Lymphoma Diagnosis

JAN DELABIE
OSLO UNIVERSITY HOSPITAL
UNIVERSITY OF OSLO
W.H.O. CLASSIFICATION OF LYMPHOMA: HISTORY

1994: Revised European-American Lymphoma Classification

2001: W.H.O. Lymphoma Classification

2005: W.H.O.-E.O.R.T.C. Classification for skin lymphomas

2008: W.H.O. Lymphoma Classification including skin lymphomas
W.H.O CLASSIFICATION OF CUTANEOUS LYMPHOMA

- PRIMARY CUTANEOUS LYMPHOMAS THAT REPRESENT ENTITIES ALSO RECOGNIZED OUTSIDE THE SKIN
  - FOLLICULAR LYMPHOMA
  - MARGINAL ZONE LYMPHOMA
  - DIFFUSE LARGE B CELL LYMPHOMA, LEGG TYPE
  - ANAPLASTIC LARGE CELL LYMPHOMA
W.H.O CLASSIFICATION OF CUTANEOUS LYMPHOMA

- PRIMARY CUTANEOUS LYMPHOMAS THAT HAVE NO NODAL COUNTERPART
  - MYCOSIS FUNGOIDES
  - SéZARY SYNDROME
  - SUBCUTANEOUS PANNICULITIS-LIKE T CELL LYMPHOMA
  - GAMMA-DELTA T-CELL LYMPHOMA
  - CD8 POSITIVE AGGRESSIVE EPIDERMOTROPIC CYTOTOXIC T CELL LYMPHOMA
  - CD4 POSITIVE SMALL/MEDIUM T CELL LYMPHOMA
W.H.O CLASSIFICATION OF CUTANEOUS LYMPHOMA

- PRIMARY CUTANEOUS MYELOID NEOPLASIA
  - BLASTIC PLASMACYTOID DENDRITIC CELL NEOPLASM
A. LYMPHOMAS ARE CLASSIFIED ACCORDING TO THE MATURATION STAGES OF NORMAL LYMPHOCYTES (‘frozen stage model’)
FROZEN STAGE MODEL

Cortical Thymus
- TdT
- HLA-DR
- CD34
- cCD-3
- CD7
- CD2
- CD5
- CD4
- CD8

Medullary Thymus
- Tdt
- HLA-DR
- CD34
- cCD-3
- CD7
- CD2
- CD5
- CD1
- CD4
- CD8

Blood, lymphoid tissue
- Helper-Inducer T-cell
- Regulatory T-cells (CD25+/FoxP3+)
- Follicular helper T-cell (PD1+)
- Suppressor-Cytotoxic T-cells

TCRδ
TCRγ
TCRβ
TCRα
FROZEN STAGE MODEL

Cortical Thymus

Medullary Thymus

Blood, lymphoid tissue

MYCOSIS FUNGOIDES
ADULT T-CELL LEUKEMIA-LYMPHOMA
ANGIO-IMMUNOBLASTIC T-CELL LYMPHOMA
PERIFOLLICULAR T-CELL LYMPHOMA
ENTEROPATHY-ASSOCIATED T-CELL LYMPHOMA
SUBCUTANEOUS PANNICULITIS LIKE T-CELL LYMPHOMA
FROZEN STAGE MODEL

Cortical Thymus

- TdT
- HLA-DR
- CD34
- cCD-3
- CD7
- CD2
- CD5
- TCRδ
- TCRγ

Medullary Thymus

- TdT
- HLA-DR
- CD34
- CD7
- CD2
- CD5
- CD1
- CD8-/+
- CD56 -/+ sCD3
- CD7
- CD2
- CD5
- TCR
- TCRδ
- TCRγ

Blood, lymphoid tissue

γδ T-cell
FROZEN STAGE MODEL

Cortical Thymus → Medullary Thymus → Blood, lymphoid tissue

HEPATOSPLENIC TCL
CUTANEOUS γδ TCL
FROZEN STAGE MODEL

Bone marrow  Blood  Lymphoid tissues

Myeloid precursor?  Lymphoid precursor?  CD34+  CD117-  CD94-
CD34+  CD117-  CD94-
CD34+  CD117-  CD94-
CD34-  CD117+  CD94-
cCD3  sCD3-  CD8-/-  TCR-  CD2  CD7  CD11b  CD16  CD56  CD57  KIR

NK cell
FROZEN STAGE MODEL

Bone marrow  Blood  Lymphoid tissues

EXTRANODAL NK/TCL, NASAL TYPE
FROZEN STAGE MODEL

- Marginal zone (MZ) - marginal zone B cells
- Follicle mantle (FM) - mantle cells
- Germinal center (FC) - germinal center cells: centrocytes, centroblasts
- T zone - immunoblasts, plasmablasts
FROZEN STAGE MODEL

- Marginal zone (MZ)  
  MARGINAL ZONE LYMPHOMA
- Follicle mantle (FM)  
  MANTLE CELL LYMPHOMA
- Germinal center (FC)  
  FOLLICULAR LYMPHOMA
- T zone  
  DIFFUSE LARGE BCL, ACT TYPE
  PLASMABLASTIC LYMPHOMA

Diagram:
- B area
- MZ
- FM
- FC
- T area
B. RECOGNITION OF BIOLOGICAL ENTITIES

- morphology
- immunophenotype
- genetics
- clinical presentation
1. architecture:
   - ① symmetry, V shape
   - ② epidermotropism
   - ③ spongiosis
   - ④ ulceration
   - ⑤ diffuse dermal
   - ⑥ perivascular
   - ⑦ superficial/deep dermal
   - ⑧ subcutaneous: septal or lobular
2. cytological features:
   ① size: small/medium/large
2. cytological features:
   ② atypia: cerebriform/anaplastic
morbidity

2. cytological features:
   ③ monomorphism / pleomorphism/ polymorphism
2. cytological features: technical aspects
1. Immunohistochemistry: panels are recommended
   ① Small cell B cell lymphoma: CD20, CD3, CD79a, PAX5, CD138, CD10, BCL6, BCL2, CD5, CD21, CD23, cyclin D1, IgK, IgL, Ki67
   ② Large cell B cell lymphoma: CD20, CD79a, CD3, CD5, PAX5, MUM1, BCL6, BCL2, CD10, CD30, EBV (ISH), Ki67
   ③ T-/NK-cell neoplasia: CD3, CD20, CD21, CD2, CD5, CD7, CD4, CD8, TIA1, granzyme B, PD1, TCL1, CD56, CD57, EBV (ISH), CD30, ALK
   ④ Blaster: CD45, CD34, MPO, PAX5, CD3, TdT, Ki67

Ref: nasjonalt handlingsprogram med retningslinjer for diagnostikk, behandling og oppfølging av maligne lymfomer (Helsedirektoratet’s nettsider)
immunophenotype

2. Flow cytometry
Genetics: antigen receptor gene rearrangements

T CELL RECEPTOR GENE REARRANGEMENTS

VARIABLE GENES

JOINING GENES

CONSTANT GENE

JUNCTIONAL GENES

CONSTANT GENE
Genetics: antigen receptor gene rearrangements
Genetics: antigen receptor gene rearrangements
Genetics: antigen receptor gene rearrangements
Genetics: antigen receptor gene rearrangements
Clinical information
Mycosis fungoides

OSLO UNIVERSITY HOSPITAL
UNIVERSITY OF OSLO
Definition and epidemiology

‘mycosis fungoides’ by Jean-Louis-Marc Alibert in 1806. ‘Descriptions des maladies de la peau à l’Hôpital Saint-Louis, et exposition des meilleures méthodes suivies pour leur traitement’
Definition and epidemiology

Represents about 50% of primary cutaneous T-cell lymphomas

With an incidence of 5/1,000,000

Male/female ration: 2/1

Affects mostly adults or elderly, but can also affect the pediatric population

The term is reserved for a cutaneous T-cell lymphoma with the typical clinical patch – plaque – tumor stages
Clinical presentation
Clinical presentation
Clinical presentation
Patients survive for decades before dissemination and morc

An erythrodermic variant is described, without leukemic cells
Histology

Patch stage
Patch stage
Histology

Plaque stage
Histology

Plaque stage
Histology

Tumor stage
Immunophenotype and immunogenotype

CD4+, CD8-, CD5+ (-), CD7- (+), CD2+, CD30- (+)

Clonal TCR gene rearrangements
Diagnosis of ‘early mycosis fungoides’

**Scoring system: four points needed**

### Clinical

**Basic**
- Persistent and/or progressive patches/thin plaque: 2 points for basic criteria and two additional criteria

**Additional**
- 1) Non-sun exposed location
- 2) Size/shape variation
- 3) Poikiloderma
- 1 point for basic criteria and one additional criterion

### Histopathologic

**Basic**
- Superficial lymphoid infiltrate: 2 points for basic criteria and two additional criteria

**Additional**
- 1) Epidermotropism without spongiosis
- 2) Lymphoid atypia
- 1 point for basic criteria and one additional criterion

### Molecular biological

- 1) Clonal TCR gene rearrangement: 1 point for clonality

### Immunopathologic

- 1) <50% CD2+, CD3+, and/or CD5+ T cells: 1 point for one or more criteria
- 2) <10% CD7+ T cells
- 3) Epidermal/dermal discordance of CD2, CD3, CD5 or CD7

# Staging of mycosis fungoides


## Skin

<table>
<thead>
<tr>
<th>Stage</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>T₁</td>
<td>Limited patches, papules, and/or plaques covering &lt; 10% of the skin surface. May further stratify into T₁a (patch only) vs T₁b (plaque ± patch).</td>
</tr>
<tr>
<td>T₂</td>
<td>Patches, papules or plaques covering 10% of the skin surface. May further stratify into T₂a (patch only) vs T₂b (plaque ± patch).</td>
</tr>
<tr>
<td>T₃</td>
<td>One or more tumors (1-cm diameter)</td>
</tr>
<tr>
<td>T₄</td>
<td>Confluence of erythema covering 80% body surface area</td>
</tr>
</tbody>
</table>

## Node

<table>
<thead>
<tr>
<th>Stage</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>N₀</td>
<td>No clinically abnormal peripheral lymph nodes; biopsy not required</td>
</tr>
<tr>
<td>N₁</td>
<td>Clinically abnormal peripheral lymph nodes; histopathology Dutch grade 1 or NCI LN₀₂</td>
</tr>
<tr>
<td>N₁ₐ</td>
<td>Clone negative#</td>
</tr>
<tr>
<td>N₁ₚ</td>
<td>Clone positive#</td>
</tr>
<tr>
<td>N₂</td>
<td>Clinically abnormal peripheral lymph nodes; histopathology Dutch grade 2 or NCI LN₃</td>
</tr>
<tr>
<td>N₂ₐ</td>
<td>Clone negative#</td>
</tr>
<tr>
<td>N₂ₚ</td>
<td>Clone positive#</td>
</tr>
<tr>
<td>N₃</td>
<td>Clinically abnormal peripheral lymph nodes; histopathology Dutch grades 3-4 or NCI LN₄; clone positive or negative</td>
</tr>
<tr>
<td>N₅</td>
<td>Clinically abnormal peripheral lymph nodes; no histologic confirmation</td>
</tr>
</tbody>
</table>

## Visceral

<table>
<thead>
<tr>
<th>Stage</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>M₀</td>
<td>No visceral organ involvement</td>
</tr>
<tr>
<td>M₁</td>
<td>Visceral involvement (must have pathology confirmation§ and organ involved should be specified)</td>
</tr>
</tbody>
</table>

## Blood

<table>
<thead>
<tr>
<th>Stage</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>B₀</td>
<td>Absence of significant blood involvement: 5% of peripheral blood lymphocytes are atypical (Sézary) cells</td>
</tr>
<tr>
<td>B₀ₐ</td>
<td>Clone negative#</td>
</tr>
<tr>
<td>B₀ₚ</td>
<td>Clone positive#</td>
</tr>
<tr>
<td>B₁</td>
<td>Low blood tumor burden: &gt;5% of peripheral blood lymphocytes are atypical (Sézary) cells but does not meet the criteria of B₂</td>
</tr>
<tr>
<td>B₁ₐ</td>
<td>Clone negative#</td>
</tr>
<tr>
<td>B₁ₚ</td>
<td>Clone positive#</td>
</tr>
<tr>
<td>B₂</td>
<td>High blood tumor burden: 1000/µL Sézary cells</td>
</tr>
</tbody>
</table>
Staging of mycosis fungoides


Dutch grading system:

Grade 1: dermatopathic lymphadenopathy

Grade 2: dermatopathic lymphadenopathy with the presence of atypical cerebriform cells (>7.5µm)

Grade 3: partial effacement of the lymph node with the presence of atypical cerebriform cells

Grade 4: total effacement of the lymph node with the presence of atypical cerebriform cells
Staging of mycosis fungoides

Dutch grade 1
Staging of mycosis fungoides

Dutch grade 3
Variants of mycosis fungoides

1. Pagetoid reticulosis (Woringer-Kolopp disease)

   Predominantly adult males
   Acral distribution, solitary lesions, mostly scaly patches or plaques
   Completely benign disease
Variants of mycosis fungoides

1. Pagetoid reticulosis (Woringer-Kolopp disease)
1. Pagetoid reticulosis (Woringer-Kolopp disease)

CD4+ or CD8+, often CD30+
2. Folliculotropic mycosis fungoides

Head and neck regions
Mostly plaques and tumors with some peculiar features
More aggressive clinical course than typical mycosis fungoides
Clinical information

2. Folliculotropic mycosis fungoides
KASUS 1. Kvinne, 50 år.


Pasient får egnet behandling etter at diagnosen foreligger, men får ny lesjon fire år etter på høyre ankel. Det blir tatt ny biopsi (SNITT 2)

Diagnose: 1. mycosis fungoides, follikulotrop variant, 2. mycosis fungoides, tumor stadium
Variants of mycosis fungoides

2. Folliculotropic mycosis fungoides
Variants of mycosis fungoides

2. Folliculotropic mycosis fungoides

1. CD3+, CD2+, CD5+, CD7-, CD4+, CD8-, CD30-/+ 
2. CD3+, CD2+, CD5+, CD7-, CD4+, CD8-, CD30+/-
Variants of mycosis fungoides

2. Folliculotropic mycosis fungoides

BIOPSY 1

BIOPSY 2
Variants of mycosis fungoides

3. Granulomatous slack skin
   Mostly trunk region
   Slowly progressive with the development of large skin folds
3. Granulomatous slack skin
Sézary syndrom
Sézary syndrome was first described by Albert Sézary in 1938. He noted the presence of ‘cellules monstrueuses’ in the blood. Today the disease is defined by: 1. erythroderma, 2. generalized lymphadenopathy, 3. circulating Sézary cells.
Definition and epidemiology

Rare disease, less than 5% of cutaneous T-cell lymphomas
Clinical presentation

The disease has an aggressive clinical course with a 5-year OS of 10-20%
Histology
Blood smear
Immunophenotype

CD3+, CD4+, CD8-, CD5+, CD7-, CD26-
Immunophenotype

- **FITC-A**: CD7
- **PerCP**: CD3
- **PE-A**: CD7
- **695/40-A**: CD3 PerCP

Graphs showing the distribution of cells based on CD2 and CD7 markers.
Staging of Sézary syndrome


Skin
- **T₁**: Limited patches, papules, and/or plaques covering < 10% of the skin surface. May further stratify into T₁a (patch only) vs T₁b (plaque ± patch).
- **T₂**: Patches, papules or plaques covering 10% of the skin surface. May further stratify into T₂a (patch only) vs T₂b (plaque ± patch).
- **T₃**: One or more tumors (1-cm diameter)
- **T₄**: Confluence of erythema covering 80% body surface area

Node
- **N₀**: No clinically abnormal peripheral lymph nodes; biopsy not required
- **N₁**: Clinically abnormal peripheral lymph nodes; histopathology Dutch grade 1 or NCI LN₀-2
  - **N₁a**: Clone negative*
  - **N₁b**: Clone positive*
- **N₂**: Clinically abnormal peripheral lymph nodes; histopathology Dutch grade 2 or NCI LN₂
  - **N₂a**: Clone negative*
  - **N₂b**: Clone positive*
- **N₃**: Clinically abnormal peripheral lymph nodes; histopathology Dutch grades 3-4 or NCI LN₃; clone positive or negative
  - **N₃a**: Clone negative*
  - **N₃b**: Clone positive*
- **Nₓ**: Clinically abnormal peripheral lymph nodes; no histologic confirmation

Visceral
- **M₀**: No visceral organ involvement
- **M₁**: Visceral involvement (must have pathology confirmation* and organ involved should be specified)

Blood
- **B₀**: Absence of significant blood involvement: 5% of peripheral blood lymphocytes are atypical (Sézary) cells
  - **B₀a**: Clone negative*
  - **B₀b**: Clone positive*
- **B₁**: Low blood tumor burden: > 5% of peripheral blood lymphocytes are atypical (Sézary) cells but does not meet the criteria of B₂
  - **B₁a**: Clone negative*
  - **B₁b**: Clone positive*
- **B₂**: High blood tumor burden: 1000/μL Sézary cells with positive clone*
Blastic plasmacytoid dendritic cell neoplasm
Is a very rare tumor from precursor cells of plasmacytoid dendritic cells, that has a similar immunophenotype: CD4+, CD123+, CD68+
Clinical presentation

Usually affects skin first with one or more nodules, then bone marrow and peripheral blood. 20% of cases develop a myelomonocytic leukemia or acute leukemia

Kasus 4
Jente, 13 år.

Diagnose: blastisk plasmacytoid dendritisk celle neoplasie
Histology
Immunophenotype

CD4

CD56
Immunophenotype

CD4+, CD56+, CD68+, CD123+, MPO-, CD16-, CD5-, CD7-, CD3-, CD20-, TdT+, CD34-
Benmargen av pasienten viste 0,3% blaster, med samme immunfenotype ved flow cytometrisk analyse
Langerhans cell histiocytosis
Definition and epidemiology

Is a neoplasm that derives from Langerhans cells, and has a similar morphology and immunophenotype

Incidence: 5/1,000,000

Male/female ratio: 4/1

Unifocal and multifocal variants of the disease are known
Kasus 10

Kvinne, 84 år
Pasient opplyser om vekttap på 10 kg. det siste året, og har fått i løpet av siste måned multiple papler i hals regionen og rundt venstre øre. Disse papler har tendens til å komme og gå. Det blir tatt biopsi for histopatologisk diagnose.

Diagnose: Langerhans celle histiocytose
Histology
Histology
Immunophenotype

CD1A