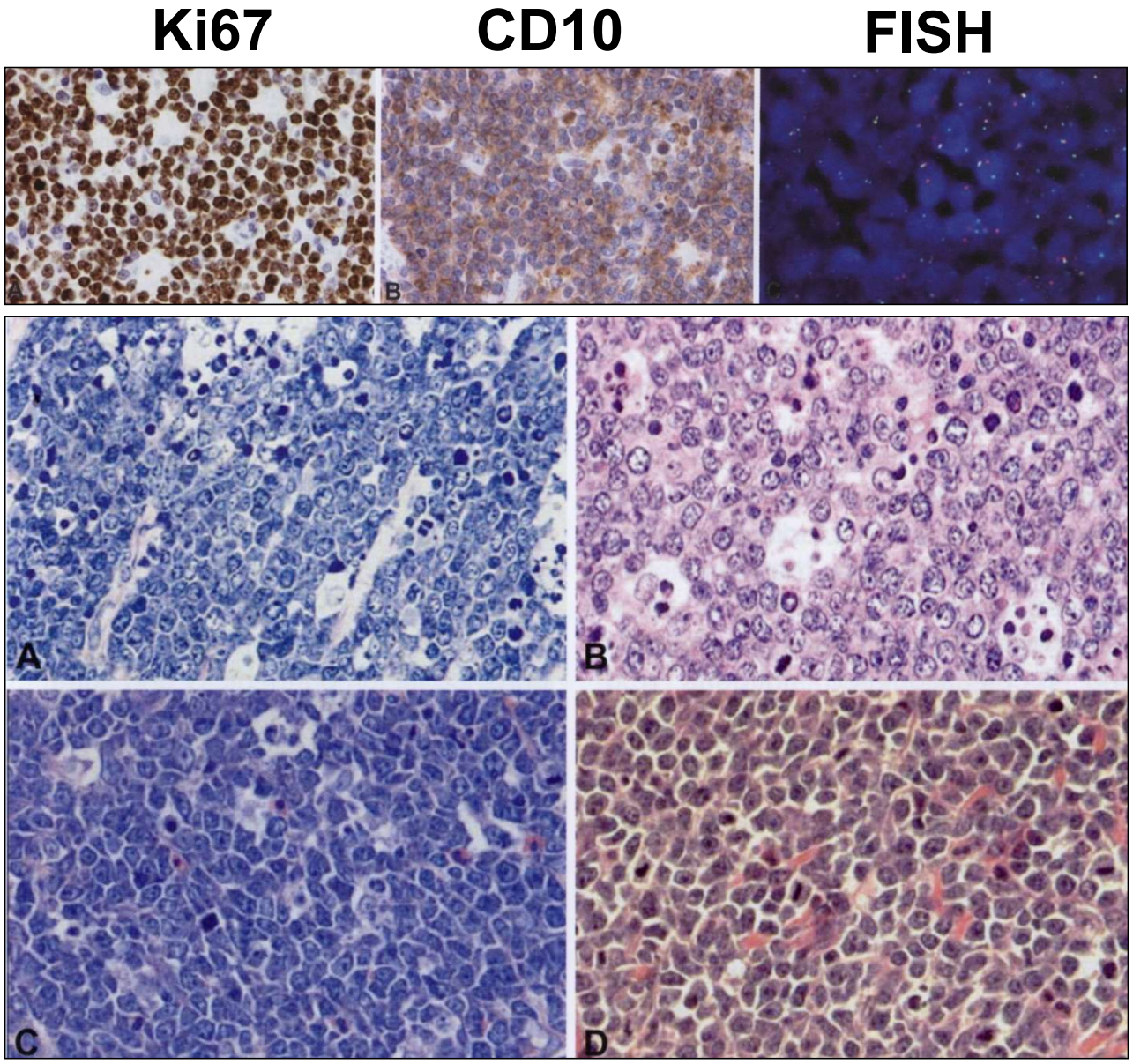
- **Morphology:**
  - Medium-sized monotonous cohesive cells, with deeply basophilic cytoplasm and multiple basophilic paracentrally located nucleoli
- **Extremely high mitotic and apoptotic activity *starry sky pattern**
- **Occasionally seen: Florid granulomatous reaction, greater nuclear pleomorphism with more prominent nucleoli, plasmacytoid differentiation (in association with immunodeficiency)**

- **Immunophenotype:**
  - pan-B cell markers+
  - germinal centre markers (CD10/bcl6)+
  - bcl2 - (or faint expression)
  - Tdt -
  - Ki67 >98%
- **Genotype: IgH/MYC translocation (or rarely IGK or IGL) with no BCL2 or BCL6 translocations**

- **~10% of classic BL show MYC-R(-) at FISH (BAP) → MYC/IG fusion probes**

- **EBV+ and EBV- BL cases are two different biologic groups based on molecular findings, irrespective epidemiologic or geographic features**

- **Prognosis: highly aggressive but curable lymphoma**

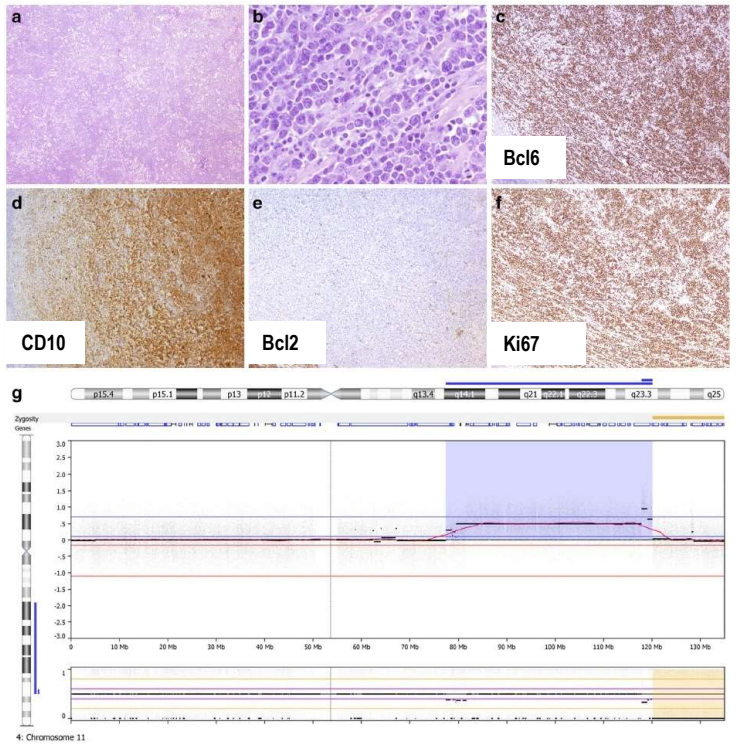

Ki67
CD10
FISH
A
B
C
A
B
C
D

High grade B-cell lymphoma with 11q aberrations (Burkitt-like) (HGBL-11q, WHO 2022)/Large B cell Lymphoma with 11q aberrations(ICC 2022 provisional entity)

- ✓ An aggressive MYC-R negative mature B-cell lymphoma with immunophenotype typical of BL
- ✓ 13% of all BL, DLBCL and HGBL
- ✓ Lower MYC expression by IHC compared to BL
- ✓ Blastoid or intermediate morphology
- ✓ Nodular pattern in some cases
- ✓ Nodal presentation in most cases
- ✓ 11q aberrarions (proximal gain and telomeric loss of 11q sequences simultaneously)
- ✓ GEP: GC-derived
- ✓ Asses 11q status in:

-MYCR-HGBL or BL type morphology-

-BL-type immunophenotype (No BCL2 expression)



Salaverria, Blood 2014; Ott, BJH 2017; Au-Yeung, BJH 2020; Gonzalez-Farre, Haematologia 2020; Rimsza, Virch Arch 2017; Alaggio, Leukemia 2022; Campo, Blood 2022

LBCL with IRF4 rearrangements

New entity in WHO 2022 and ICC 2022 for DD DLBCL and paediatric FL

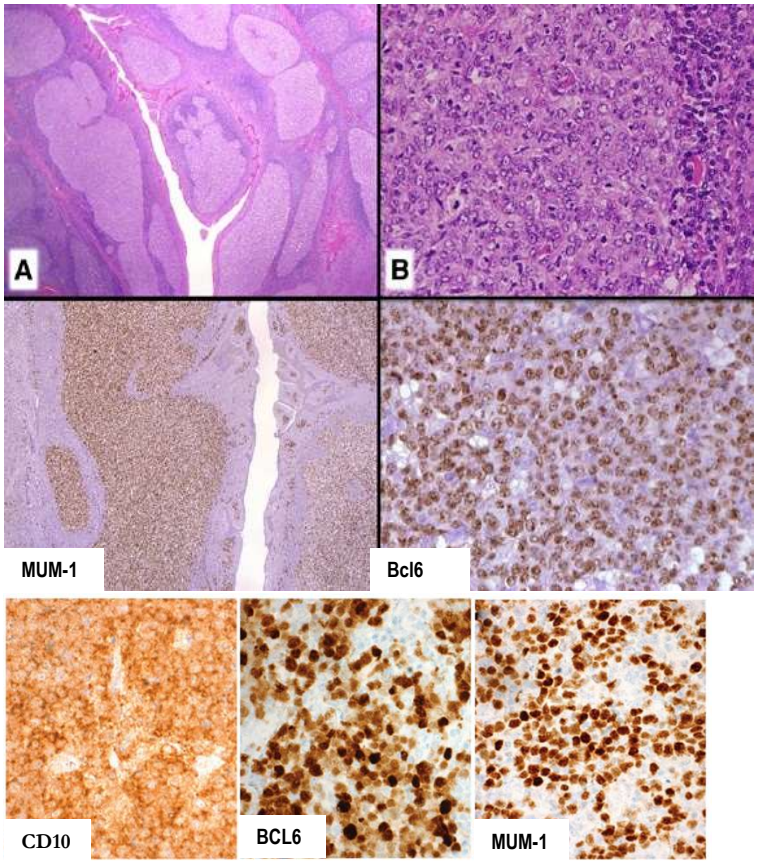
- ✓ **Mostly in children/young adults**
- ✓ 6-20% of DLBCL/FL3 in children, more rare in adults
- ✓ Low stage at presentation, typically confined to **Waldeyer's ring and/or cervical lymph nodes**

- ✓ Nodular and/or diffuse growth **resembling FLBCL or DLBCL**
- ✓ **Cytology: Large cell +/- blastoid**
- ✓ Intense **MUM-1+** (100%), **Bcl6+** , **CD10 and** bcl2+ (>50%), rarely CD5+, very high Ki67

- ✓ ***Genetics: IRF4-R (with IGH in most cases), Bcl6-R (often), no Bcl2-R***

- ✓ **Better prognosis when treated**

- ✓ Concurrent IRF4 mutations, along with IRF4-R



INTERMEDIATE MORPHOLOGIC FEATURES' CATEGORIES

1. B CELL LYMPHOMA UNCLASSIFIED, WITH MORPHOLOGIC FEATURES INTERMEDIATE BETWEEN DLBCL AND BURKITT LYMPHOMA

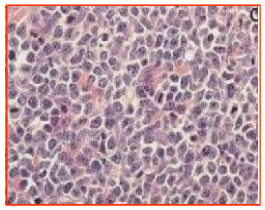
2. B CELL LYMPHOMA UNCLASSIFIED, WITH MORPHOLOGIC FEATURES INTERMEDIATE BETWEEN DLBCL AND CLASSIC HODGKIN LYMPHOMA

INTERMEDIATE MORPHOLOGIC FEATURES' CATEGORIES

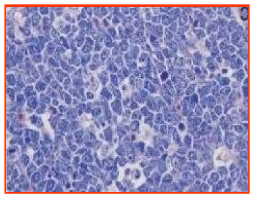
B CELL LYMPHOMA UNCLASSIFIED, WITH MORPHOLOGIC FEATURES INTERMEDIATE BETWEEN DLBCL AND BURKITT LYMPHOMA

Morphology

Intermediate between DLBCL, NOS and BL



BL type



Immunophenotype

BL-type
Bcl6+ CD10+ bcl2- Mum-1-

Ki-67>90%

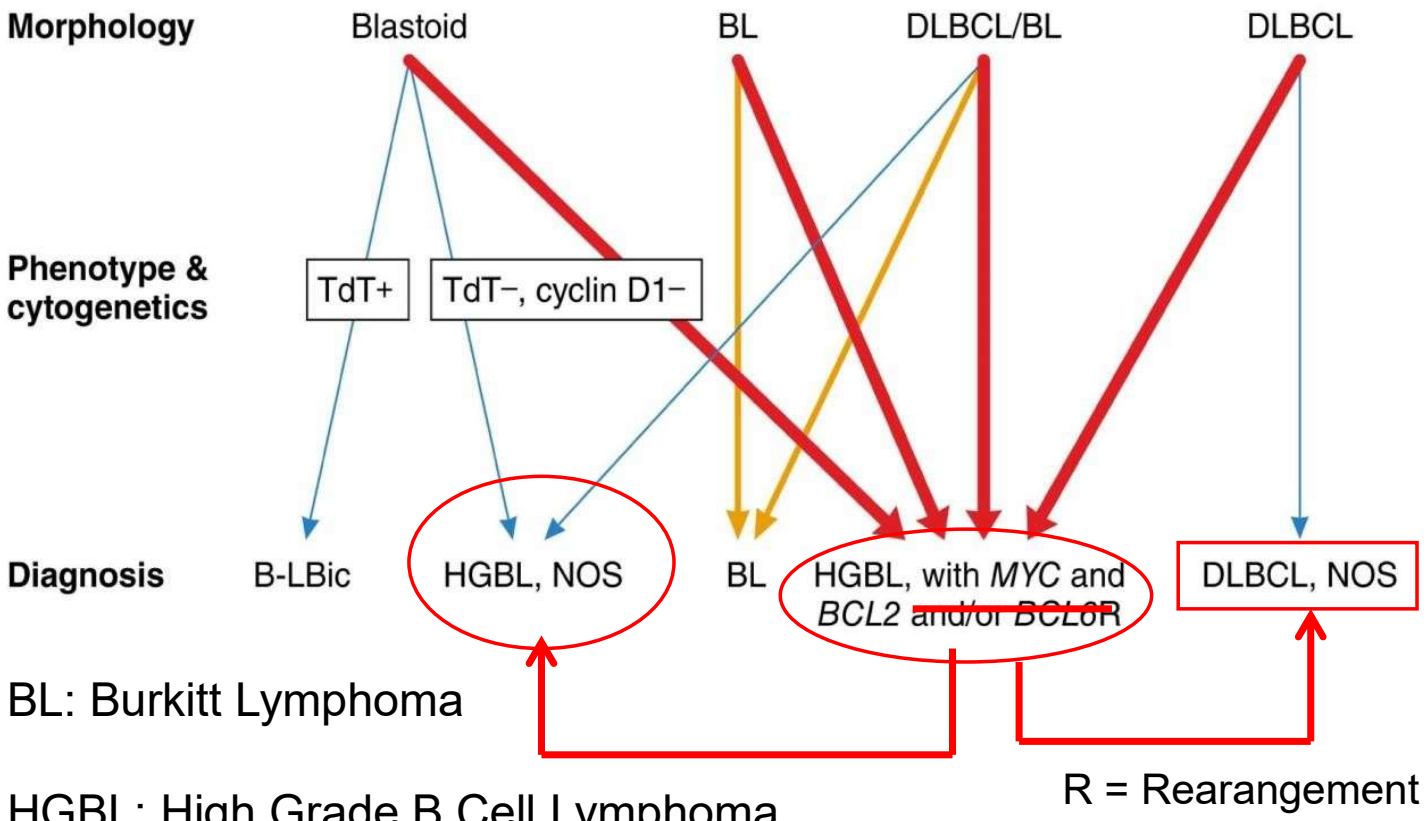
Not compatible with BL
bcl6+ CD10+ bcl2+ Mum-1+/-

karyotypic features

- **Complex karyotypes and MYC-R**
-**Concurrent MYC-R and BCL2-R (15%, "double hit") or MYC, BCL6 +/- BCL2 (rarely, "triple hit")**

Poor prognosis

Chemotherapy resistant
Overall median survival 4 mo



- BL: Burkitt Lymphoma
- HGBL: High Grade B Cell Lymphoma
- DLBCL: Diffuse Large B Cell Lymphoma
- B-LB: B – Lymphoblastic Lymphoma
- NOS: Not otherwise specified

R = Rearangement

Kluin P. et al. Blood 2016
Alaggio et. al, Leukemia 2022

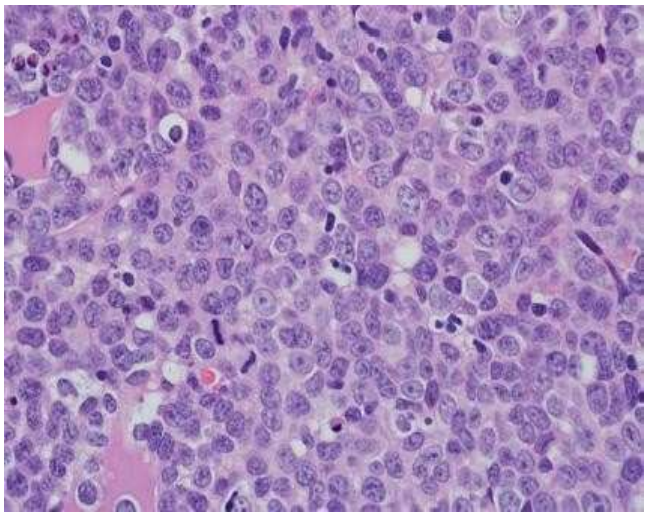
“Double hit” lymphomas

(High grade B-cell lymphoma with MYC and bcl-2 rearrangements, WHO 2022, HGBL with MYC και BCL2 (ICC 2022))

- **Morphology: unclassified DLBCL/Burkitt, DLBCL, blastoid, BL**
- **bcl-2+, CD10+, Ki-67 ~90%**
- **“double hit” lymphomas’ prognosis very poor, irrespective of morphologic features**
- **“double hit” AND GC immunophenotype → asses «double hit» status in DLBCL/BL unclassified and DLBCL-GC type**

R.Pillai, AJSP 2013 ,M.Pedersen, Haematology 2012 S.Li, Modern Pathology 2012

High Grade B Cell Lymphoma, NOS (in WHO 2022 including cases with concurrent MYC-R and BCL6-R)



Ott, BJH 2017

- ✓ **No double hit or triple hit**
- ✓ **There may be *MYC, BCL2 or BCL6 amplification***
- ✓ Morphology: **Blastoid or DLBCL/BL,** usually with '**starry-sky' areas**
- ✓ **High proliferation rate (Ki67)**

- ✓ **Usually GCB**, sometimes ABC type

- ❖ **DLBCLs with *MYC-R, with no Bcl2-R (MYC single hit) or those with* MYC-R/Bcl6-R are diagnosed as DLBCL-NOS (*WHO 2022*)**
- ❖ **High Grade B Cell Lymphoma, NOS diagnosis is MORPHOLOGY BASED!!!!**

High Grade B Cell Lymphoma with MYC and BCL6 rearrangements (ICC 2022)

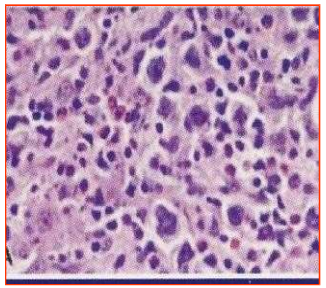
- ✓ ***MYC/BCL2/BCL6 triple hit much more prevalent in males***
- ✓ ***MYC/BCL6*** **DHL: MUM-1+, CD10+/BCL2+ not so often (in comparison with *MYC/BCL2* DHL), GCB ≈ ABC**
- ✓ **Aggressive clinical course, possibly worse than MYC/BCL2 DHL**
- ✓ **Excluded from WHO 2022 (classified as DLBCL NOS or HGBL NOS, morphology taken into account)**
- ✓ **Provisional entity in ICC 2022**
- ✓ **Cell of Origin closer to DLBCL**
- ✓ **More heterogenous disease than HGBL MYC-R and BCL2-R**

Pillai, AJSP 2013; Turakhia AJSP 2014; Wang AJSP 2015; Lin, Curr Hematol Malign Rep 2013; Li, Histopathology 2016; Landsburg Cancer 2016; Moore, AJSP 2017, Johnson Hum Pathol 2018, EAHP 2021; Alaggio, Leukemia 2022, ; Campo, Blood 2022; EAHP 2022

B CELL LYMPHOMA UNCLASSIFIED, WITH MORPHOLOGIC FEATURES INTERMEDIATE BETWEEN DLBCL AND CLASSIC HODGKIN LYMPHOMA

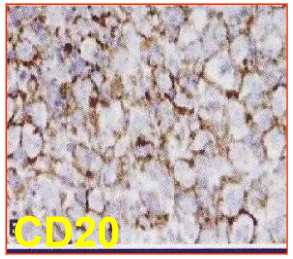
Morphology

cHL



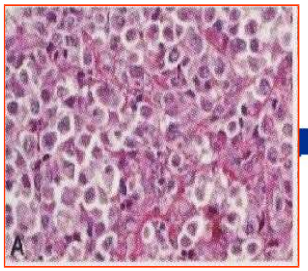
Immunophenotype

DLBCL
CD20+ CD79a+
bcl6+ OCt2+
CD30-/+ CD15-



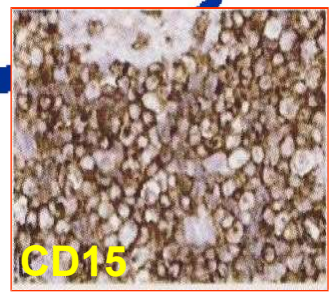
Morphology

DLBCL
(PMBCL)



Immunophenotype

cHL
CD30+ CD15+/- CD20+/-
CD79+/- bcl6-Oct2-



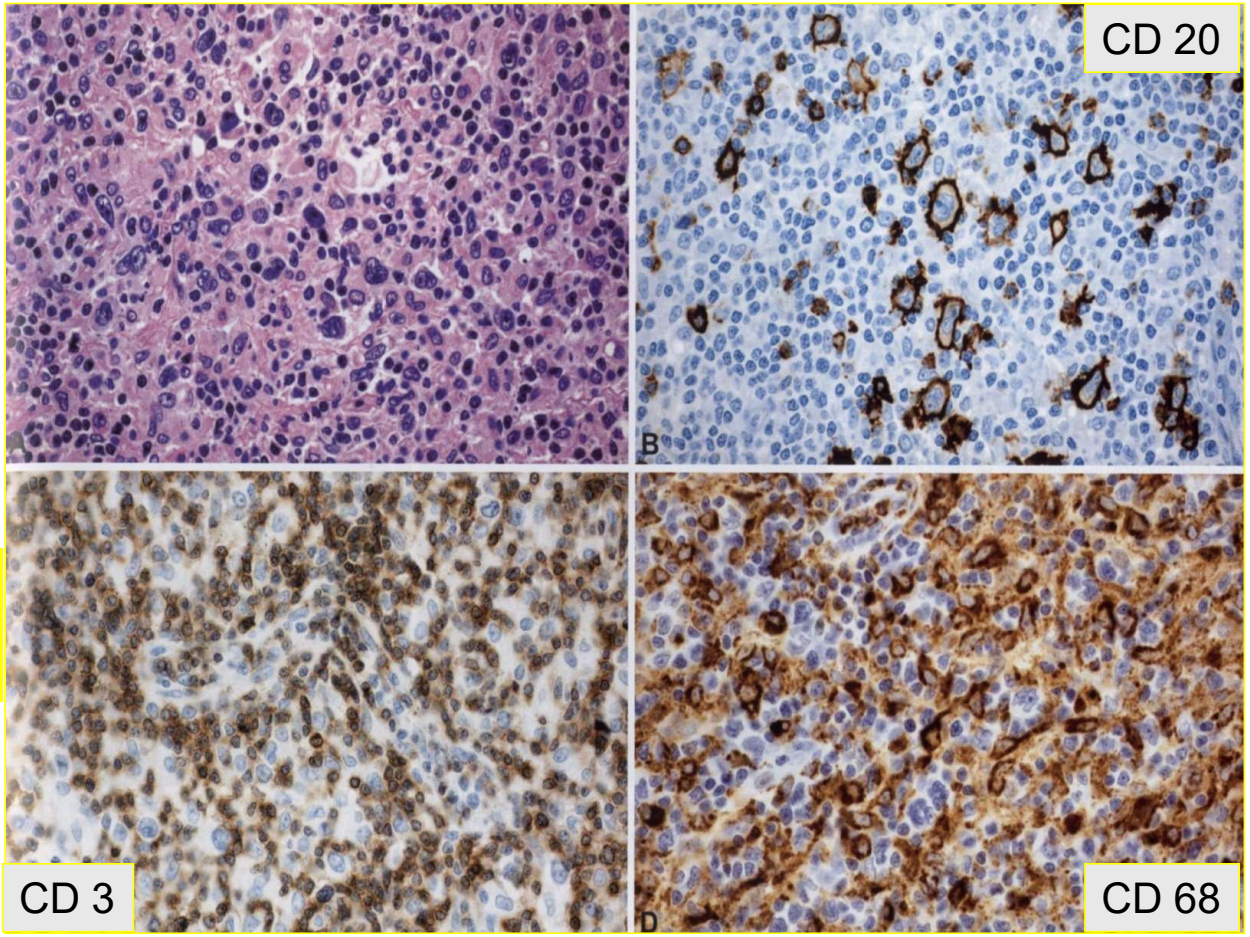
Prognosis

Poorer than → cHL DLBCL → Necessary to distinguish

- No uninamity in therapeutic decisions
- Usually treated as aggressive B cell lymphoma

T-cell/histiocyte-rich large B-cell lymphoma (THRLBCL)

T-cell/histiocyte-rich large B-cell lymphoma is an aggressive B-cell lymphoma with <10% large neoplastic B cells, scattered in a diffuse background rich in T cells and histiocytes, and with virtual absence of small B cells. A subset of cases shows marked clinical, immunophenotypic, and molecular overlap with nodular lymphocyte predominant Hodgkin lymphoma (NLPHL).

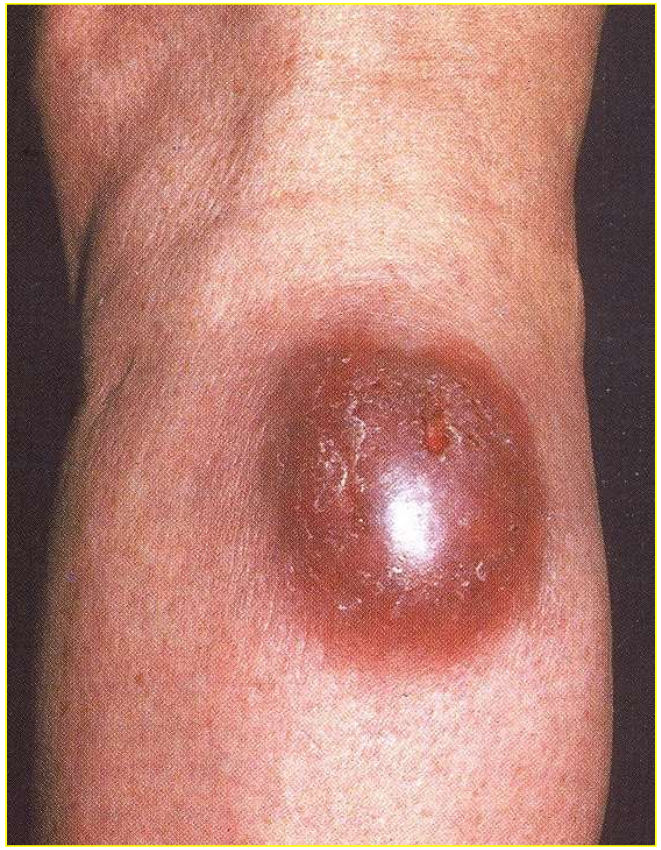


Small numbers of randomly scattered neoplastic large B cells

Abundant T cells CD8+

Primary Cutaneous Large B-Cell Lymphoma (PCLBCL) - Leg Type

Definition: Primary cutaneous diffuse large B-cell lymphoma, leg type (PCLBCL-LT) is a lymphoma composed exclusively of centroblasts and immunoblasts, most commonly arising in the leg.



Clinical features

- Red or bluish-red, often rapidly growing and ulcerating, localized or multiple skin tumours
- Usually in elderly women

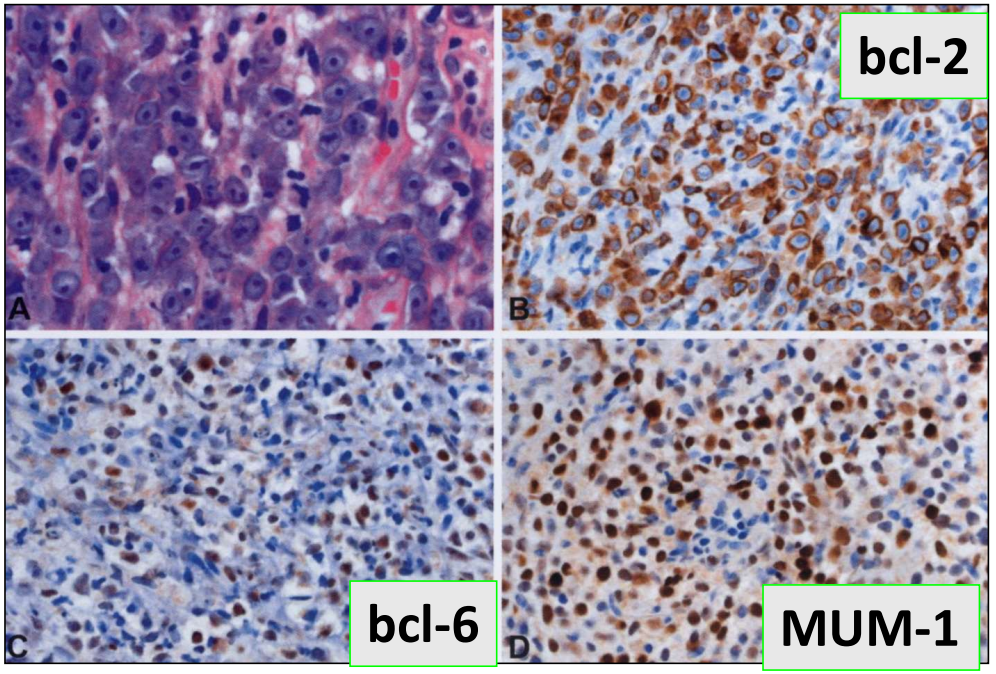
Primary Cutaneous Large B-Cell Lymphoma (PCLBCL) - Leg Type

Immunophenotype

- CD20/CD79a^{+}
- bcl-2^{+}
- bcl-6^{+}
- CD10^{-}
- MUM-1/IRF4^{+}

Genotype

- ABC GEP [MYD88 mut. (69%) (Fam-Ledard, JID 2012)
- 5-year overall survival rate of approximately 50%



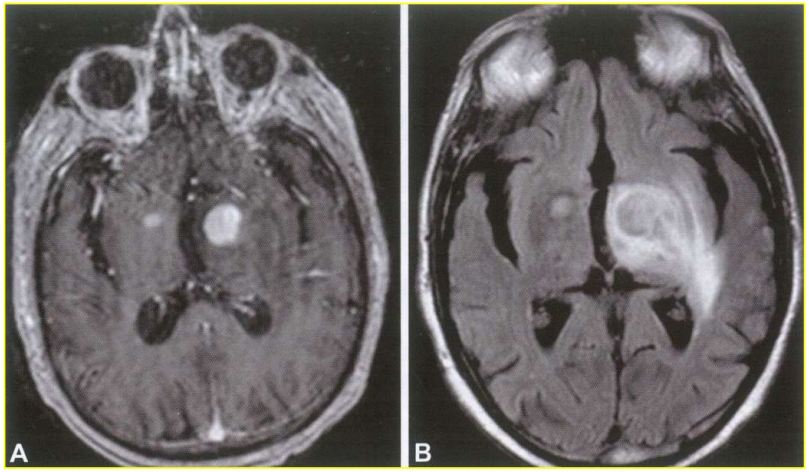
Large B-Cell Lymphomas in immune-privileged sites

Subtypes	**Large B cell Lymphoma arising in the central nervous system (CNS) and vitreoretina of immunocompetent patients**
	Large B cell Lymphoma arising in the testis of immunocompetent patients
Clinical features	Usually in adults over age of 60 years
	Lymphoma tends to “home” to other immune privileged sites: vitreoretina tumour may occur concurrently with or follow CNS tumour; testicular tends to relapse in CNS or contralateral testis
	Aggressive tumours with poor prognosis
Morphology	Large B cell
Immunophenotype	Activated B-cell: usually CD10-, MUM1+, BCL6+
	EBV-
Mutational Profile	**Concomitant MYD88 and CD79B mutations**
	Immune evasion: genetic inactivation of MHC class I and II and B2M (β 2-microglobulin) with subsequent loss of protein expression
Grouped as one category because they arise in immune sanctuaries , created by the respective anatomical structures (blood-brain, blood-testinal, blood-testicular barriers) and target organ immune regulation	

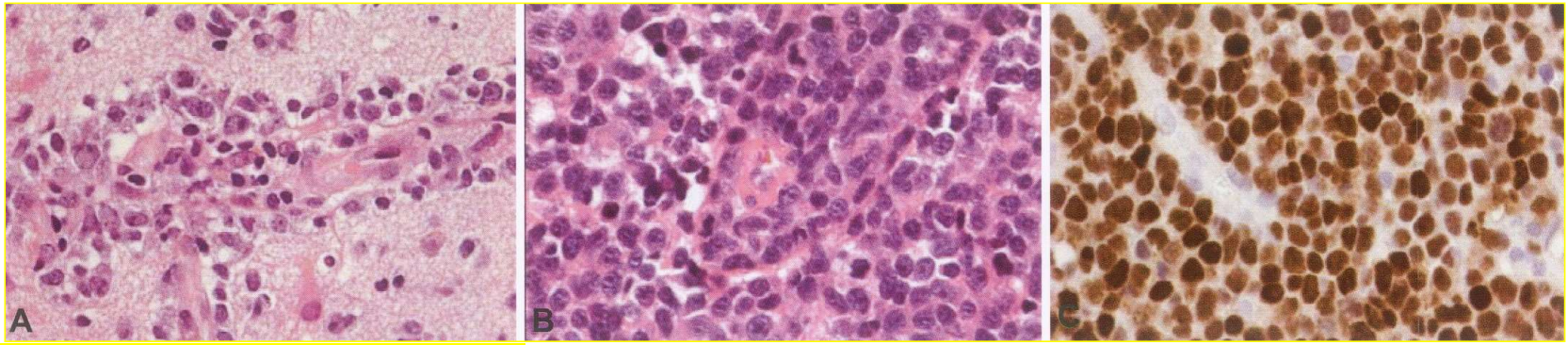
Alaggio, Leukemia 2022

Large B cell Lymphoma of the CNS

Multiple brain lesions



MUM-1



Perivascular infiltration pattern

EBV+ DLBCL

EBV+ LBCL arising in immunocompetent patients

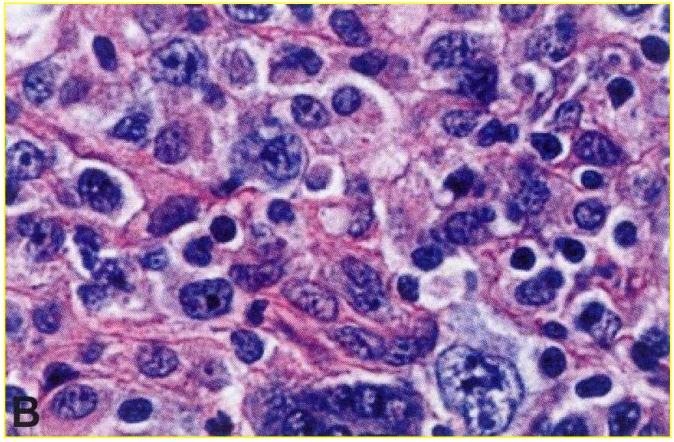
- ✓ **Higher prevalence in East Asia and South America (8-10%) than in Western countries (<5%)**
- ❖ **>80% EBER positive cells required for diagnosis**
- ❖ **Oncogenetic mechanisms of EBV are attributed to LMP1**

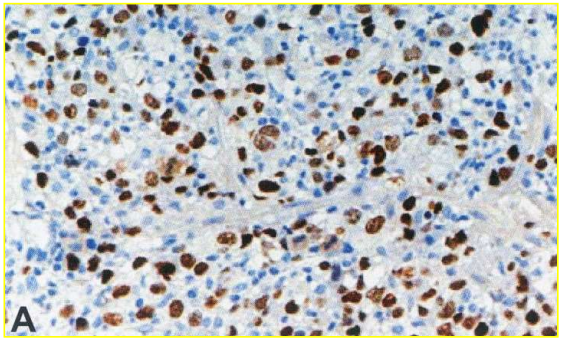
- ✓ **Both nodal (especially in young patients) and extranodal sites (lungs, GI) involved**
- ✓ **Prognosis better in younger patients or children**

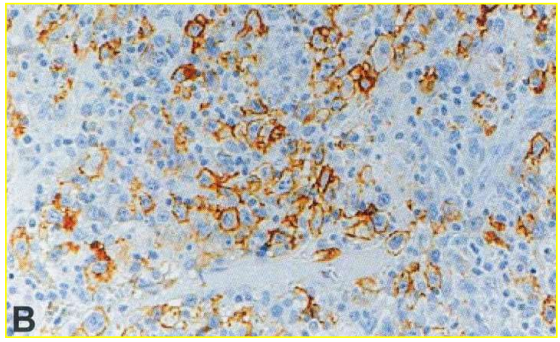
- ✓ **Rearrangements of MYC, BCL2, BCL6 rare to absent**

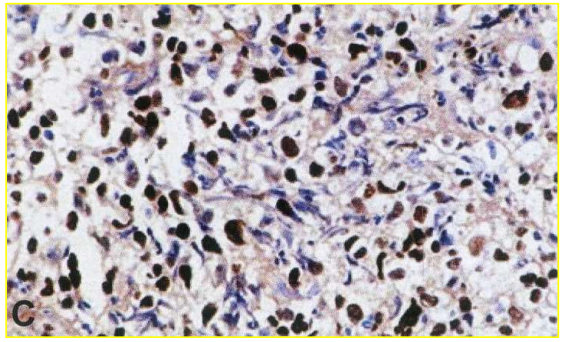
Ok, Blood 2013; Dojinov, Blood 2011; Hong, Ann Oncol 2015; Swerdlow, Blood 2016; Piris, Exp Rev Anticancer Ther 2016; Castillo, Am J Hematol 2018; Yin, Ann Transl Med 2018

EBV+ DLBCL









EBNA 2

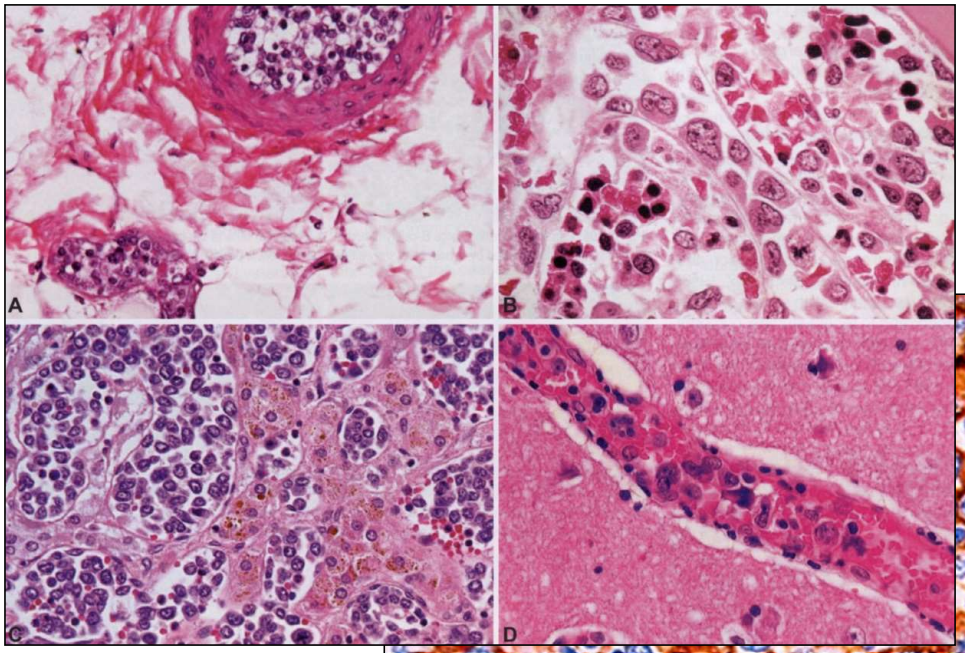
CD 20

EBER

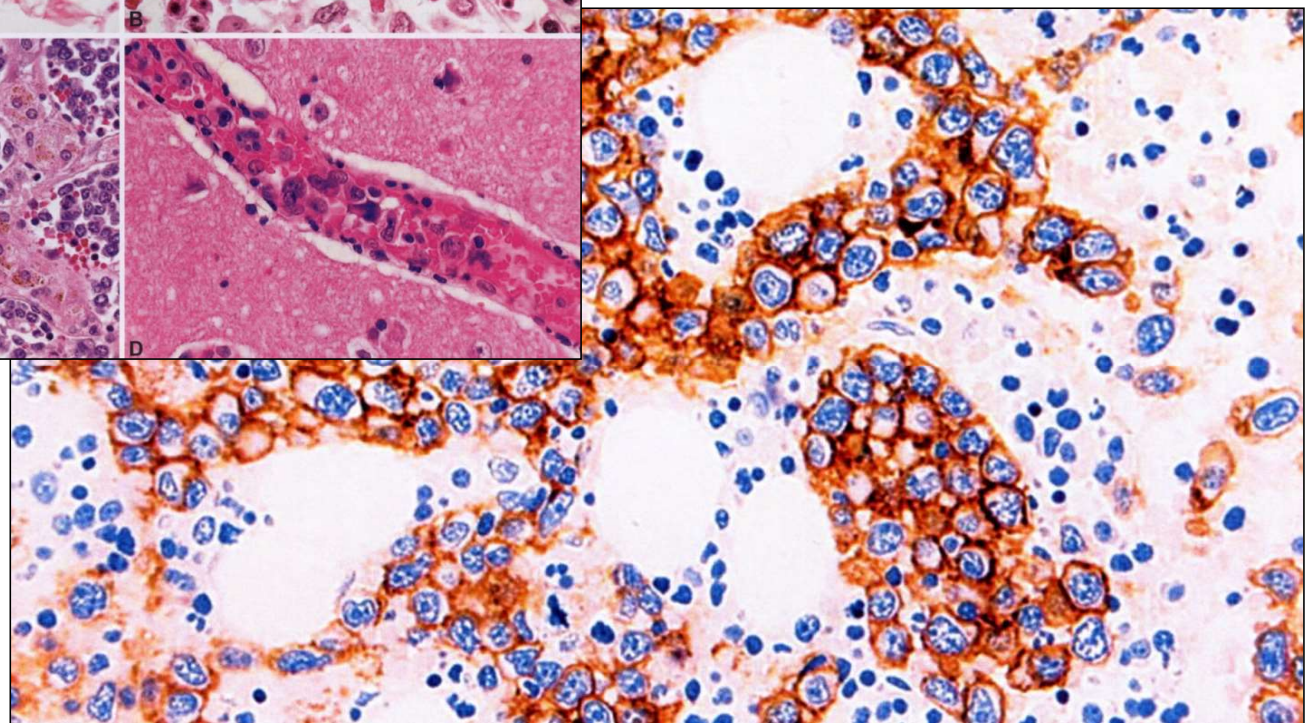
Special DLBCL subtypes - Intravascular DLBCL

- Special subtype with poor prognosis
- Extranodal localization: liver, spleen, bone marrow, skin, CNS, etc
- Morphology:
  – Large or medium sized lymphoid cells with prominent nucleoli
  – Intrasinusoidal/intravasclular infiltration pattern
  – Extravascular growth absent to minimal
  – Tissue necrosis
- Immunophenotype:
  – Pan-B markers, MUM-1+, may be CD5+
  – Cell adhesion molecule markers are negative
- Genetic features, such as NF-κB pathway gene mutations (MYD88, CD79B) and immune evasion, similar to PCNSL and cutaneous LBCL leg-type
- Normal cell counterpart: peripheral B-cell of non-GC immunophenotype (83%)

Intravascular DLBCL

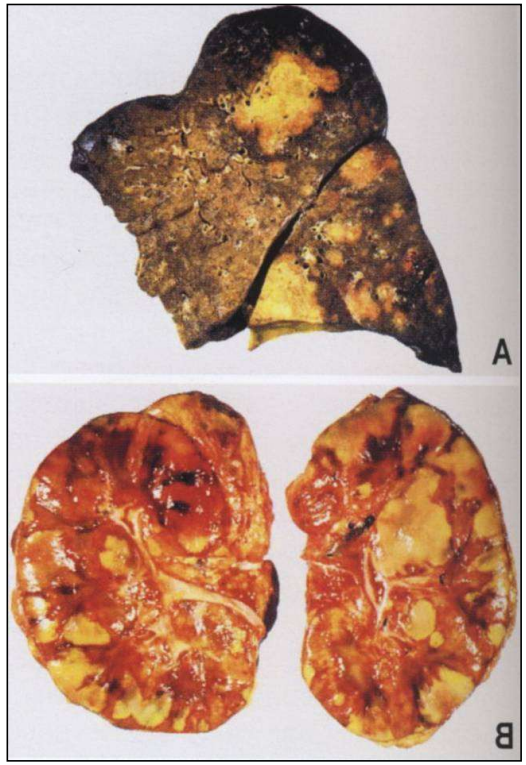


CD 20



Special DLBCL subtypes - Lymphomatoid Granulomatosis (LYG)

- Usually in adults with overt respiratory symptoms (cough, dyspnoea and chest pain)
- Localization: lung (100%), skin, upper respiratory tract, GI tract, kidney, CNS
- Pathogenesis related to Epstein-Barr virus
- Impaired immunity is regarded as a predisposing factor.
- Nodular lesions with central necrosis



Lymphomatoid Granulomatosis (LYG)

- **Morphology**

✓ Polymorphous infiltrate comprising small and large lymphoid cells (immunoblast or HRS-like)

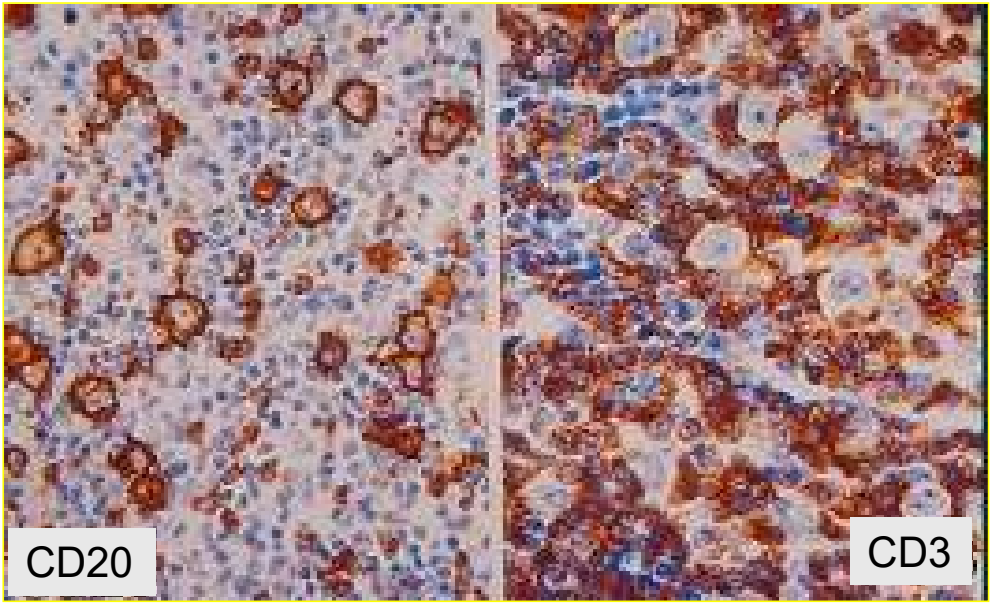
✓ No acute inflammation

✓ Vascular invasion by small lymphocytes may result in infarct-like necrosis, vascular destruction

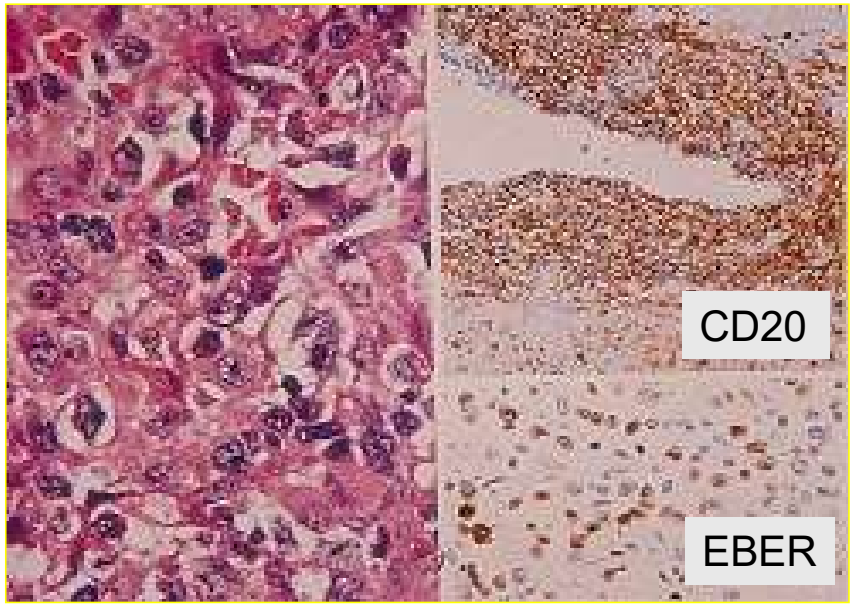
- **Immunophenotype**

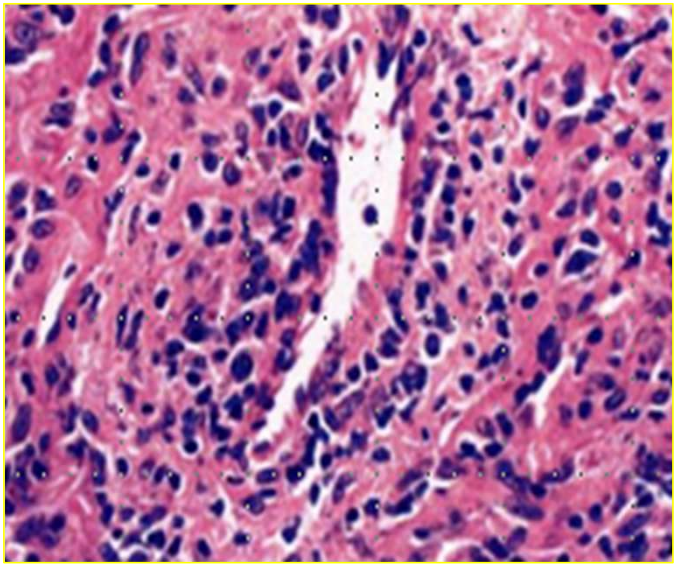
✓ Large B cells: CD20 + / EBV + / CD30 -(+) / CD15 -

✓ Small lymphocytes: CD3 + (CD4 > CD8)

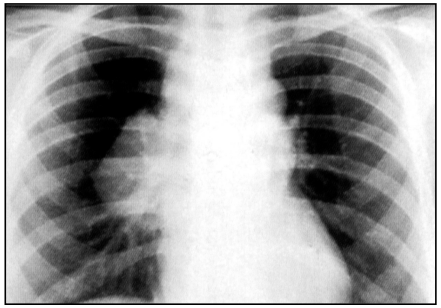


Lymphomatoid Granulomatosis





Primary mediastinal (thymic) large B-cell lymphoma (PMBCL)



Epidemiology

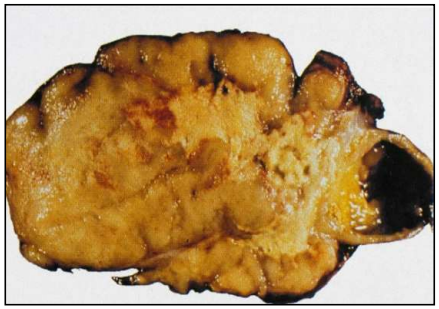
2-4% of non-Hodgkin lymphomas,
median age 35 yrs, F/M (2:1)

Localization

- **Anterior mediastinum** thymus, fat, lung, ribs,pericardium, supraclavicular lymph nodes
- **Stage I-II** at dianosis (bone marrow infiltration very rare)
- Extension or relapse **in extranodal sites**

Cell of Origin

Thymic asteroid B cell



Clinical features

- Vena cava syndrome
- Respiratory tract obstruction
- Pleural/pericardial effusion

Primary mediastinal (thymic) large B-cell lymphoma (PMBCL)

Morphology

- Diffuse infiltration pattern
- Neoplastic cells with round or pleomorphic nuclei and clear or pale cytoplasm
- Variable compartmentalizing sclerosis
- HRS like cells occasionally
- Variable inflammatory cells

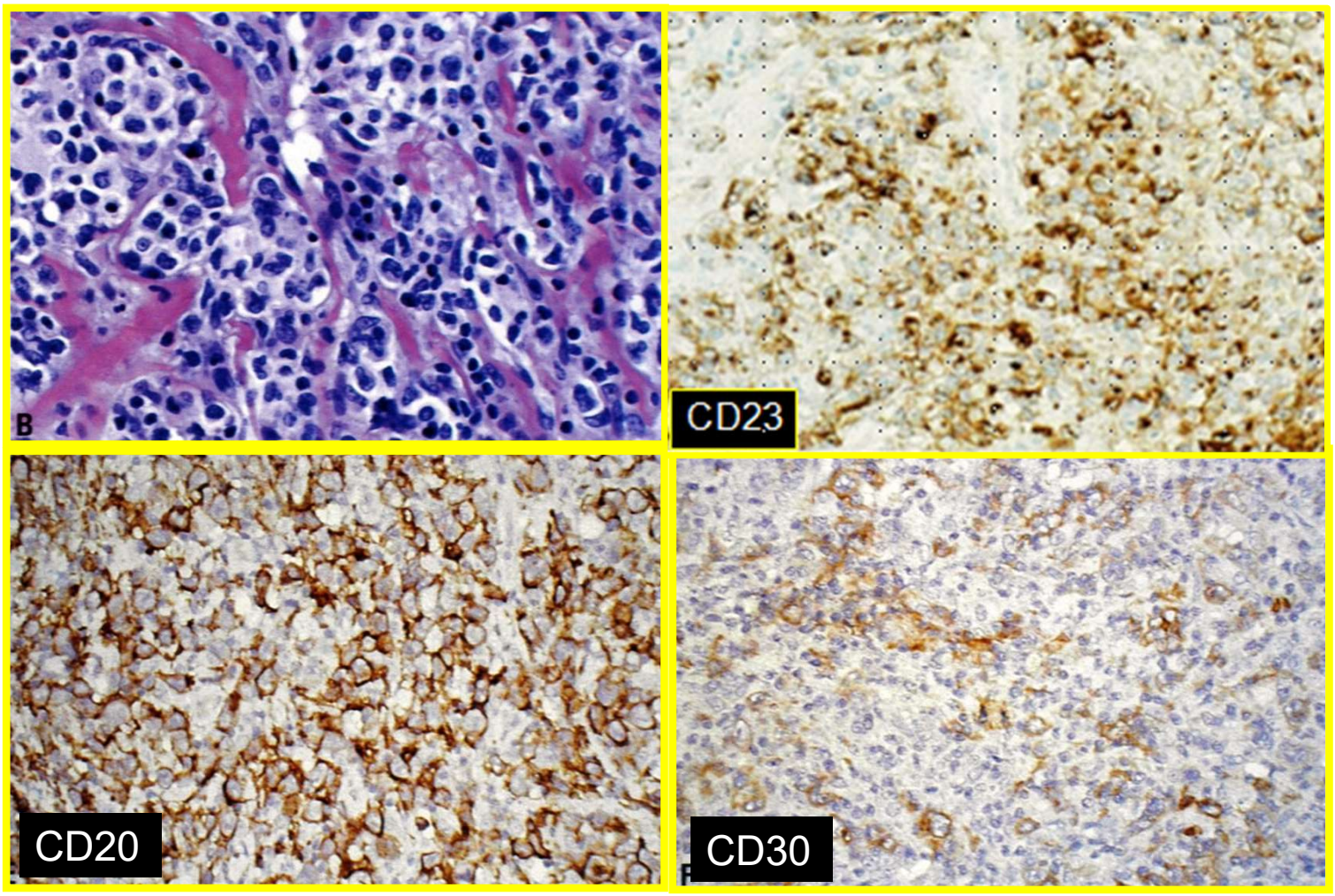
Immunphenotype

- CD20+, CD79a+, PAX-5+, CD23+, CD30+, MUM-1+, CD15-, CD45+ , p63 +

} Post-follicular B cell

Gene expression profile

Similarities with cHL [IgH rearrangements+, 2p (cREL, BCL11) and 9p (JAK2, PDL1. PDL2) gains, NF-kB nuclear overexpression]


B
CD23
CD20
CD30

DLBCL- special subtypes

Lymphomas with plasmablastic features

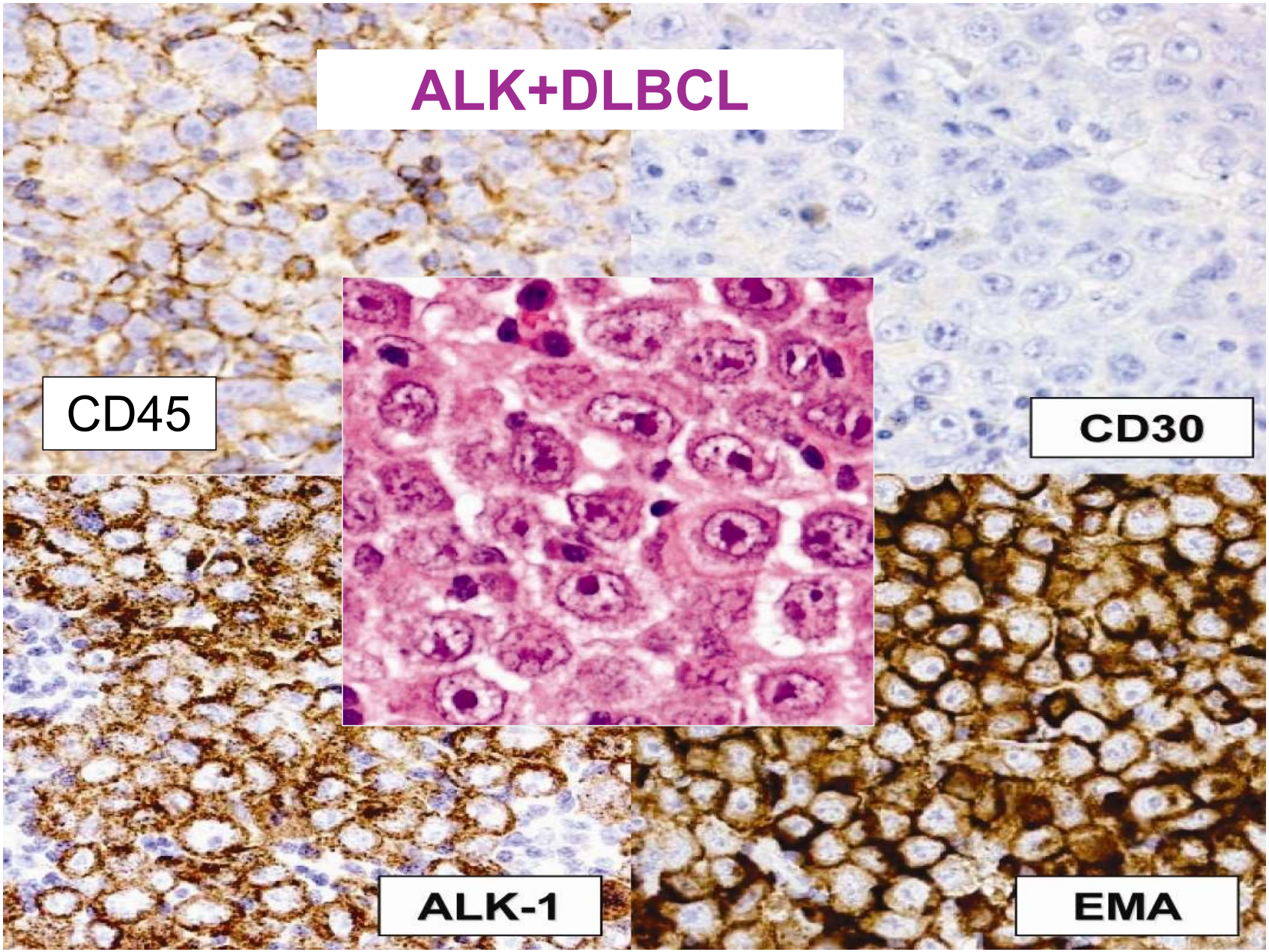
- Plasmablastic lymphoma
- Primary effusion lymphoma (PEL)
- HHV8+ DLBCL
- ALK + DLBCL

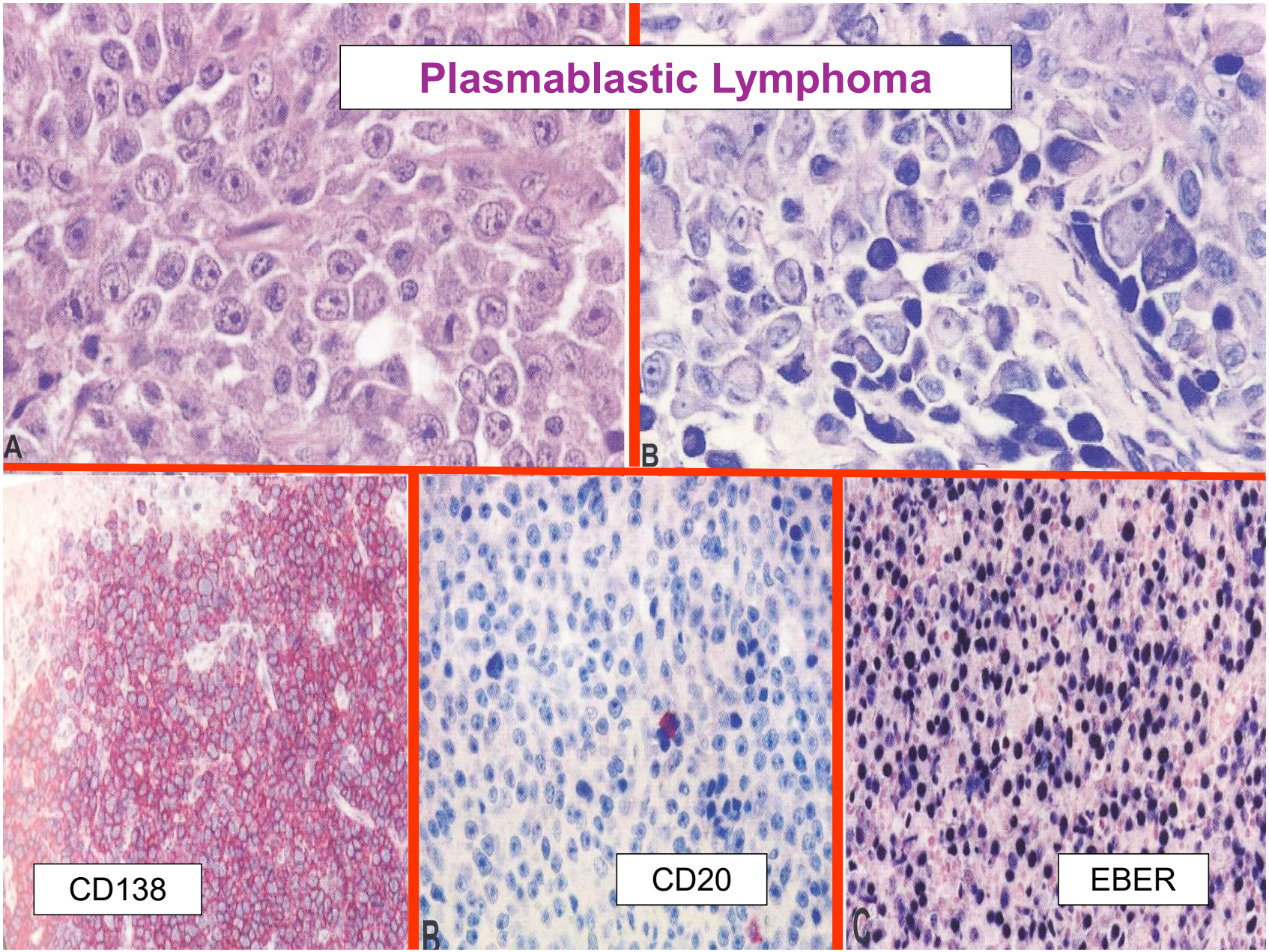
* Plasma cell marker expression
* No B cell markers

Lymphomas with plasmablastic features

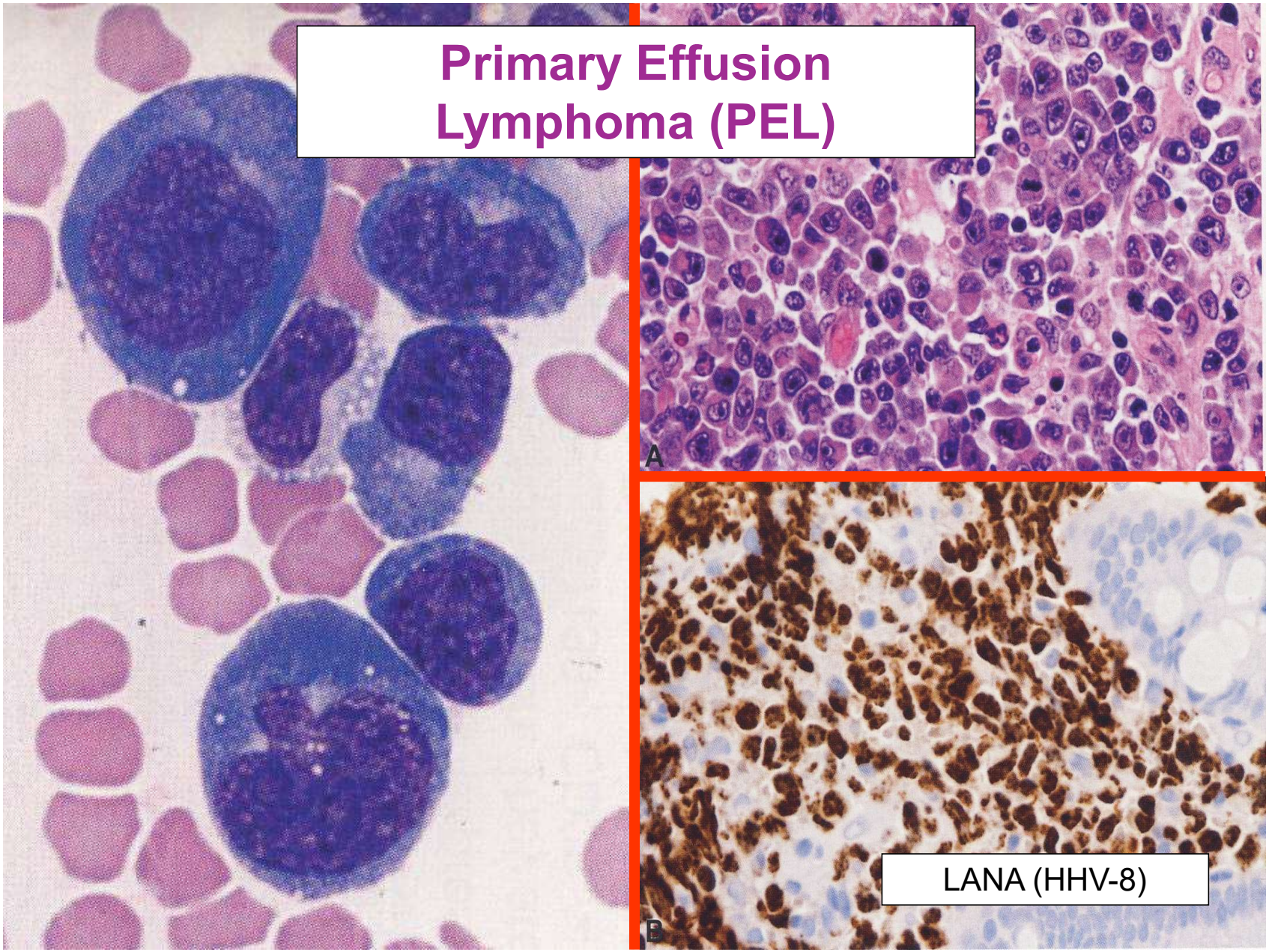
	ALK+DLBCL	Plasmablastic Lymphoma	Primary Effusion Lymphoma (PEL)	HHV8+ DLBCL
Adult men	+	+	+	+
HIV	-	+	+	+
Other immunodeficiancy	-	+/-	+/- after transplant	+
HHV-8	-	-	+	+/-
EBV	-	+	+	-
Morphology	immunoblastic +/- plasmablastic			
B-cell markers	No CD20 / PAX-5			
Plasma cell markers	Positive: CD138 / CD38 / Mum1			CD38/Mum 1 +
	CIgA+	CIgG+/-	SIg/CIg-	CIgM(lamda)+
Other important markers	ALK+	EBER	LANA (HHV-8)	
Other markers	EMA+CD30-CD45-/+	EMA+ CD30+ CD45-/+	EMA+ CD30+ CD45+	
Localization	Lymph nodes	-- Oral mucosa -- Extranodal sites	Body cavities	Lymph nodes/spleen
Prognosis	Poor			

ALK+DLBCL




Plasmablastic Lymphoma
A
B
CD138
CD20
B
EBER
C

Primary Effusion Lymphoma (PEL)



T-cell and NK-cell lymphoid proliferations and lymphomas

Definition: Neoplasms stemming from mature, post-thymic cells

- **T-cell lymphomas with leukaemic presentation**
  - T-prolymphocytic leukaemia (T-PLL)
  - T-large granular lymphocytic leukaemia (T-LGL)
  - NK-large granular lymphocytic leukaemia
  - Aggressive NK-cell leukaemia
  - Sezary syndrome
  - Adult T-cell leukaemia/lymphoma (ATLL)

- **T-cell nodal lymphomas**
  - Nodal T-follicular helper cell lymphomas (nTFH)(Nodal TFH cell lymphoma, angioimmunoblastic-type)
  - Peripheral T-cell lymphoma, NOS
  - ALK-positive anaplastic large cell lymphoma
  - ALK-negative anaplastic large cell lymphoma

- **Primary cutaneous T-cell lymphomas**
  -Mycosis fungoides
  -Primary cutaneous CD30-positive T-cell lymphoproliferative disorders
  -Primary cutaneous gamma/delta T-cell lymphoma
  -Subcutaneous panniculitis-like T-cell lymphoma

- **Other extranodal T-cell lymphomas**
  -Extranodal NK/T-cell lymphoma
  -Enteropathy-associated T-cell lymphoma
  -Monomorphic epitheliotropic intestinal T-cell lymphoma
  -Hepatosplenic T-cell lymphoma

Peripheral T/NK cell lymphomas

Under question:

- Molecular pathogenesis?
- Cell of origin?
- Drug resistance?

Signs and symptoms raising suspicion for T-cell lymphoma

Clinical features	Skin or lung involvement Hypercalcemia
Nodal architecture	**Paracortical involvement.** Follicles are intact Conspicuous **vessels** Compartmentalising fibrosis
Cell cytology	Irregular **nuclear shape** (cerebriform, multilobular, nuclear grooves) Large amounts of **clear** cytoplasm
Reactive microenvironment	Interdigitating reticular cells **Eosinophils** Epithelioid histiocytes Plasma cells
Immunophenotype	**Abnormal T-cell phenotype** (e.g. CD4+CD8+, or CD2-CD7-)

T cell lymphomas of the small bowel

1. Enteropathy-associated T-cell lymphoma (EATL)
2. Monomorphic epitheliotropic intestinal T-cell lymphoma (MEITL)

WHO 2022

Enteropathy-associated T-cell lymphoma (EATL)

- **Definition:** Primary small bowel lymphoma stemming from **intraepithelial T-cell & often occurs in individuals with coeliac disease (CD).**
- **Frequency**: The most common T-cell lymphoma of the GI tract
- **Etiology: Related to coeliac disease in 80-90% of cases**
- **Localization:** Small bowel (jejunum-ileum) – Rare in the rest GI tract
- **Clinical features**: Clinical history of coeliac disease – Small bowel perforation due to lymphoma involvement – Coeliac sprue that did not answer to gluten-free diet

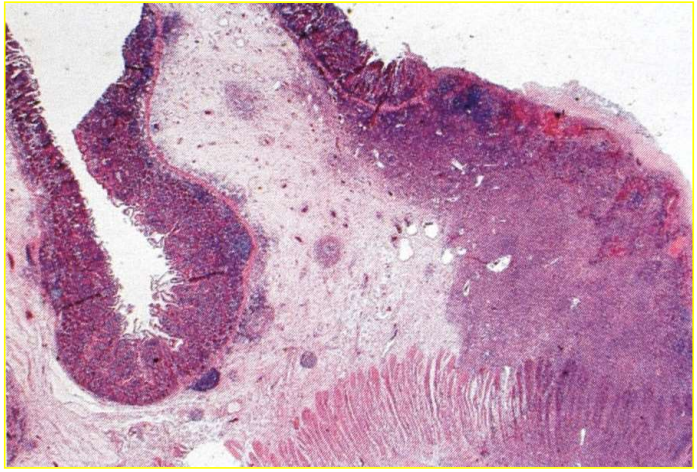
Intestinal T-cell lymphomas –Main types

	EATL	MEITL
Frequency	80-90%	10-20%
Morphology	Large-anaplastic cells, conspicuous nucleolus, abundant cytoplasm	Medium-sized cells with pyknotic nucleus
Cryptic involvement	+	+
Reactive cells	Often conspicuous	Absent
Distant mucosa	Villi are atrophic, intraepithelial T cells	Villi are normal, intraepithelial T cells
Clinical features	Coeliac disease, dermatitis herpetiform	Not related to coeliac disease
Molecular features	JAK1 and STAT3 mutations, 9q34.3 gains and 16q12.1 deletions	SETD2, JAK3, STAT5B mutations, 9q34.3 gains and 16q12.1 deletions

EATL –Morphology

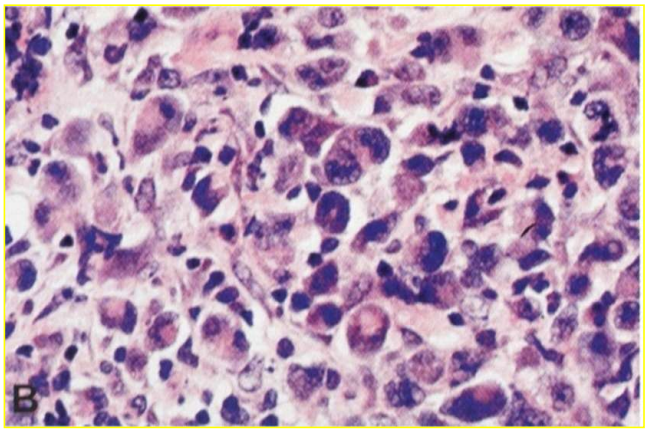
Multiple ulcers

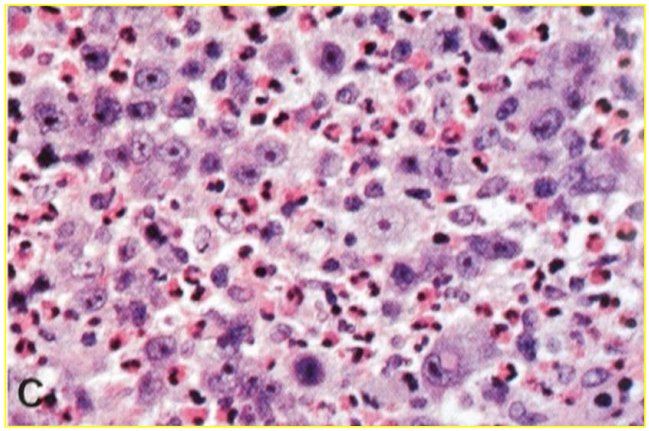
Massive small bowel wall involvement



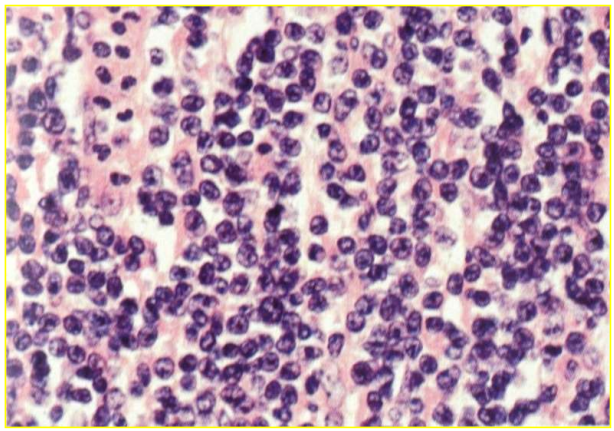
Intestinal T-cell lymphomas – Cytomorphology

EATL
(non-monomorphic)





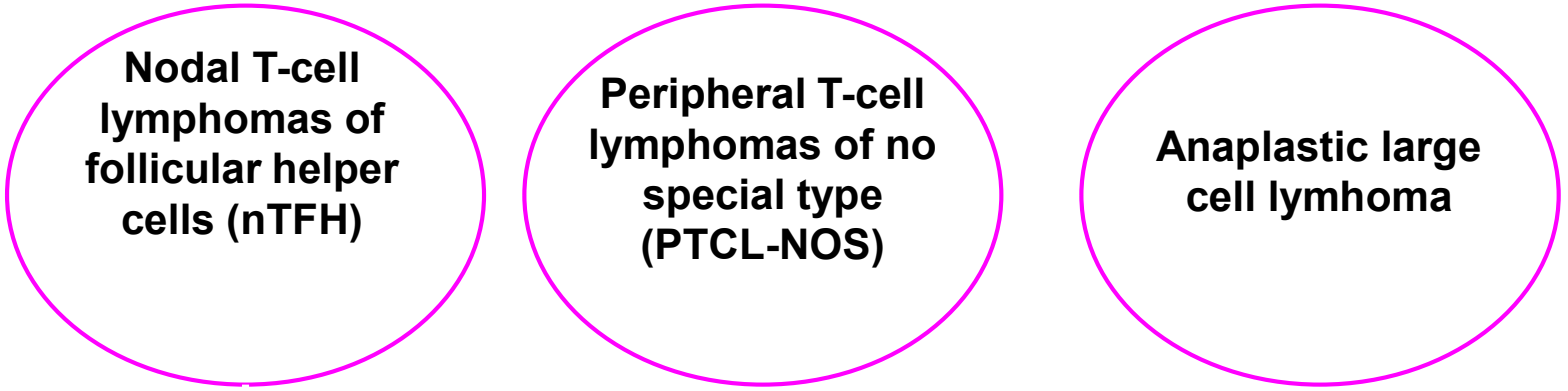
MEITL
(monomorphic)



Intestinal T-cell lymphomas –Immunophenotype

EATL	MEITL
CD3+ / CD5 - / CD7+	CD3+
CD56-	CD56+
CD8- , CD4 ±	CD8+, CD4-
TcR b ±	TcR β+
CD103 +	
Cytotoxic molecules +	Cytotoxic molecules +
CD30 +	CD30 –
Intraepithelial T-cells CD8-/CD56-	Intraepithelial T-cells CD8+/CD56+

Nodal T-cell lymphomas



Prototype:

Angioimmunoblastic lymphoma (AITL)

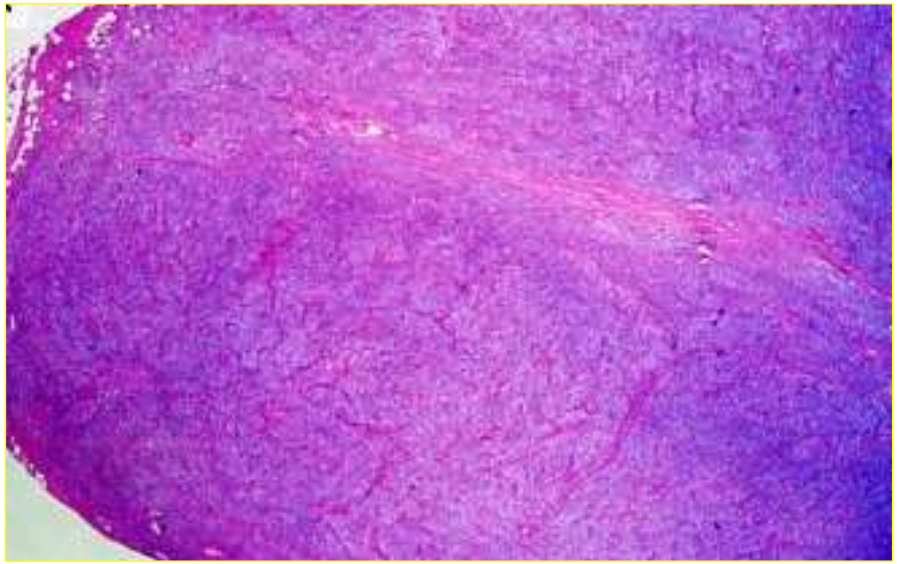
Angioimmunoblastic lymphoma (AITL)- Clinical features

- **Frequency:** 15-20% of T-cell lymphomas / 1-2% of non-Hodgkin lymphomas
- **Age:** Middle age-elderly
- **Gender:** A=F
- **Symptoms:** generalized lymphadenopathy, fever, skin rash and pruritus, oedema, pleural effusion, arthritis, ascites, hepatosplenomegaly

- **Laboratory findings**: polyclonal hyperglobulinaimia, cryoglobulinaemia ($\rightarrow$ haemolytic anaemia), Rheumatoid factor + and anti-smooth muscle antibodies

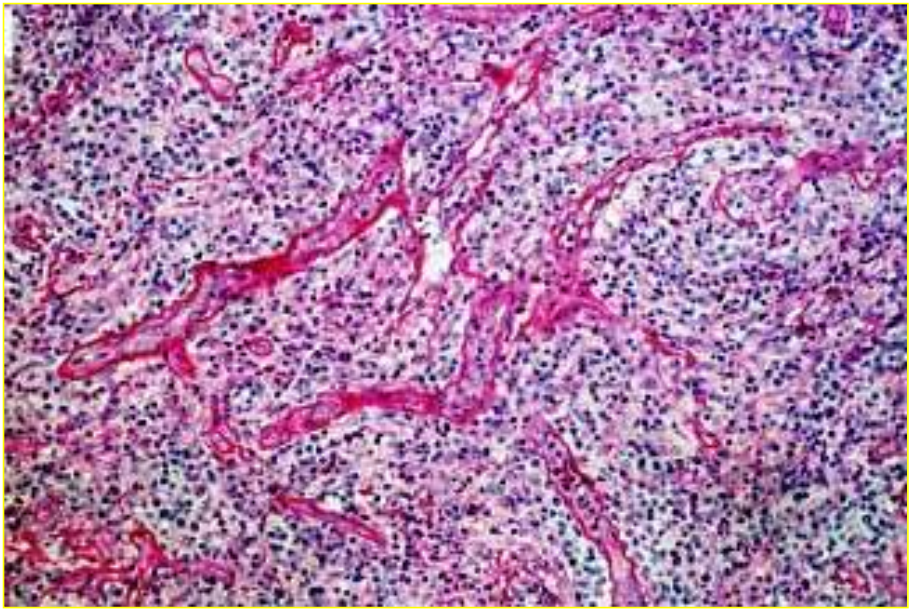
AITL- Morphology

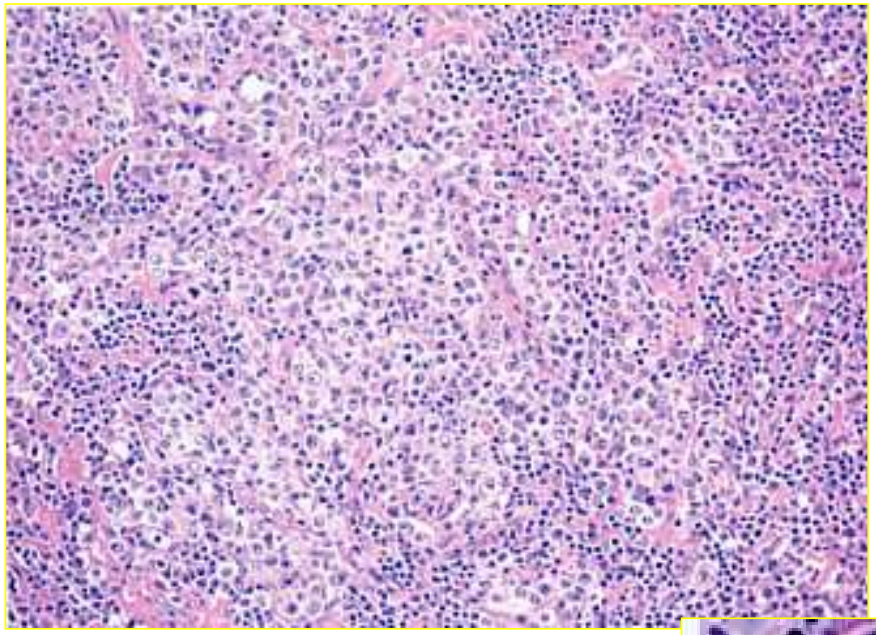
- Partial or total lymph node effacement
  - Pattern I (hyperplastic follicles)
  - Pattern II (atrophic follicles)
  - Pattern III (no follicles)
- Diffuse involvement by polymorphous cell infiltrate
- Medium-large sized neoplastic cell aggregates, often with clear cytoplasm
- Abundant High Endothelial Venules, often branching (arborizing vessels)
- Follicular dendritic cell aggregates, often around vessels
- Reactive cells: plasma cells, epithelioid histiocytes, eosinophils
- Early TET2 and DNMT3A mutations in stem cells, RHOA mutation in ~70% of AITL, IDH2 mutations in AITL but not in other nTFH lymphomas



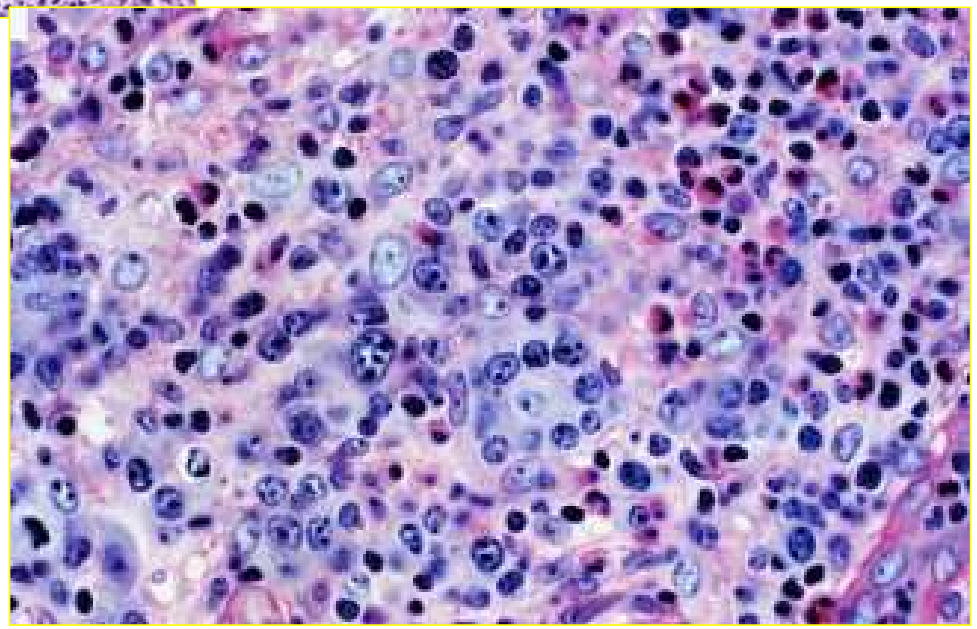
Effacement of the lymph node architecture

Marked High Endothelial Venule (HEV) proliferation

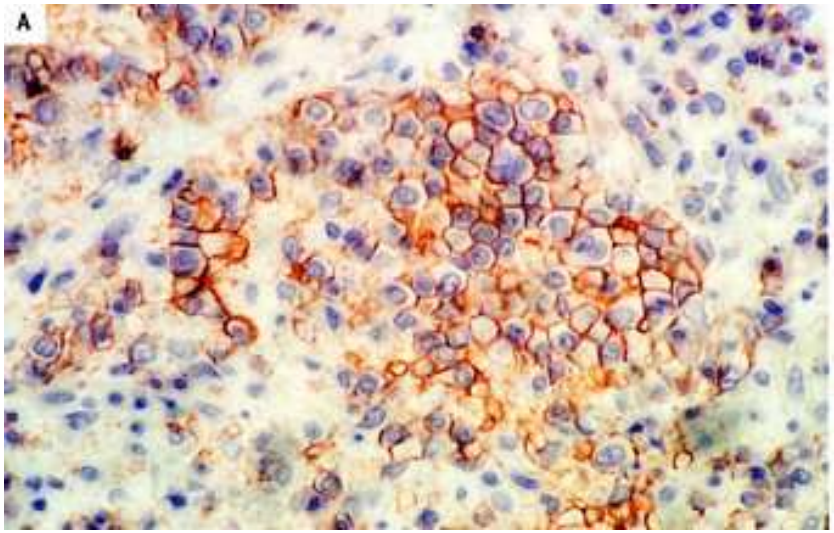




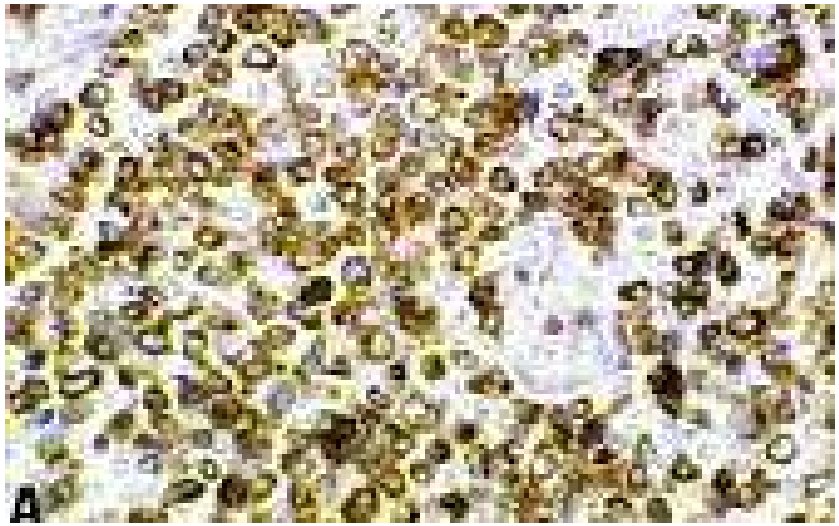
Clear cells



Polymorphous infiltrate



CD3

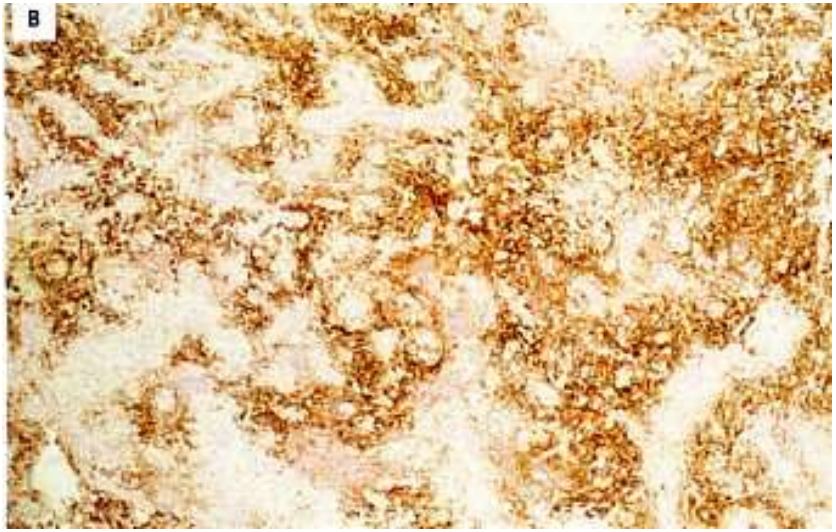


CD3

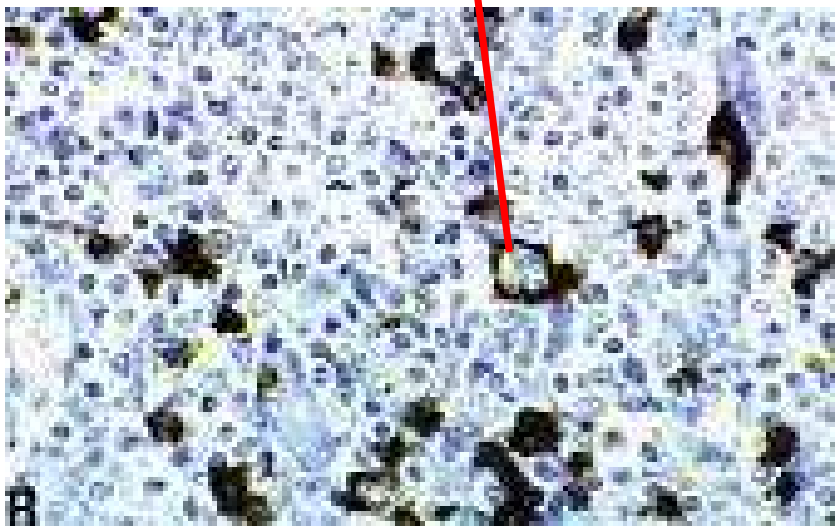
Large B-cells

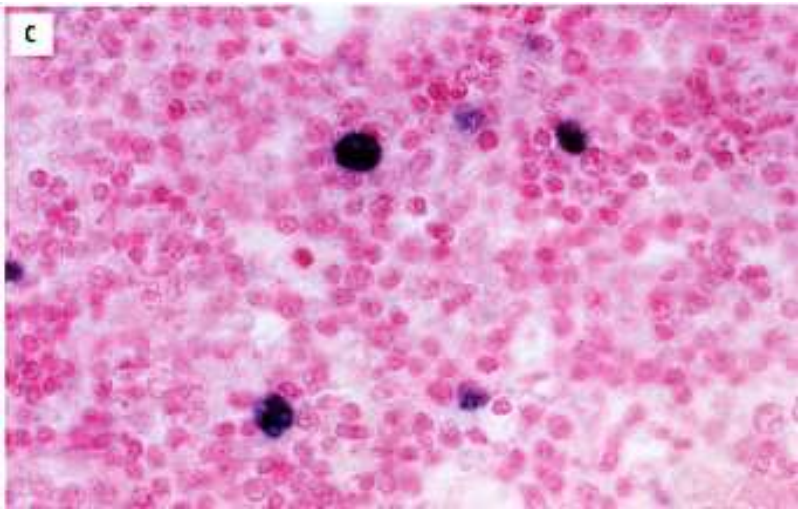
CD21

Follicular dendritic cell expansion (CD21+) entending out of follicles



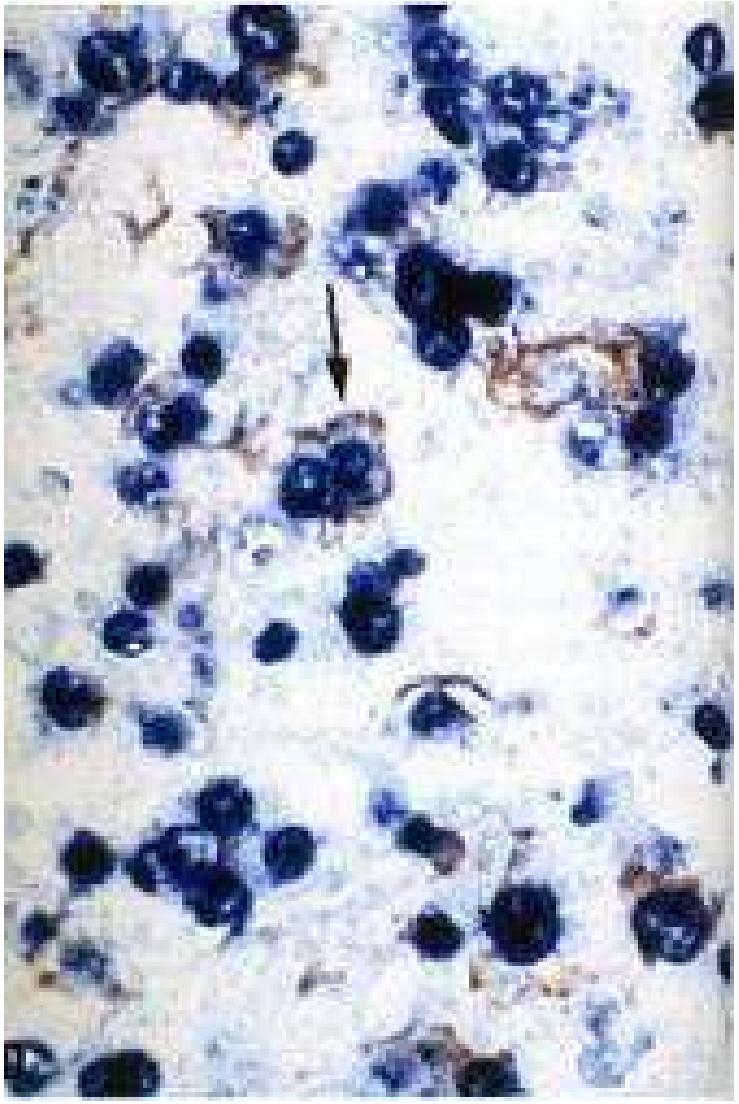
CD20





EBER

B-cells are **EBER (mRNA EBV)+**

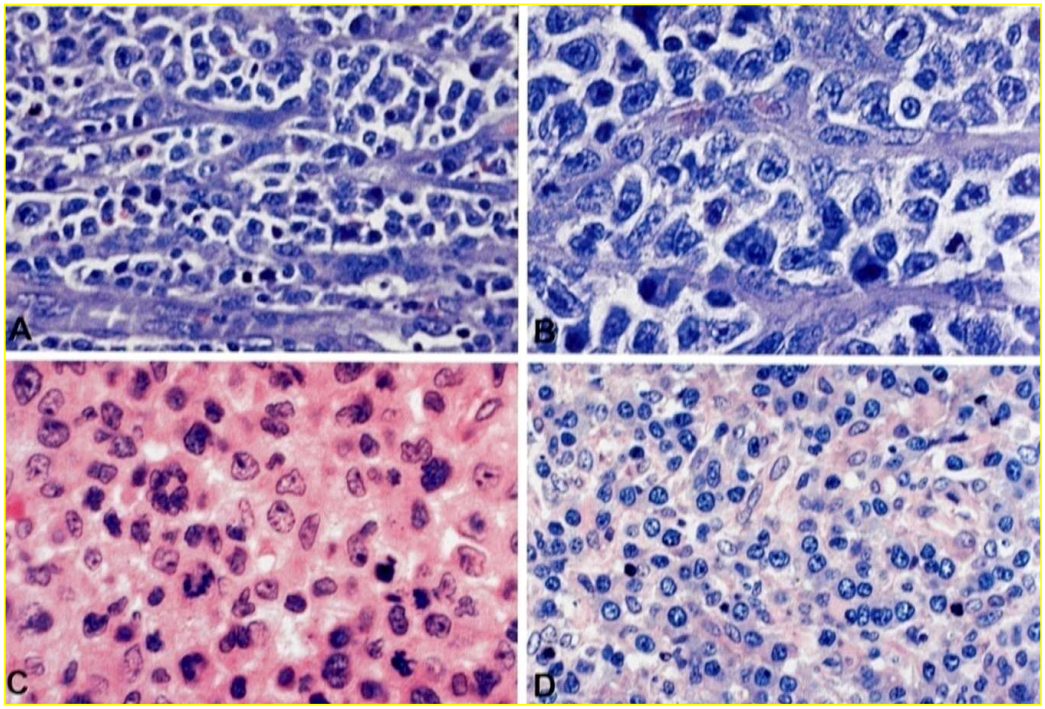


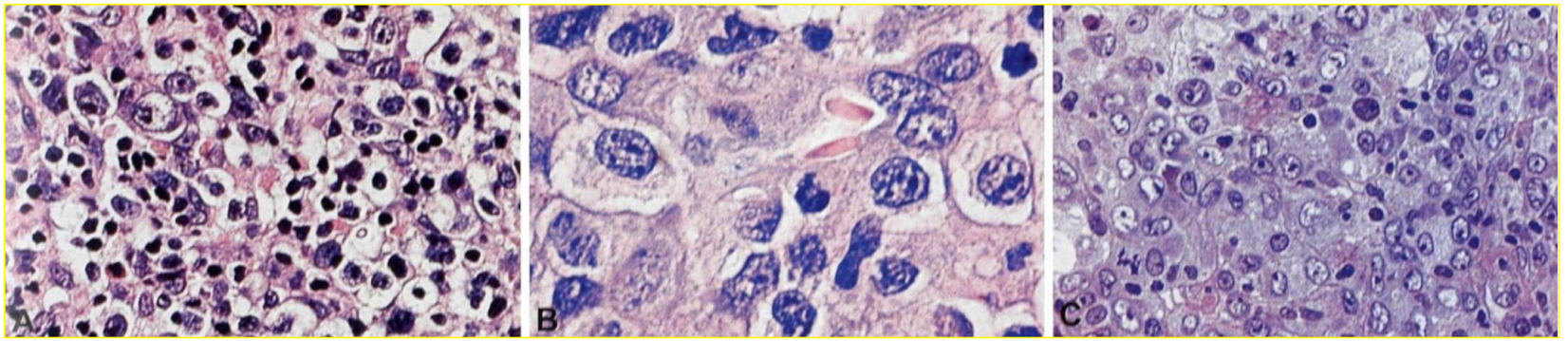
EBER/CD20

Peripheral T cell lymphoma (PTCL, NOS)

- **Definition: Heterogenous nodal and extranodal T-cell lymphoma category, not fitting in other well defined entities**
- **Frequency: 30% of T-cell lymphomas**
- **Age: Adults >>> children**
- **Gender: M/F = 2/1**
- **Localization: lymph nodes, bone marrow, liver, spleen, other extranodal sites (mainly skin, GI tract)**
- **In extranodal sites, the diagnosis requires exclusion of other defined entities**
- **Clinical features: lymphadenopathy, B symptoms, eosinophilia, pruritus**
- **Provisional classification in GATA3+ and TBX21+ subgroups according to molecular profile, with prognostic implications**

PTCL, NOS – variable histology





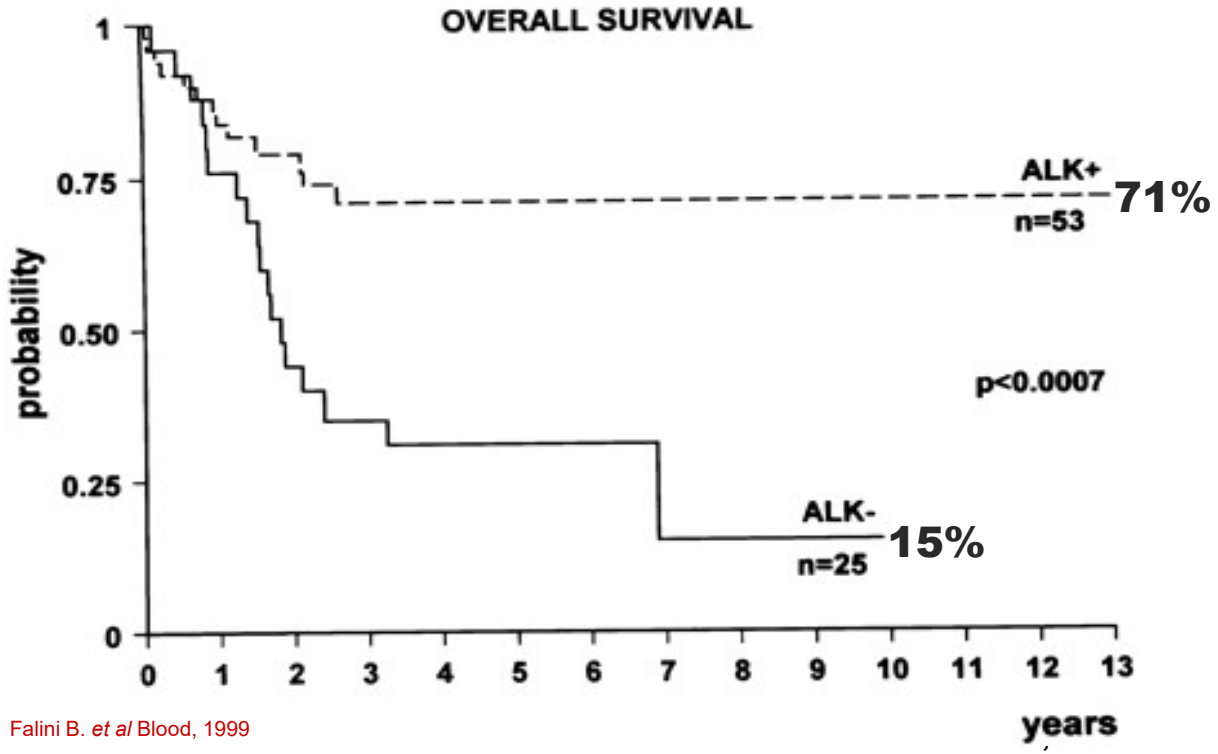
Anaplastic Large cell Lymphoma (ALCL)

Nodal lymphoma characterized by:

- **Pleomorphic large T/null -cells CD30+, often involving lymph node sinuses**
- **50-70% are bearing translocations that lead to ALK gene overexpression**

ALK expression is related to better prognosis

ALK+	ALK-
• First 3 decades	• Older age
• M:F = 6.5	• M:F = 0.9
• Favourable prognosis	• Unfavourable prognosis



Falini B. *et al* Blood, 1999

ALK+ ALCL

Morphology

Classic (80%)

Common types (»15%)

Lymphohistiocytic

Small cell

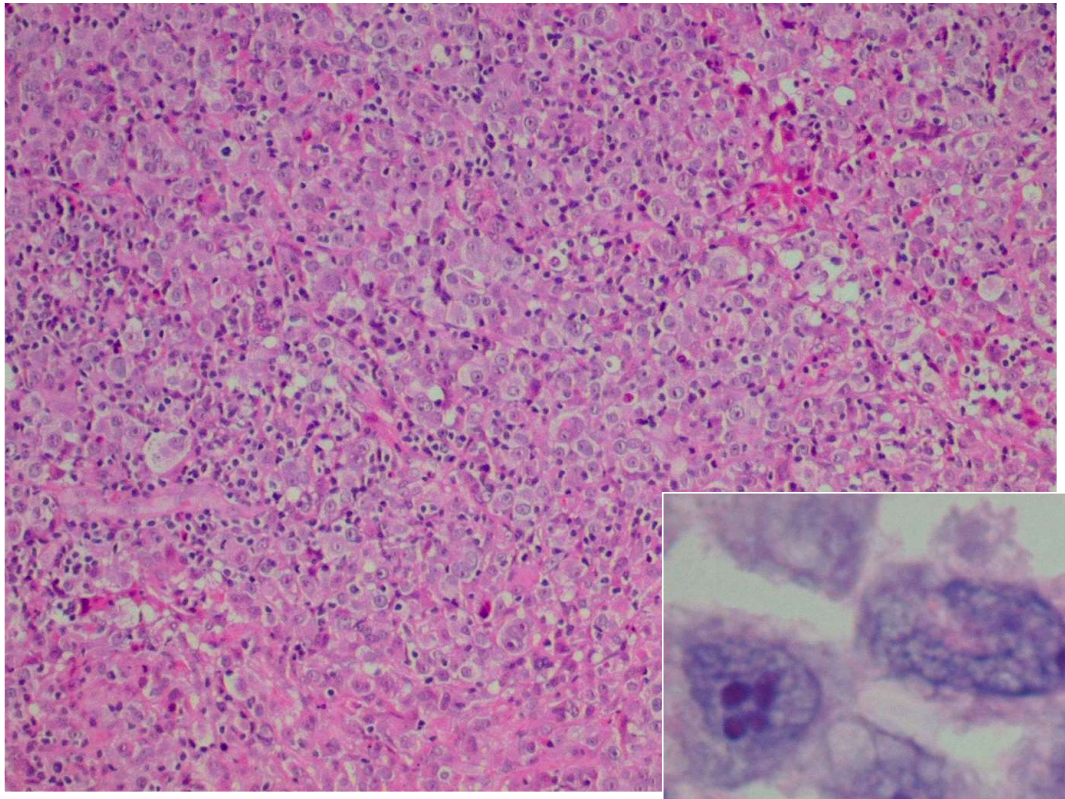
Rare types (<5%)

-with high numbers of neutrophils

-with high numbers of eosinophils

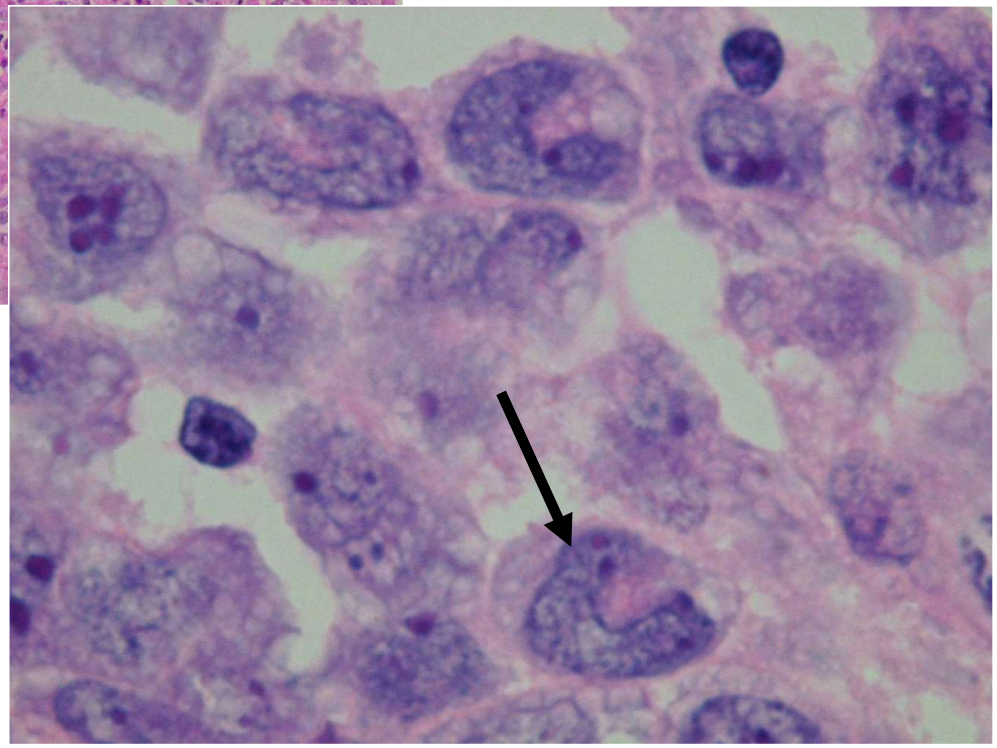
-with high numbers of giant cells

-sarcomatoid

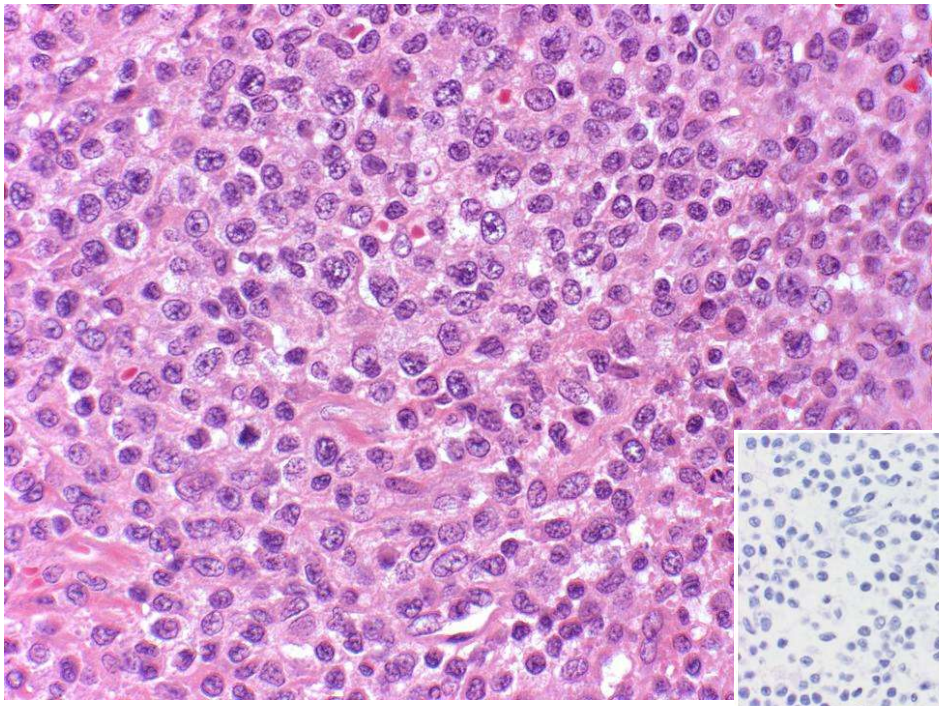


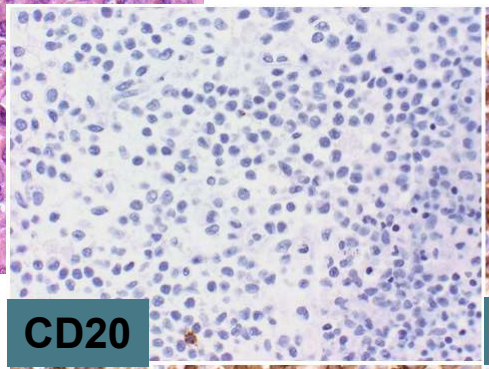
ALK+ ALCL,
Classic type

Hallmark cells

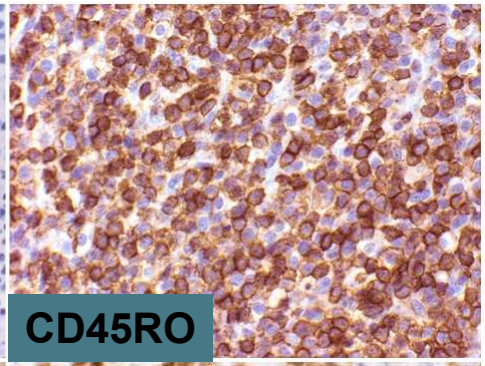


ALK+ ALCL, small cell type

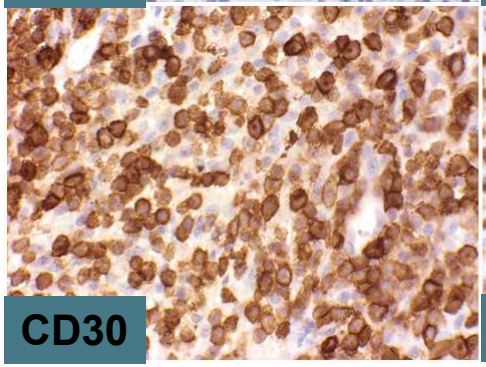




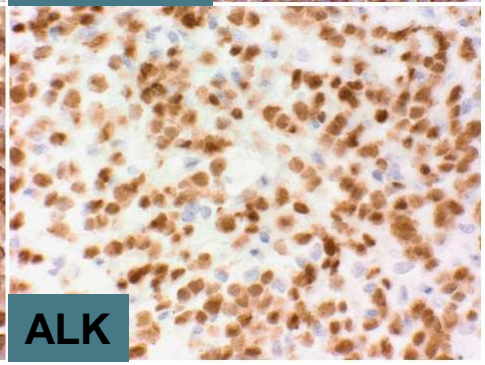
CD20



CD45RO

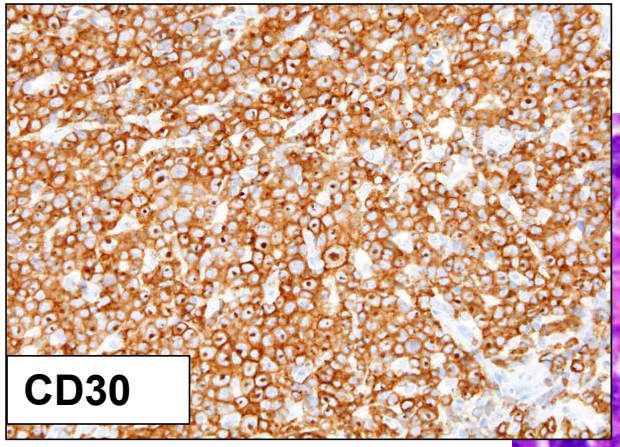


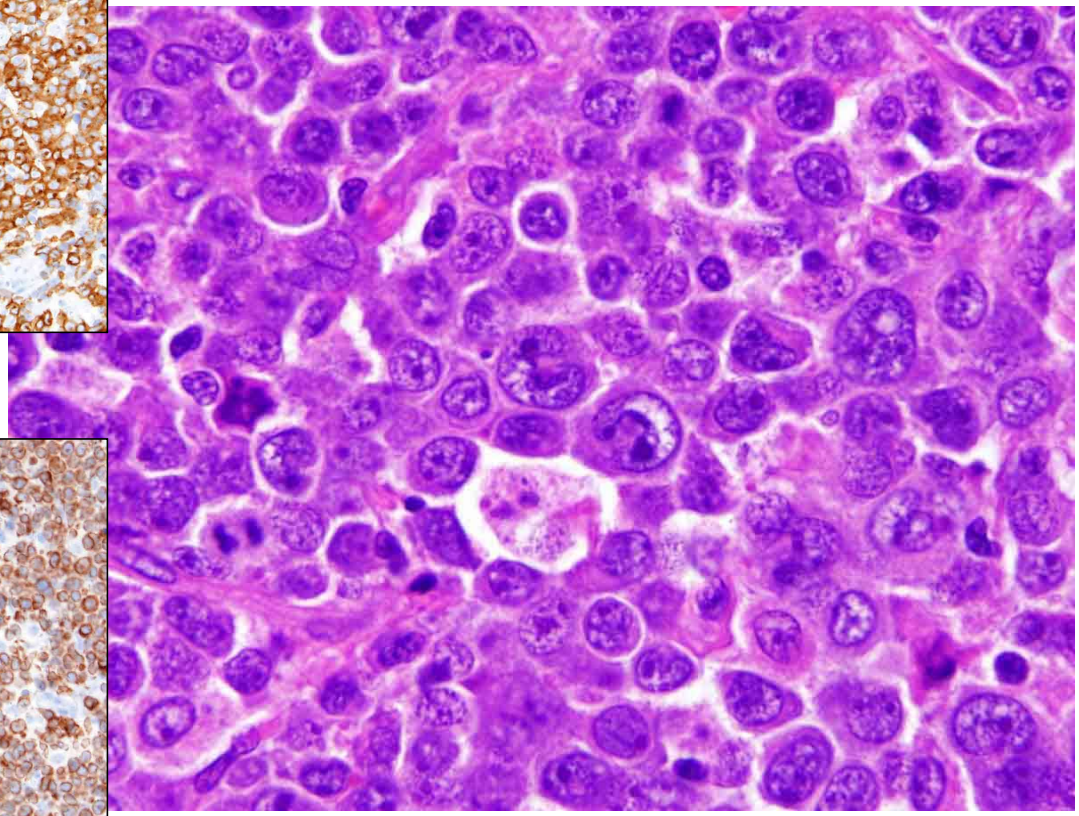
CD30

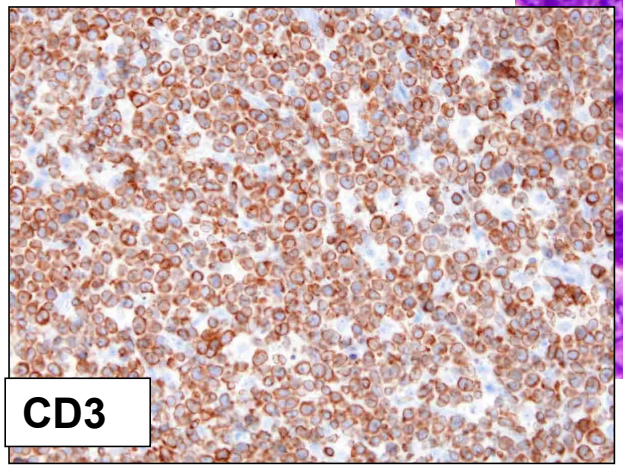


ALK

ALCL







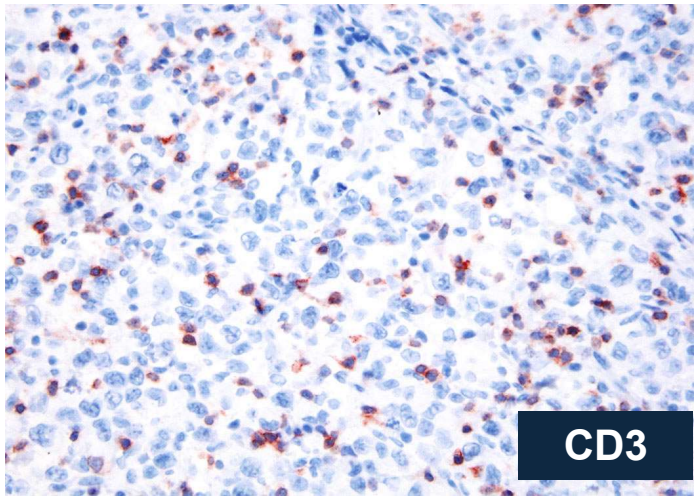
ALCL - Immunophenotype

T-cell phenotype

Positivity in T-cell antigens
(e.g. CD2, CD43, CD45RO)

Often CD3-, CD5 -

TCR rearrangement



«Null» - phenotype

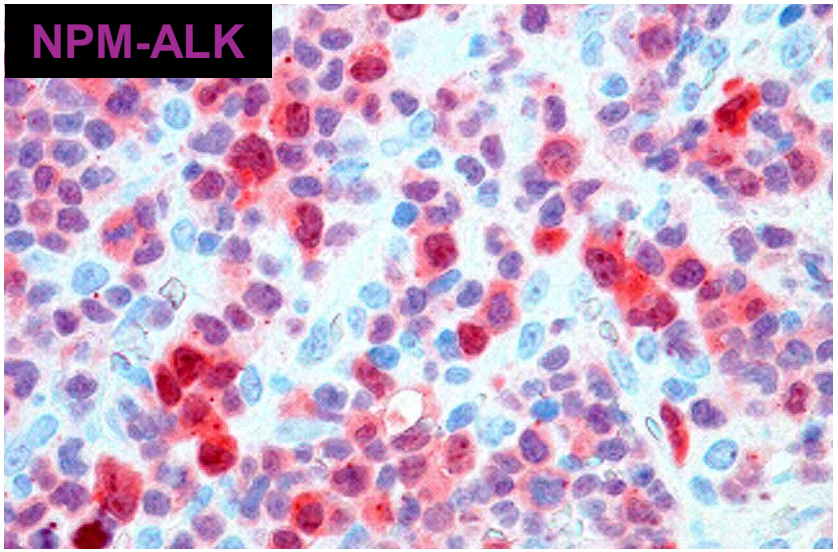
T-cell antigens negative

TCR rearrangement

ALK chimeric protein leads to STAT3 activation

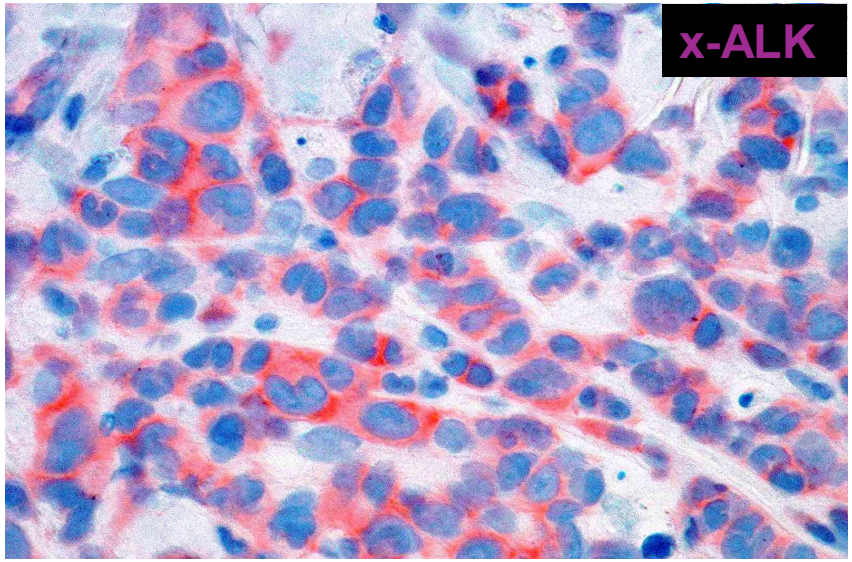
NOTCH1 mutations

ALK immunophenotypic pattern in ALCL



Cytoplasmic and nuclear staining

Cytoplasmic staining



Primary cutaneous T-cell lymphomas

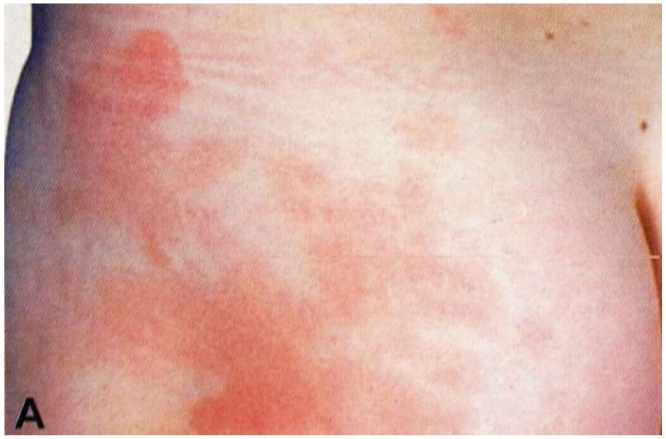
Definition: Extranodal non-Hodgkin lymphomas stemming from malignant clonal T-cells, involving the skin

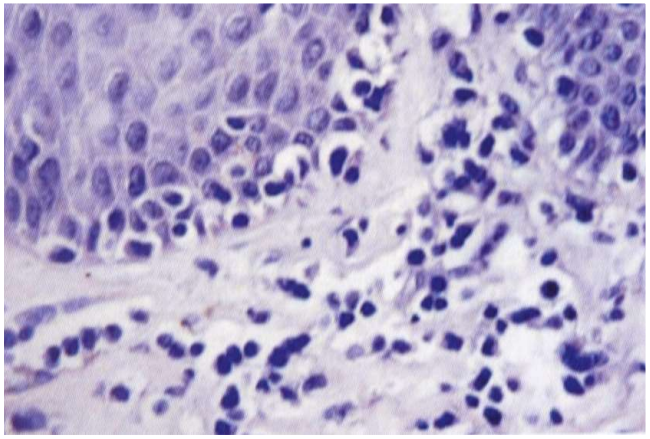
WHO – EORTC classification of primary skin T and NK lymphomas

Lymphoma type	Frequency (%)	5-yr survival (%)
Mycosis Fungoides subtypes		
Classic	**44**	**88**
Folliculotropic	**4**	**80**
Pagetoid reticulosis	**<1**	**100**
Granulomatous slack skin	**<1**	**100**
Sezary syndrome	**3**	**24**
Adult T-cell leukaemia/lymphoma		
Primary cutaneous CD30-positive T-cell lymphoproliferative disorders	**20**	**95-100**
Subcutaneous panniculitis-like T-cell lymphoma	**1**	**82**
Extranodal NK/T-cell lymphoma	**<1**	**Επιθετικό**
Rare primary skin T-cell lymphoma types, not further classified	**<6**	**16-75**

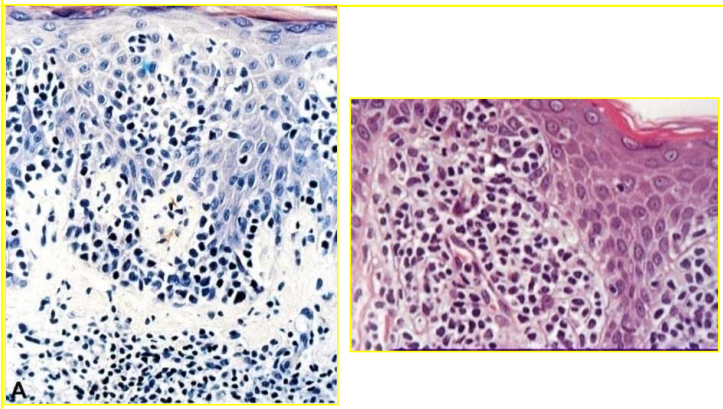
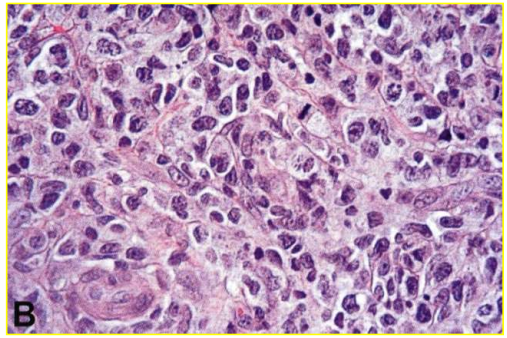
Mycosis Fungoides

- **Most common type of primary cutaneous T-cell lymphoma**
- **Adults, usually elderly, M>F**
- **Pruritic dermal lesions: patches → plaques → tumours**
- **Morphology**
- **Patches: Upper dermis and basal epidermal layer infiltrated by small lymphoid cells with cerebriform (hyperchromatic hyperconvoluted) nuclei**





Mycosis Fungoides – Morphology cont'd

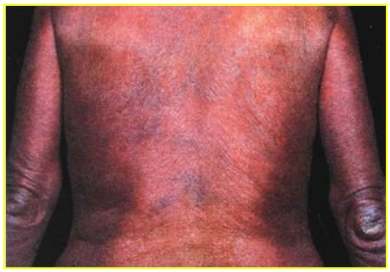
Plaques:	**Tumours**:
Conspicuous epidermotropism **Pautrier microabscesses** **Denser dermal infiltrates**	**No conspicuous epidermotropism** **Dense dermal infiltrate and extension to subcutis** **Increase in large cells (<25%)**
	

immunophenotype: CD2, 3, 4, 5+ /CD7-

CD8- (rarely +)

CD56 & cytotoxic molecules rarely +

Sezary Syndrome



- **Clinical features**

Adults, usually elderly

Pruritic erythroderma

Generalized lymphadenopathy

- **Clonal T-cell population in peripheral blood, skin and lymph nodes**
- **At least 1 of the following:**

CD4/CD8 >10 in peripheral blood

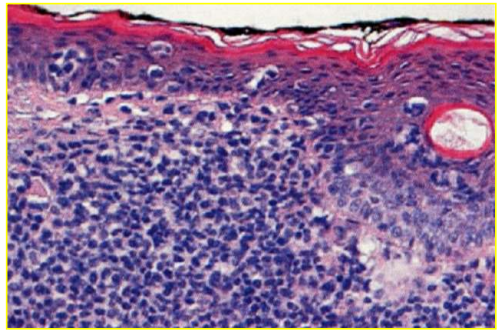
Sézary cells in peripheral blood >1000/mm^2

Loss of T antigens (CD7)

- **Clinical Features**

Epidermal/dermal involvement by small atypical lymphoid cells

1/3 of cases: no specific findings



- **Immunophenotype**

CD2, CD3, CD4, CD5+/CCR4+

CD7, CD8-

Primary cutaneous anaplastic large T-cell lymphoma (C-ALCL)

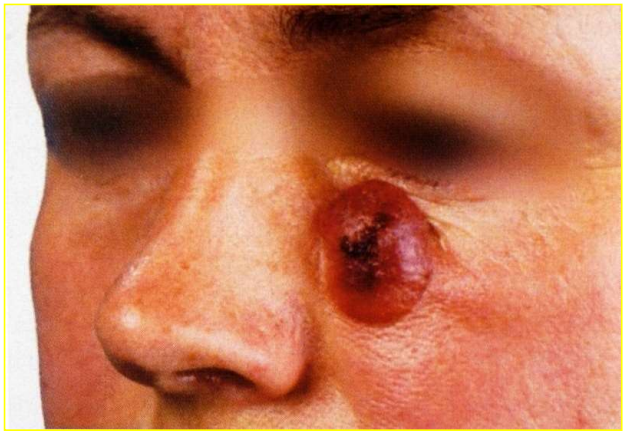
Definition:

- **Large anaplastic cells**
- ***>75% of cells CD30+***
- **No clinical history or clinical features of mycosis fungoides (→ tumour stage) or other lymphoma (→ secondary ALCL)**
- **No nodal ALCL involving the skin**

C-ALCL

Clinical features

- Adults, M>F
- Single or localized lesions, often ulcerated >2cm
- Multifocal lesions 20%
- Partial/full regression is possible

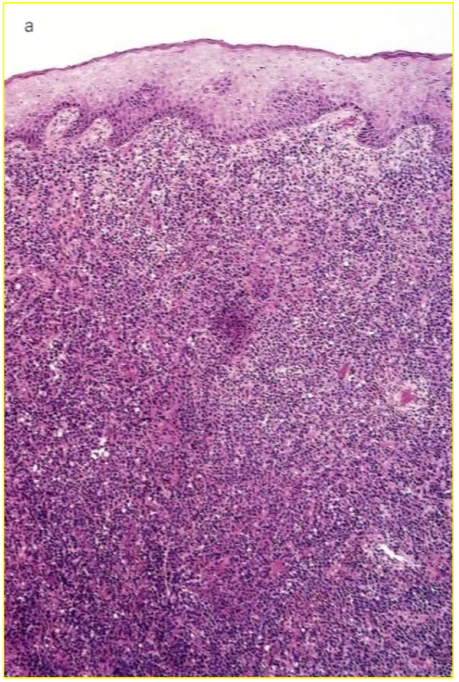


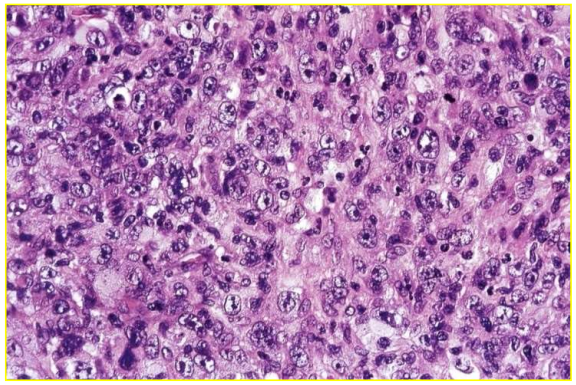
C-ALCL

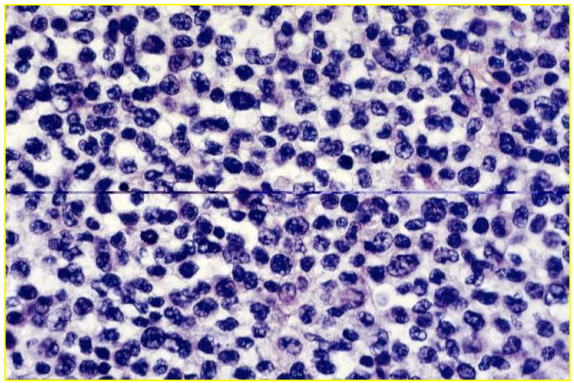
Morphology

- **Nodular/diffuse infiltration by large/anaplastic cells**
- **Usually no epidermotropism**

  DD. from LyP (lymphomatoid papylosis)
- **Reactive infiltrates (sometimes conspicuous): neutrophils, eosinophils, histiocytes, small lymphocytes**
- **Sometimes epidermis shows hyperplasia**





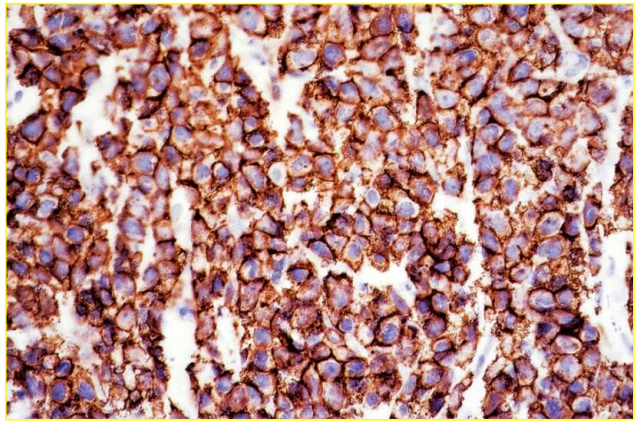


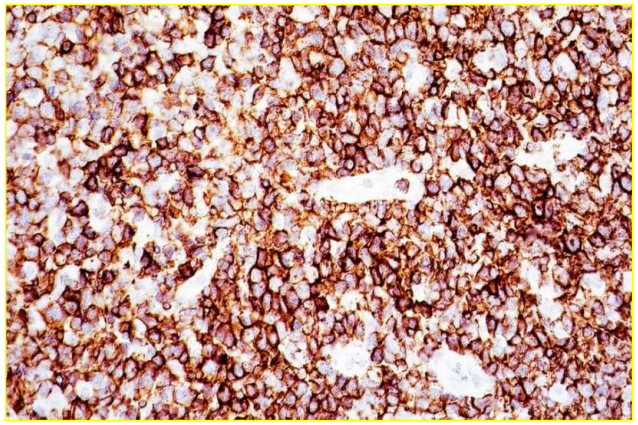
C-ALCL

Immunophenotype -Genotype

- CD2/CD5/CD3+ (variable expression)
- CD4 + >>> CD8+
- CD30+
- CD15- / EMA- / ALK-
- Cytotoxic molecules: usually +
- CD56: rarely +
- No t (2;5) – Presence of TcR rearrangements

CD30





CD4