Beyond morphology: emerging diagnostic tools for mycosis fungoides

Pr Maxime Battistella

Pathology department, Dermatopathology program director
Hôpital Saint Louis - Université Paris Cité
Inserm U1342

09-07-2026

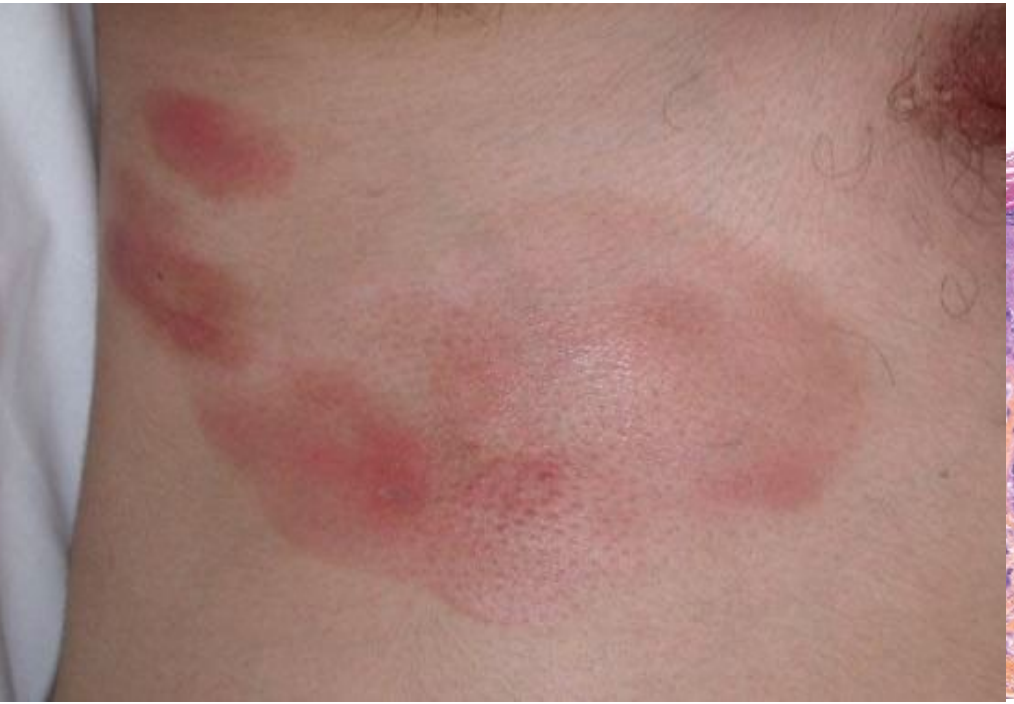
Disclosures

- **Consulting:** Innate Pharma, Cerba Research
- **Board:** BMS, Regeneron, Takeda, Kyowa Kirin, HAC pharma
- **Speaker:** Kyowa Kirin, Recordati, BMS, MSD, Takeda, Sanofi
- **Research funding:** Kyowa Kirin, Takeda, Innate Pharma
- **Congress/travel fees:** Recordati, Kyowa Kirin

Agenda

1. **Today's situation**
2. **IHC markers** from Sézary cell biology studies
3. **Clonality**: new techniques, new limits
4. **Advanced molecular methods**: utopia or reality
5. **AI** in the game for MF diagnosis

Mycosis fungoides: clinico-pathological correlation is key



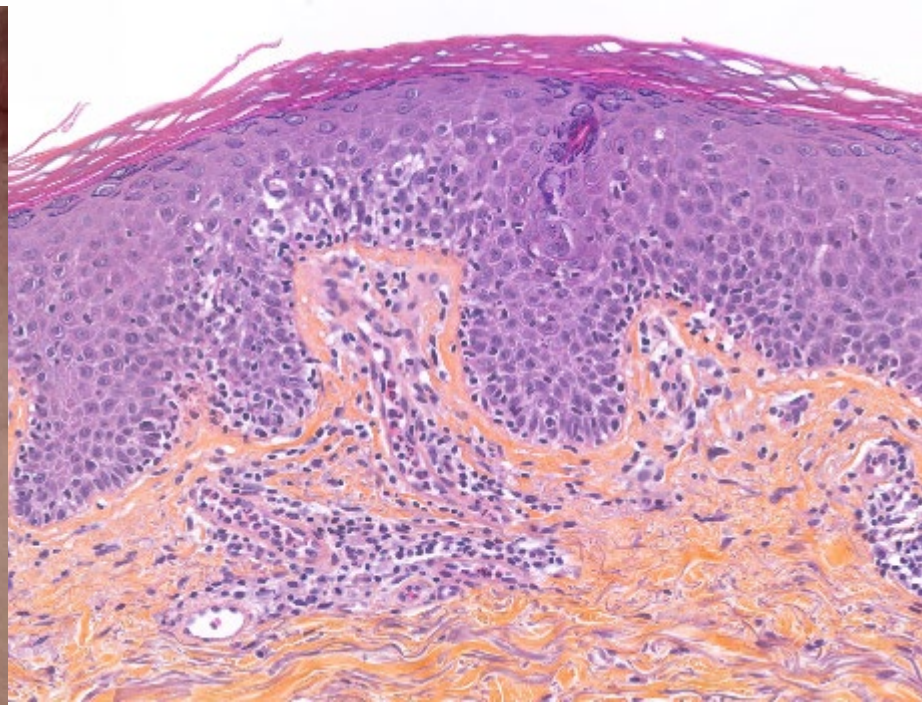


Table I. Algorithm for diagnosis of early MF*

Criteria	Scoring system
Clinical	
Basic	2 points for basic criteria and two additional criteria
Persistent and/or progressive patches/thin plaques	1 point for basic criteria and one additional criterion
Additional	
1) Non-sun exposed location	
2) Size/shape variation	
3) Poikiloderma	
Histopathologic	
Basic	2 points for basic criteria and two additional criteria
Superficial lymphoid infiltrate	1 point for basic criteria and one additional criterion
Additional	
1) Epidermotropism without spongiosis	
2) Lymphoid atypia†	
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‡	

MF, Mycosis fungoides; *TCR*, T-cell receptor.
*A total of 4 points is required for the diagnosis of MF based on any combination of points from the clinical, histopathologic, molecular biological, and immunopathologic criteria.
†Lymphoid atypia is defined as cells with enlarged hyperchromatic nuclei and irregular or cerebriform nuclear contours.
‡T-cell antigen deficiency confined to the epidermis.

Pimpinelli al. JAAD 2005

Table I. Algorithm for diagnosis of early MF*

Criteria	Scoring system
Clinical	
Basic	2 points for basic criteria and two additional criteria
Persistent and/or progressive patches/thin plaques	1 point for basic criteria and one additional criterion
Additional	
1) Non-sun exposed location	
2) Size/shape variation	
3) Poikiloderma	
Histopathologic	
Basic	2 points for basic criteria and two additional criteria
Superficial lymphoid infiltrate	1 point for basic criteria and one additional criterion
Additional	
1) Epidermotropism without spongiosis	
2) Lymphoid atypia†	
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‡	

MF, Mycosis fungoides; *TCR*, T-cell receptor.
*A total of 4 points is required for the diagnosis of MF based on any combination of points from the clinical, histopathologic, molecular biological, and immunopathologic criteria.
†Lymphoid atypia is defined as cells with enlarged hyperchromatic nuclei and irregular or cerebriform nuclear contours.
‡T-cell antigen deficiency confined to the epidermis.

Validation study

Results: Among the 24 cases of MF, 21 cases achieved four or more points through application of the algorithm. Among the 10 cases of MF mimics, only four achieved four or more points. This difference was significant (Fisher's exact test, $p = 0.009$). The sensitivity of the 4-point threshold for a diagnosis of MF was 87.5% and the specificity was 60%.

				95% Confidence Interval	
Measure	AUC	Std. error*	p-Value†	Lower bound	Upper bound
Total score	0.819	0.076	0.004	0.670	0.967
Clinical score	0.679	0.112	0.104	0.460	0.898
Persistent and progressive plaques	0.500	0.110	>0.999	0.284	0.716
Non-sun exposed	0.579	0.114	0.473	0.356	0.803
Size/shape variability	0.579	0.114	0.473	0.356	0.803
Poikiloderma	0.700	0.093	0.070	0.518	0.882
Histopathology score	0.675	0.101	0.112	0.476	0.874
Superficial lymphoid infiltrate	0.550	0.114	0.650	0.327	0.773
Epidermotropism	0.588	0.113	0.427	0.366	0.809
Atypia	0.642	0.104	0.199	0.437	0.846
Immunohistochemistry score	0.579	0.107	0.473	0.369	0.789
<50% CD2, CD3 and/or CD5	0.542	0.106	0.705	0.333	0.750
<10% CD7	0.579	0.107	0.473	0.369	0.789
Discordance	0.542	0.106	0.705	0.333	0.750
TCR rearrangement	0.538	0.109	0.734	0.324	0.751
Density table 3	0.619	0.106	0.281	0.411	0.826
Epidermotropism table 3	0.642	0.101	0.199	0.444	0.840
Atypia table 3	0.646	0.101	0.186	0.449	0.843

Pimpinelli al. JAAD 2005
Vandergriff et al. J Cutan Pathol 2015

Mycosis fungoides : histopathology

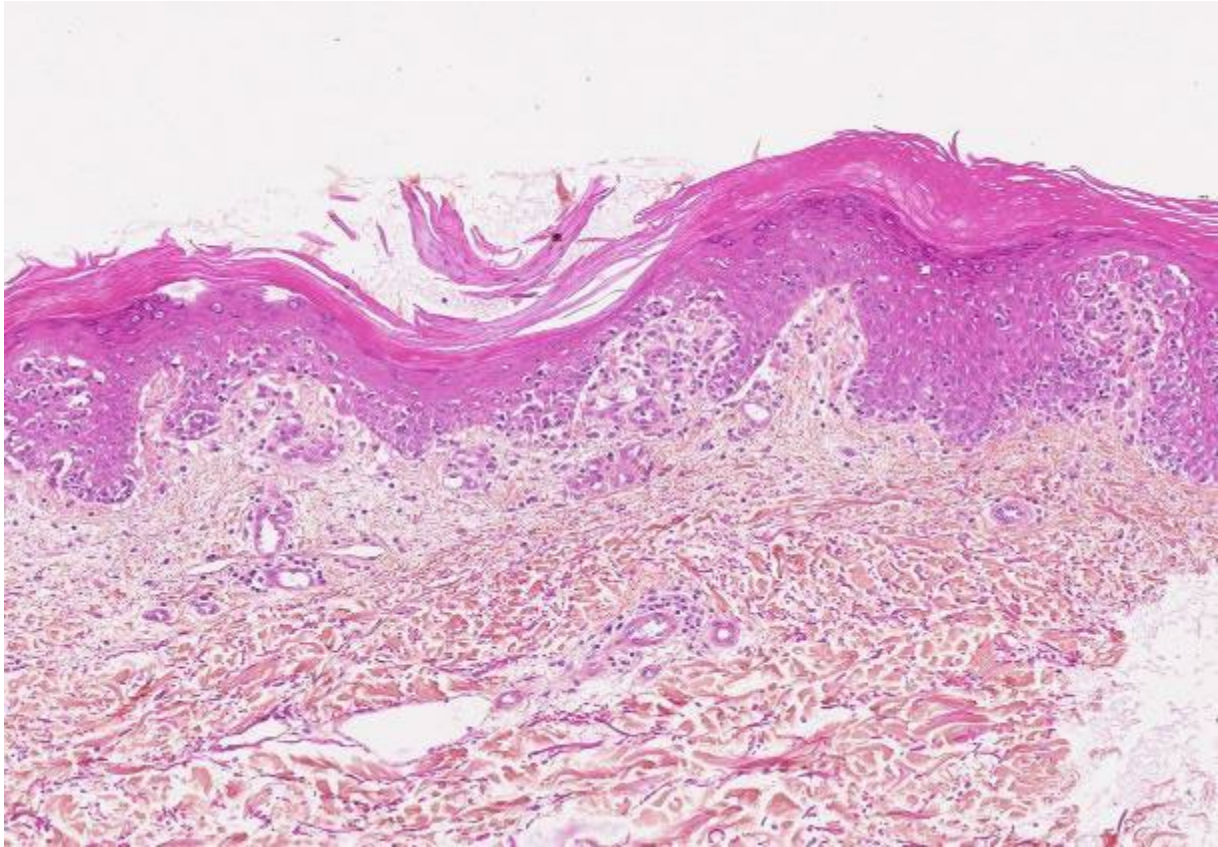
Early patch/plaque mycosis fungoides

- **Intra-epidermal atypical lymphoid infiltrate (epidermotropism)**
- **Variable band-like infiltrate in the superficial dermis with few atypical lymphoid cells**

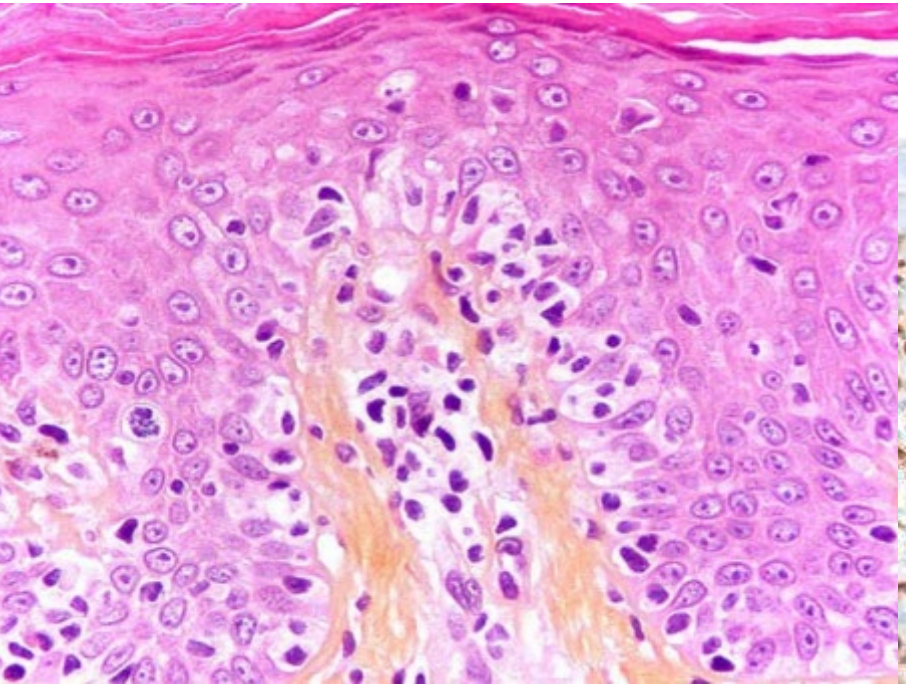
- **Cytology**
  - Atypical small to medium lymphoid cells
  - Variable amount of reactive infiltrate
    - Reactive lymphoid cells
    - Histiocytes
    - Plasma cells
    - Neutrophils, eosinophils

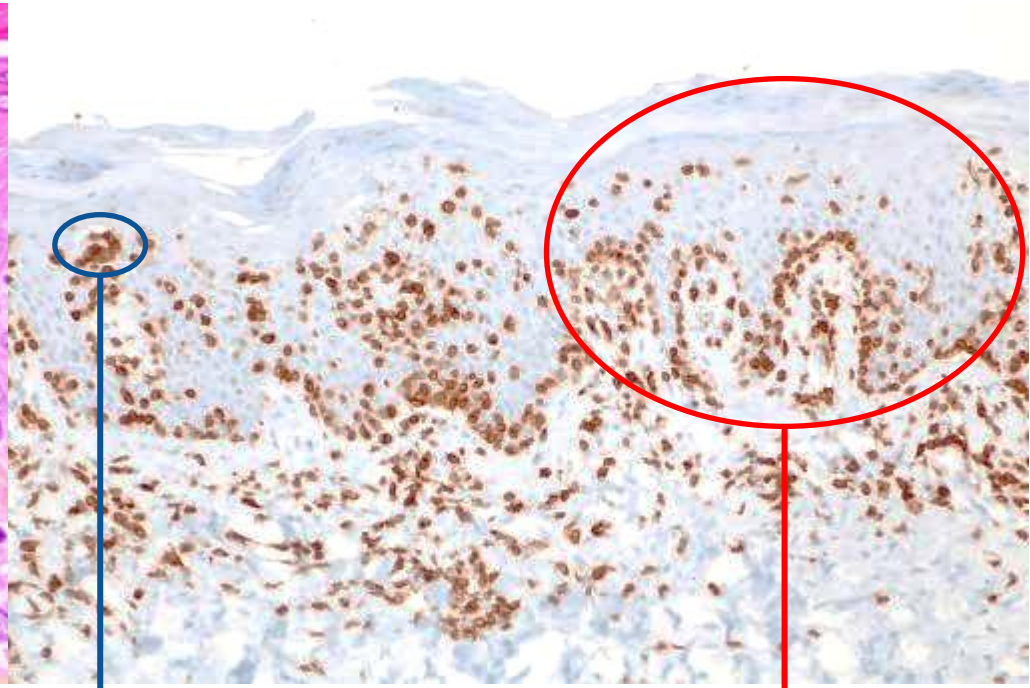
- **Epidermis**
  - Typically normal, slightly hyperkeratotic and parakeratotic
  - May show psoriasiform, spongiform or lichenoid changes

- **Papillary dermis : fibrosis (especially in pretreated, long-lasting lesions)**



Mycosis fungoïdes : peculiar intraepidermal distribution of atypical T-cells





Atypical lymphoid cells
- Hyperchromatic
- Angular or folded nuclei
- Larger than ajdacent keratinocyte nuclei
- Pericellular halo (haloed cells)

Pautrier's microabscess (rare finding in MF)
collection of atypical T-cells

Basilar epidermotropism
Basal contiguous alignment predominating over supra-basal lymphoid cells

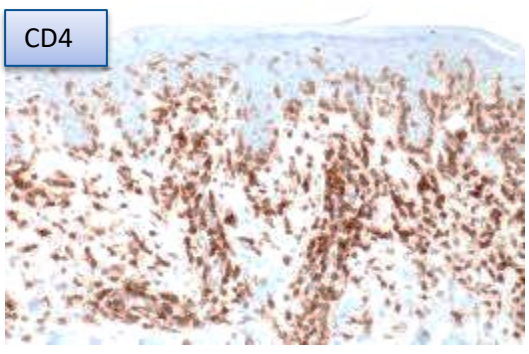
Mycosis fungoides: phenotype

- **Typical phenotype of neoplastic cells**
  - CD2 + CD3+ CD5+ CD4+ TCRβ+
  - CD8- TCRγ or TCRδ-
  - CD7 frequently lost
  - CD30 expression variable (increasing with stage)
  - **Less typically**
    - CD8+ CD4- with cytotoxic markers (more frequent in paediatric MF and hypopigmented MF)
    - TCRδ+
    - Null (CD4- CD8-) phenotype

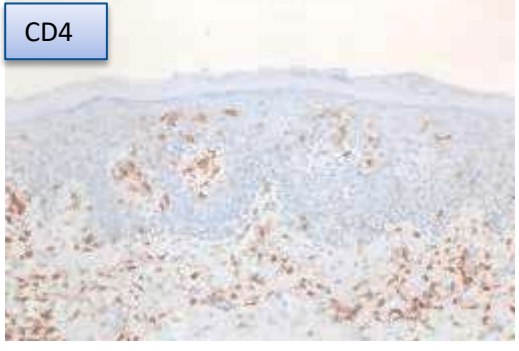
- **Advanced MF**
  - More frequent T-cell antigen loss (CD2, CD3, CD5, CD7, CD4…)
  - May express cytotoxic granules
  - May aberrantly express CD20 (PAX5 negativity differentiates from admixed B-cells)

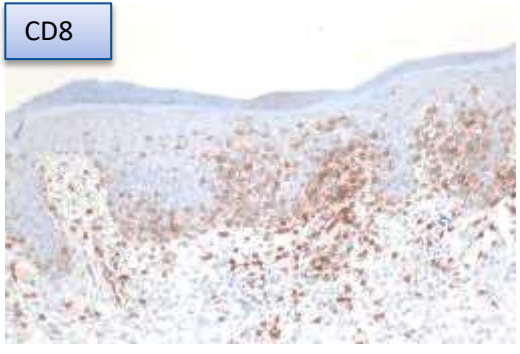
- **Markers useful for diagnosis in routine basis**
  - **Early MF**
    - CD3, CD4, CD8, CD7, CD30
  - **Advanced (tumor) MF**
    - CD3, CD4, CD5, CD7, CD8, CD30, CD56
    - Granzyme B, EBER, TCRβ, TCRδ
    - Ki67 proliferative index

Classical early CD4+ MF



Early CD8+ MF





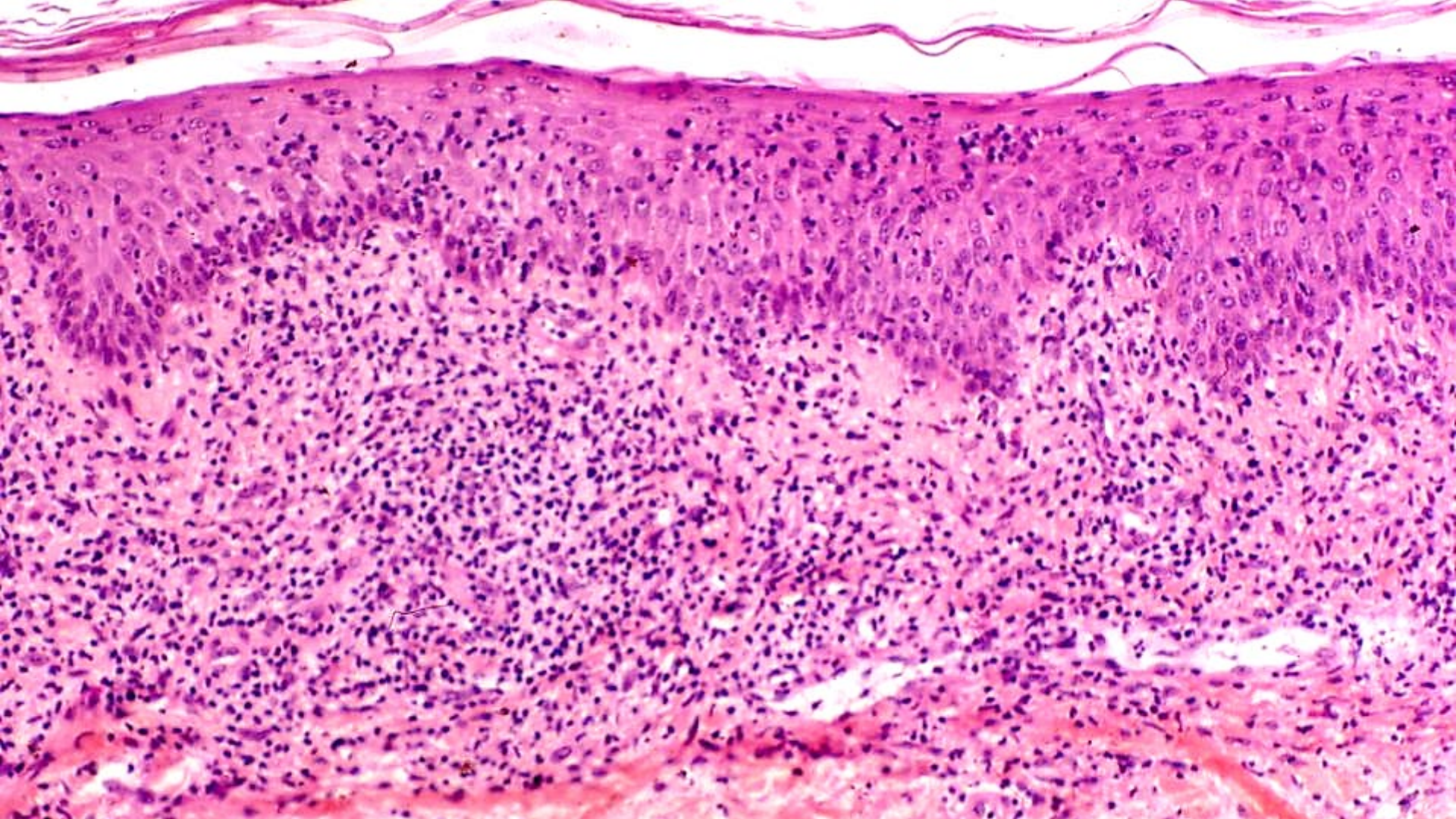
MF-like reactive T-lymphoid proliferation and other MF mimickers

MF-like rT-LP

- Drug-induced
  - Lymphomatoid drug reaction
  - DRESS
- Allergen-induced
  - Contact allergens
- UV-induced
  - Actinic reticuloid
- HIV-induced
  - CD8+ pseudo-MF
- Borrelia-induced
- Idiopathic

Other MF mimickers for dermpath

- Interface dermatosis (lupus, lichen planus)
- Inflammatory morphea, LS
- Inflammatory vitiligo
- Interstitial granuloma annulare
- Chronic spongiotic dermatitis
- Inflammatory psoriasis
- Purpuric pigmented dermatosis
- Pityriasis lichenoides
- Small plaque parapsoriasis



Practice survey among pathologists of the GFELC

IHC

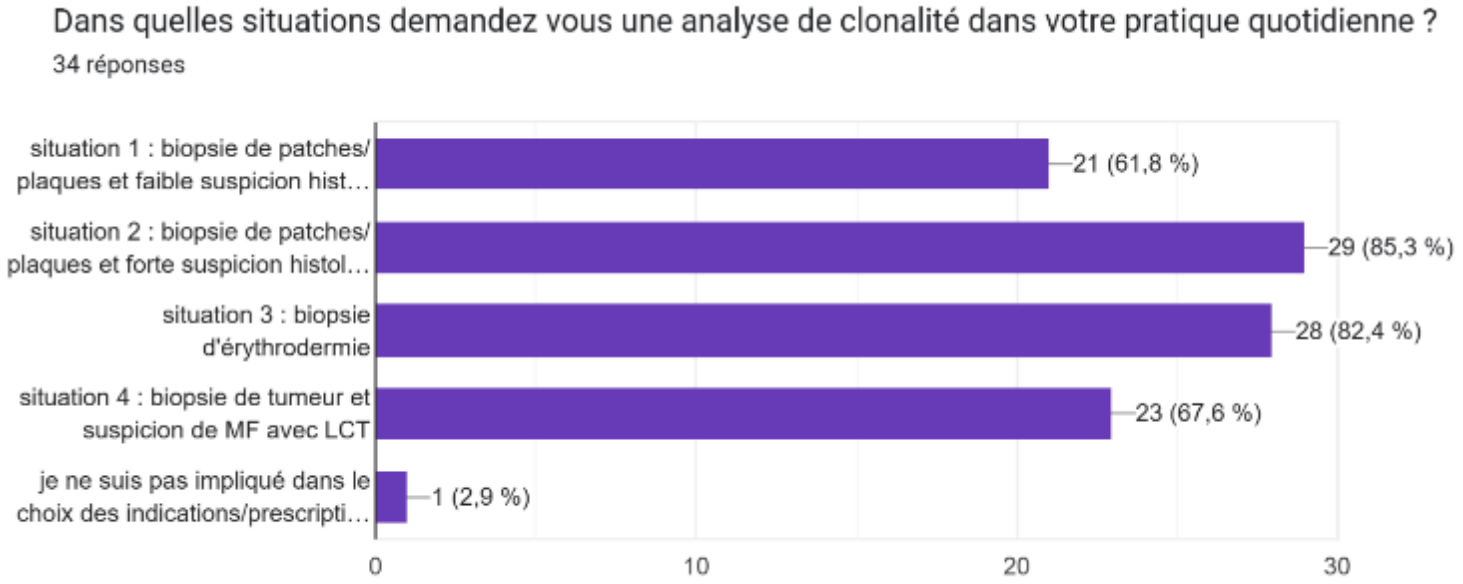
Situation	1st intention IHC	2nd intention IHC
Low suspicion of MF	**CD3, CD4**, **CD8**, CD7, CD20	CD2, CD5, CD30, PD1 (TRBC1)
High suspicion of MF	**CD3, CD4, CD5, CD7, CD8**, CD2, CD20, CD30, PD1 (TRBC1)	Ki67, CD25
Erythroderma	**CD3, CD4**, **CD7, CD8**, CD5, CD20, PD1	CD2, CD30, Ki67, CD25 (TRBC1)
Suspicion of MF with LCT	**CD2, CD3, CD4, CD5, CD7, CD8, CD20, CD30, Ki67**, PD1 (TRBC1)	(betaF1), (TCRdelta), Bcl6, CXCL13, ICOS, EBER, TIA1, GrB, Perforine

Bold (>75% consensus)

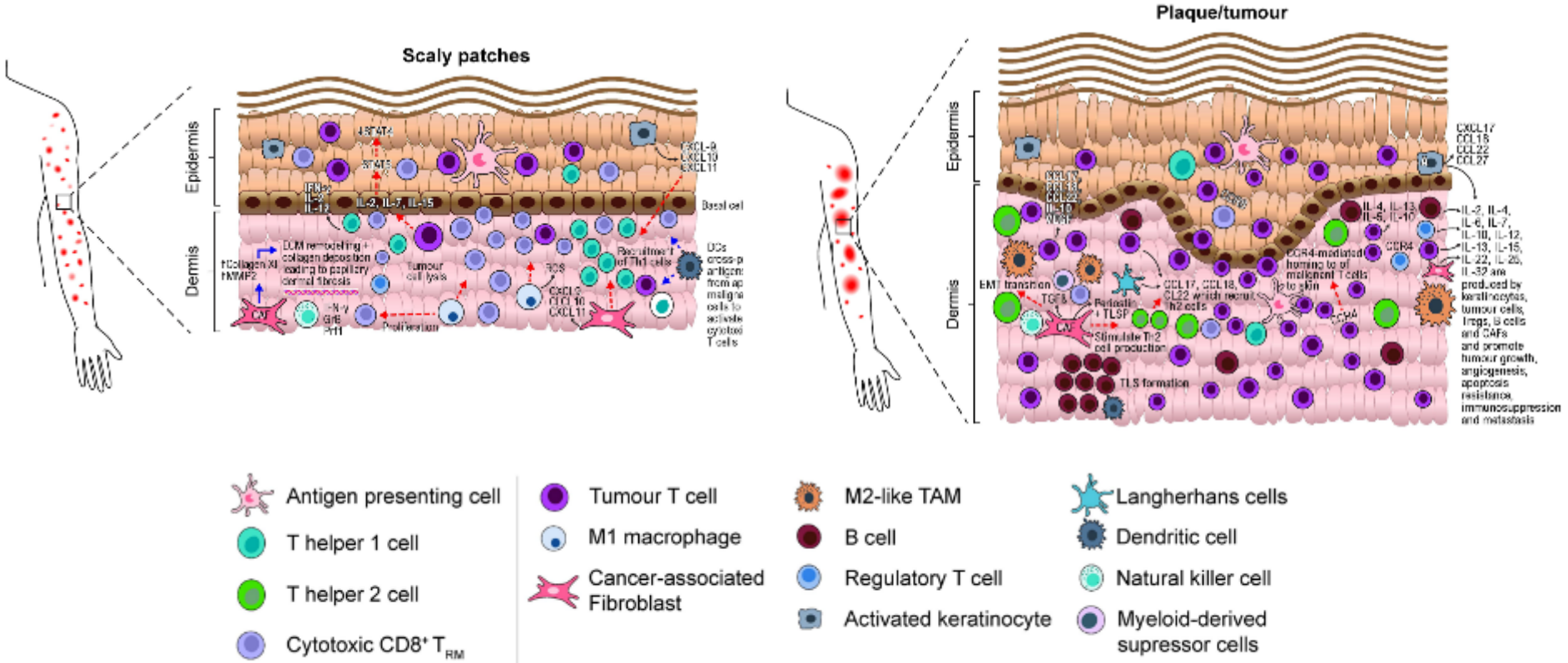
GFELC

Practice survey among pathologists of the GFELC

TCR Clonality



MF: tumor cells among their TME



De Masson A, Battistella M, et al. In press

IHC markers from Sézary cell biology studies

Surface markers

CD158k / KIR3DL2

CD164 · FcRL3

PD-1 · CLA · CCR4

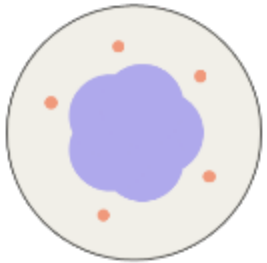
Transcription factors

STAT3 · STAT4 · STAT5

TWIST1 · TOX

GATA3 → KLHL42

Sezary cell (SC)



Therapeutic targets

CD158k (anti-KIR3DL2)

PD-1 · CTLA-4

CCR4 (mogamulizumab)

scRNA-seq targets

AIRE (top upregulated)

CD74 · KLHL42

KIR2DL3 · KIR3DL1

MET · GATA6 · S100A8/9

Malignant T-cell subtypes (scRNA-seq)

MTC-CM

Central memory-like (dominant, Th2)

MTC-E/EM

Effector/effector-memory (Th1)

MTC-Reg

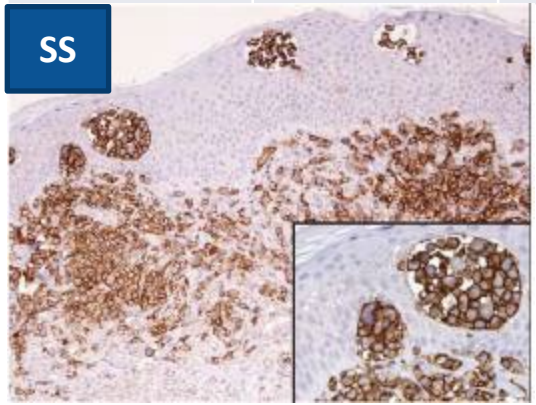
Regulatory-like, exhausted

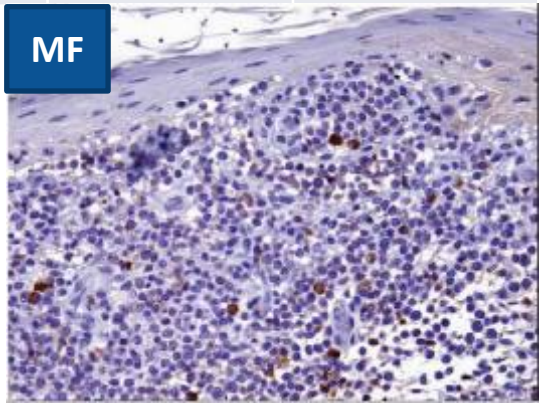
TN-, TCM- and TSCM-like surface phenotypes converge into one transcriptional clone (Roelens et al., 2017)

Sources: Nicolay et al., JDDG 2016 · Roelens et al., Blood 2017 · Mains-Teisserenc et al., J Invest Dermatol 2015 · Childs et al., JCADV 2025

Differential PD-1 expression in SS and MF

		PD-1+ neoplastic T cells, n (%)		
Type of lymphoma	**Cases, n**	**> 50%**	**11%–50%**	**< 10%**
SS	27[a,b]	24 (89)	1 (4)	2 (7)
MF	60[a]	8 (13)	4 (7)	48 (80)
Patch/plaque	30	4 (13)	2 (7)	24 (80)
Tumour	22	3 (14)	1 (4)	18 (82)
Erythrodermic MF	8[b]	1 (12)	1 (12)	6 (75)

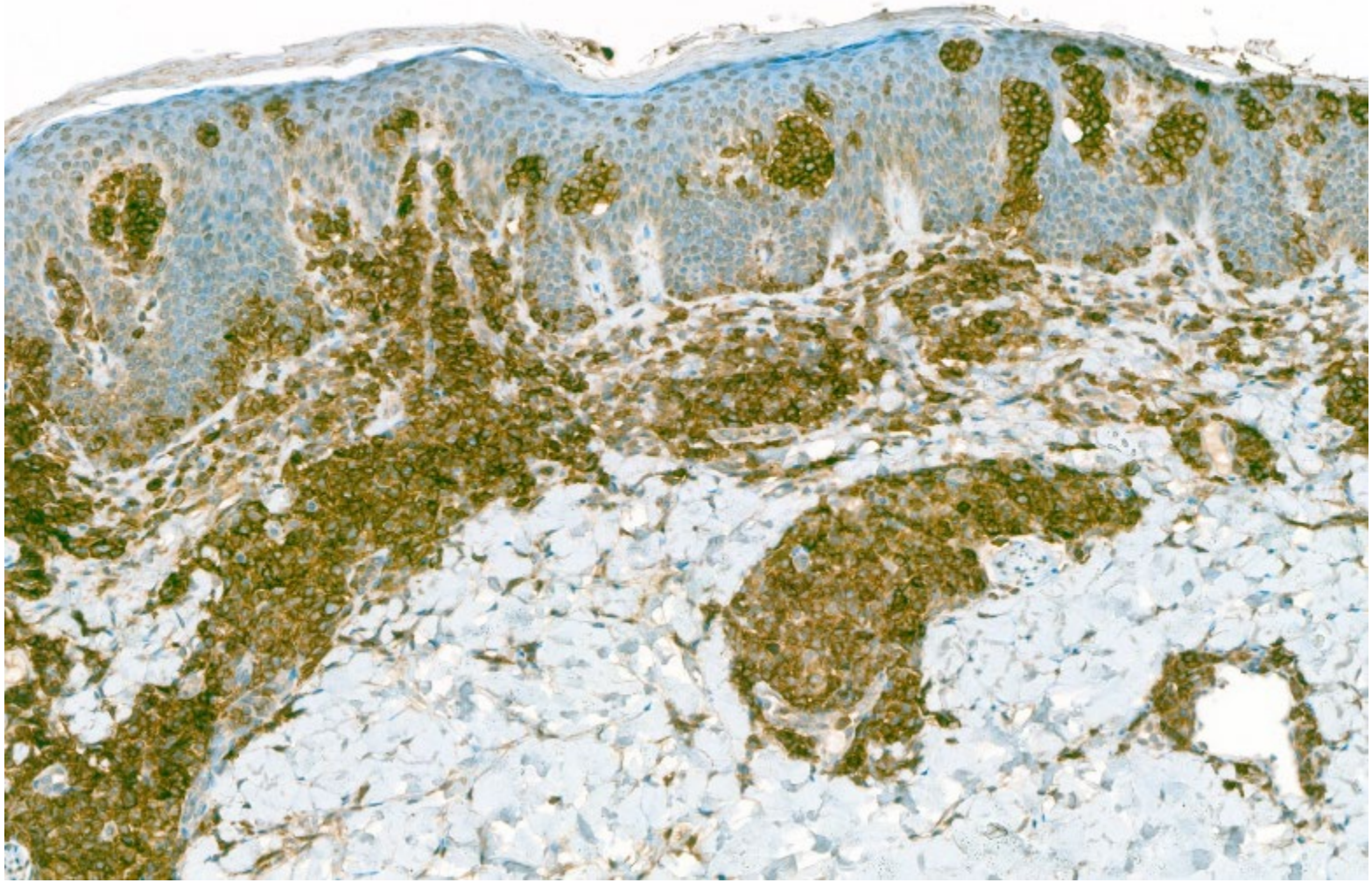




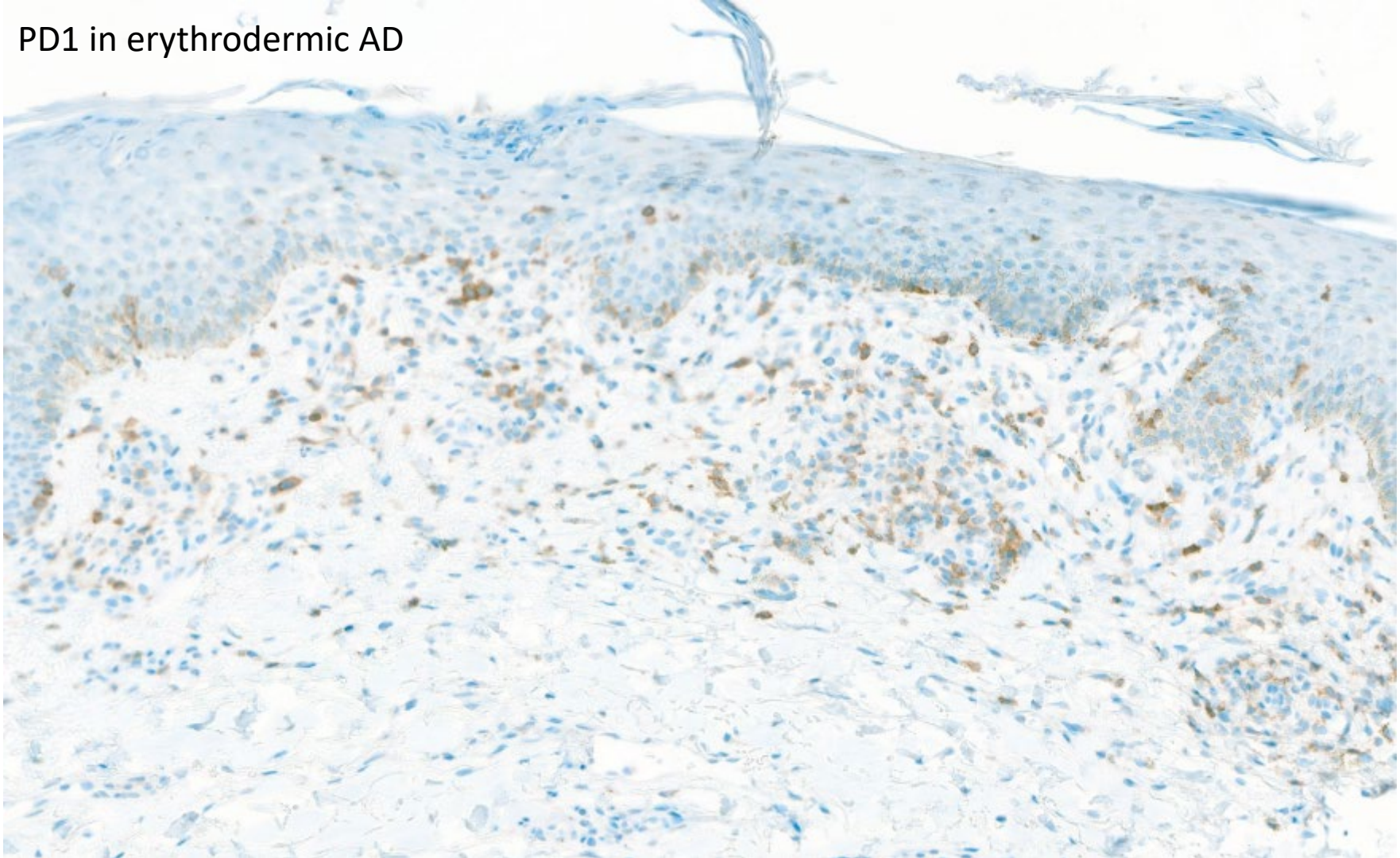
[a] Patients with SS (n = 27) vs those with MF (n = 60); $p < 0.001$.
[b] Patients with SS (n = 27) vs those with erythrodermic MF (n = 8); $p < 0.001$.

Cetinözman F, et al. Arch Dermatol. 2012;148:1379-85.

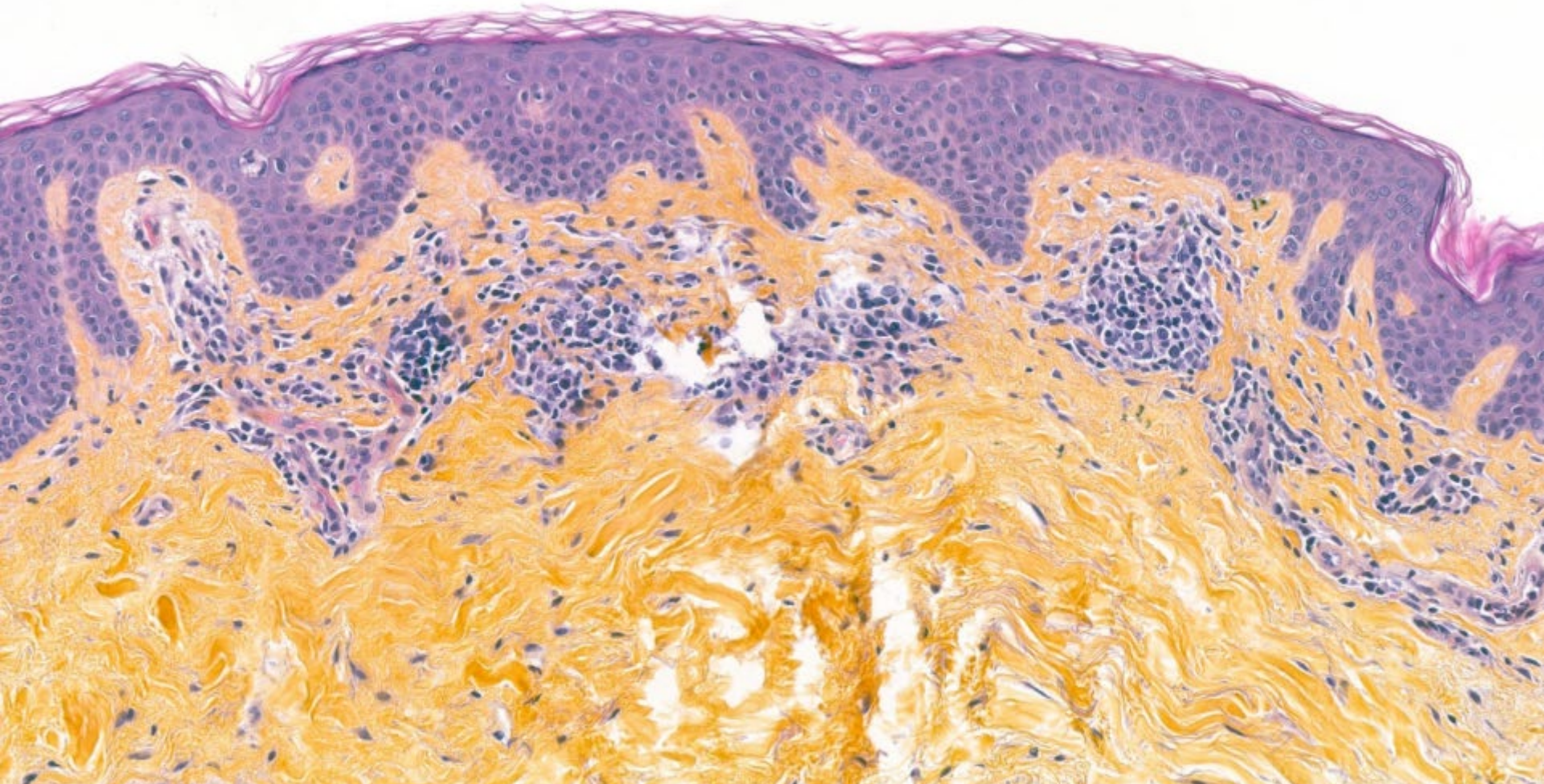
PD1 in Sézary syndrome



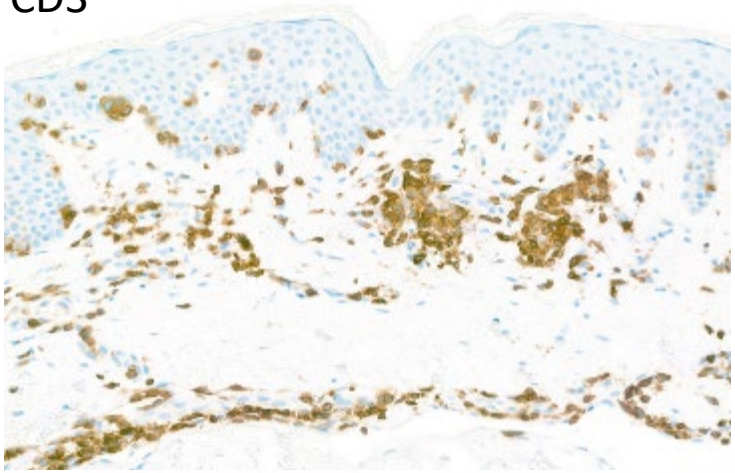
PD1 in erythrodermic AD



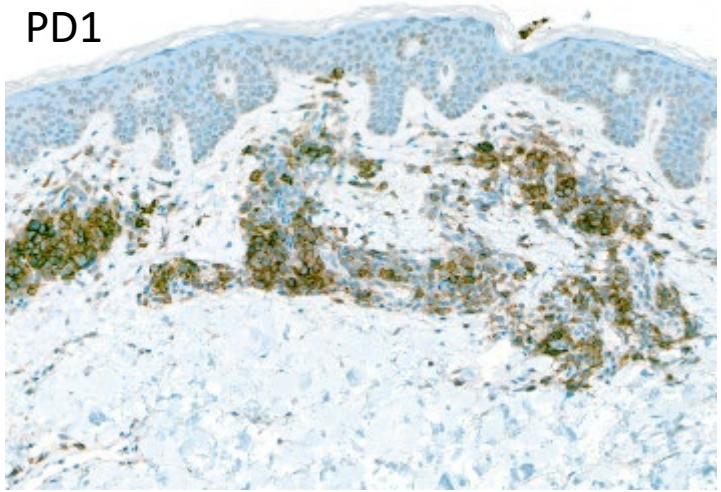
MF plaques in the axillae and abdomen with hypochromic center (Blood B0)



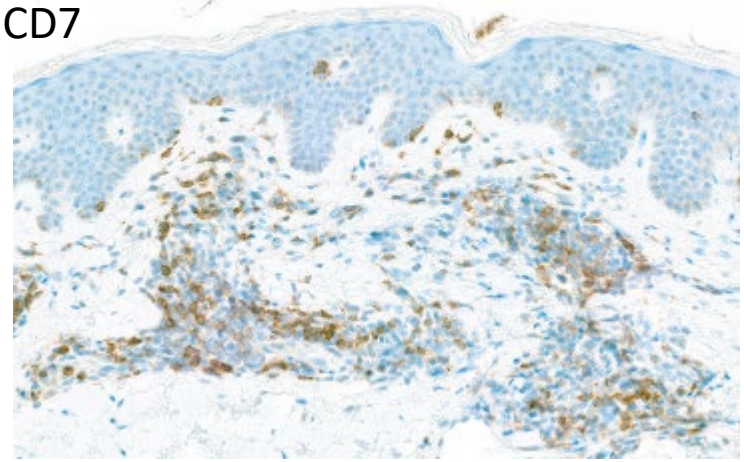
CD3



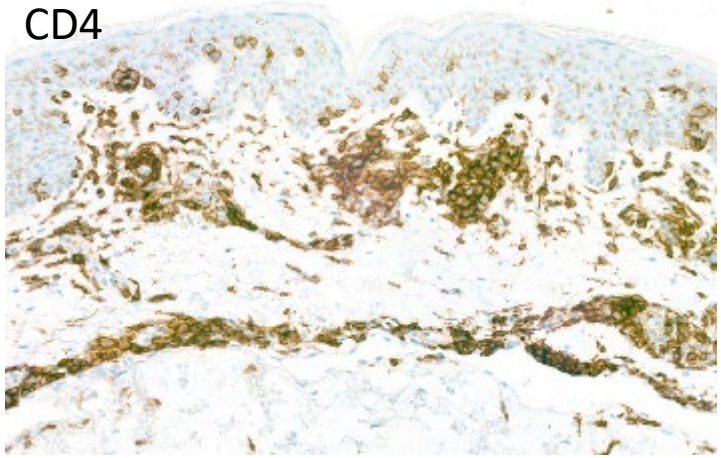
PD1



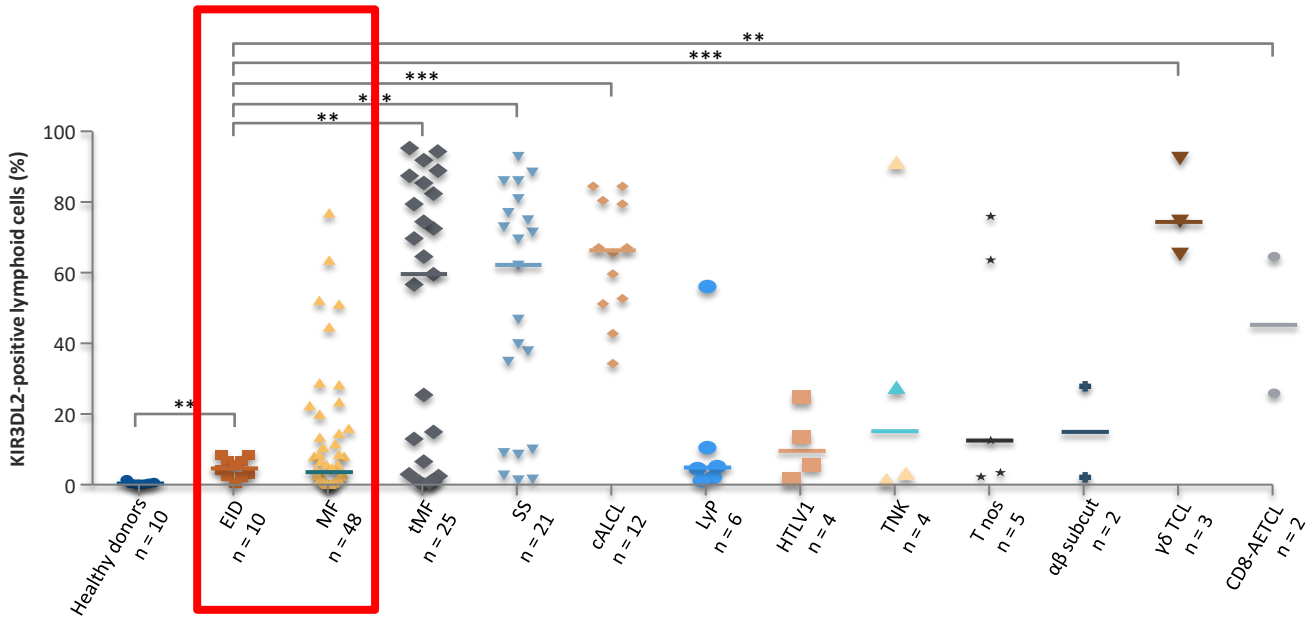
CD7



CD4



KIR3DL2 (CD158k) is not frequently expressed in early MF



- KIR3DL2 significantly increases with MF/SS disease stage
- Age > 60 years, extra-cutaneous involvement, and KIR3DL2 > 10% are associated with OS (multivariate analysis)

Figure from: Battistella M, et al. Blood. 2017;130:2900-2.
Bagot M, et al. Blood. 2001;97:1388-91.
Poszepczynska-Guigné E, J Invest Dermatol. 2004;122:820-3. Ortonne N, et al. Exp Dermatol. 2012;21:461-3.
Hurabielle C, et al. Clin Cancer Res. 2017;23:3619-27. Wechsler J, et al. J Pathol. 2003;199:77-83.
Bouaziz JD, et al. Br J Dermatol. 2010;162:123-8. Battistella M, et al. Br J Dermatol. 2016;175:325-33.

OS, overall survival.

GENERAL DERMATOLOGY

BJD
British Journal of Dermatology

TOX expression in cutaneous T-cell lymphomas: an adjunctive diagnostic marker that is not tumour specific and not restricted to the $CD4^+$ $CD8^-$ phenotype

A.M.R. Schrader,[1] P.M. Jansen[1] and R. Willemze[2]

[1]Department of Pathology and [2]Department of Dermatology, Leiden University Medical Centre, Leiden, the Netherlands

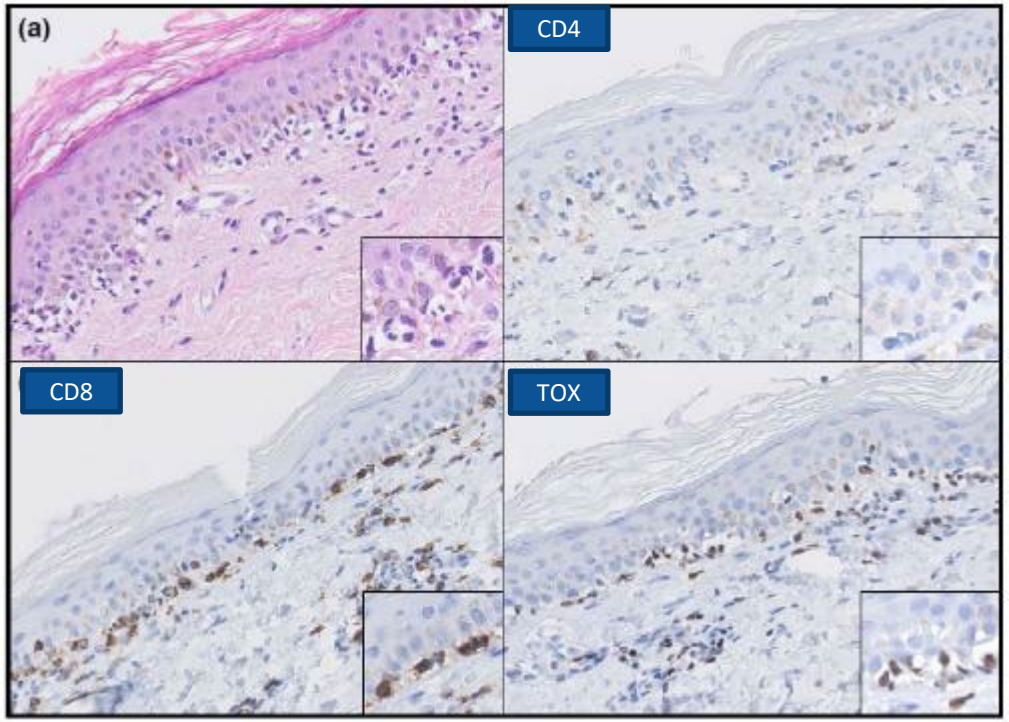
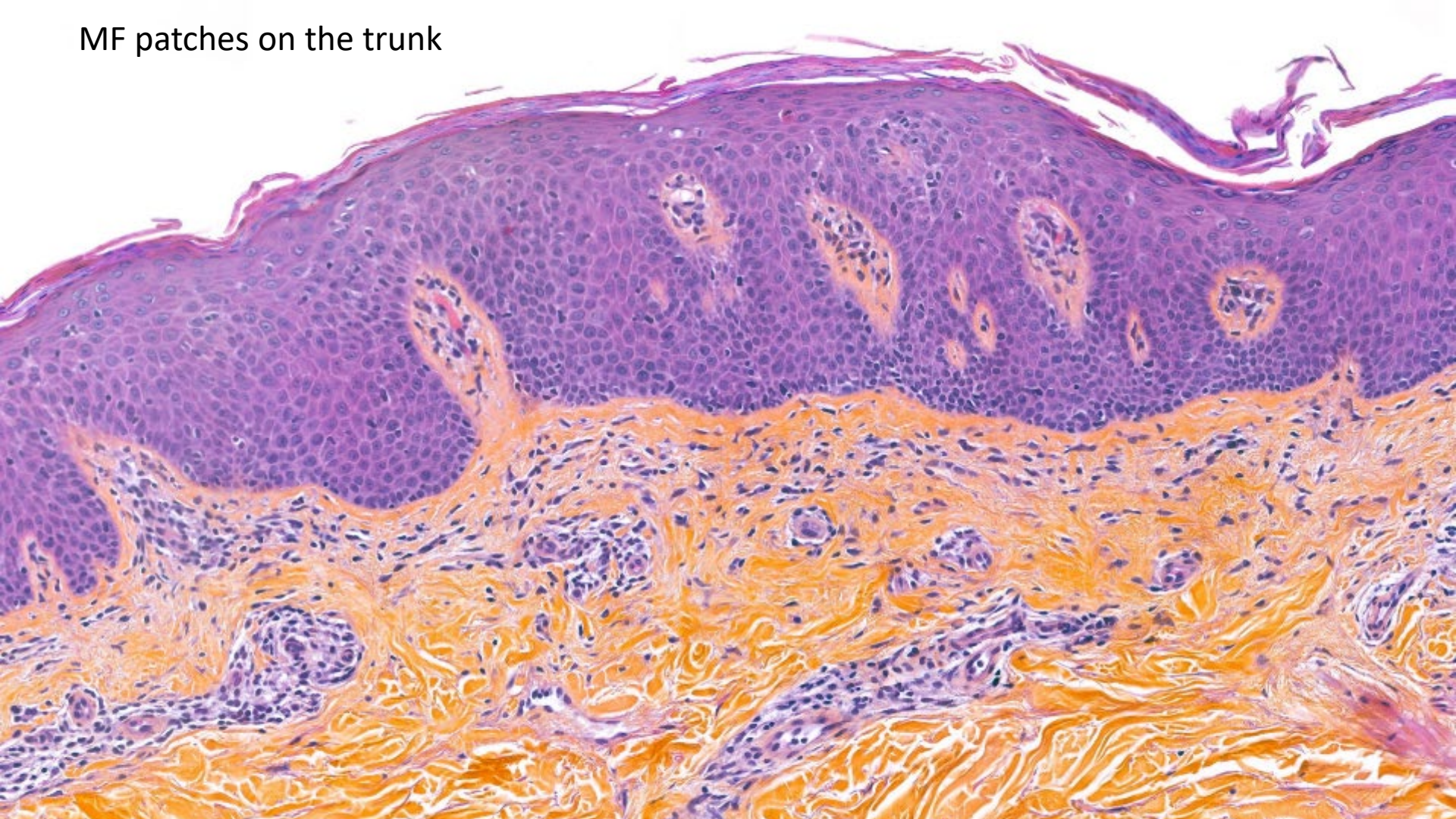


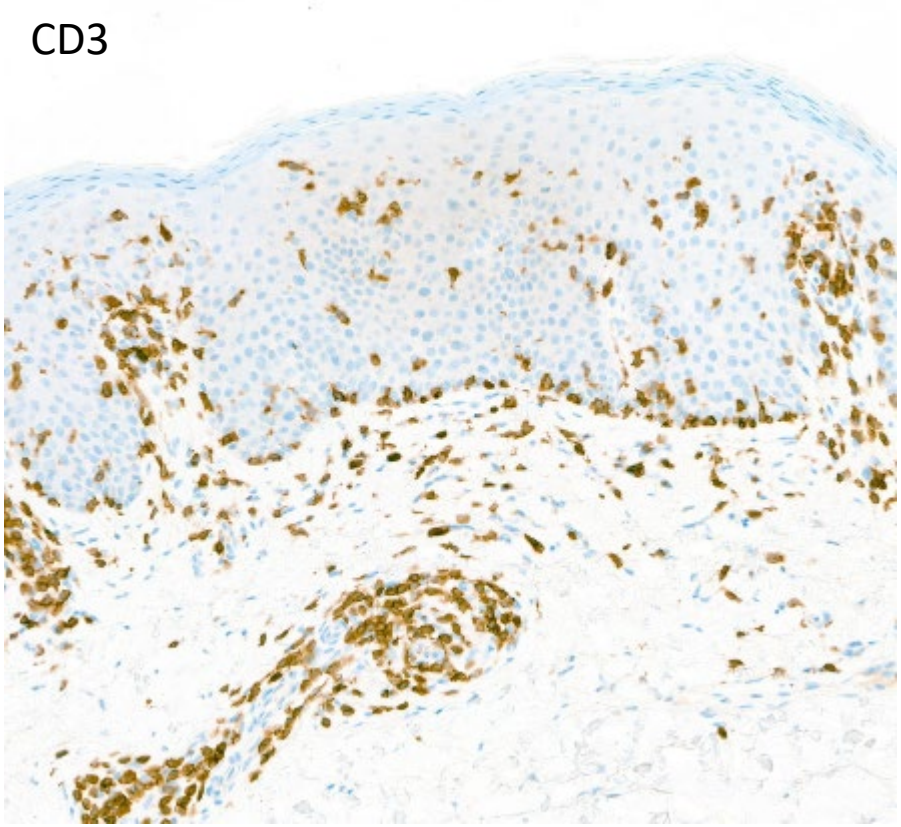
Table 1 TOX expression in different types of cutaneous T-cell lymphoma and benign inflammatory dermatoses

	Amount of TOX expression		
	< 10%	11–50%	> 50%
Mycosis fungoides (n = 59)	3 (5)	7 (12)	49 (83)
Early stage (n = 41)	2 (5)	7 (17)	32 (78)
Advanced stage (n = 18)	1 (6)	0	17 (94)
Sézary syndrome (n = 22)	0	3 (14)	19 (86)
$CD30^+$ LPDs (n = 22)	5 (23)	2 (9)	15 (68)
C-ALCL (n = 11)	4 (36)	1 (9)	6 (55)
Lymphomatoid papulosis (n = 11)	1 (9)	1 (9)	9 (82)
SPTCL (n = 2)	1 (50)	1 (50)	0
PTCL, unspecified and rare subtypes (n = 27)	2 (8)	1 (4)	23 (88)
PTCL-NOS (n = 7)	0	1 (14)	6 (86)
CGDTCL (n = 2)	0	0	2 (100)
AETCL (n = 3)	2 (67)	0	1 (33)
PCSM-TCL (n = 15)	0	0	15 (100)[a]
BPDCN (n = 3)	3 (100)	0	0
Benign inflammatory dermatoses (n = 60)	24 (40)	35 (58)	1 (2)
Atopic dermatitis (n = 12)	4 (33)	8 (67)	0
Psoriasis (n = 10)	5 (50)	5 (50)	0
Drug-induced dermatitis (n = 9)	5 (56)	3 (33)	1 (11)
Idiopathic erythroderma (n = 9)	3 (33)	6 (67)	0
Lichen planus (n = 7)	0	7 (100)	0
Lupus erythematosus (n = 6)	4 (67)	2 (33)	0
Other[b] (n = 7)	3 (43)	4 (57)	0

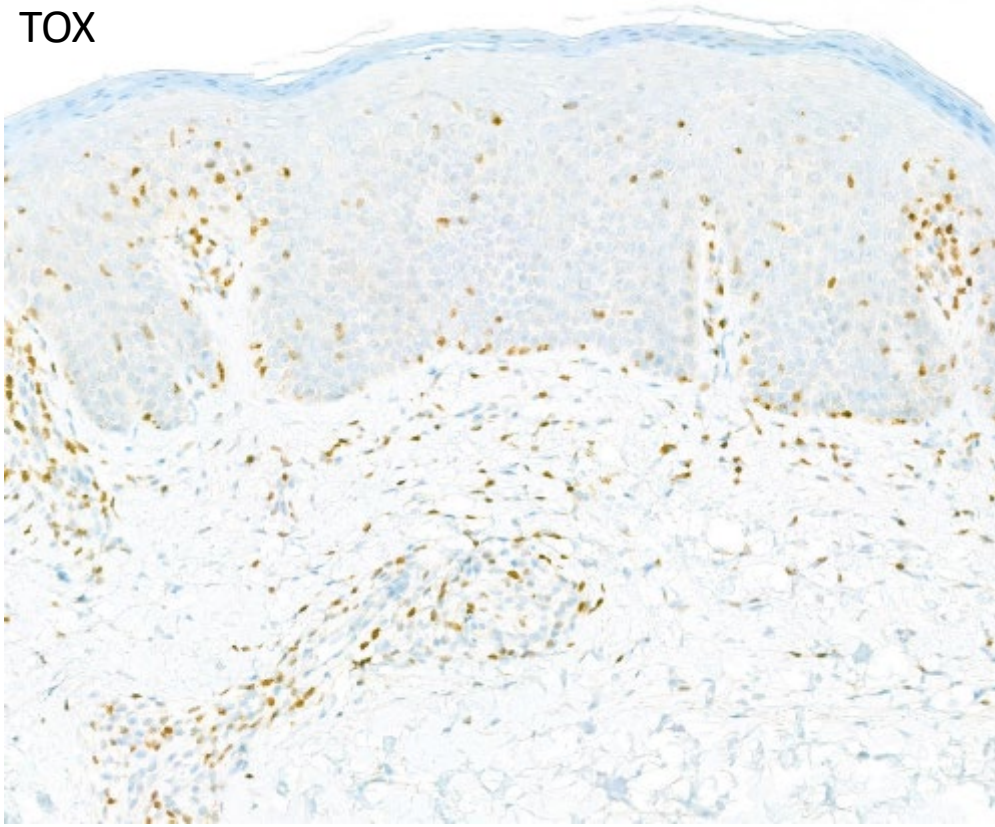
MF patches on the trunk

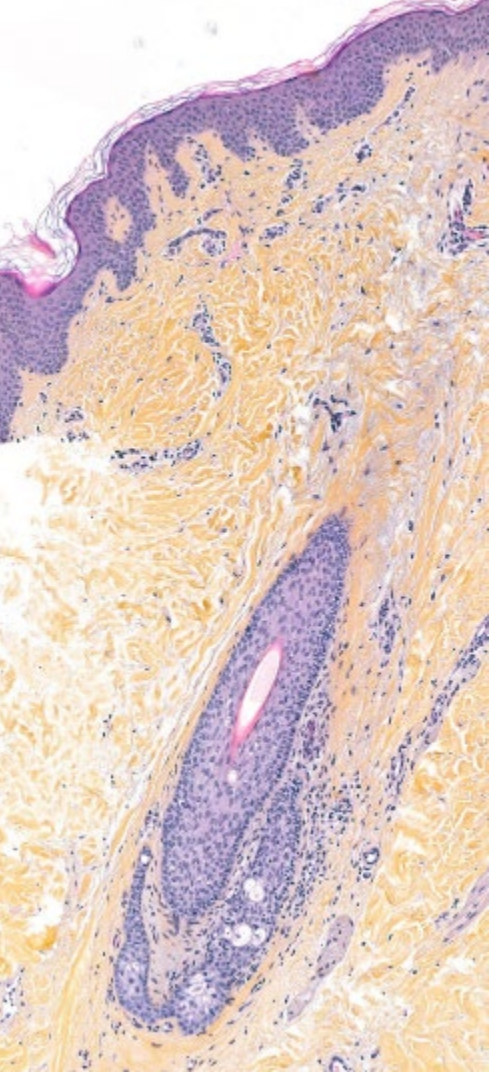


CD3

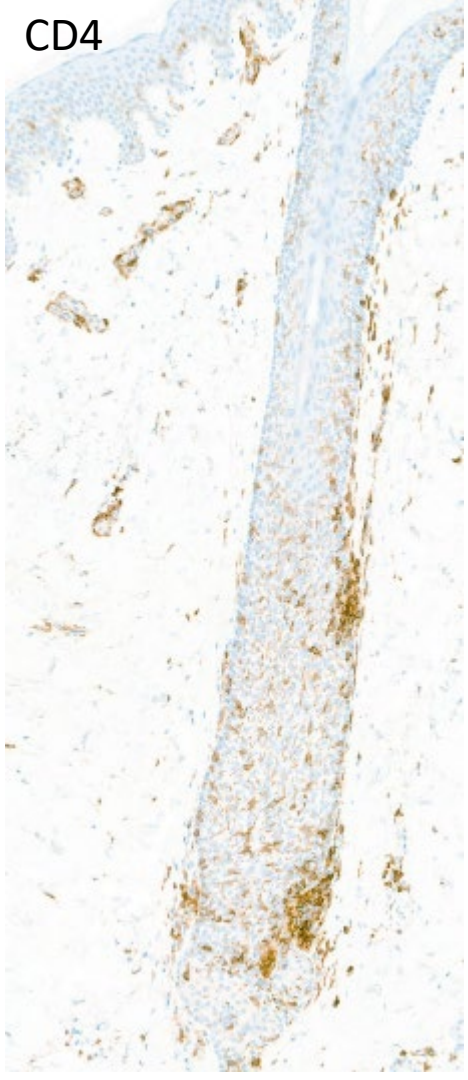


TOX

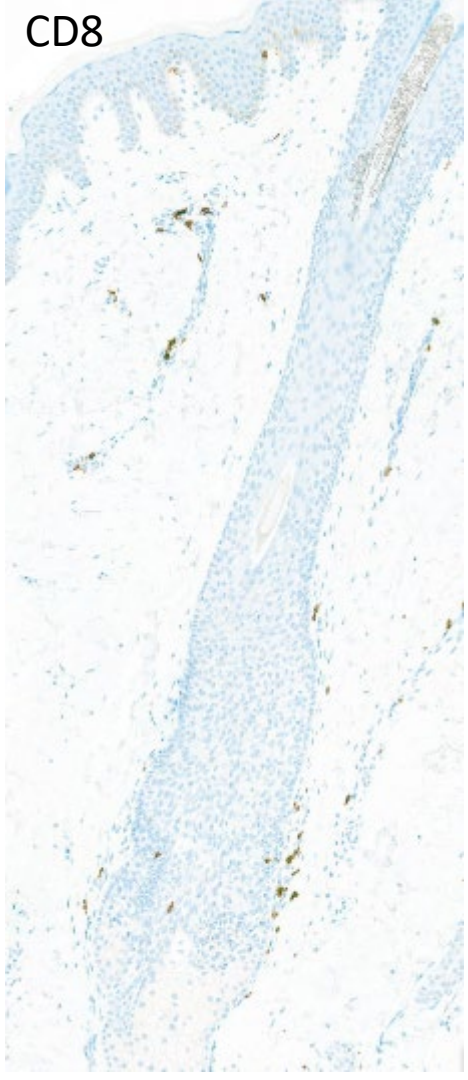




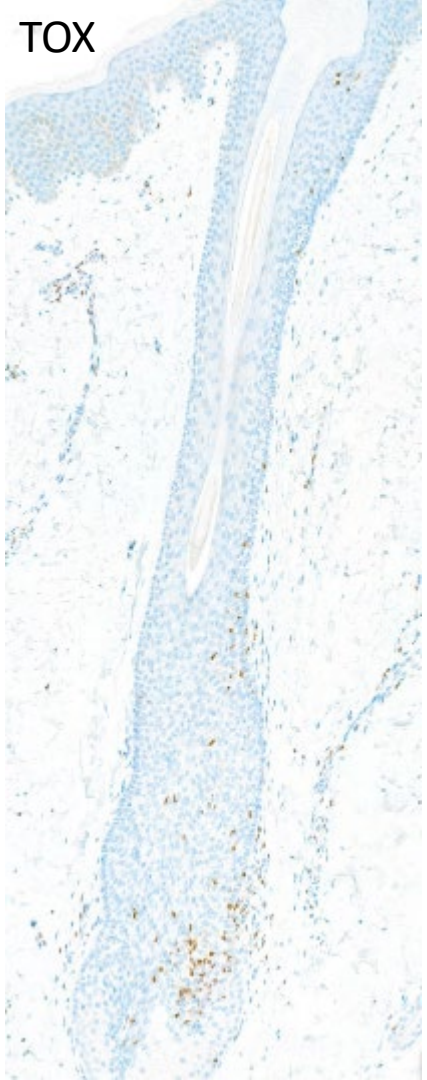
CD4

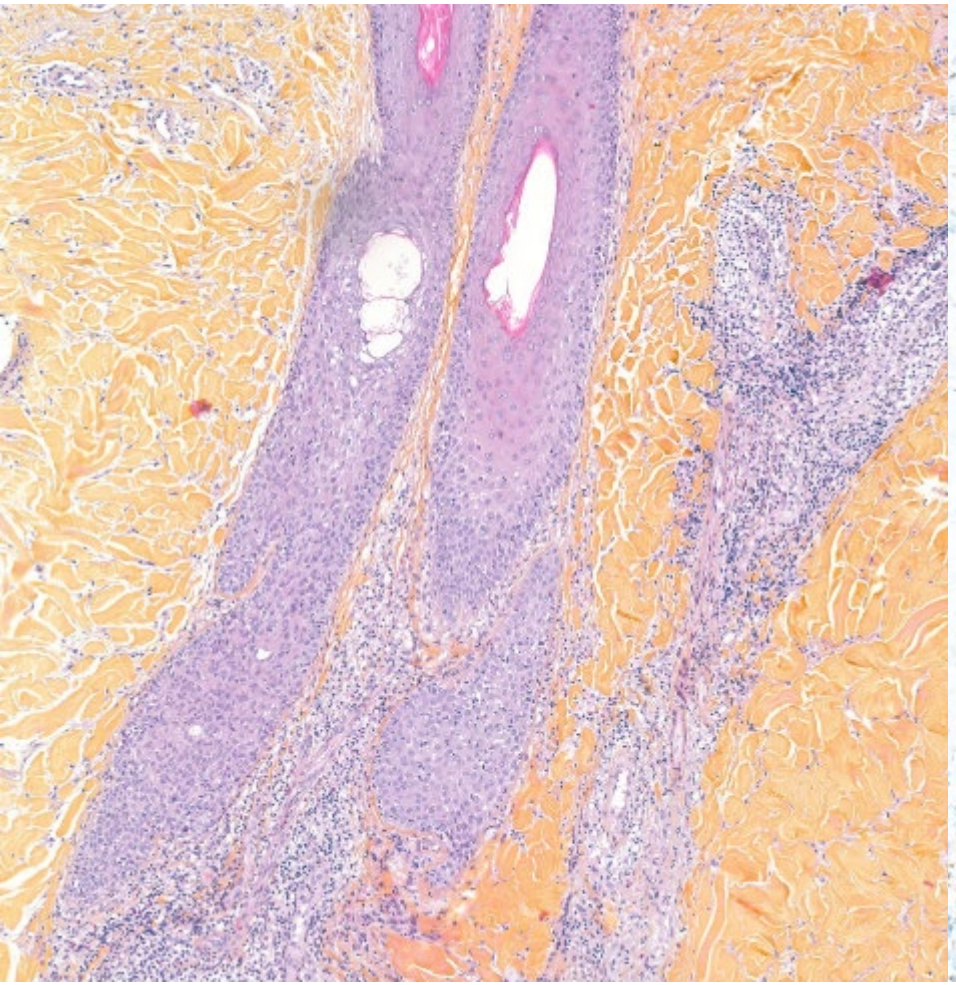


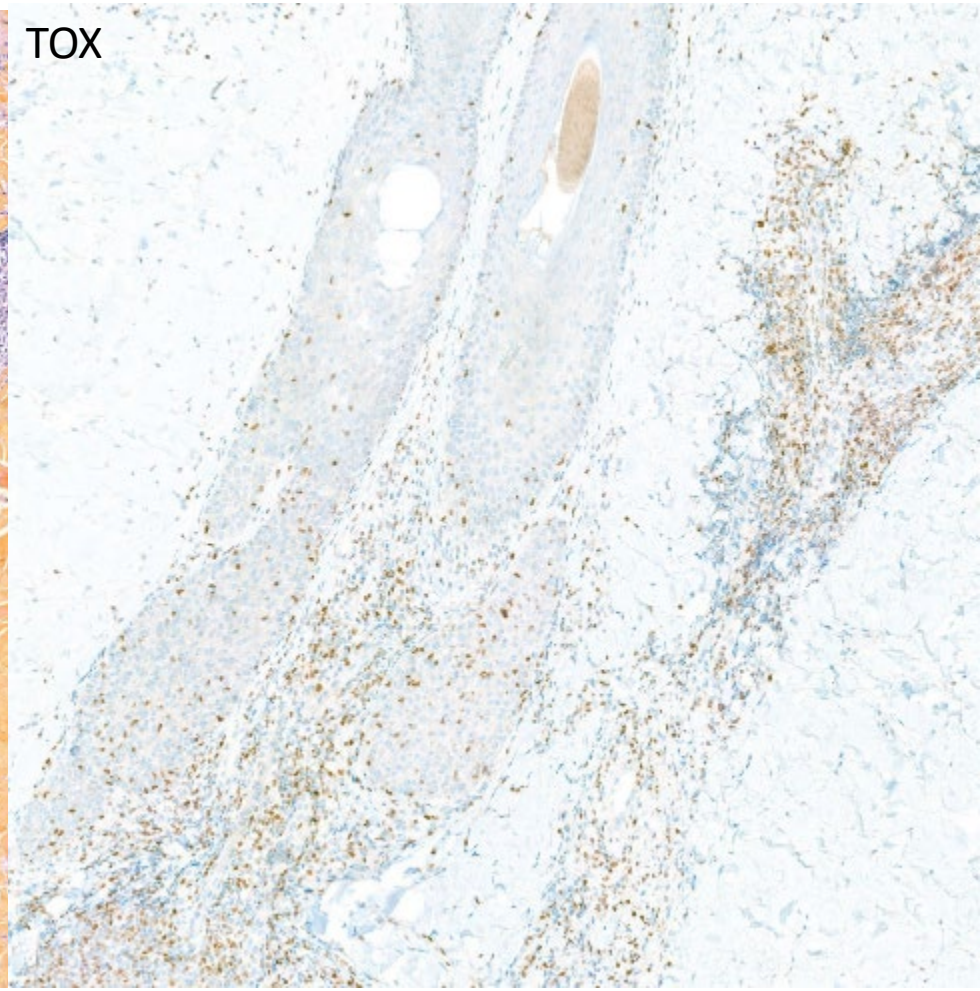
CD8



TOX






TOX

Lessons from PD1, KIR3DL2, TOX IHC in real life

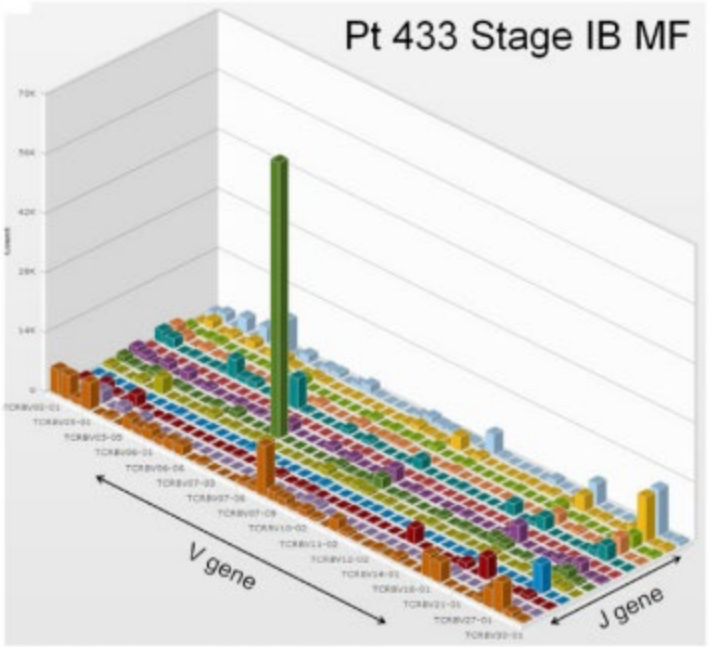
- **PD1**
  - Use a high threshold for % and intensity of staining to suspect SS/MF
  - Useful in erythroderma
  - In patch/plaques rarely strongly positive (rarely useful for diagnosis)

- **KIR3DL2**
  - H5 clone (FFPE compatible) is not commercialized

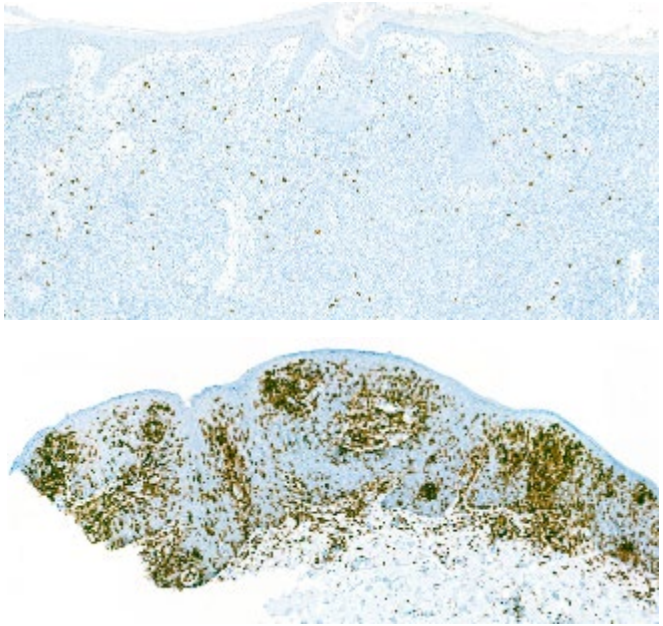
- **TOX**
  - Use a high threshold for % staining / localization of positive cells to favor MF/SS
  - Reactive dermatoses frequently between 25-50% positive
  - Difficult to use for early MF
  - May be better for FMF versus reactive ?

Clonality: new techniques, new limits

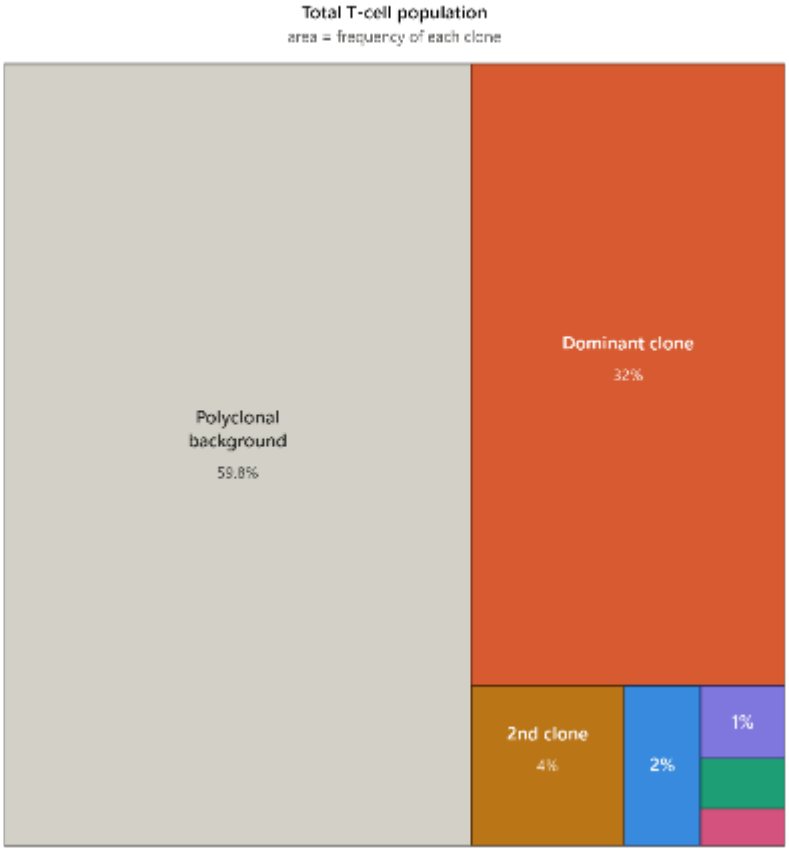
HTS/NGS techniques



TRBC1 immunohistochemistry



HTS of the TCRB or TCRG gene (CDR3 region)



TCF – tumor clone frequency
Rank 1 reads / total productive reads (in %)

Dominant clone TCF : 32%

RPF – ratio of productive frequencies
Rank 1 frequency / Rank 2 frequency

Dominant clone RPF : 32/4= 8

Value of HTS-TCR for early MF diagnosis

- **PCR of TCR gamma**

  Sensitivity : 72% Specificity : 88%

- **HTS-TCRB**
  - Sensitivity : 68-89% Specificity : 95-100%
  - Need consensus for threshold validation
    - % of tumor clone frequency : 5% to 17.5% in the studies ?
    - Ratio of productive frequencies ?

Rea B et al, J Clin Pathol 2018
Kirsch et al, Sci Transl Med 2015
Sufficool KE et al, JAAD 2015

Diagnostic performance of high-throughput sequencing of the T-cell receptor beta gene for the diagnosis of cutaneous T-cell lymphoma

Zimmermann et al, Br J Dermatol 2021

MF diagnosis vs ID

TCF threshold 5%
RPF threshold 3.7

Sensitivity: 89%
Specificity: 95%

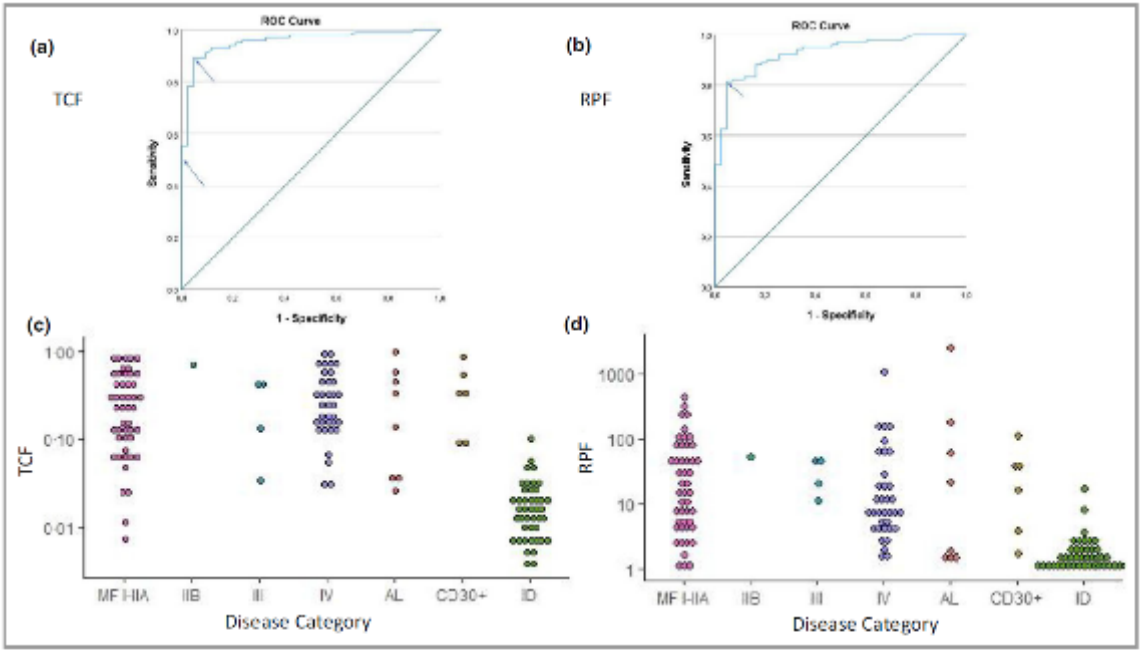
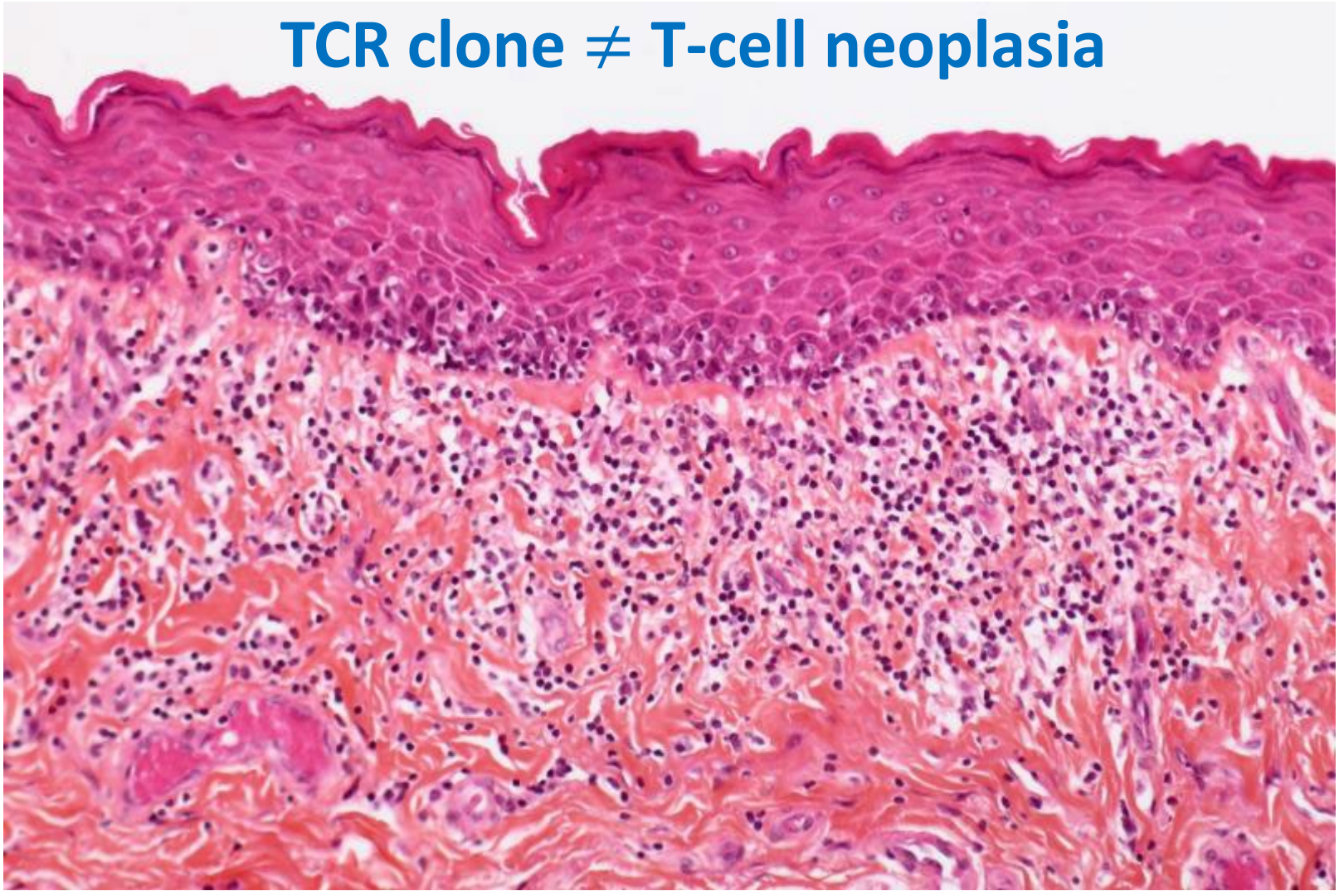
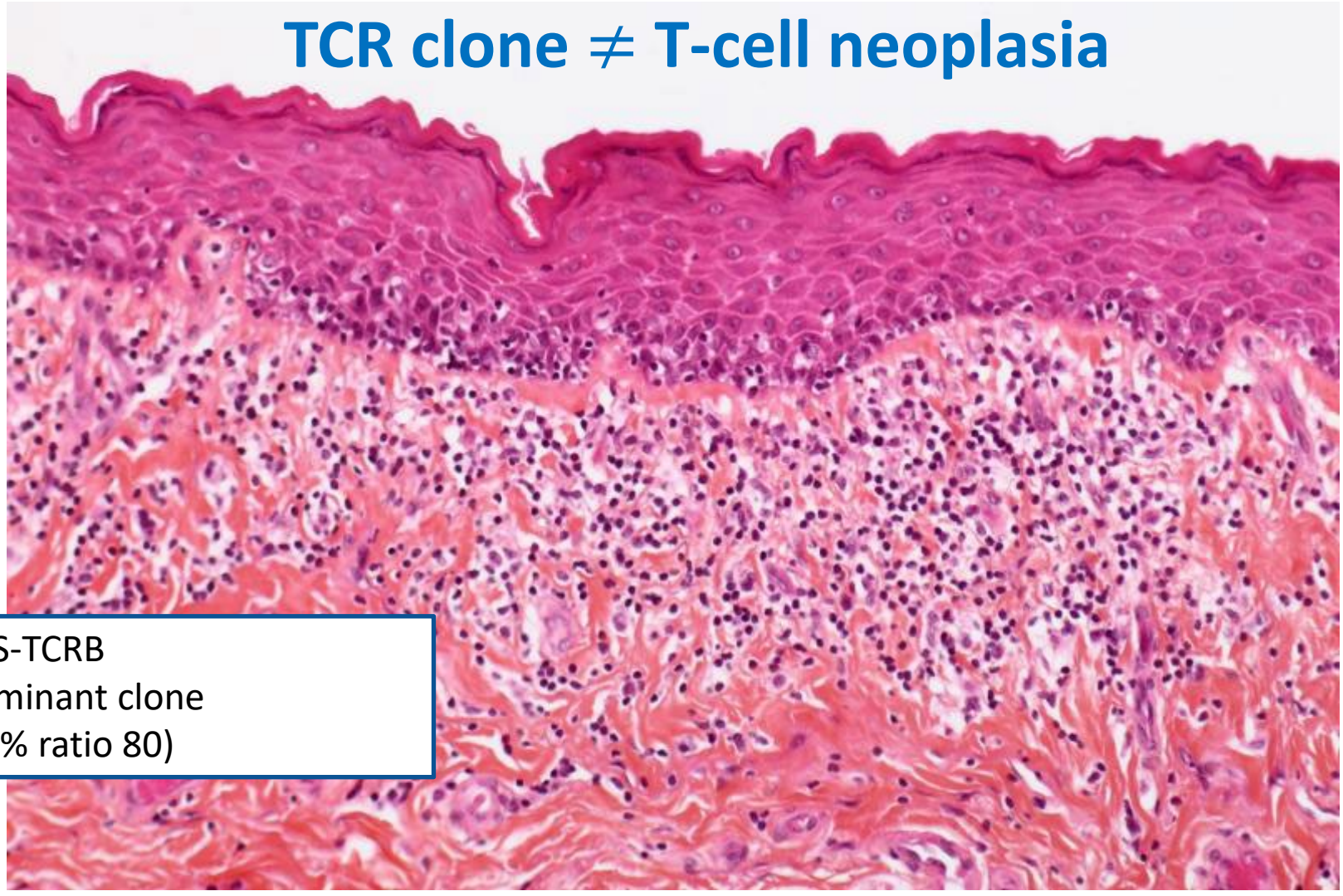


Figure 1 (a) Sensitivity, specificity and receiver operating characteristic (ROC) curve of the tumour clone frequency (TCF), defined as the dominant T-cell clone frequency as a percentage of all T-cell clones in lesional skin by high-throughput sequencing of TRB in skin. The blue arrows show values for TCF of 5% and 25%. (b) Sensitivity, specificity and ROC curve of the ratio of productive frequencies (RPF), defined as the ratio between the first and second T-cell clone frequencies in lesional skin by high-throughput sequencing of TRB in skin. The best result is obtained for an RPF of 3·7 (blue arrow). (c) TCFs according to the diagnosis. (d) RPFs according to the diagnosis. AL, aggressive T-cell lymphoma; $CD30^+$, CD30-positive lymphoproliferative disorders; ID, inflammatory skin disease; MF, mycosis fungoides. Subtypes: AL (seven peripheral T-cell lymphomas not otherwise specified, one primary cutaneous $CD8^+$ T-cell lymphoma); ID (10 atopic dermatitis, 10 contact dermatitis, five psoriasis, three lichen planus, seven drug reactions, two cutaneous lupus erythematosus, six other IDs).

TCR clone ≠ T-cell neoplasia

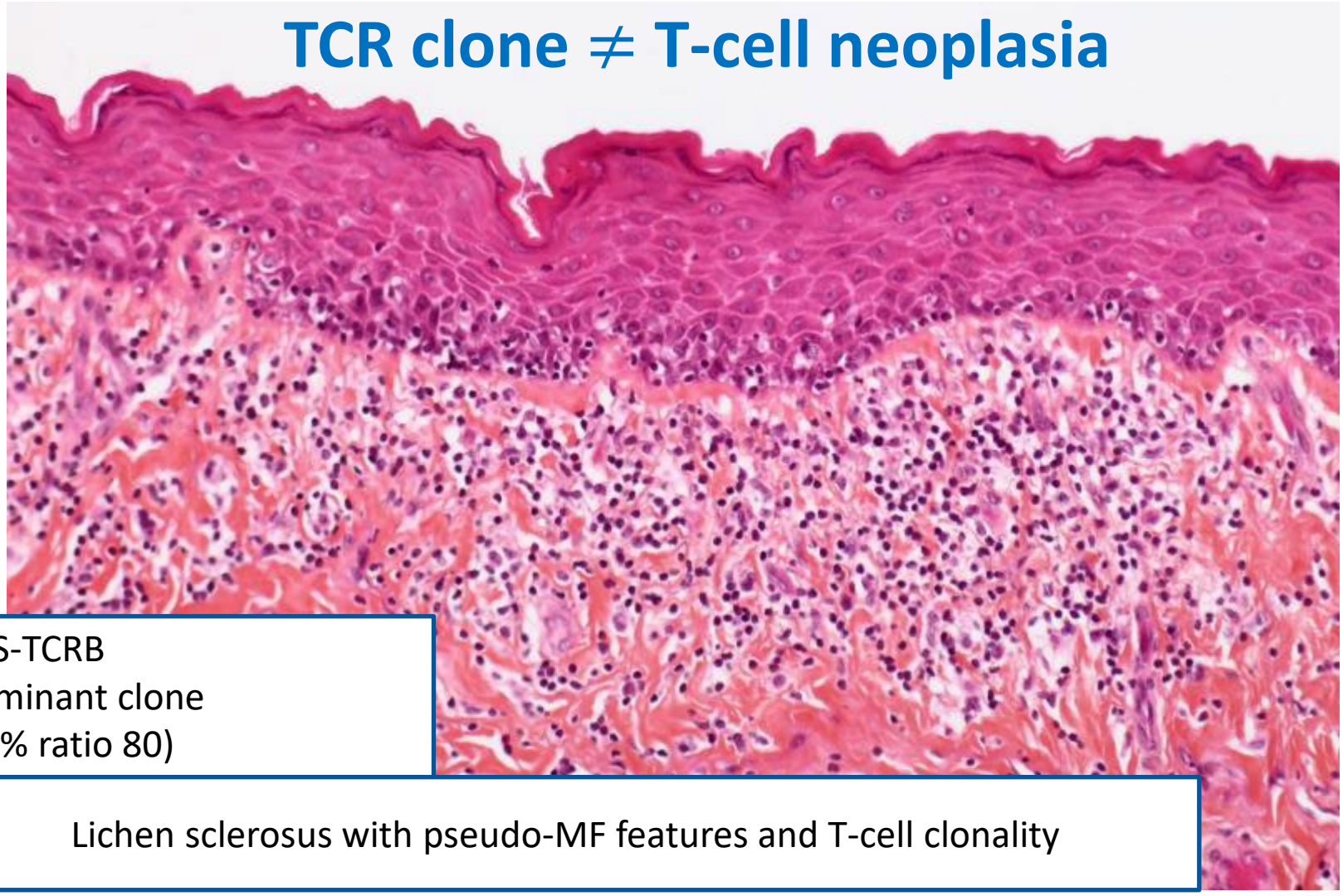


TCR clone ≠ T-cell neoplasia



HTS-TCRB
Dominant clone
(64% ratio 80)

TCR clone ≠ T-cell neoplasia



HTS-TCRB
Dominant clone
(64% ratio 80)

Lichen sclerosus with pseudo-MF features and T-cell clonality

When do I use HTS-TCRB ?

- **At diagnosis (MF/SS)**
  - Enables skin/blood/lymph node clone comparison
  - Enables clone quantification
    - Skin at diagnosis (prognostic value ?)
    - LN (help for staging in LN core needle biopsies)

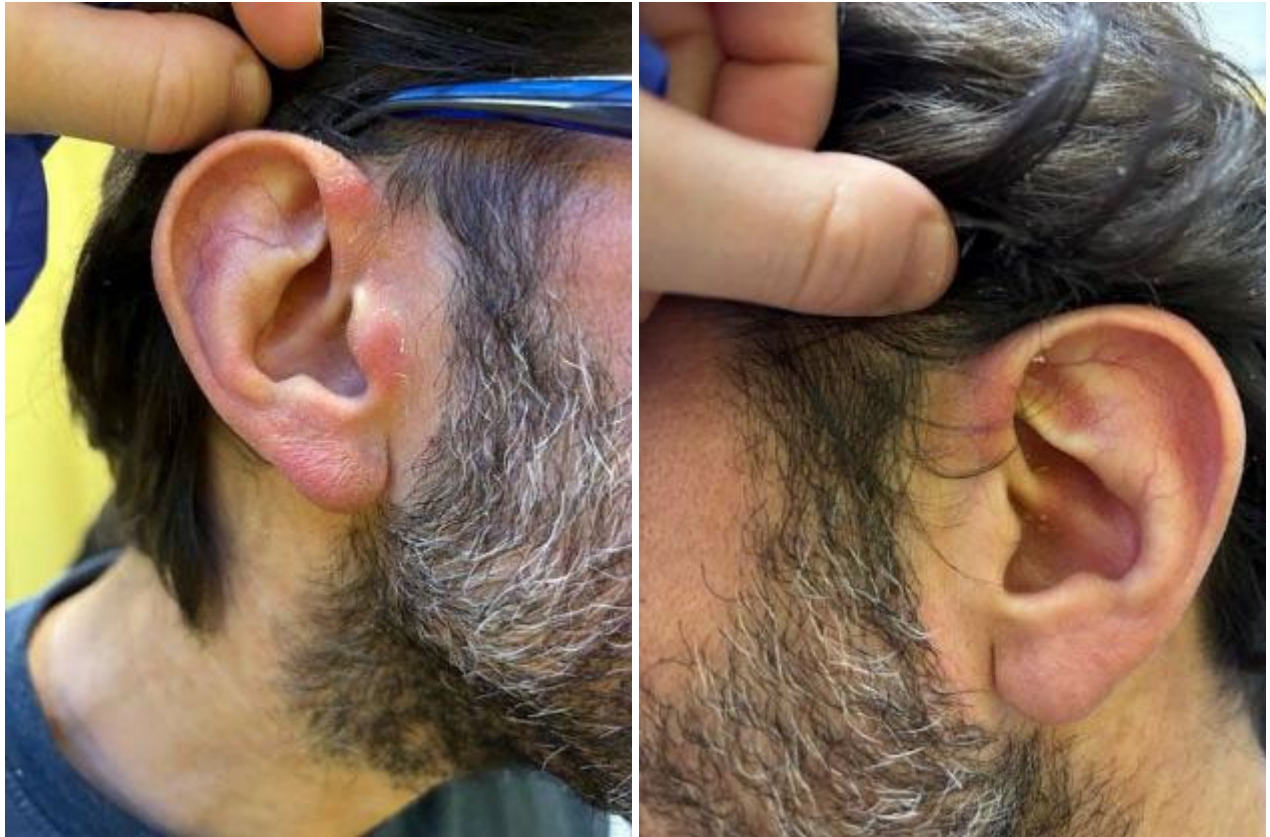
- **During follow-up**
  - Recurrences
  - MRD
  - Differential diagnosis
    - Mogamulizumab-associated rash vs recurrence
    - GVHD vs recurrence

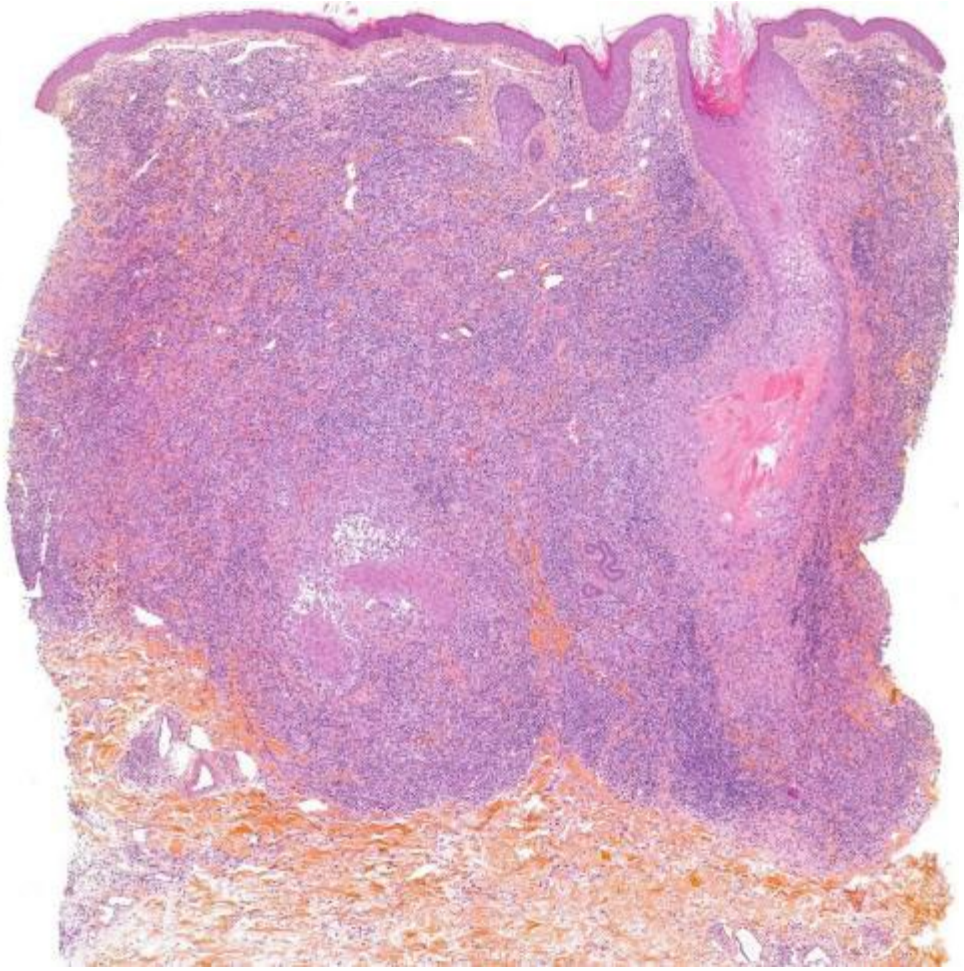
Granulomatous and folliculocentric mogamulizumab-associated rash

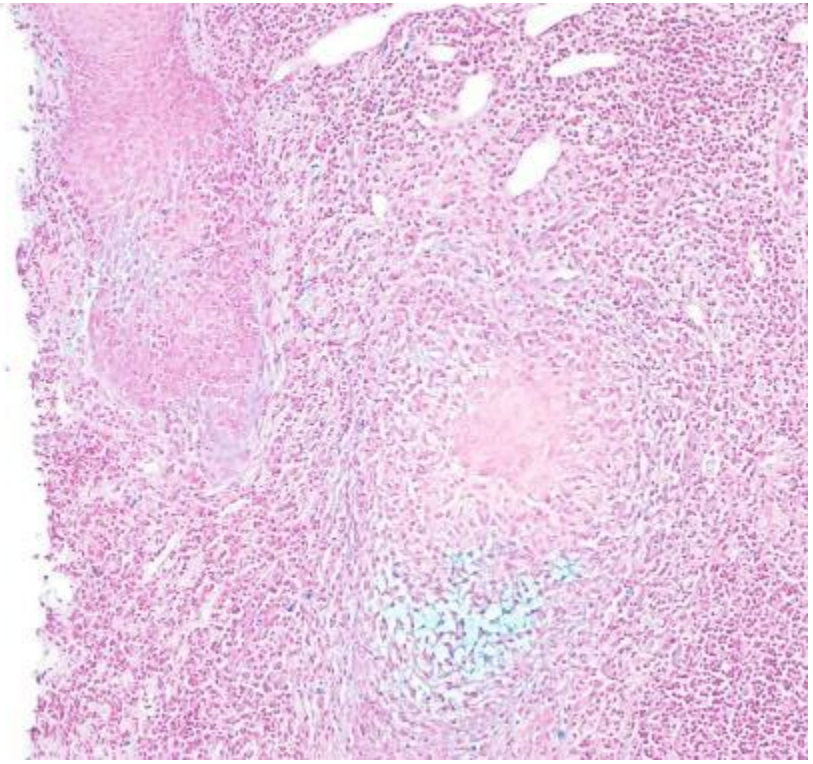
Sezary syndrome

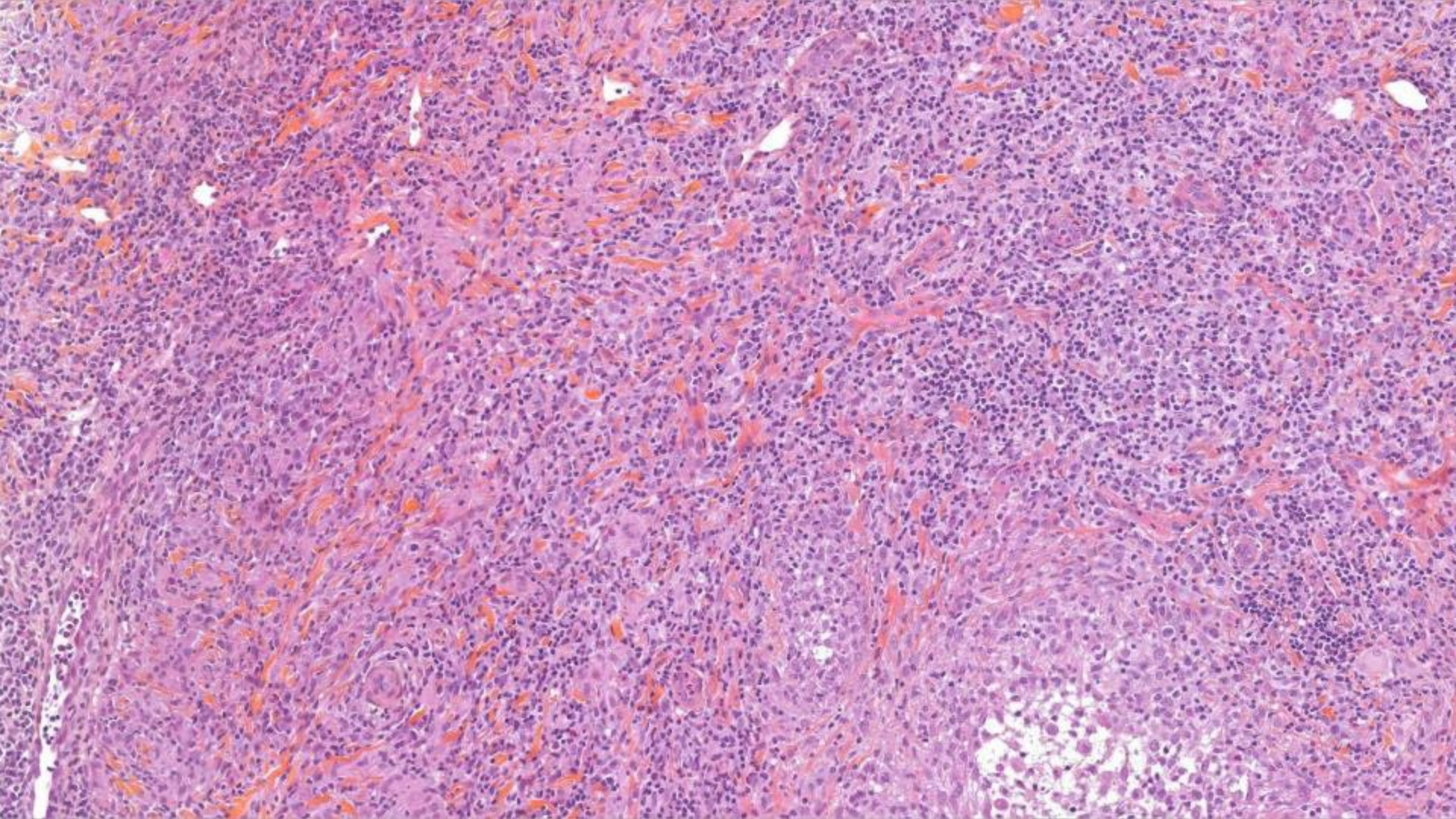
CR under MGZ

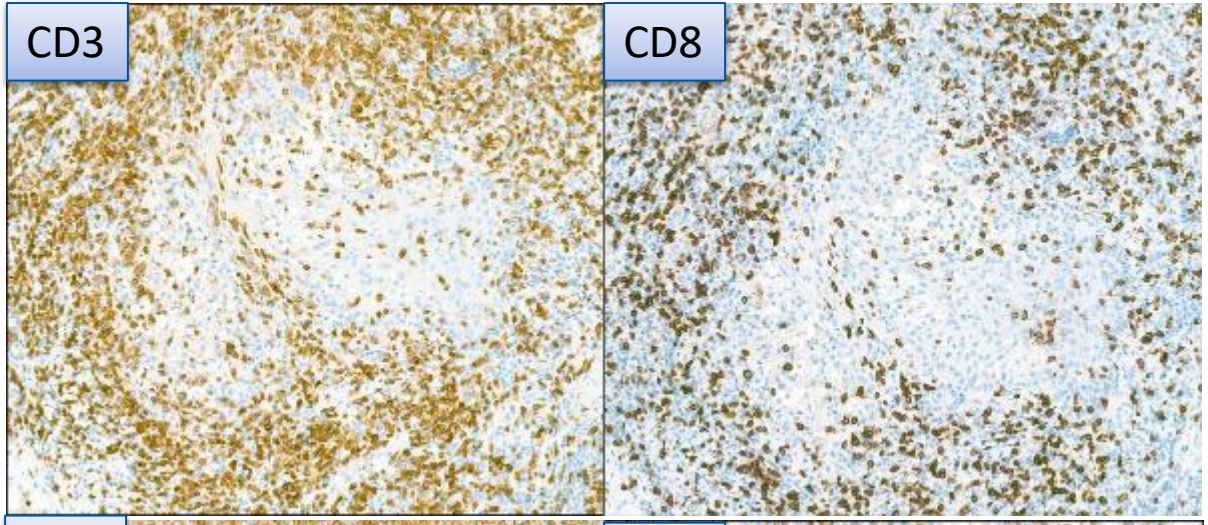
Novel eruption after 4 months of MGZ

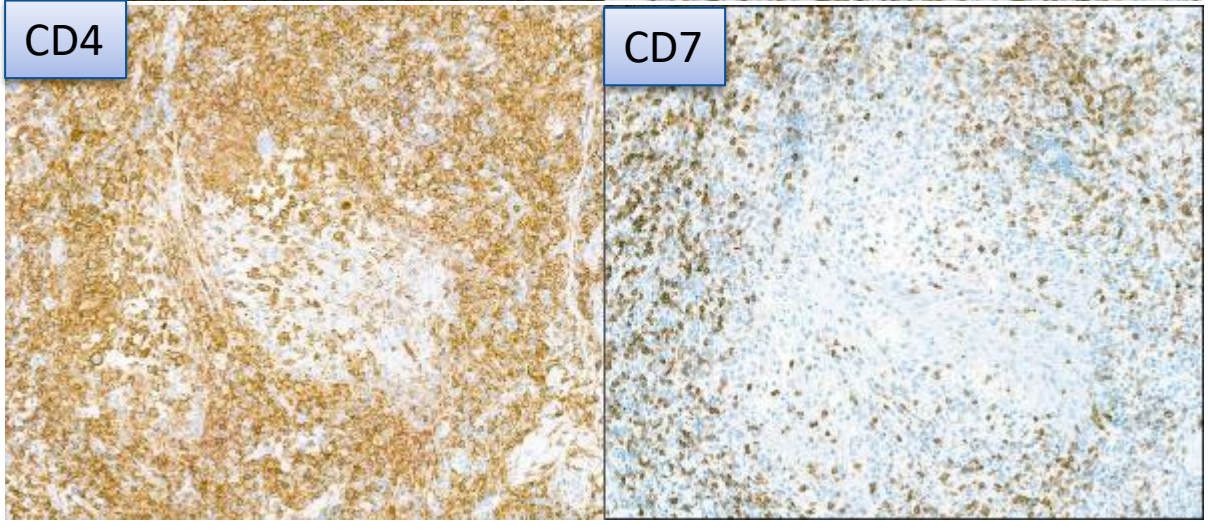










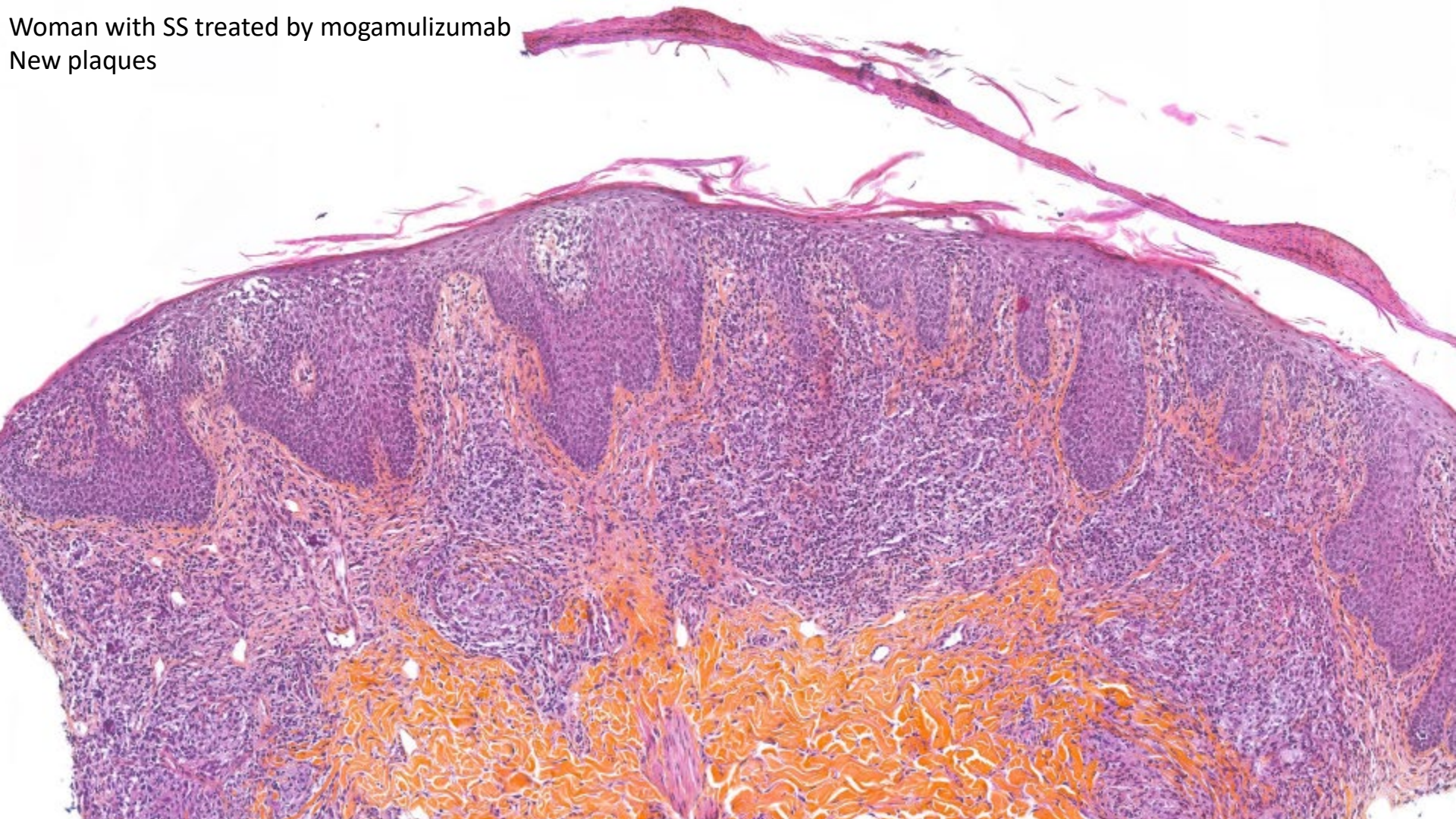


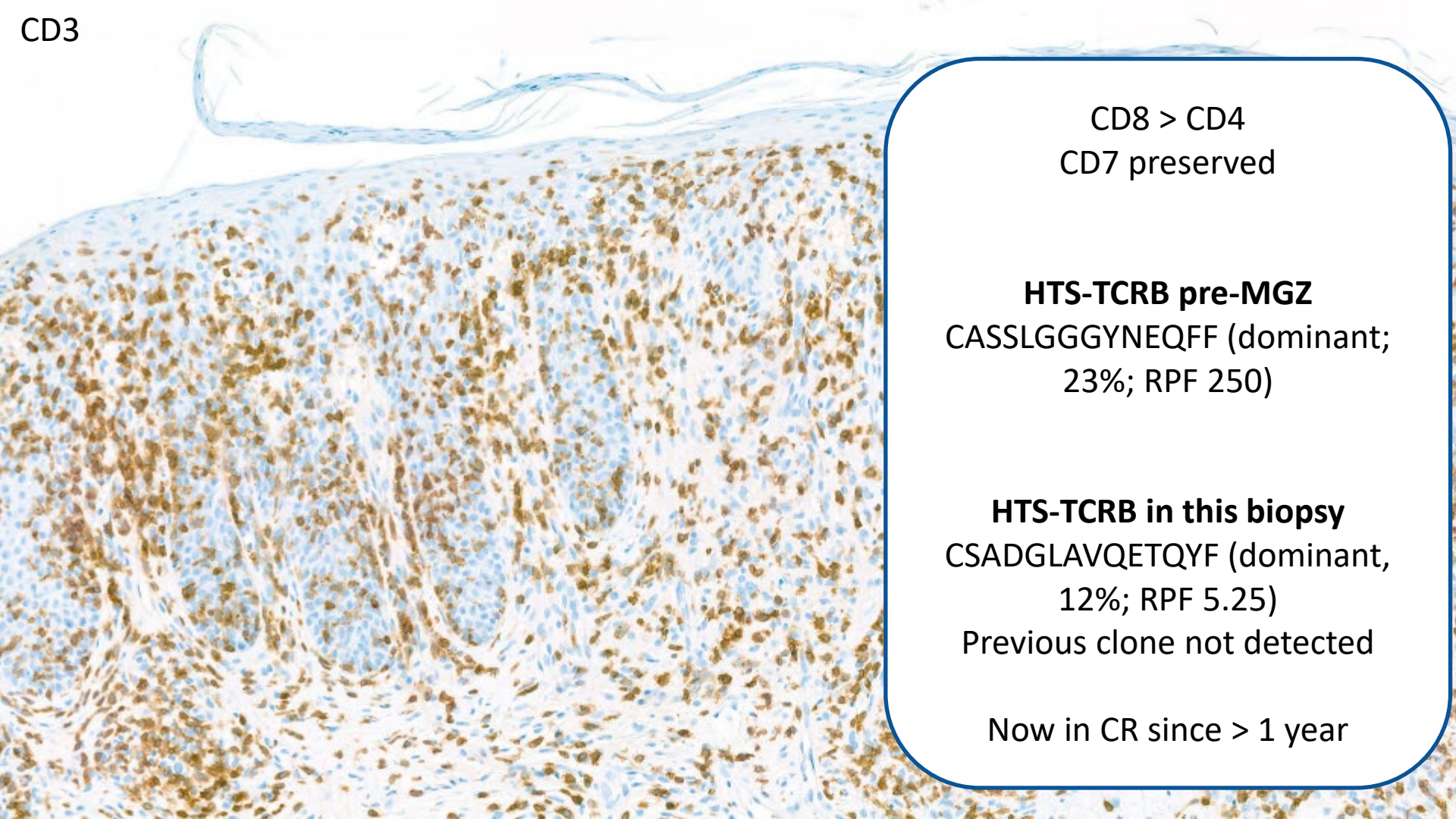
HTS-TCRB

Pre-MGZ biopsy
CASSLVGYGYTF clone 77% (ratio 52)

MAR biopsy
- No dominant clone
- CASSLVGYGYTF clone not present in the 1000 first clones

Woman with SS treated by mogamulizumab
New plaques





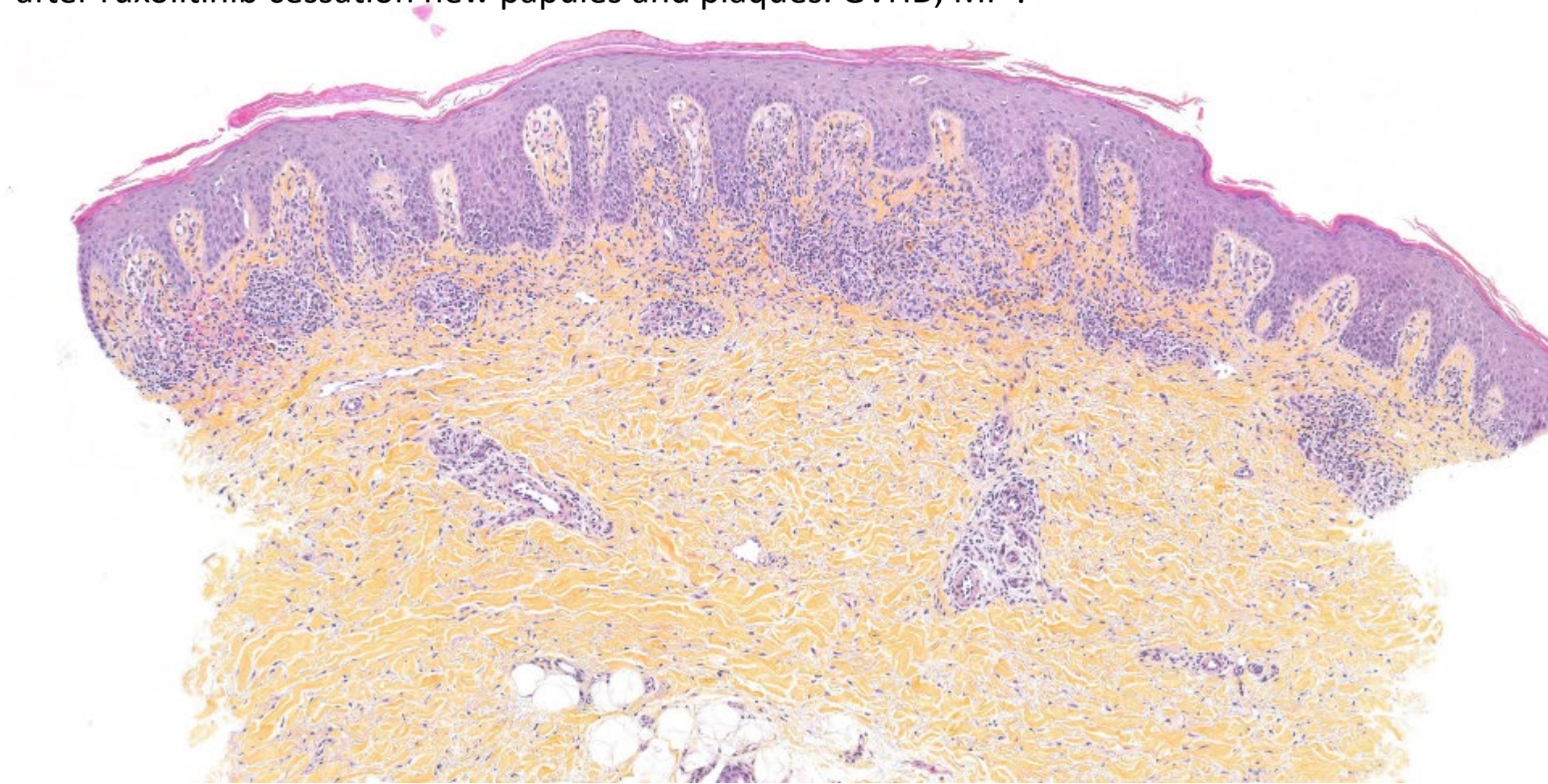
CD8 > CD4
CD7 preserved

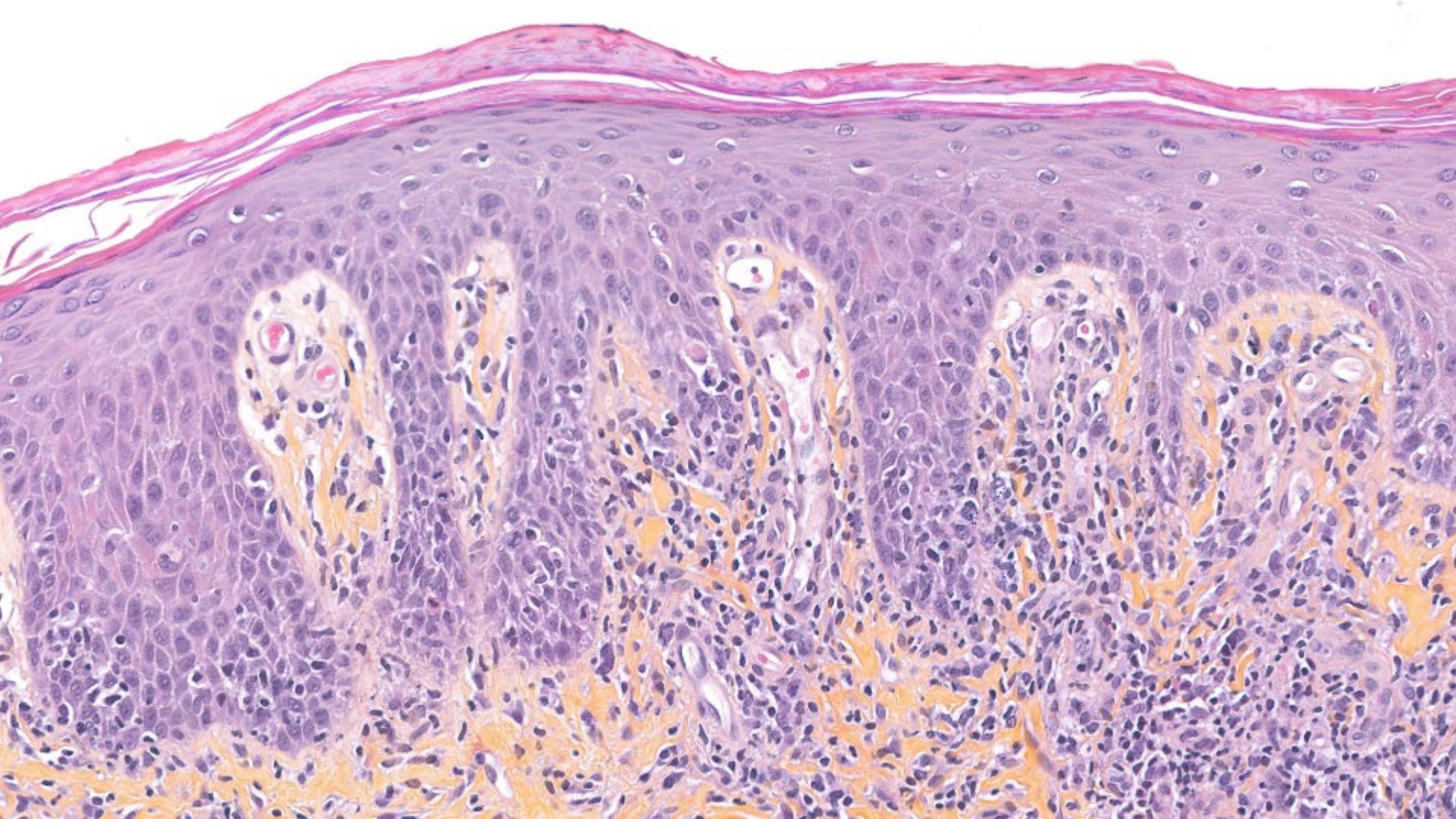
HTS-TCRB pre-MGZ
CASSLGGGYNEQFF (dominant; 23%; RPF 250)

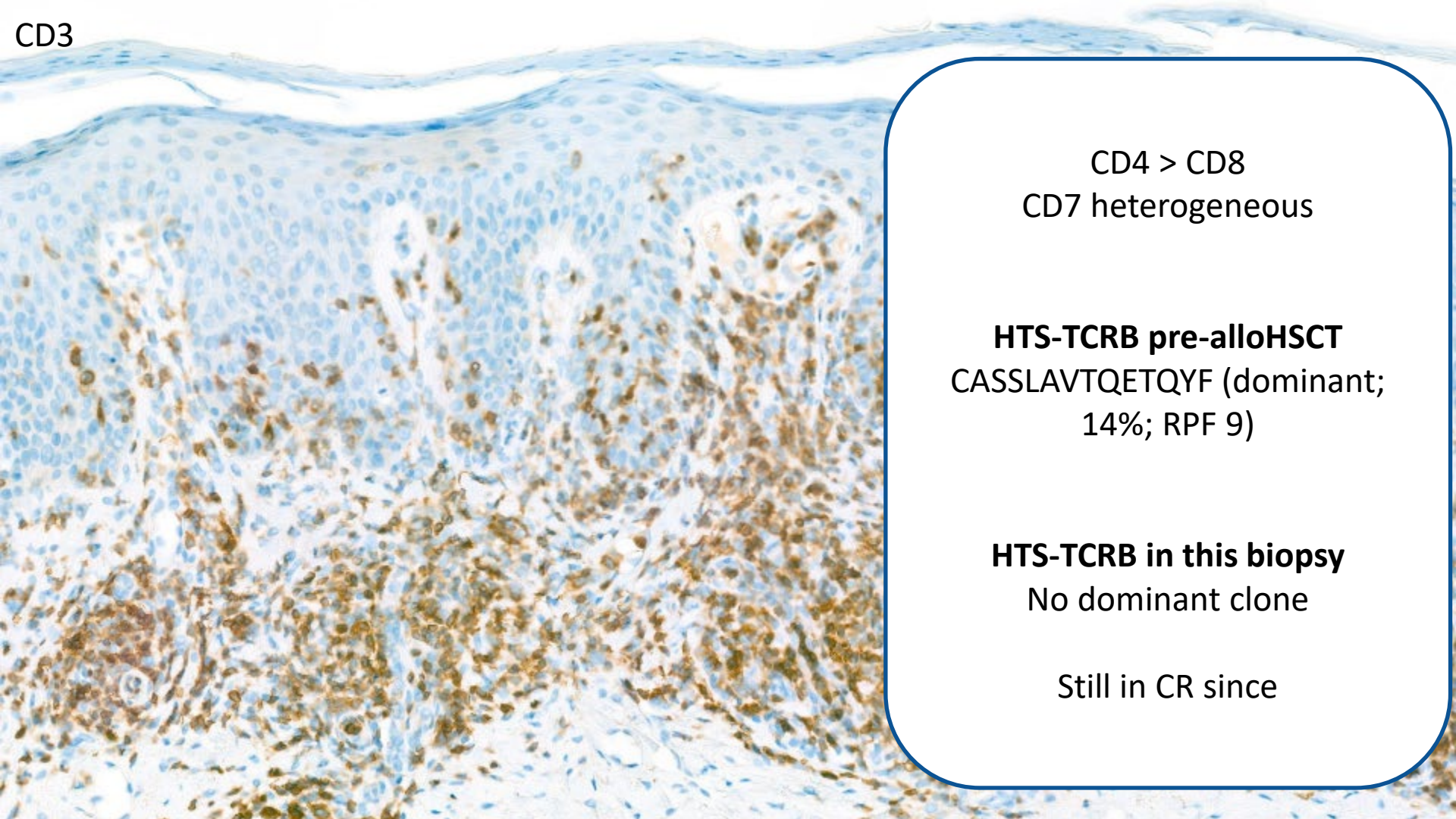
HTS-TCRB in this biopsy
CSADGLAVQETQYF (dominant, 12%; RPF 5.25)
Previous clone not detected

Now in CR since > 1 year

39 yo man ; 18months after allo-SCT for MF ; history of GVHD treated by ruxolitinib ; 3 months after ruxolitinib cessation new papules and plaques. GVHD, MF ?







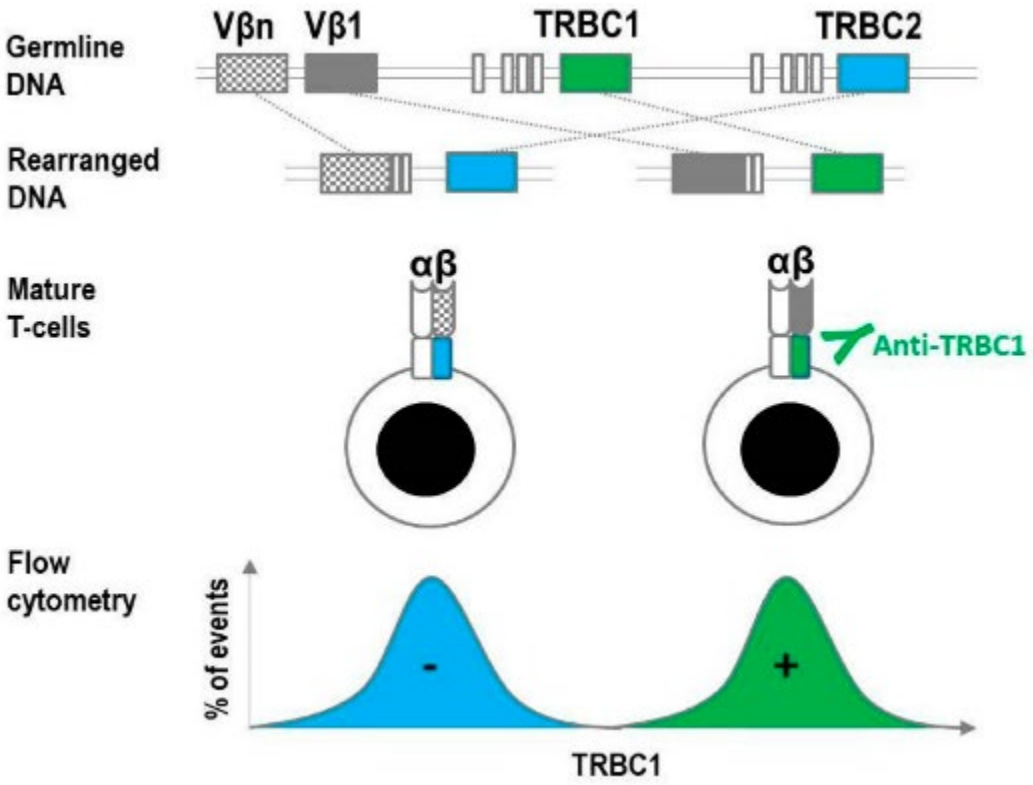
CD4 > CD8
CD7 heterogeneous

HTS-TCRB pre-alloHSCT
CASSLAVTQETQYF (dominant; 14%; RPF 9)

HTS-TCRB in this biopsy
No dominant clone

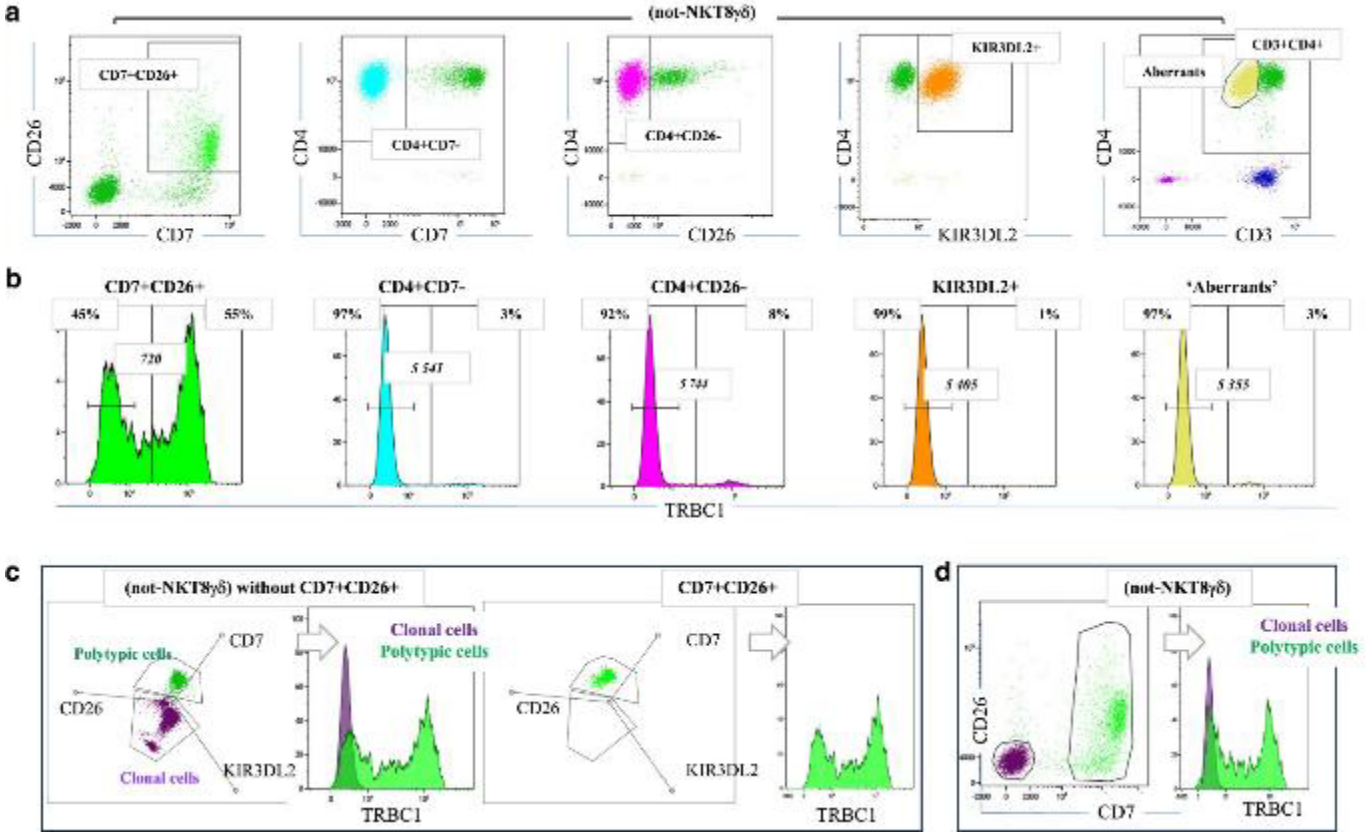
Still in CR since

TRBC1 for clonality detection



Horna et al, Int J Mol Sci 2021

TRBC1 flow-cytometry for clonality detection



Ta VA et al, J Invest Dermatol 2025

TRBC1 immunohistochemistry for clonal detection

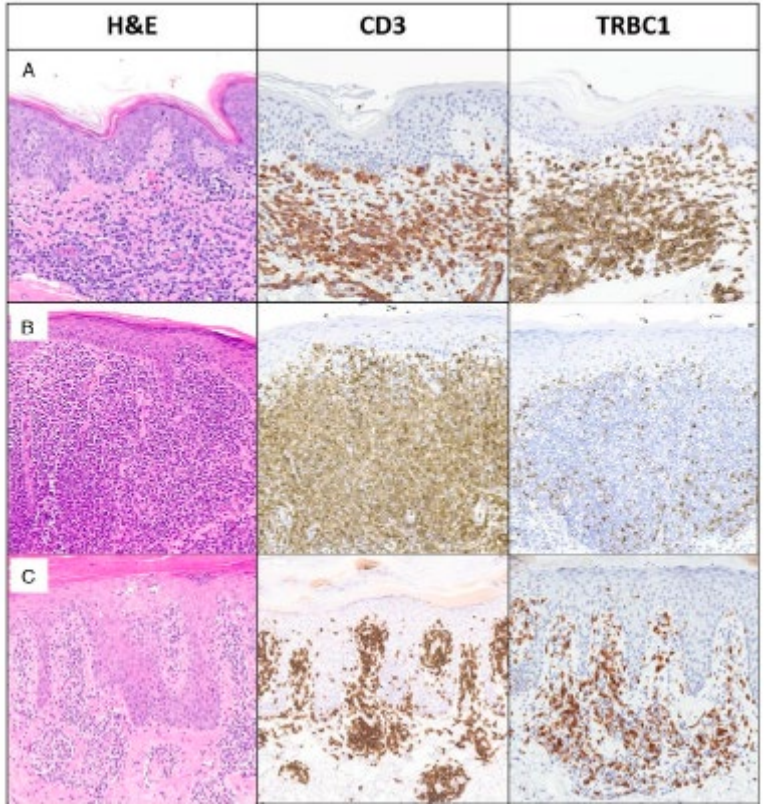


FIGURE 1. Dot plot showing the distribution of TRBC1%, quantified as TRBC1+ cells as a percentage of CD3-positive cells, stratified by consensus interpretation as monoclonal (positive) or polyclonal/indeterminate (negative). Each dot represents an individual case. Dot color indicates T-cell receptor (gold standard) clonality status, with green denoting monoclonality and black denoting polyclonality.

Sensitivity 72% in MF (50% if only intraepithelial cells scored) ; Specificity 97%

Suggested threshold to diagnose clonality : ≤20% or ≥80%

Grand et al, AJSP 2026

Performance of TRBC1 Immunohistochemistry Versus High-Throughput Sequencing of TCRB for Clonality Assessment in CTCL

Andy Li, Maxime Mouthon, Maxence Mancini, Caroline Ram-Wolff, Baptiste Louveau, Fanélie Jouenne, Adèle de Masson, Samia Mourah, Maxime Battistella.

Saint-Louis Hospital, AP-HP
INSERM UMRS-1342, Université Paris Cité
Paris, France

Hôpital Saint-Louis AP-HP

Inserm

IRSL Institut de Recherche Saint-Louis

Université Paris Cité

Methods

Study design

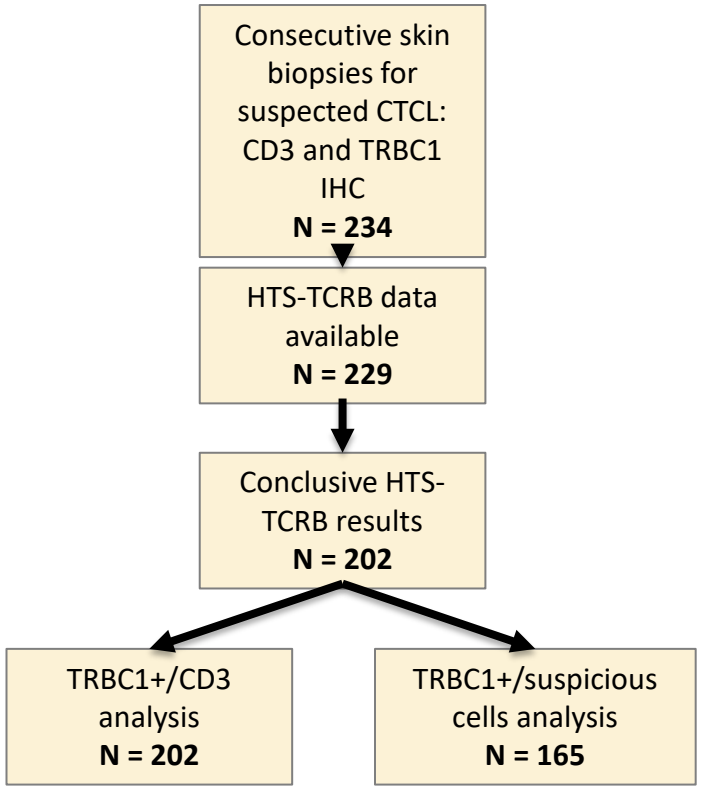
- Prospective, single-center

Clinical context

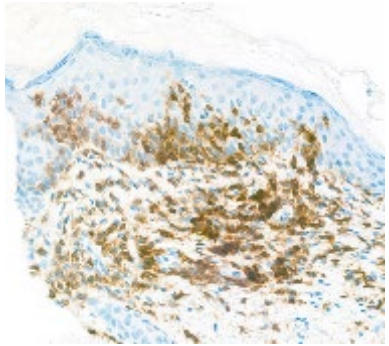
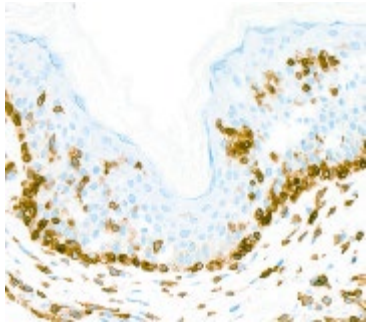
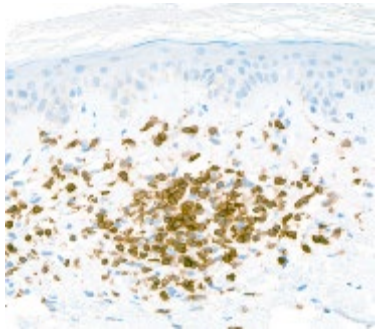
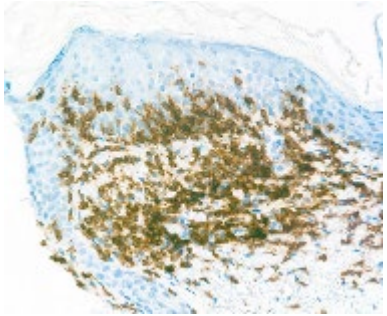
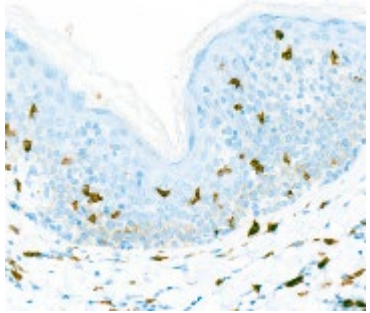
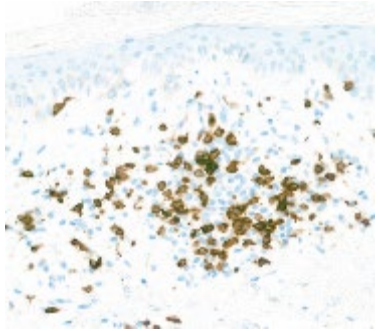
- MF suspicion (patches-plaques)
- Erythroderma
- Mogamulizumab-associated rash vs CTCL
- Graft-vs-Host Disease vs CTCL

Methods

- **Sequential** CD3 and TRBC1 IHC, assessed by expert pathologist
  - %TRBC1 in CD3+ population global assessment (TRBC1+/CD3)
  - % TRBC1 in morphologically suspicious cells
- HTS-TCRB considered clonal if **TCF>5%** & **RPF>3.7**

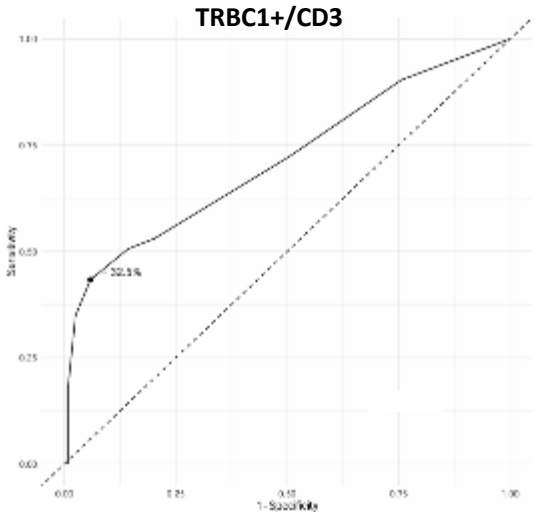


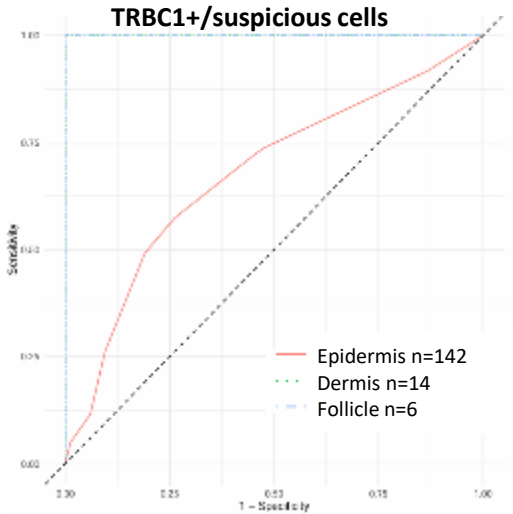
Results: Examples

CD3			
TRBC1			
TRBC1+/CD3	95%	10%	40%
HTS-TCRB	**Clonal** ; TCF 82% ; RPF 203	**Clonal** ; TCF 43% ; RPF 58	**Polyclonal**

Results: Overall Performance

Method	N	AUC	Se (%)	Spe (%)	PPV (%)	NPV (%)	Acc (%)
TRBC1/CD3	234	0.71	**43.4**	**94.1**	83.7	70.4	**73.3**
TRBC1/susp cells	165	0.73	**64.0**	**75.6**	69.0	71.6	**70.5**



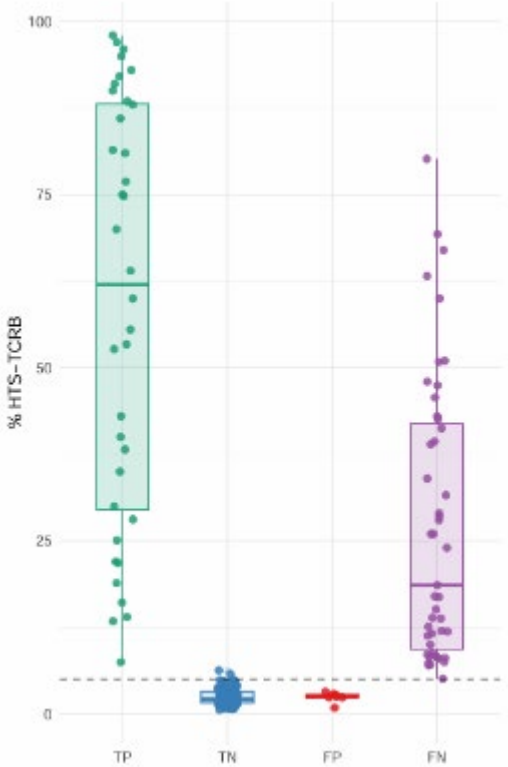


- **Symmetrical cut-off : <17.5% or >82.5%**

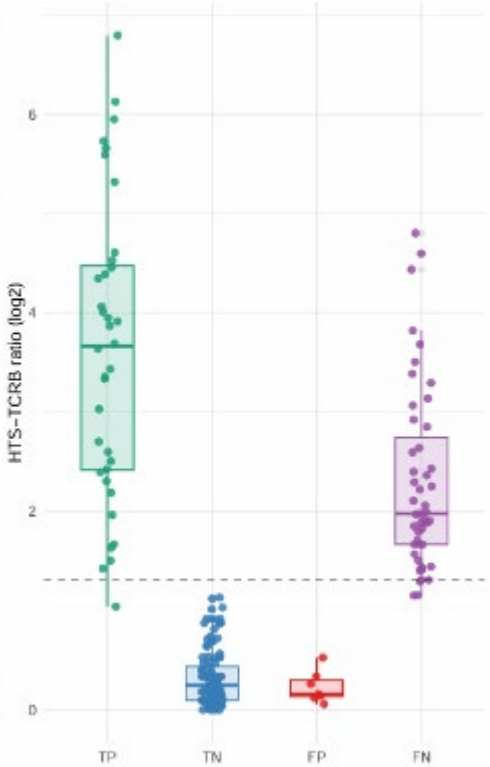
- TRBC1/CD3 : **high specificity for clone detection**
- Morphology-guided assessment favored **sensitivity**
- **Comparable overall accuracy** between both strategies.
- But sensitivity/specificity trade-offs

Analysis of Discordant Cases

HTS-TCRB Tumour Clone Frequency (%)



HTS-TCRB Ratio of Productive Frequencies

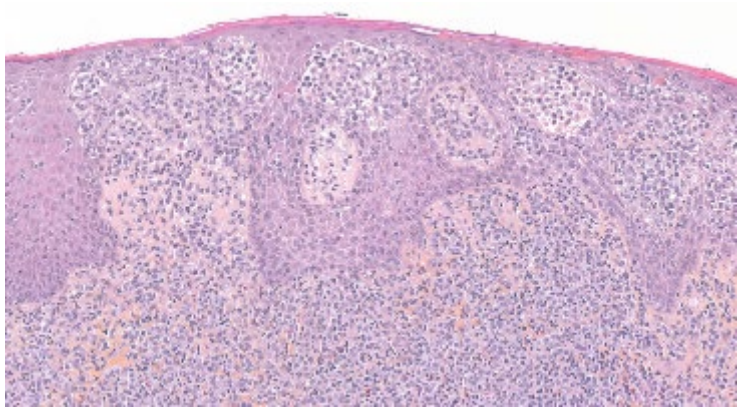


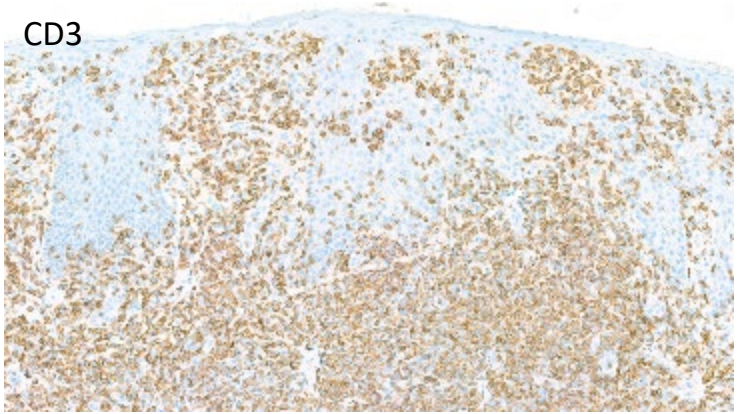
True Positives : 17.8%	True Negatives : 55.4%

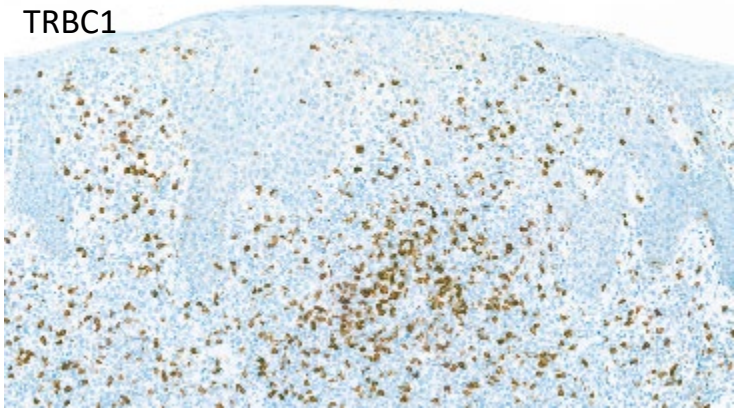
- False Negative (HTS-TCRB clonal not detected in IHC)
  - **23.3% of cases**
  - **Low-intermediate clonal burdens**
  - **Limited sensitivity of IHC for these clonal burdens**

- False Positive (HTS-TCRB non clonal with clonal IHC pattern)
  - **3.5% of cases,**
  - **Technical limits of HTS-TCRB (some cases were clonal with PCR TCRG)**
  - **MF with γδ-phenotype (without TRCB clone)**

« False-positive » TRBC1 pattern without TCRB clone detected, indicating MF-γδ

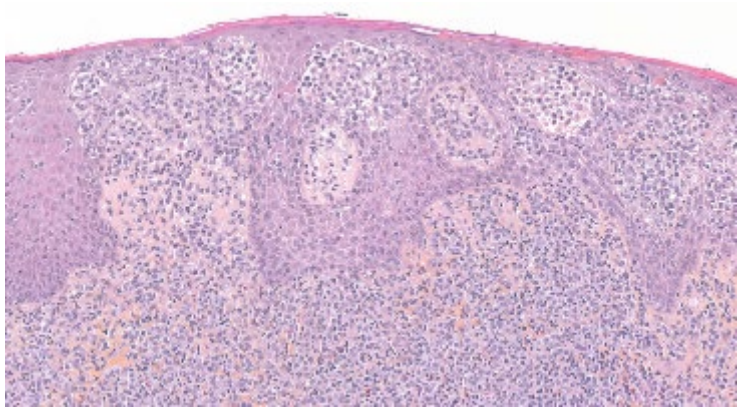


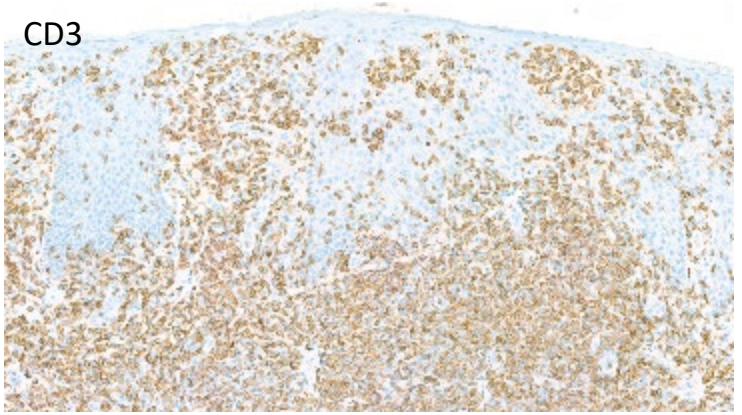


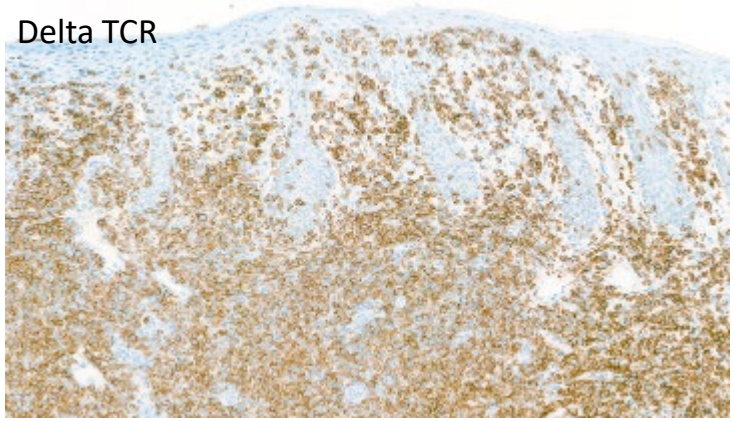


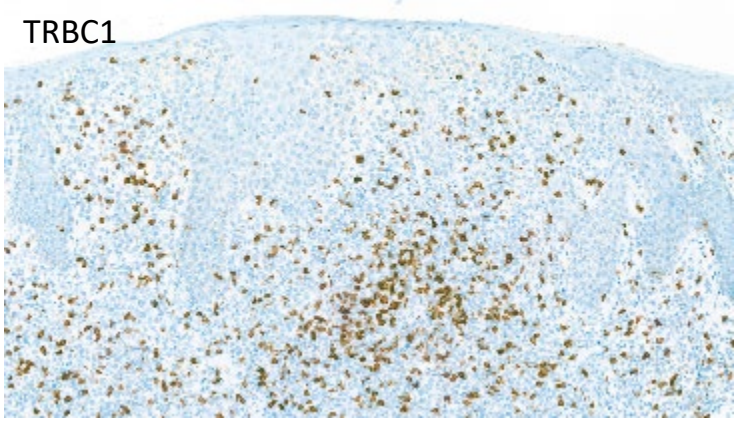
HTS-TCRB : no dominant clone

« False-positive » TRBC1 pattern without TCRB clone detected, indicating MF-γδ

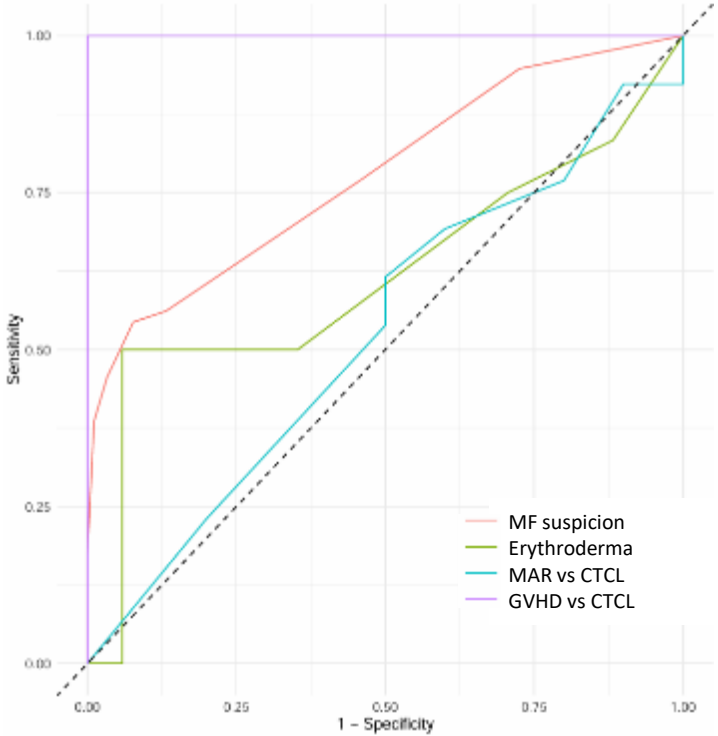








Performance by Clinical Indication - TRBC1/CD3



Indication	N	AUC	Se (%)	Spe (%)	Acc (%)
MF suspicion	**148**	0.78	54.4	92.3	77.7
Erythroderma	**29**	0.62	50.0	94.1	75.9
MAR vs CTCL	**23**	0.53	61.5	50.0	56.5
GVHD vs CTCL	**2**	1	100	100	100

- **Highest** diagnostic performance: **suspicion of MF at diagnosis**,
- **Intermediate** performance: **erythroderma**,
- **Lowest** performance observed in the **MAR vs CTCL** setting.

TRBC1 IHC in CTCL

TRBC1 IHC is a rapid, accessible, and reproducible tool for T-cell clonality assessment in CTCL

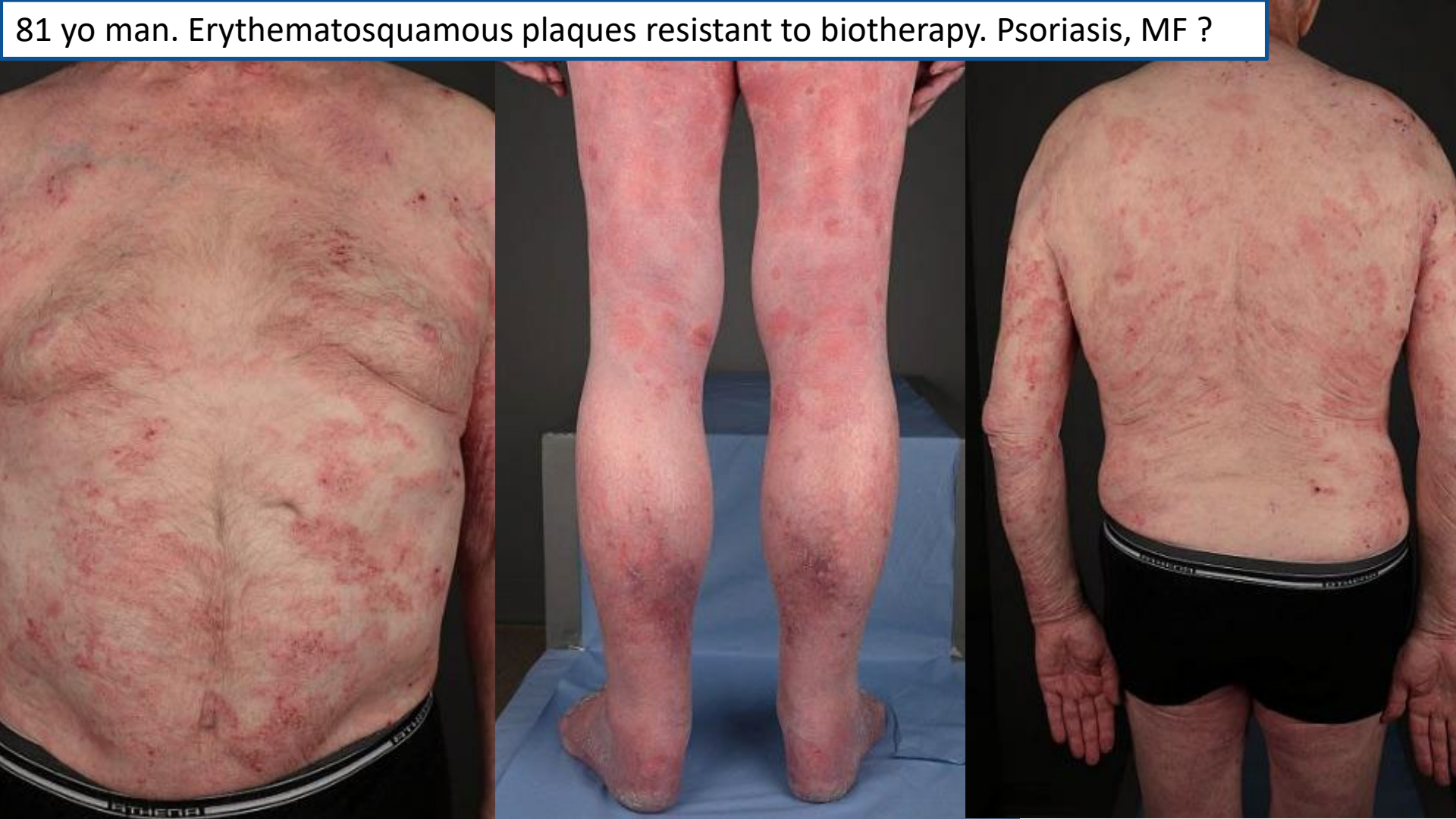
High specificity (94%) but moderate sensitivity (44%) for clonal detection using <17.5% and >82.5% thresholds: reliable rule-in test, suitable for routine diagnostic integration

Can be incorporated into routine workflows, especially when other clone detection techniques are unavailable or delayed

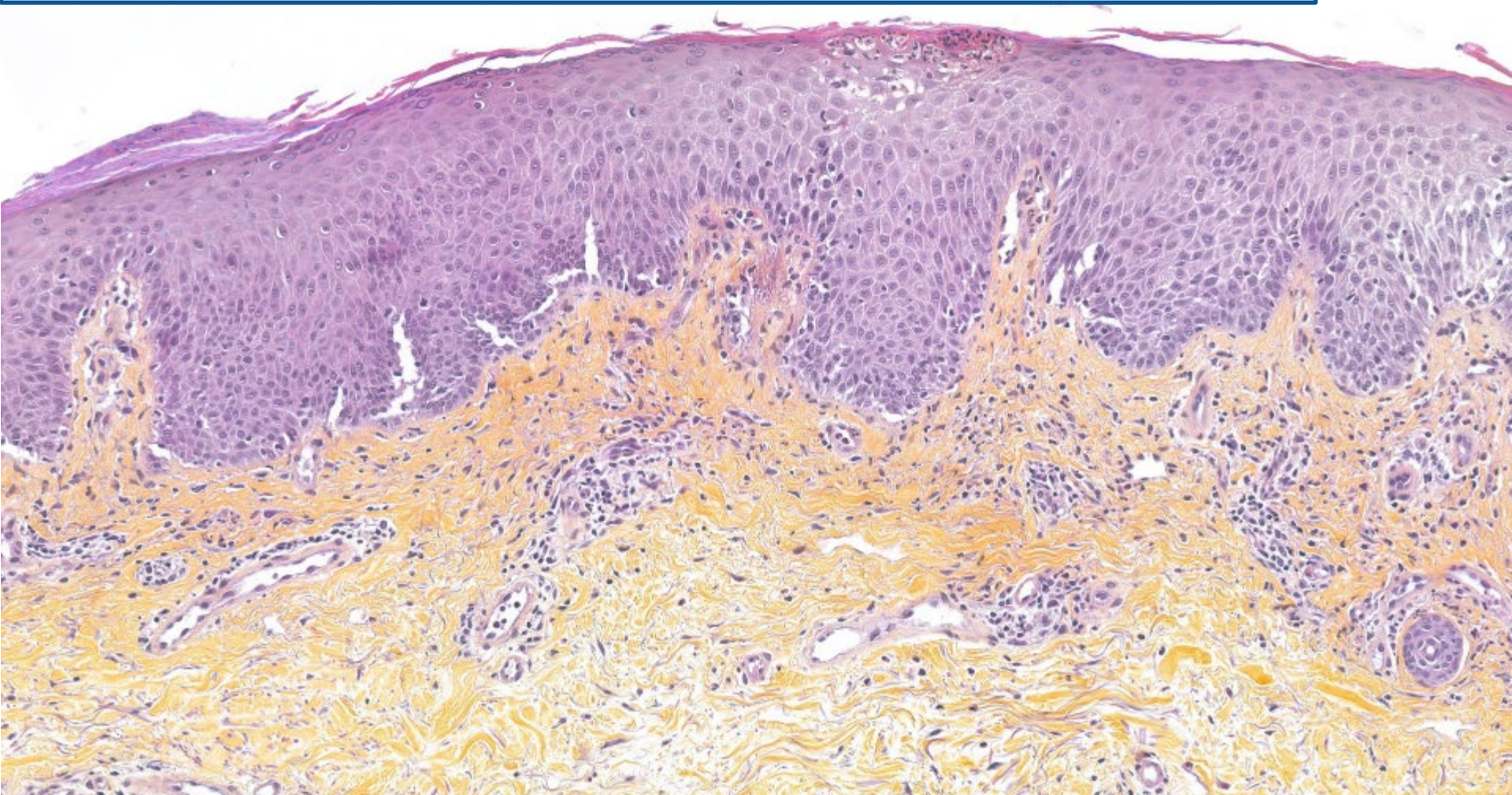
Key limits:

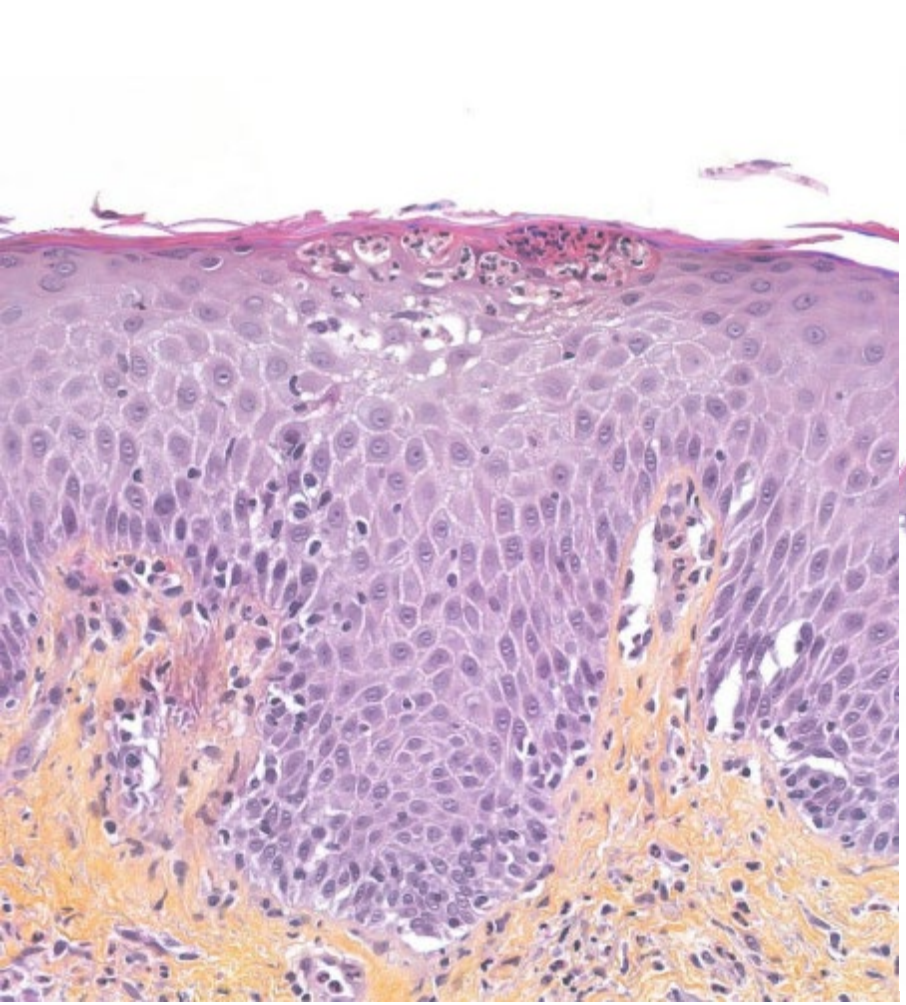
- Low-intermediate clonality burden & mogamulizumab rash: molecular techniques (HTS or PCR) remains standard in these settings
- TRBC1 IHC may help spot cases with TCRG or TCRD clonal rearrangement (“false-positive TRBC1 pattern”)

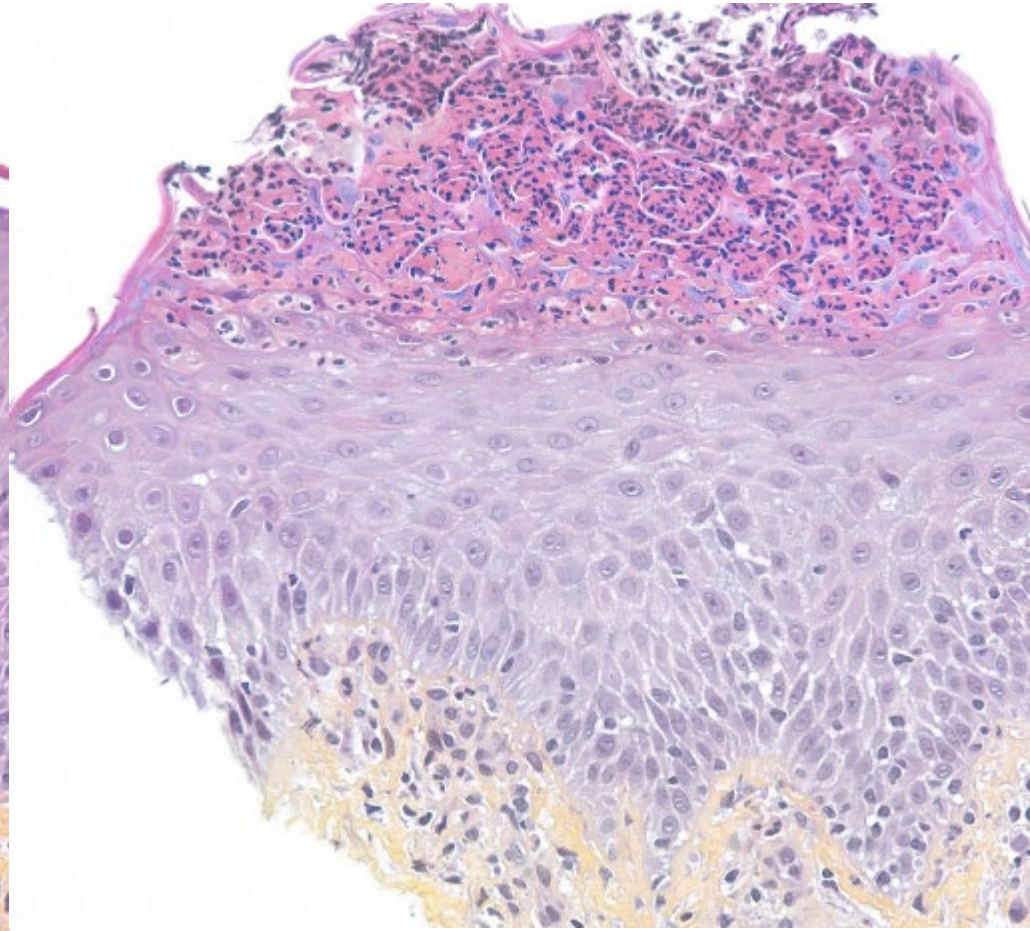
81 yo man. Erythematosquamous plaques resistant to biotherapy. Psoriasis, MF ?



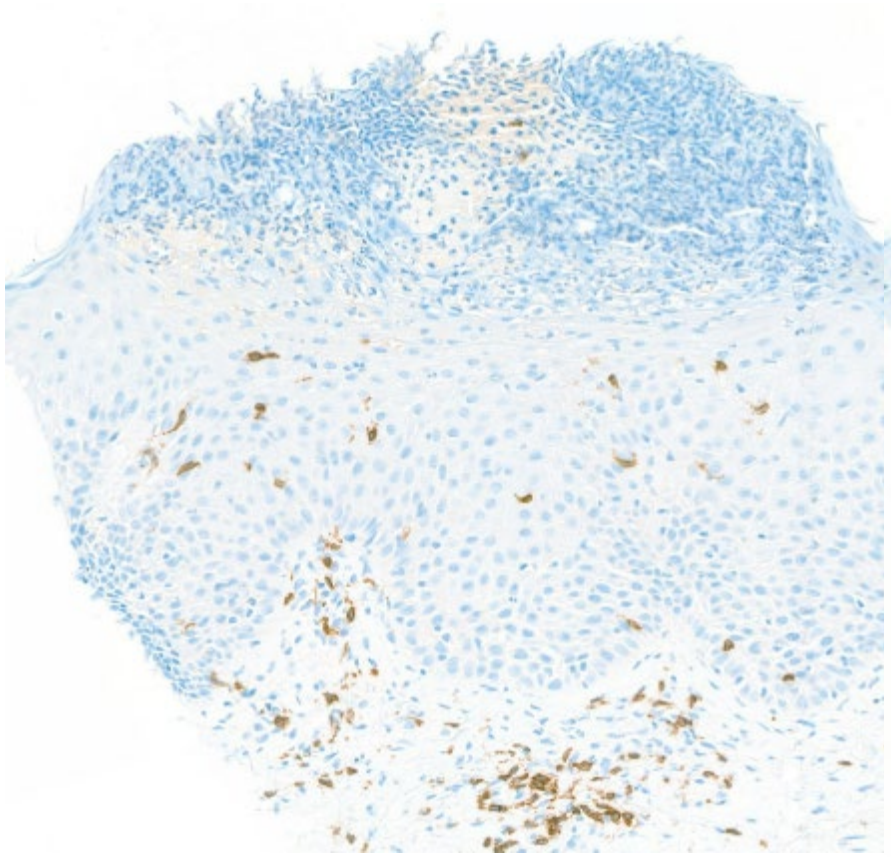
81 yo man. Erythematosquamous plaques resistant to biotherapy. Psoriasis, MF ?



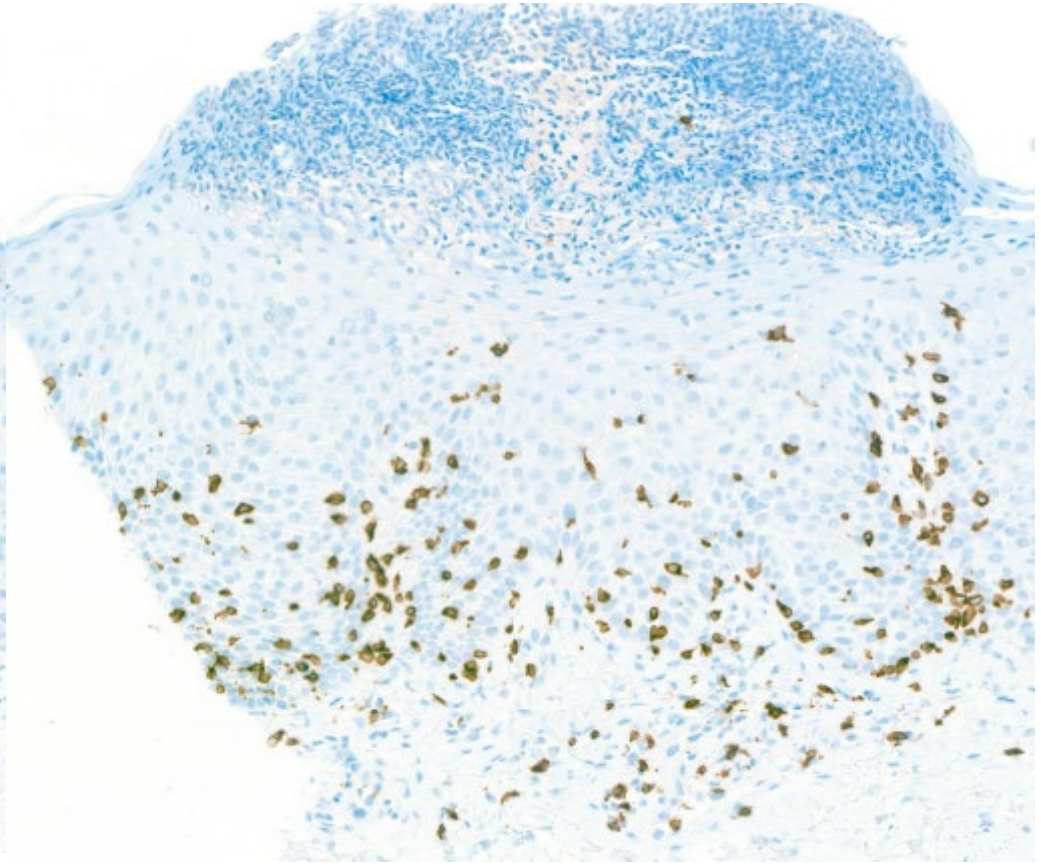




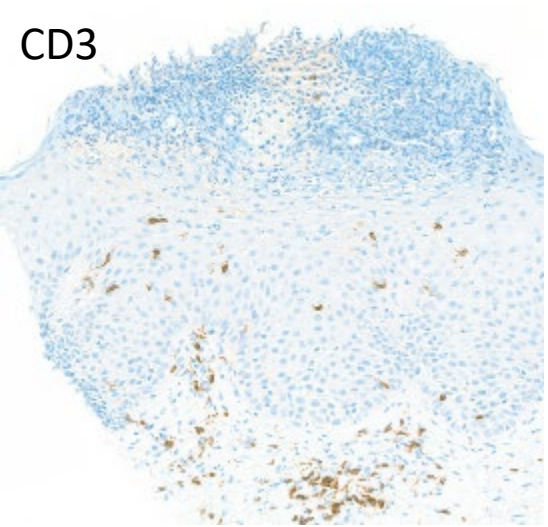
CD3



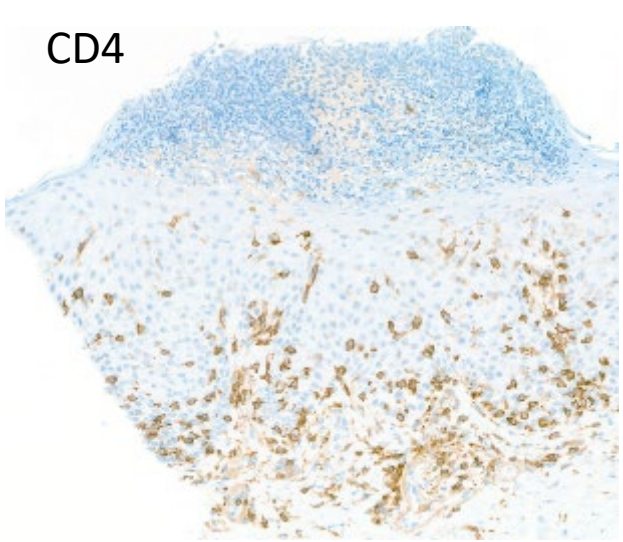
TRBC1



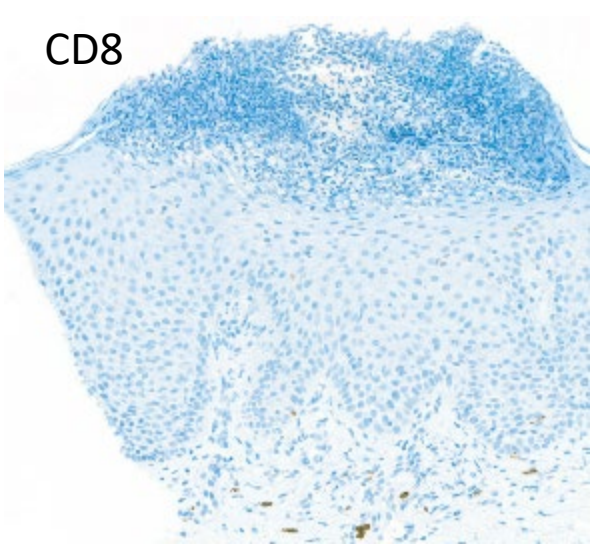
CD3

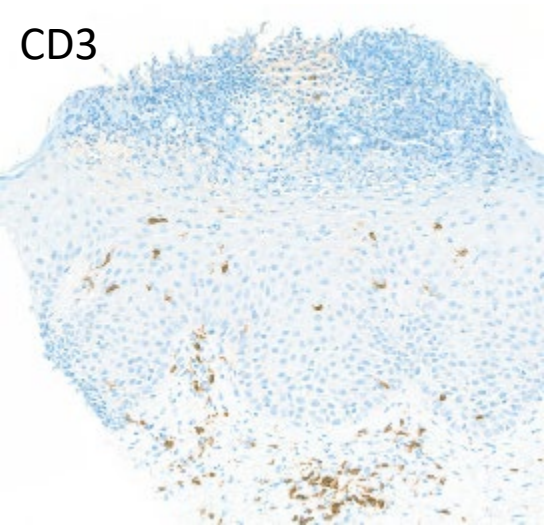


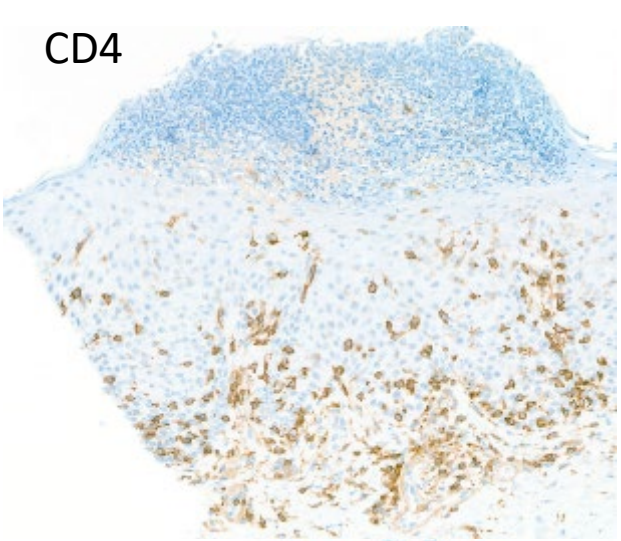
CD4

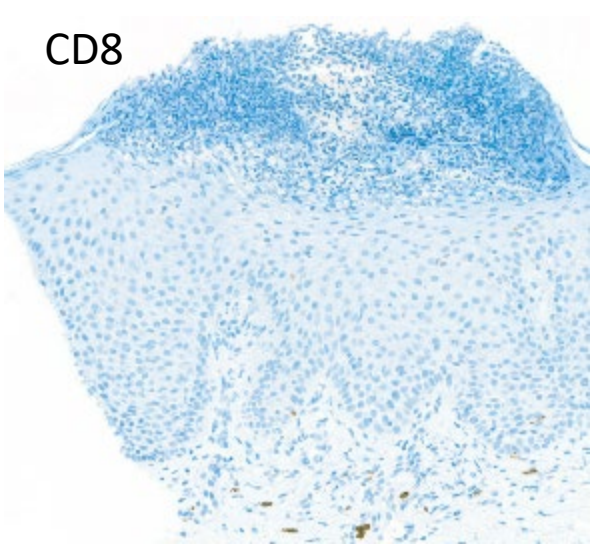


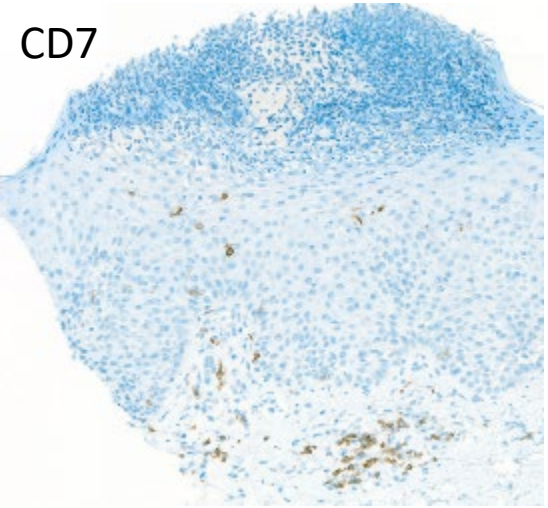
CD8

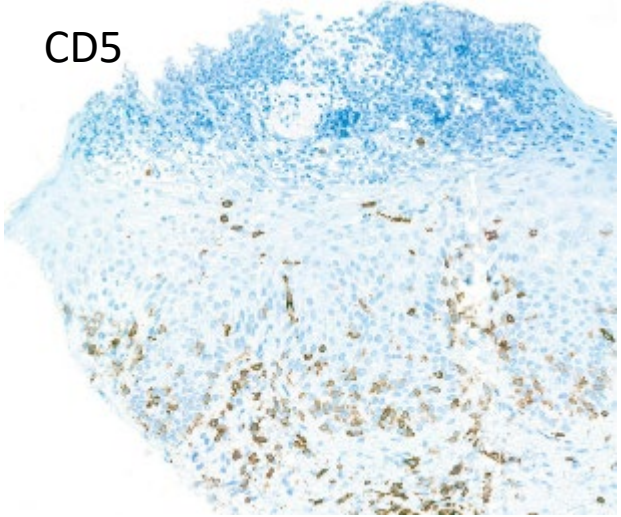


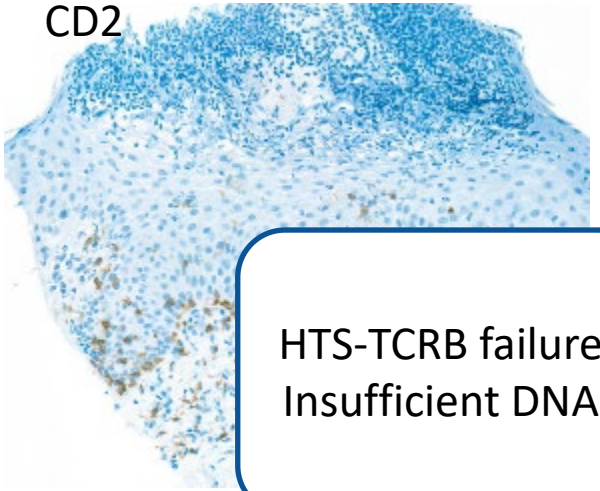






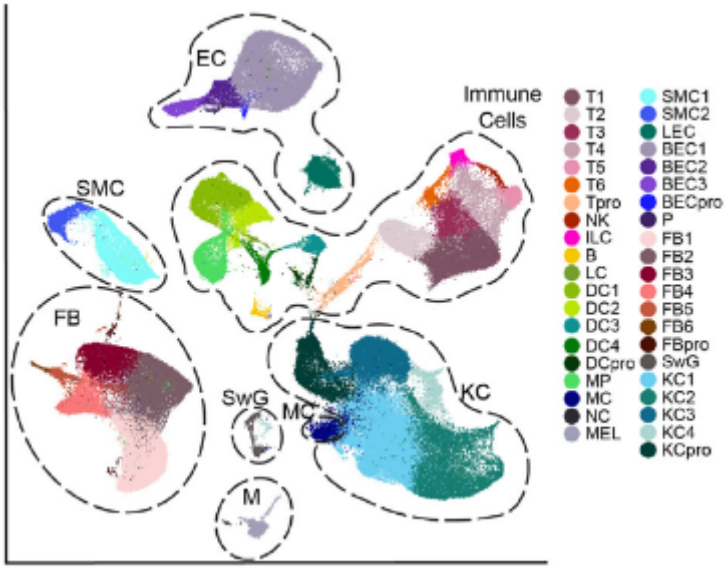


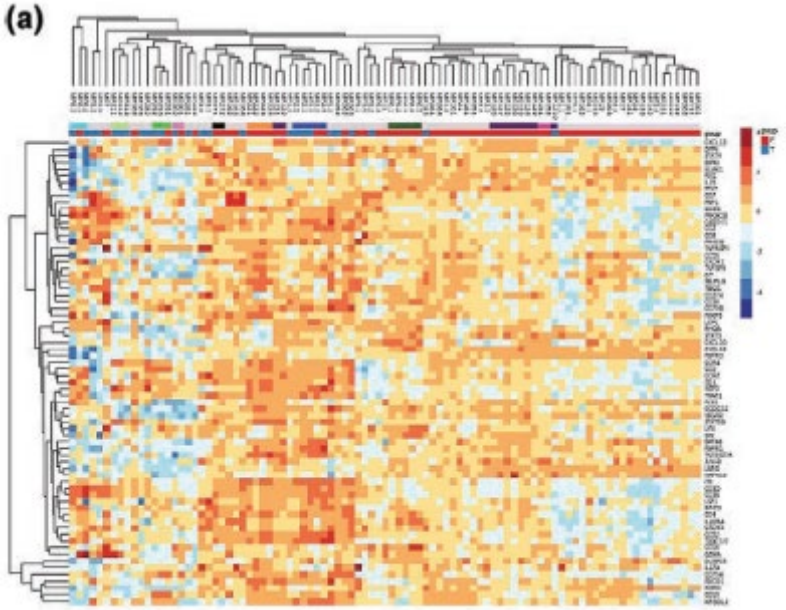




HTS-TCRB failure
Insufficient DNA

Advanced molecular methods: utopia or reality

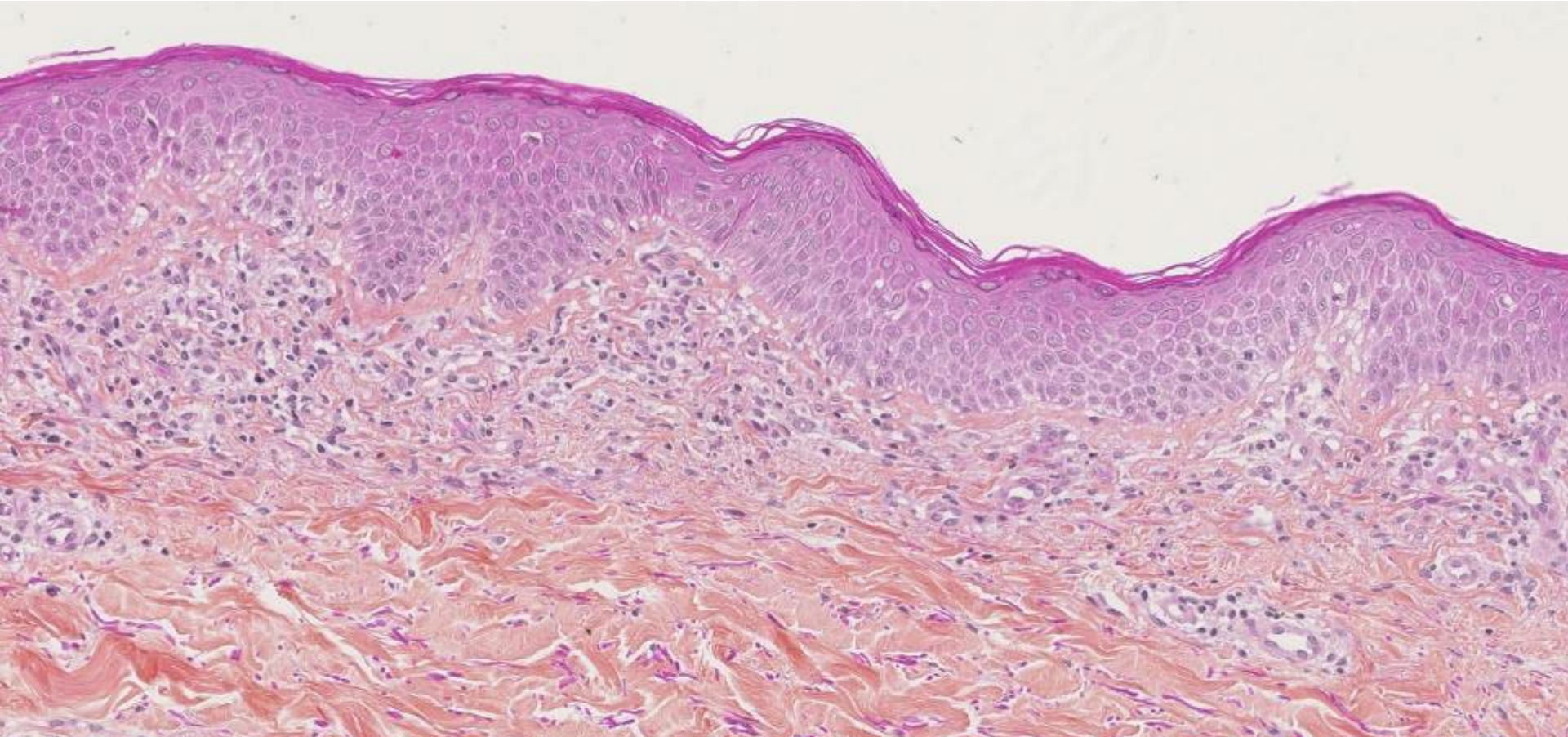


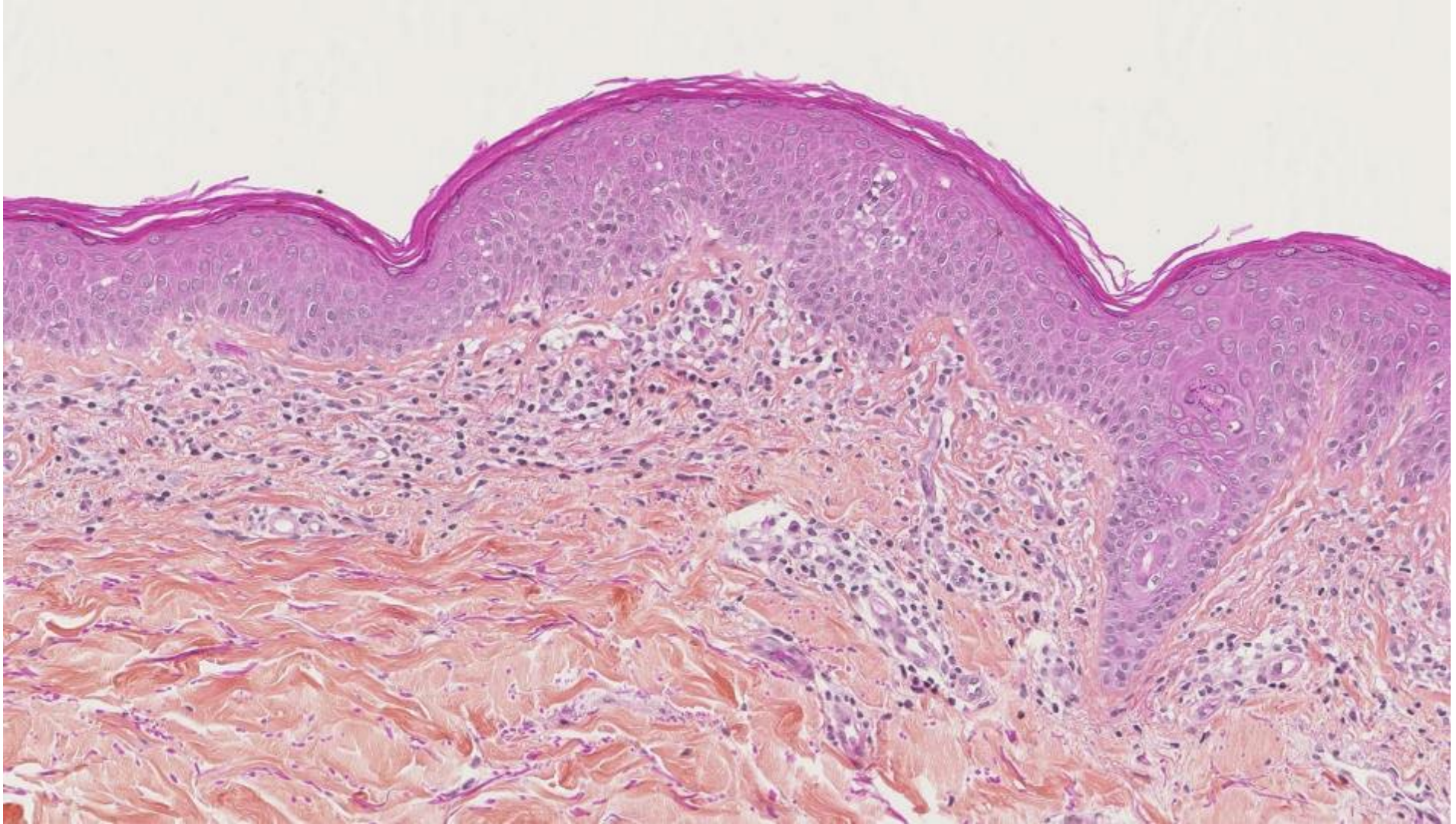


57 yo man

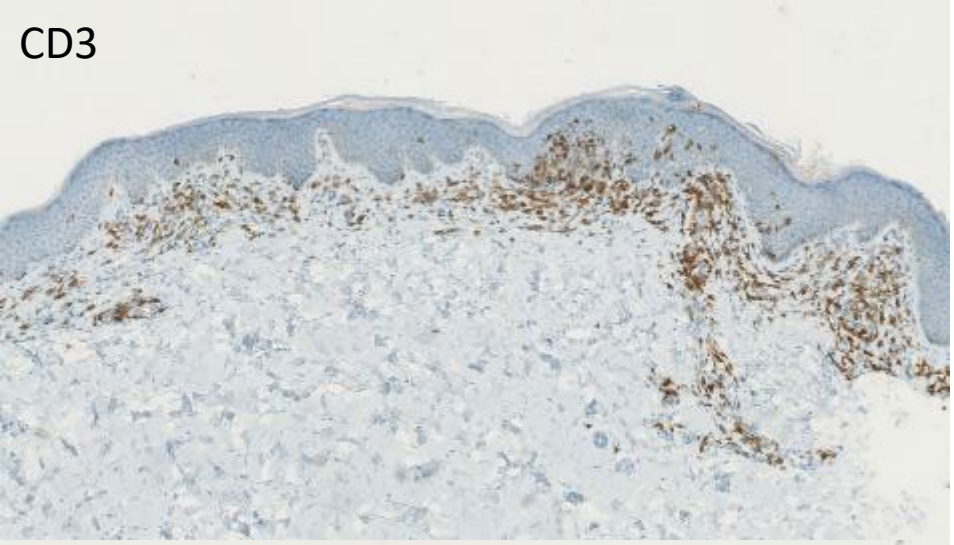
Since 4 years, extensive small erythematous patches and non-infiltrated plaques on the trunk and limbs.

Mycosis fungoides, Small plaque parapsoriasis (digitate dermatosis) ?

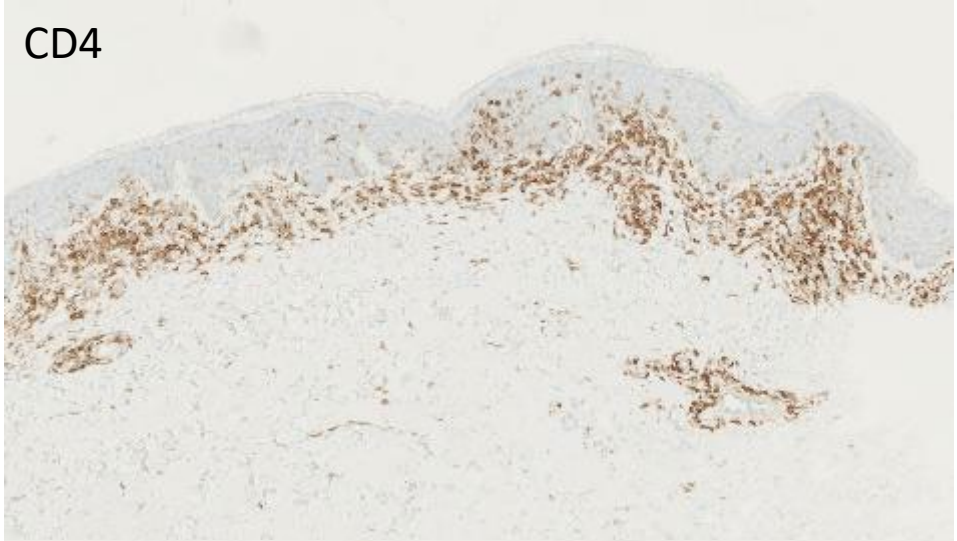




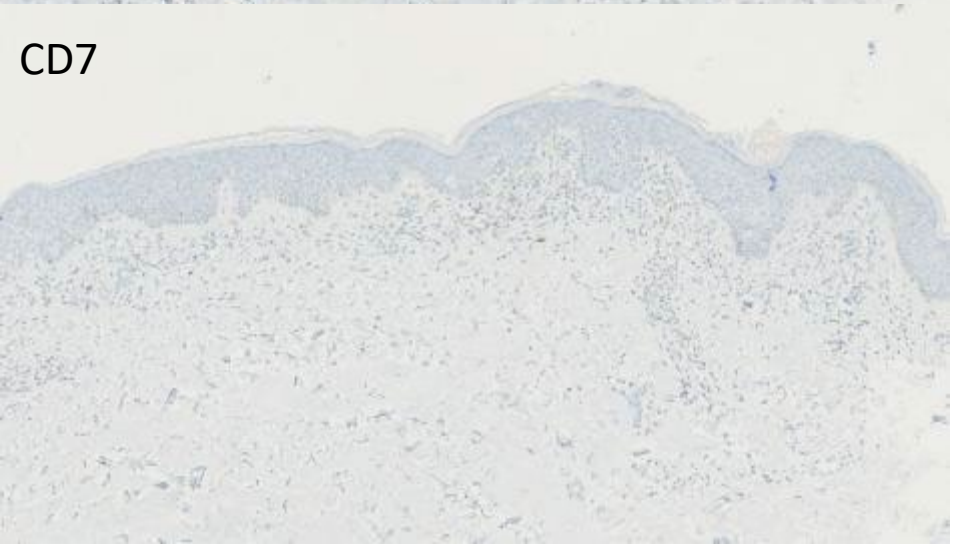
CD3



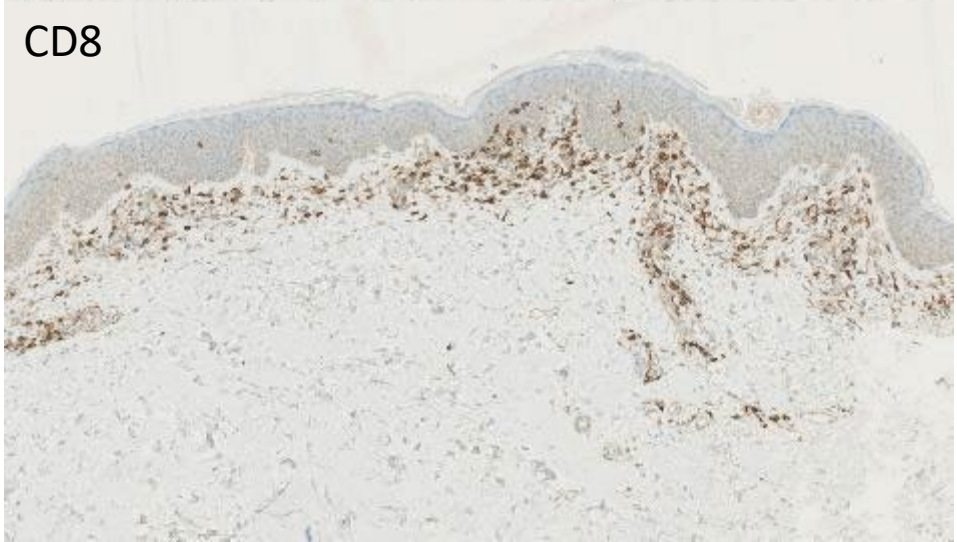
CD4



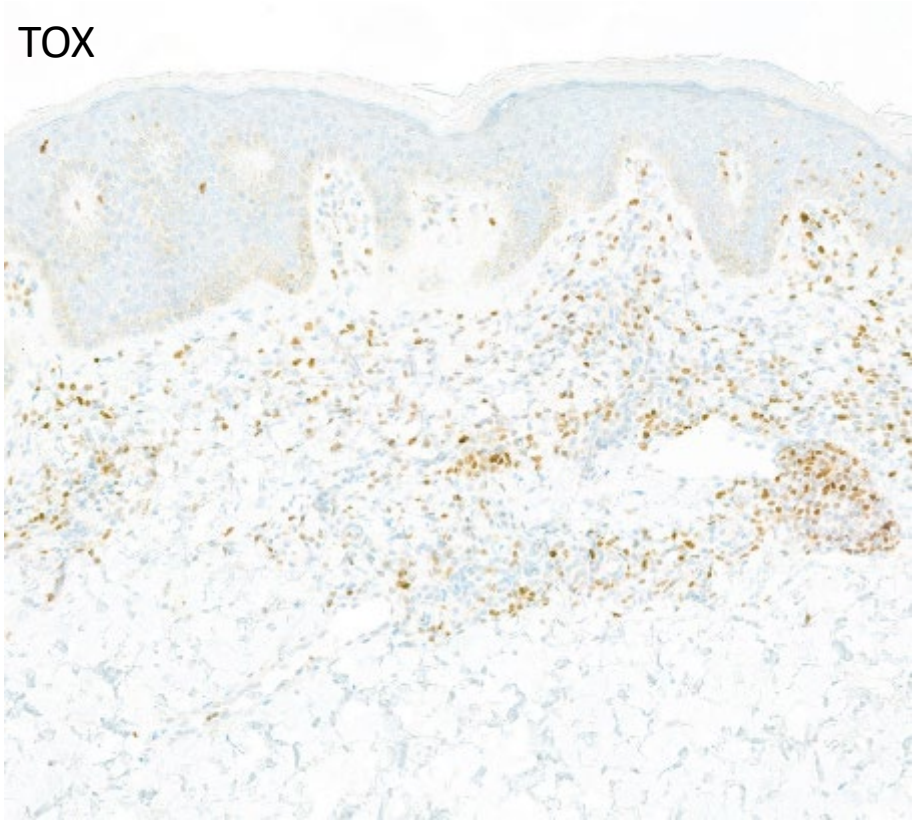
CD7



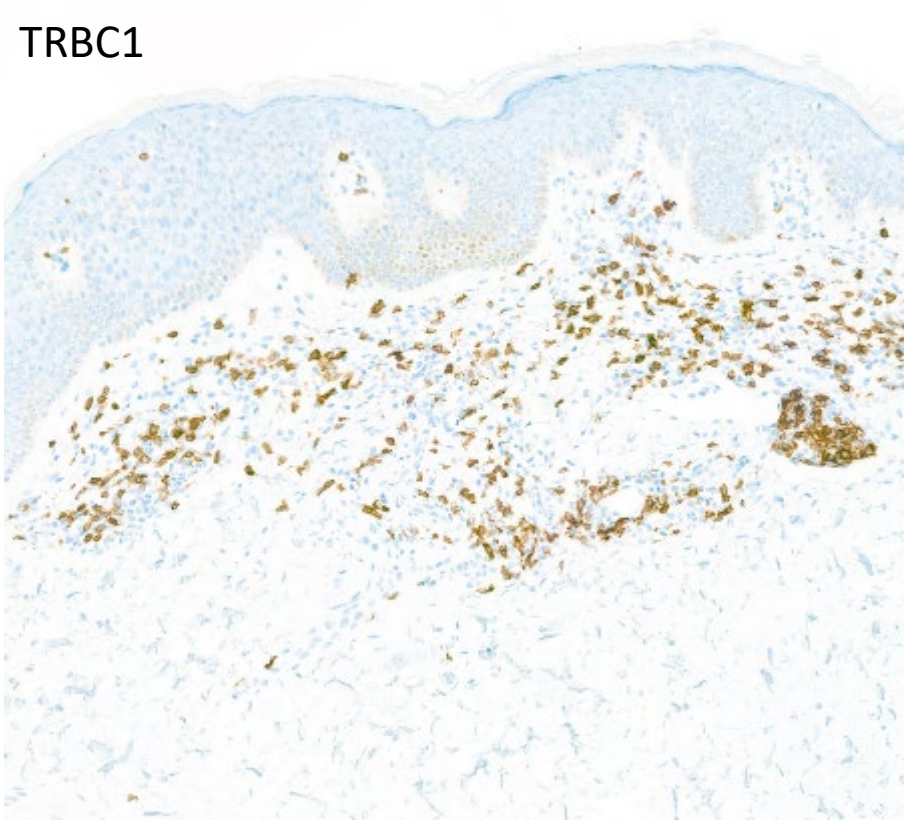
CD8

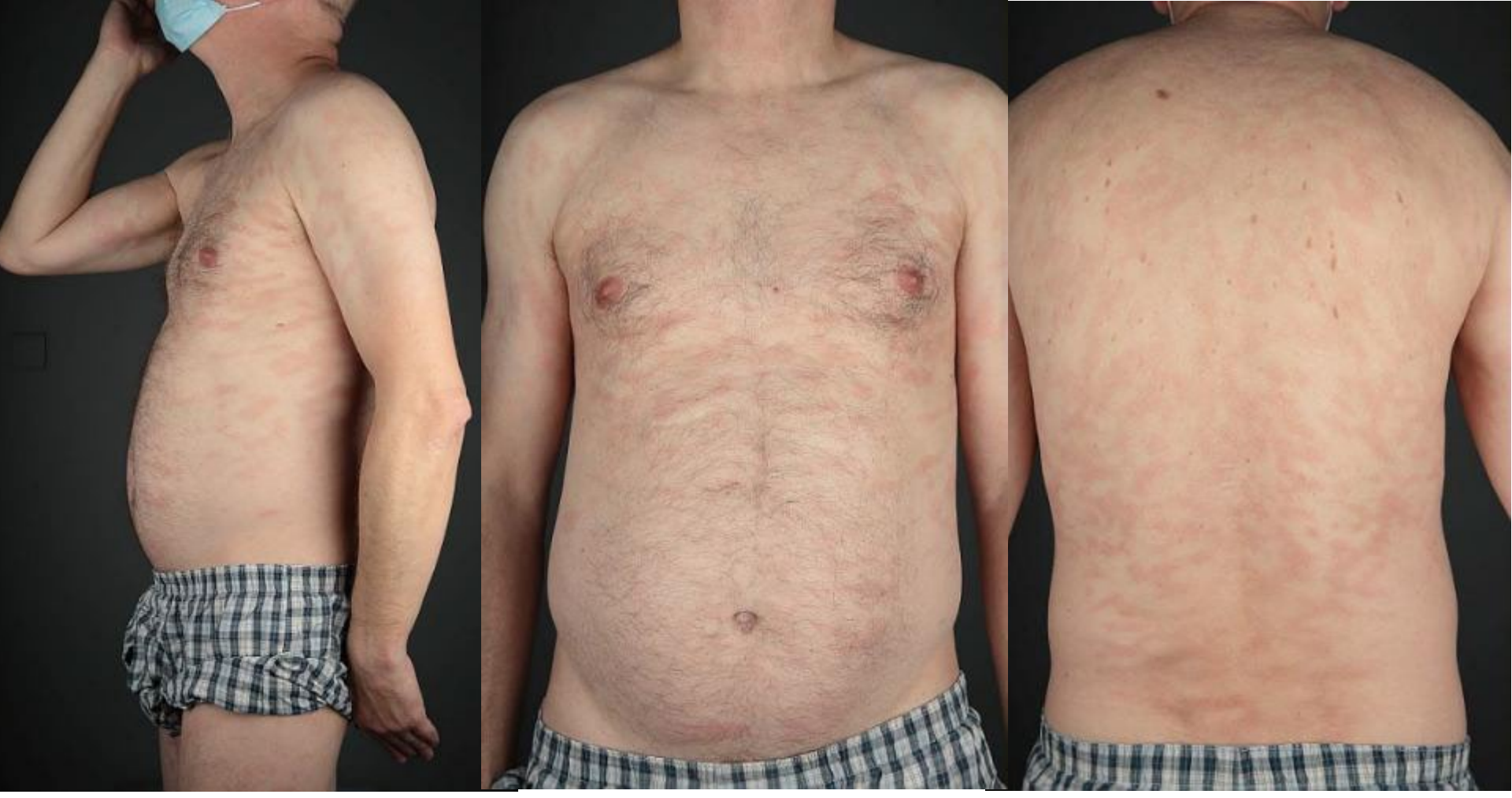


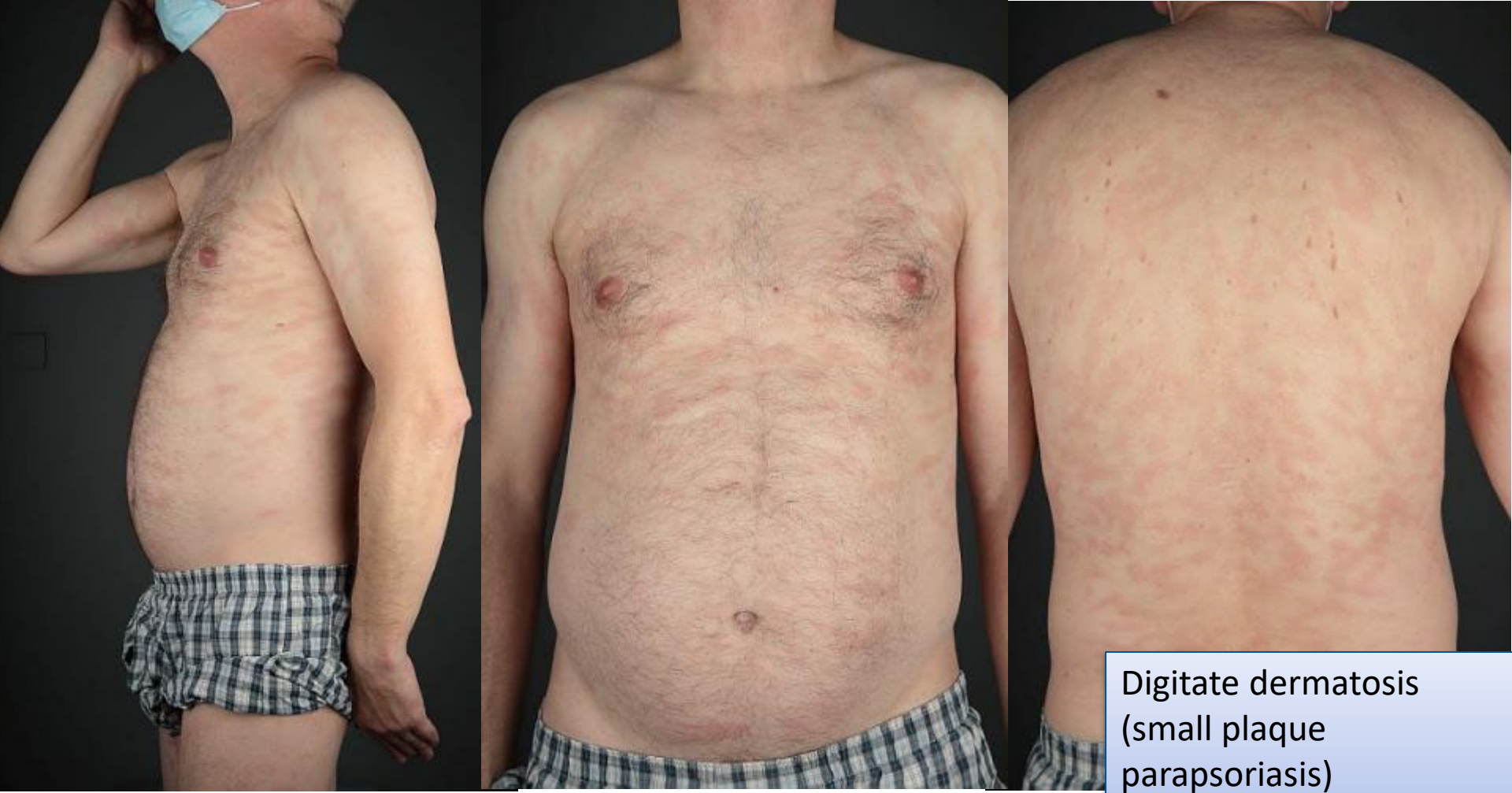
TOX



TRBC1







No dominant T-cell clone (HTS-TCRB)

Digitate dermatosis (small plaque parapsoriasis)
Steroids and follow-up

Single-Cell RNA-Seq exploration of the Parapsoriasis-to-MF Spectrum

Alkon, Chennareddy, et al. J Allergy Clin Immunol 2025;155:461-78 — 15 patients (SPD n=7, LPD n=8) + AD (n=5) + healthy skin (n=4) — scRNA-seq + TCR-seq, 184,133 cells / 41 clusters

Clinical lesion

Small-plaque (<5 cm, "fingerprint dermatitis") or large-plaque (>5 cm) chronic dermatosis

→

scRNA-seq + TCR-seq

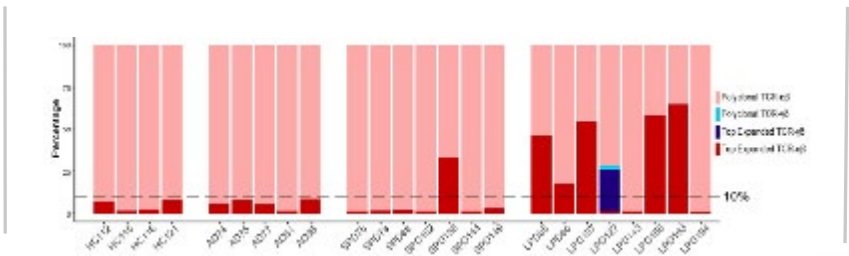
Skin biopsy dissociated to single cells; paired transcriptome + TCR clonotype per cell

→

TCR clonality gate

Is the single most abundant clone ≥10% of all TCR+ cells in the sample?

polyclonal (<10%) *monoclonal (≥10%)*



"Polyclonal Parapsoriasis"

benign, non-lymphomatous

- 6/7 SPP + 2/8 LPP samples
- CD7 / CD27 preserved
- NPY+ ILC1-like population (unique)
- Low cytokine / "quiet" microenvironment

Early-Stage Mycosis Fungoides

clonal, lymphomatous

- 6/8 LPP + 1/7 SPP samples
- CD7↓ / CD27↓ in the expanded clone
- Type-1 skewing: IFNG, GNLY, GZMB, IL26

Take-home: TCR clonality — not plaque size — separates true parapsoriasis from early MF.

Polyclonal Parapsoriasis Has Its Own Molecular Signature

Cross-referencing skin biomarkers separates polyclonal parapsoriasis (PP) from its principal clinical mimickers

Atopic Dermatitis

chief clinical mimicker

T cells

CD3+CRTH2+IL17RB+ “TH2A” cells — absent in PP, esMF, HC

Cytokines

Strong type-2 skewing: IL13, CCL17/18/22, TSLPR

Stroma

FB3 upregulates COL6A5/6, POSTN, TNC (type-2 fibroblast)

Polyclonal Parapsoriasis

proposed distinct entity

ILCs

Expanded NPY+ / CRISP3+ ILC1-like population — not described elsewhere

Cytokines

Low overall inflammatory milieu, close to healthy skin

Stroma

Fibroblasts/sweat glands: complement (C6, C7, CFD) + antimicrobial genes (DCD, PIP)

Early-Stage MF

the diagnosis to exclude

Clonal T cells

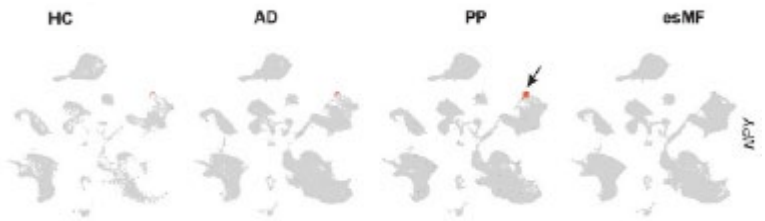
Single TCR clone ≥10%; CD7↓, CD27↓ vs bystander cells

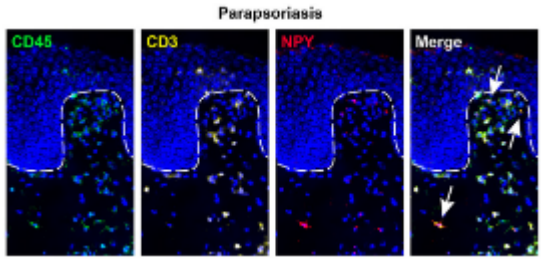
Cytokines

Type-1 skewing: IFNG, GNLY, GZMB, plus IL26 in the clone

Myeloid/KC

Strongest S100A7/8/9, FABP5 keratinocyte activation





Exploring GEP/transcriptomic as a diagnostic tool

Key studies, bulk microarray/panel era through single-cell and spatial transcriptomics

Study	Platform / method	Main result
Tracey et al., Blood 2003	cDNA microarray (29 MF vs 11 BID)	6-gene subset distinguishes MF from inflammatory dermatoses (97% accuracy)
Nebozhyn et al., Blood 2006	Oligonucleotide microarray	5-gene panel (STAT4, GATA3, PLS3, CD1D, TRAIL) distinguishes MF from benign dermatoses
Ralfkiaer et al., Blood 2011	miRNA microarray (n=198)	5-miRNA classifier separates CTCL from psoriasis/dermatitis (>90% accuracy)
Kim et al., J Invest Dermatol	Whole-genome microarray vs normal skin + BID	349 DEGs in early MF; TH1/TH2, Lck/Fyn, antigen-presentation, apoptosis pathways enriched
Benoit et al., miRNA/FOXP3 study	miRNA array + RT-PCR	miR-26a/-222/-181a/-146a differentiate early MF from inflammatory dermatoses via FOXP3
Litvinov et al., 2015 / 2017	Targeted RNA panel (RT-PCR / TruSeq)	17-gene signature (IL2RA, CCR4, STAT5A, TOX) distinguishes and risk-stratifies CTCL vs benign skin
Alonso-Alonso et al., Br J Dermatol 2023	nCounter/NanoString panel (70 genes), 109 FFPE samples (31 MF, 93 serial + 16 inflammatory dermatitis)	Fig. S1 volcano (inflammatory vs. MF): CCDC32, LCP1, CD3D/E/G, CD52, CXCR3, CARD11, CXCL13, CD79A/B up in MF; FGFR3, MAPK13, LARG up in inflammatory; pattern confirmed for inflammatory vs. early MF (Fig. S2)
Alkon et al., Mol Cancer 2021	scRNA-seq	Defines markers of CTCL disease progression
Gaydosik et al. 2019 + reanalyses	scRNA-seq (public datasets)	Clonal/transcriptional CTCL landscape; stromal TME distinct from AD and healthy skin
Li et al., 2024	scRNA-seq + TCR-seq (45 pts, largest CTCL atlas)	B cells enriched in CTCL; malignant T cells express CXCL13, absent in AD/psoriasis/HC
Bangert, Brunner et al., JACI 2024	scRNA-seq + TCR-seq	Erythrodermic CTCL: CD4+CCR7+SELL+ clonal expansion vs CD8A+KLRK1+ in chronic erythroderma
Alkon, Brunner et al., JACI 2025	scRNA-seq + TCR-seq	Polyclonal parapsoriasis marked by NPY+ ILC1-like cells; distinct from esMF and AD
CosMx spatial study, J Cutan Pathol 2024	Spatial single-cell transcriptomics (CosMx)	GNLY and FYB1 distinguish early MF from psoriasis/spongiotic dermatitis (IHC-validated)
LCK/HOMER1 classifier, medRxiv 2025	Gene-expression classifier	LCK/HOMER1 classifier distinguishes early MF from eczema and psoriasis

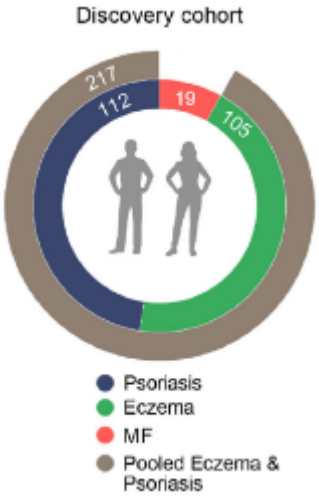
across two decades and every platform (microarray, targeted panels, miRNA, scRNA-seq, spatial transcriptomics), the same axis keeps separating MF from its mimickers — cytotoxic/TCR-signaling genes (GNLY, GZMB, TOX, IL26, CCR4/STAT) mark the malignant clone, while benign mimickers are defined by their polarized cytokine milieu (type 2 in AD, type 17 in psoriasis) rather than a lymphoma signature.

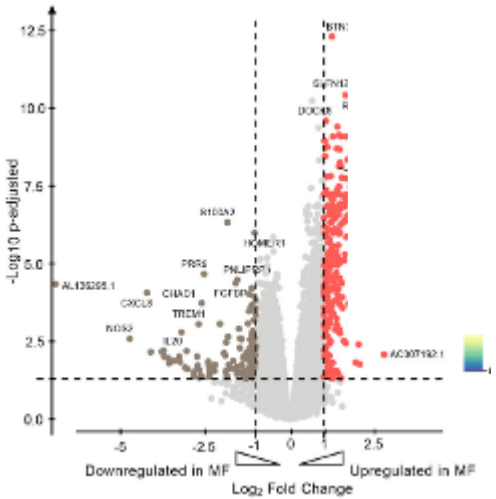
LCK and HOMER1 gene expression-based classifier distinguishes early mycosis fungoides from eczema and psoriasis

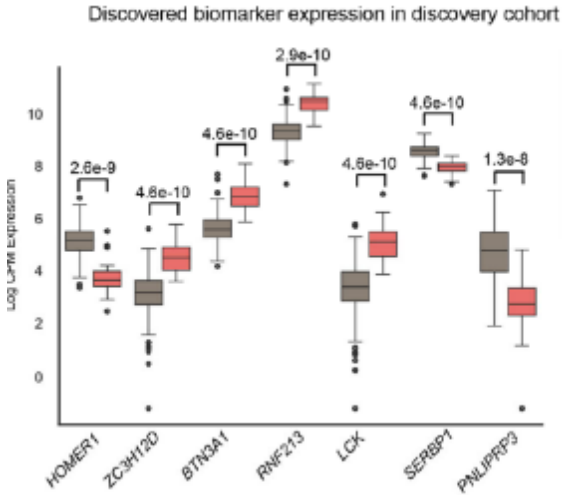
Posted October 23, 2025.

Martin Meinel, Nora Langreder, Andrea Schmitt, Kristin Technau-Hafsi, Christina Hillig, Sophie Roenneberg, Adriana Bernklauová, Jan Říčař, Denisa Kacerovská, Marion Wobser, Caroline Mann, Beate Weidenthaler-Barth, Yalda Ghoreishi, Reem El Bahtimi, Leonidas Marinos, Evangelia Papadavid, Marie Charlotte Schuppe, Christina Mitteldorf, Werner Kempf, Emmanuella Guenova, Kilian Eyerich, Michael P. Menden, Stefanie Eyerich, Natalie Garzorz-Stark

doi: https://doi.org/10.1101/2025.10.18.25338021





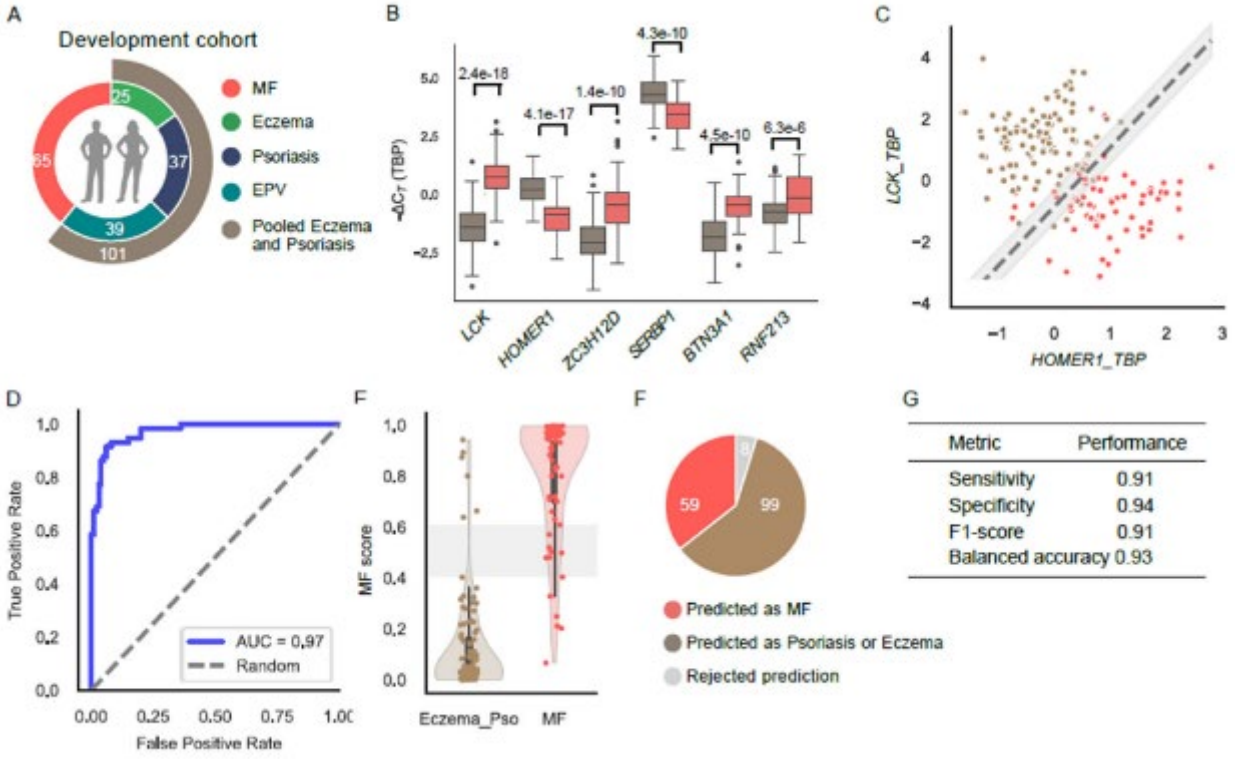


LCK and HOMER1 gene expression-based classifier distinguishes early mycosis fungoides from eczema and psoriasis

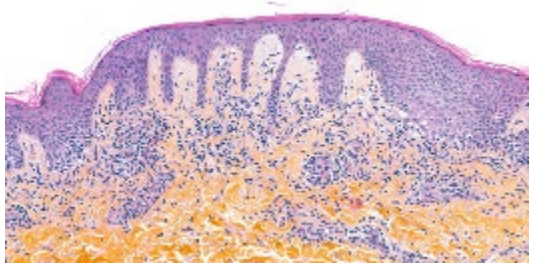
Posted October 23, 2025.

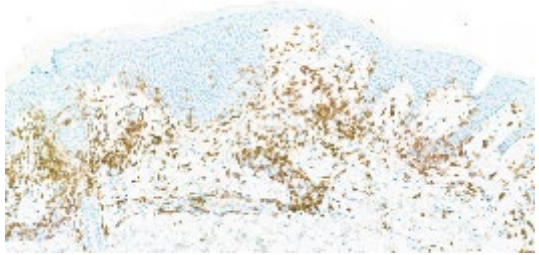
Martin Meinel, Nora Langreder, Andrea Schmitt, Kristin Technau-Hafsi, Christina Hillig, Sophie Roenneberg, Adriana Bernklauová, Jan Říčař, Denisa Kacerovská, Marion Wobser, Caroline Mann, Beate Weidenthaler-Barth, Yalda Ghoreishi, Reem El Bahtimi, Leonidas Marinos, Evangelia Papadavid, Marie Charlotte Schuppe, Christina Mitteldorf, Werner Kempf, Emmanuella Guenova, Kilian Eyerich, Michael P. Menden, Stefanie Eyerich, Natalie Garzorz-Stark

doi: https://doi.org/10.1101/2025.10.18.25338021



20yo. Small plaques. MF or small plaque PP ?



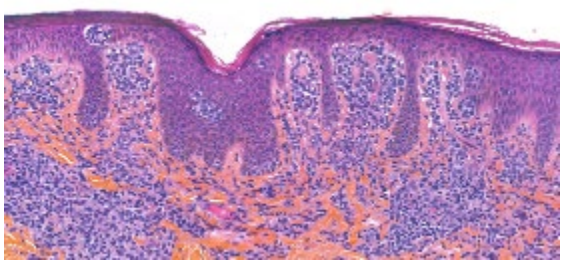


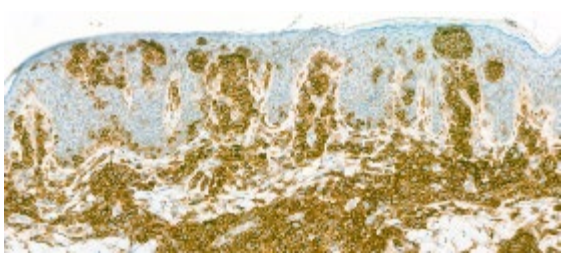
CD3+ CD7+ CD4=CD8
TOX 40% TRBC1 60%

HTS-TCRB
No dominant clone

LCK/HOMER1 expression test
MF score 97%

55yo. Erythroderma and lymphadenopathies. SS ?



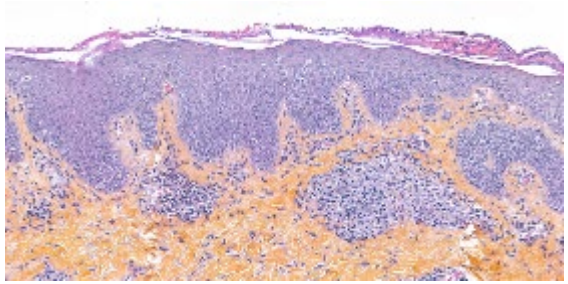


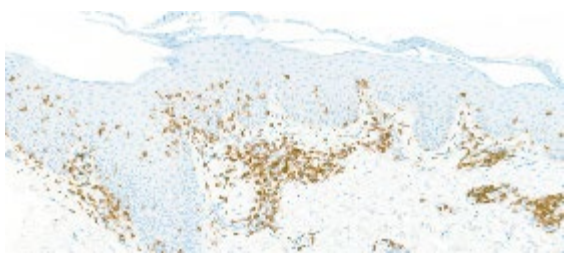
CD3+ CD4+ CD7- CD8-
PD1 100% TOX 100% TRBC1 100%

HTS-TCRB
Dominant clone 93% ratio 97

LCK/HOMER1 expression test
MF score 97%

35yo. AD worsening under dupilumab treatment. MF ?



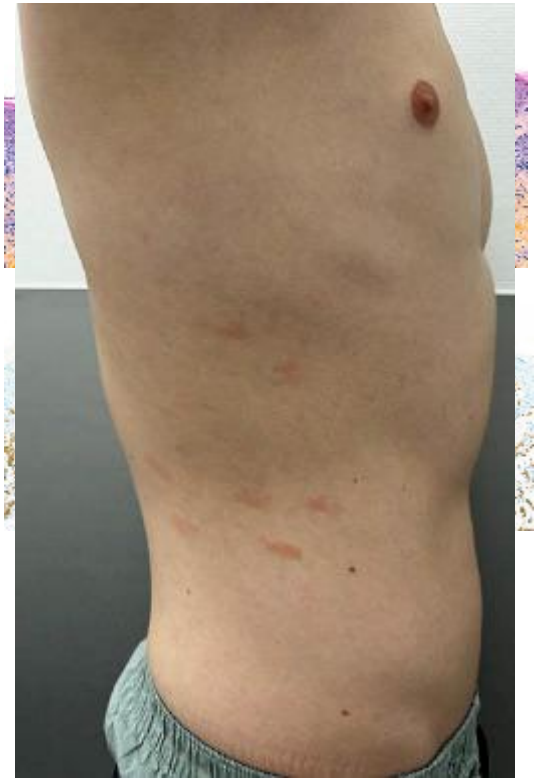


CD3+ CD7+ CD4>CD8
TRBC1 20-30%

HTS-TCRB
2 dominant clones 6.2% and 5.7% ratio 3.7

LCK/HOMER1 expression test
MF score 18%

20yo. Small plaques. MF or small plaque PP ?

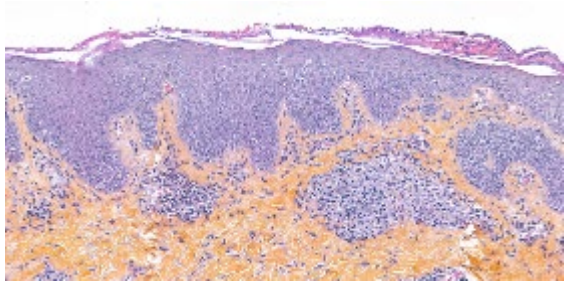


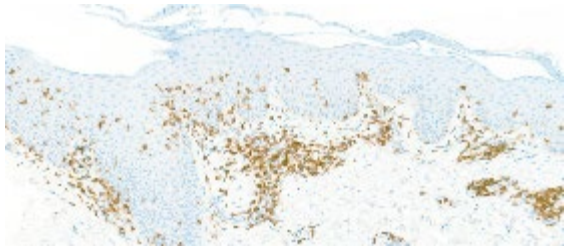
LCK/HOMER1 expression test
MF score 97%

55yo. Erythroderma and lymphadenopathies. SS ?

LCK/HOMER1 expression test
MF score 97%

35yo. AD worsening under dupilumab treatment. MF ?





CD3+ CD7+ CD4>CD8
TRBC1 20-30%

HTS-TCRB
2 dominant clones 6.2% and 5.7% ratio 3.7

LCK/HOMER1 expression test
MF score 18%

AI in the game for MF/SS patients

- **Earlier and more confident diagnosis**
  - Pattern recognition based on patient file
  - AI-informed clinical diagnosis (image analysis; skin in vivo imaging)
  - **AI-informed pathological diagnosis**

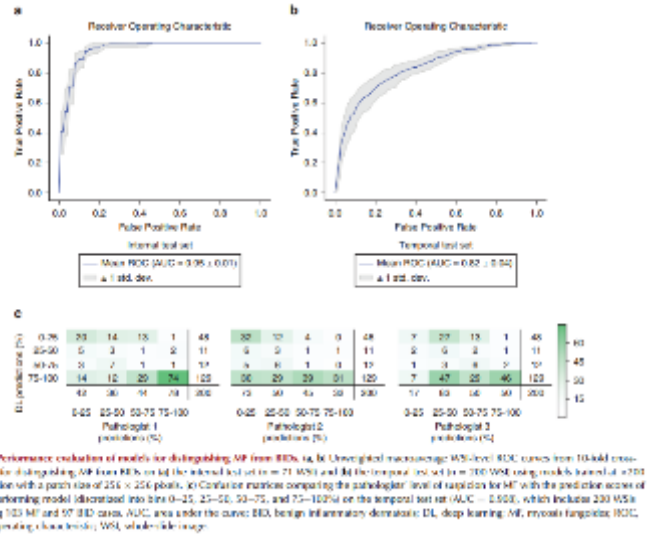
- **Staging**
  - Automated/reproducible skin burden quantification
  - AI radiological nodal staging
  - AI recognition and quantification of abnormal circulating cells

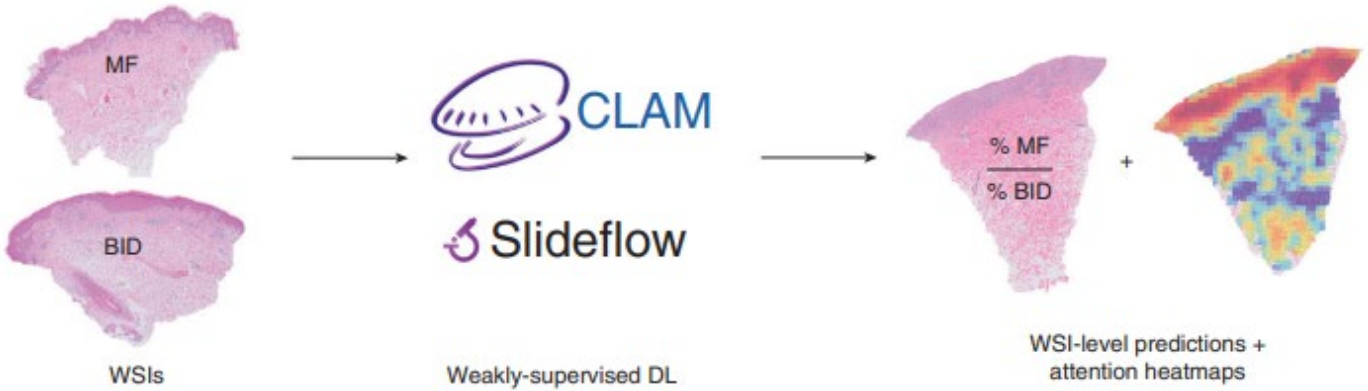
- **Prognosis**
  - AI prognostic model based on histopathologic features
  - AI multimodal prognostic model integrating clinic/biologic/histopathologic/molecular features

Deep Learning–Based Classification of Early-Stage Mycosis Fungoides and Benign Inflammatory Dermatoses on H&E-Stained Whole-Slide Images: A Retrospective, Proof-of-Concept Study

Thom Doeleman[1,2], Siemen Brussee[1], Liesbeth M. Hondelink[1], Daniëlle W.F. Westerbeek[1], Ana M. Sequeira[1], Pieter A. Valkema[1,3], Patty M. Jansen[1], Junling He[1], Maarten H. Vermeer[4], Koen D. Quint[4], Mariike R. van Diik[2], Fons J. Verbeek[5], Jesper Kers[1,3] and Anne M.R. Schrader[1]

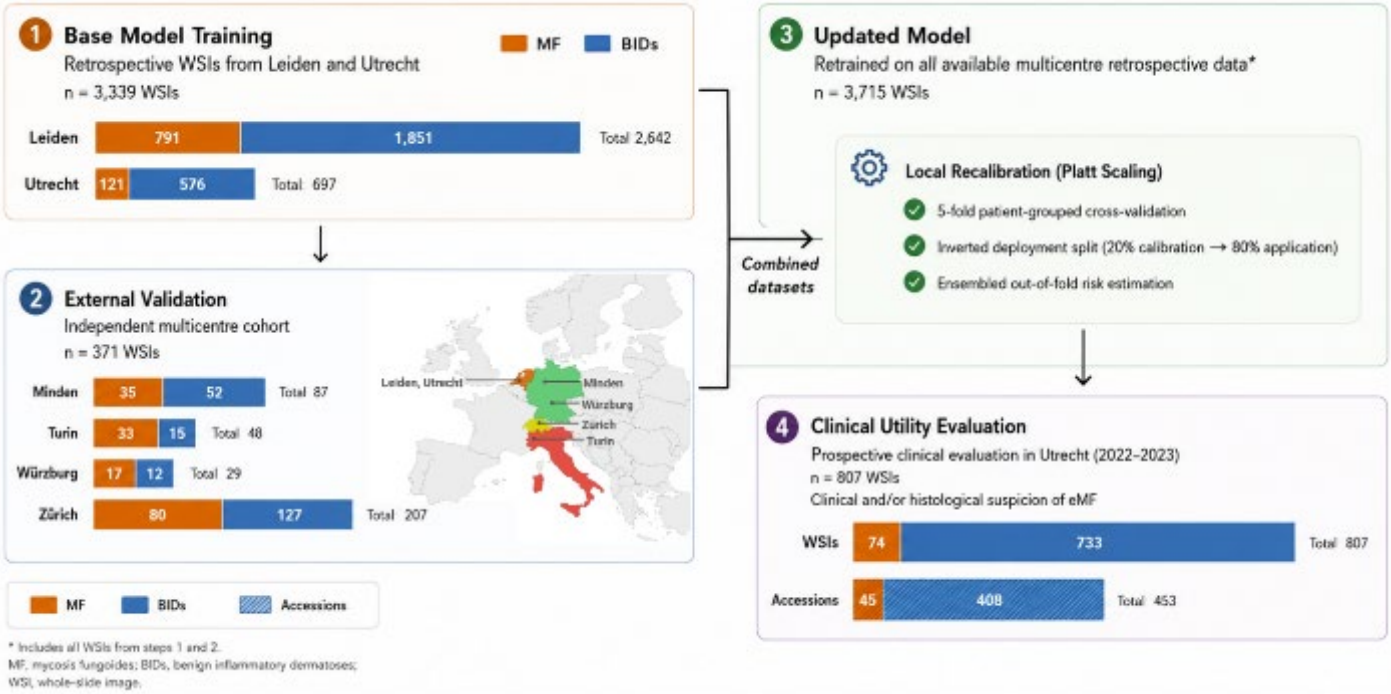
Journal of Investigative Dermatology (2025) **145**, 1127–1134; doi:10.1016/j.jid.2024.07.036





Histological triage of early-stage mycosis fungoides using a weakly supervised deep learning-based model: a multicentre, external validation, and clinical utility study

Thom Doeleman[1,2,†], Siemen Brussee[1,†], Pieter A. Valkema[1], Werner Kempf[3], Maarten H. Vermeer[4], Jesper Kers[4,5,6], Liliane C.D. Wynaendts[7], Kelly G.P. Kerckhoffs[8], Marthe M. de Jonge[9], Anh H. Nguyen[10], Elke E.M. Peters[11], Marion Wobser[12], Andreas Rosenwald[13], Rudolf Stadler[14], Patty M. Jansen[1], Maxime Battistella[15], Gabriele Roccuzzo[16], Pietro Quaglino[16], Anne M.R. Schrader[1]



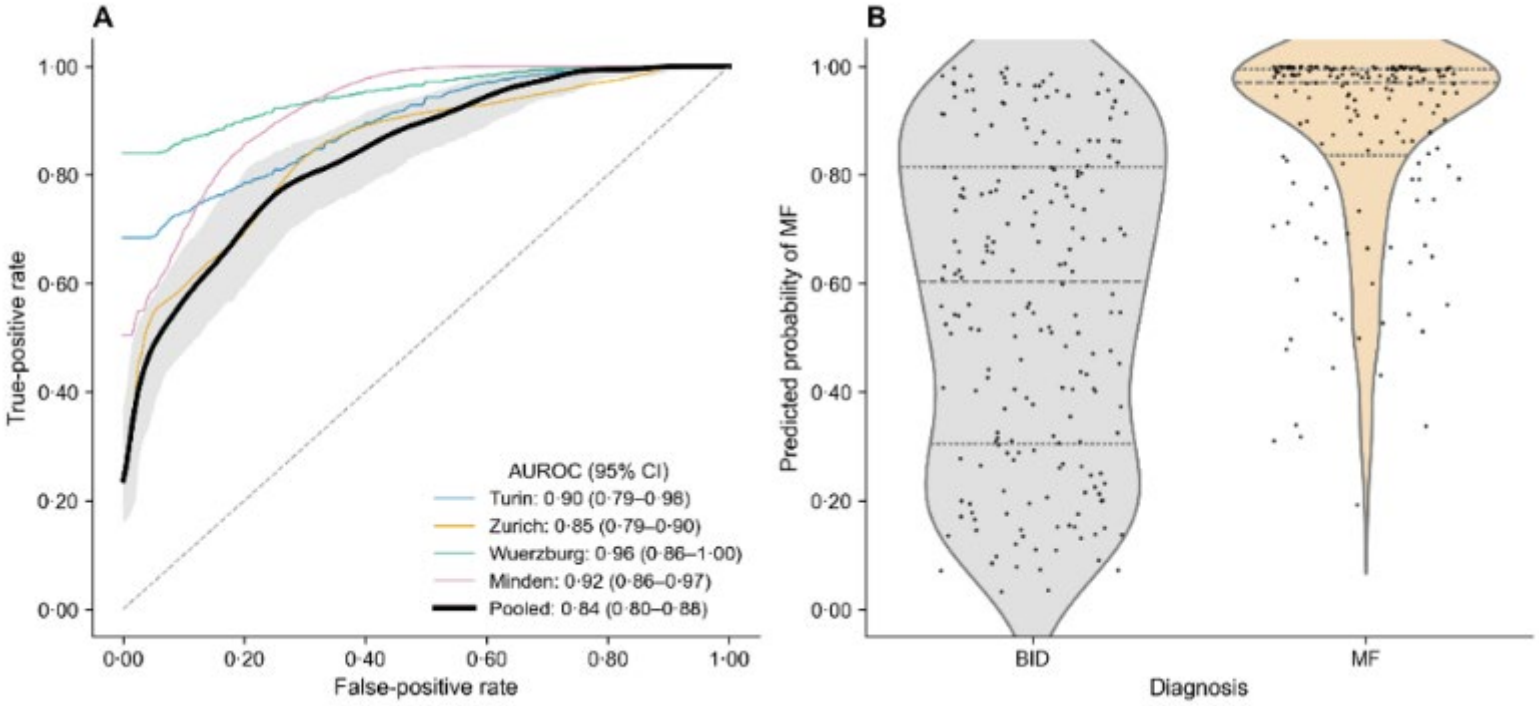


Figure 2. External validation of the base model across four European centres.

Doelemann et al, under review


IBM
GARRY
KASPAROV
DEEP
BLUE

Is delay in MF diagnosis clinically meaningful ?

Survey Response		Non-Specialist (n = 296)	Specialist (n = 86)	*p* value
Do you think the diagnosis of early-stage MF is delayed?	Yes, n (%)	228 (78.9%)	73 (91.3%)	0.02*
	No, n (%)	35 (12.1%)	6 (7.5%)	
	Don't know, n	26 (9.0%)	1 (1.3%)	
Do you think that a patient's prognosis is negatively affected by a delayed diagnosis of early-stage MF?	Yes, n (%)	93 (34.1%)	16 (21.1%)	0.02*
	No, n (%)	119 (43.6%)	47 (61.8%)	
	Don't know, n (%)	61 (22.3%)	13 (17.1%)	
Do you think that therapeutic success is negatively affected by a delayed diagnosis of early-stage MF?	Yes, n (%)	114 (41.9%)	25 (32.9%)	< 0.001*
	No, n (%)	96 (35.3%)	45 (59.2%)	
	Don't know, n (%)	62 (22.8%)	6 (7.9%)	
Do you think that a patient's health-related quality of life is impaired by a delayed diagnosis of early-stage MF?	Yes, n (%)	172 (63.2%)	58 (76.3%)	0.09
	No, n (%)	68 (25.0%)	11 (14.5%)	
	Don't know, n (%)	32 (11.8%)	7 (9.2%)	
Do you think that a timely diagnosis of early-stage MF can result in cure?	Yes, n (%)	48 (17.6%)	12 (15.8%)	0.05*
	No, n (%)	151 (55.5%)	53 (69.7%)	
	Don't know, n (%)	73 (26.8%)	11 (14.5%)	

Beatty et al, JDDG 2025

MF diagnosis: wrap up

- Clin-path correlation remains GOLD-STANDARD

- IHC for tumor cell detection/characterization
  - Practices are heterogeneous
  - PD1 and TOX may have interest (context+++)
- Clonality
  - HTS techniques are more precise than PCR
  - TRBC1: interest ; limitations
- Molecular tests: we are not there yet
- AI: triage tool?

Thanks

Dermatology Department, Cutaneous Lymphoma Clinic
Pr Martine Bagot, Pr Adèle de Masson, Dr Caroline Ram-Wolff
Dr Maelle Dumont, Dr Gilles Battesti, Dr Alizée Bozonnat,
Dr Jérémie Delaleu

Pathology Department
Dr Bénédicte Cavelier-Balloy, Dr Florian da Silva, Dr Yoan Ditchi, Dr Amélie Osio, Dr Amel Hasni, Dr Maxime Mouthon, Dr Véronique Meignin

Tumor Genomics Department
Pr Samia Mourah, Dr Fanélie Jouenne, Dr Baptiste Louveau

Hemato-Immunology
Pr Hélène Moins

INSERM U1342
Anne Marie-Cardine, Pr Jean-David Bouaziz

Hôpital Saint-Louis AP-HP

GFELC

EORTC Cutaneous Lymphoma Tumours Group

International Society for Cutaneous Lymphomas ISCL

CLIC CUTANEOUS LYMPHOMA INTERNATIONAL CONSORTIUM

maxime.battistella@aphp.fr