COACHES

Current Opinions, Advances, Controversies in HEmatology in Salerno

Updates in **Chronic Lymphocytic Leukemia** and **Lymphomas**

Salerno | **14 aprile 2025** | Grand Hotel Salerno

Novel prognostic biomarkers and risk stratification systems

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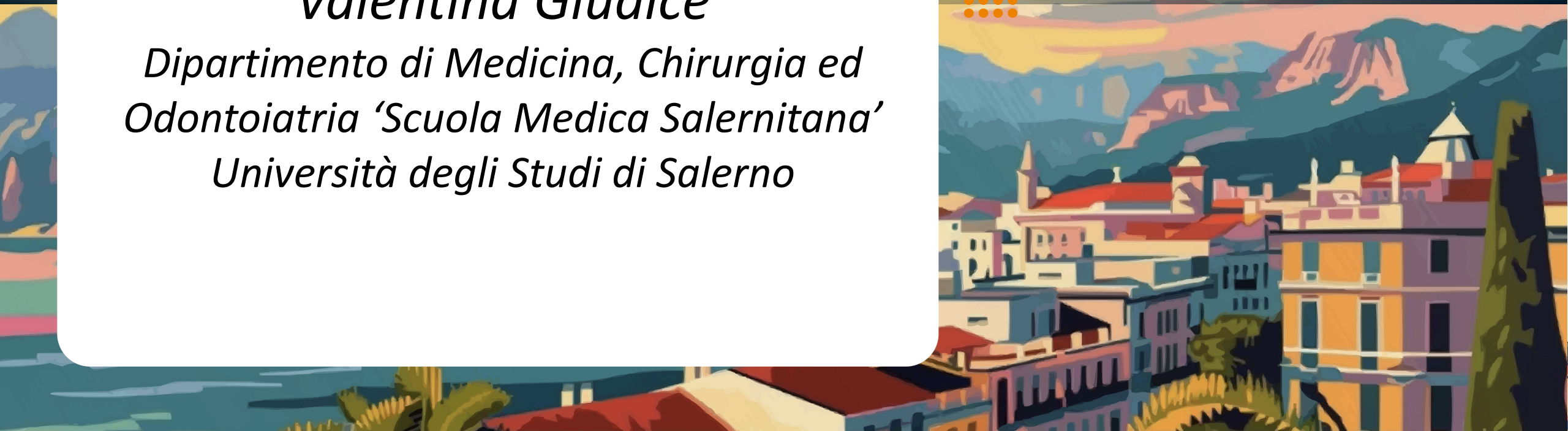
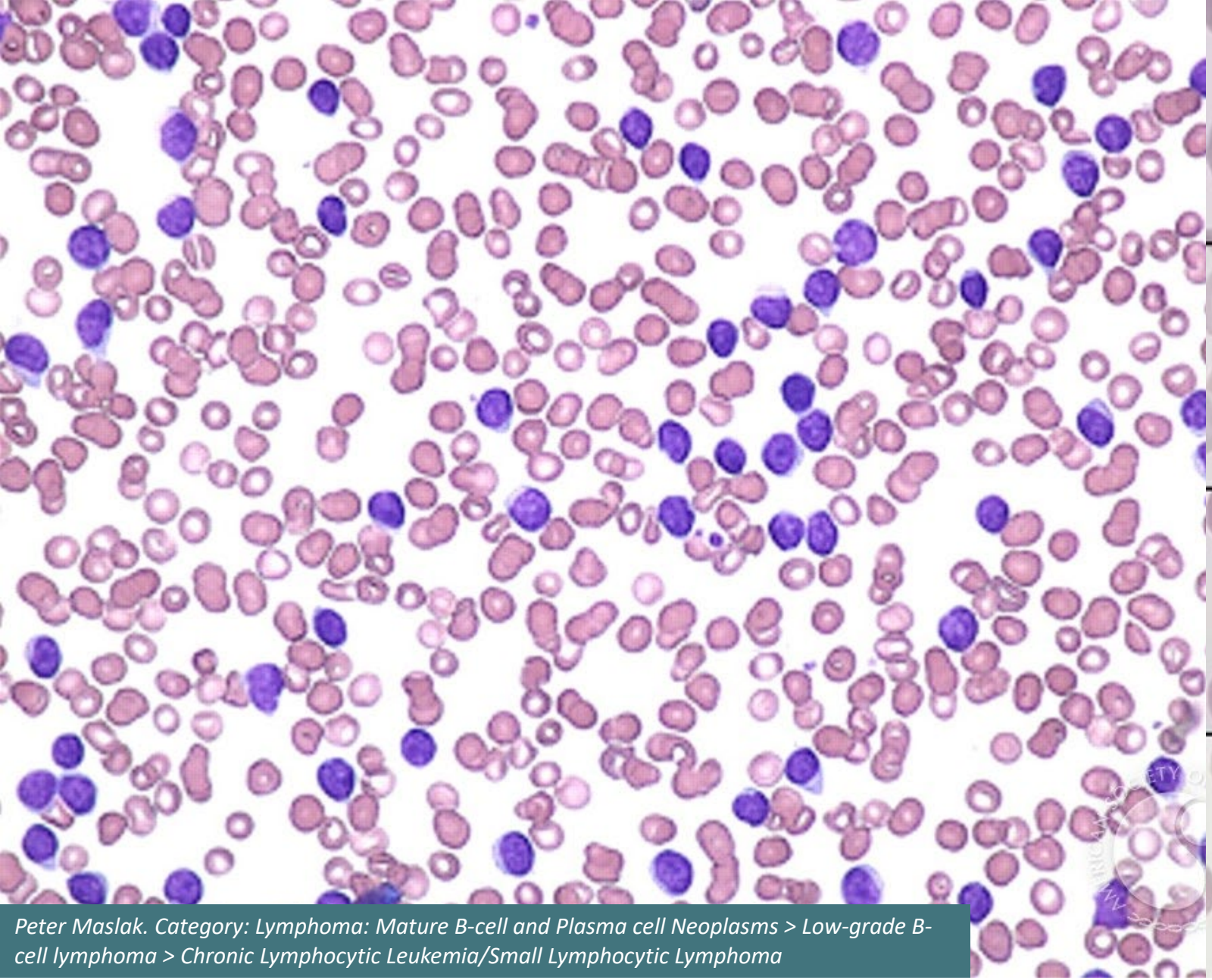
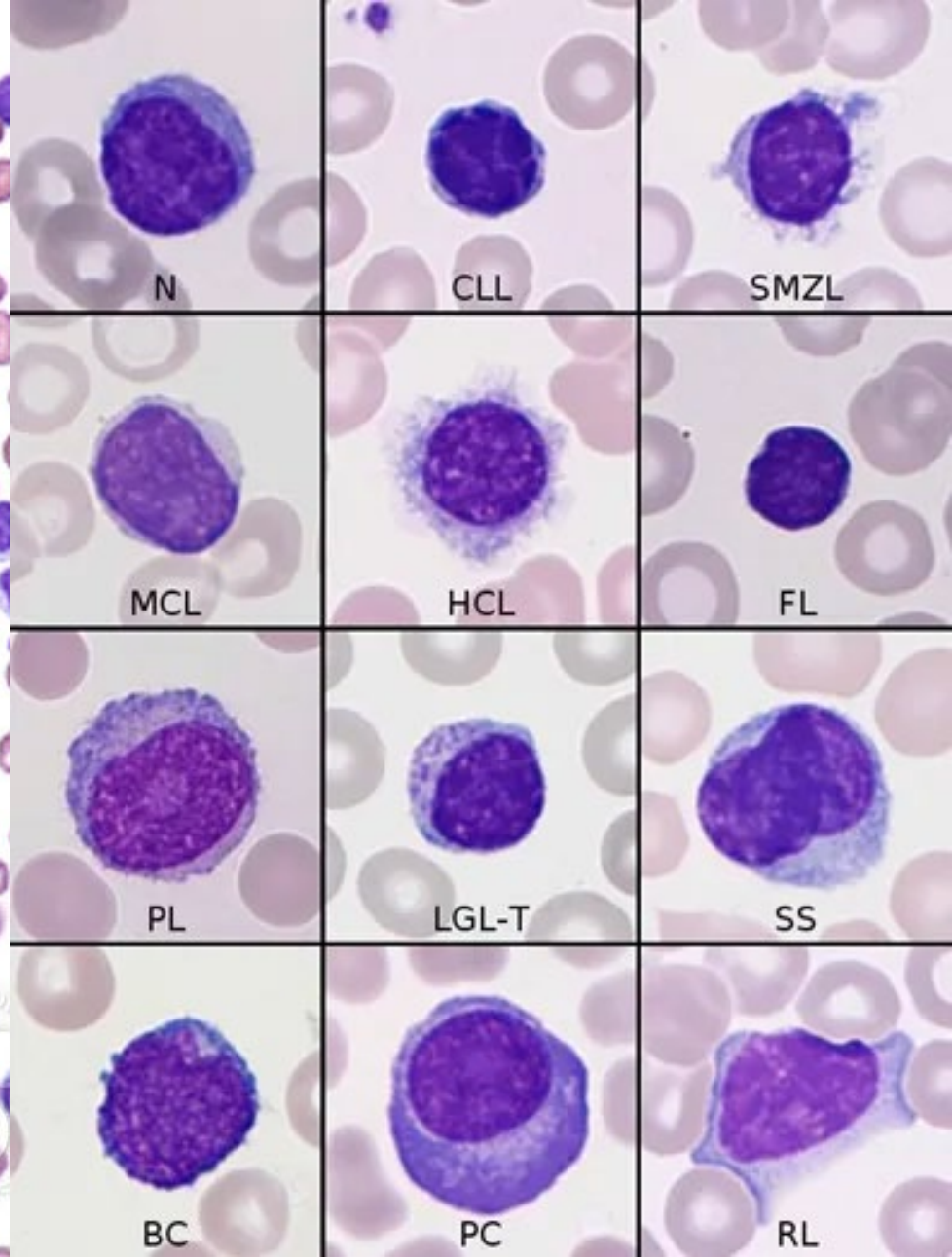


Disclosures of Valentina Giudice

Company name	Research support	Employee	Consultant	Stockholder	Speakers bureau	Advisory board	Other





Peter Maslak. Category: Lymphoma: Mature B-cell and Plasma cell Neoplasms > Low-grade B-cell lymphoma > Chronic Lymphocytic Leukemia/Small Lymphocytic Lymphoma

Chronic lymphocytic leukemia

Do we need novel prognostic markers?

An old question still unanswered?

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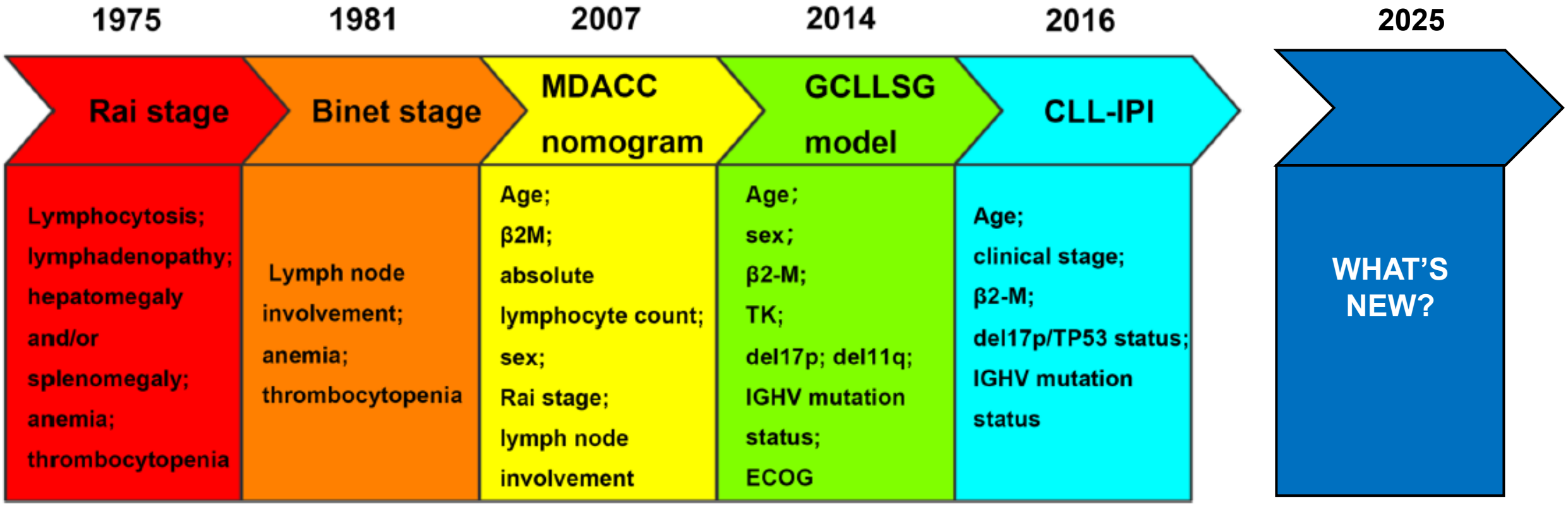
Hematology Education: the education program for the annual congress of the European Hematology Association

2013;7:121-130

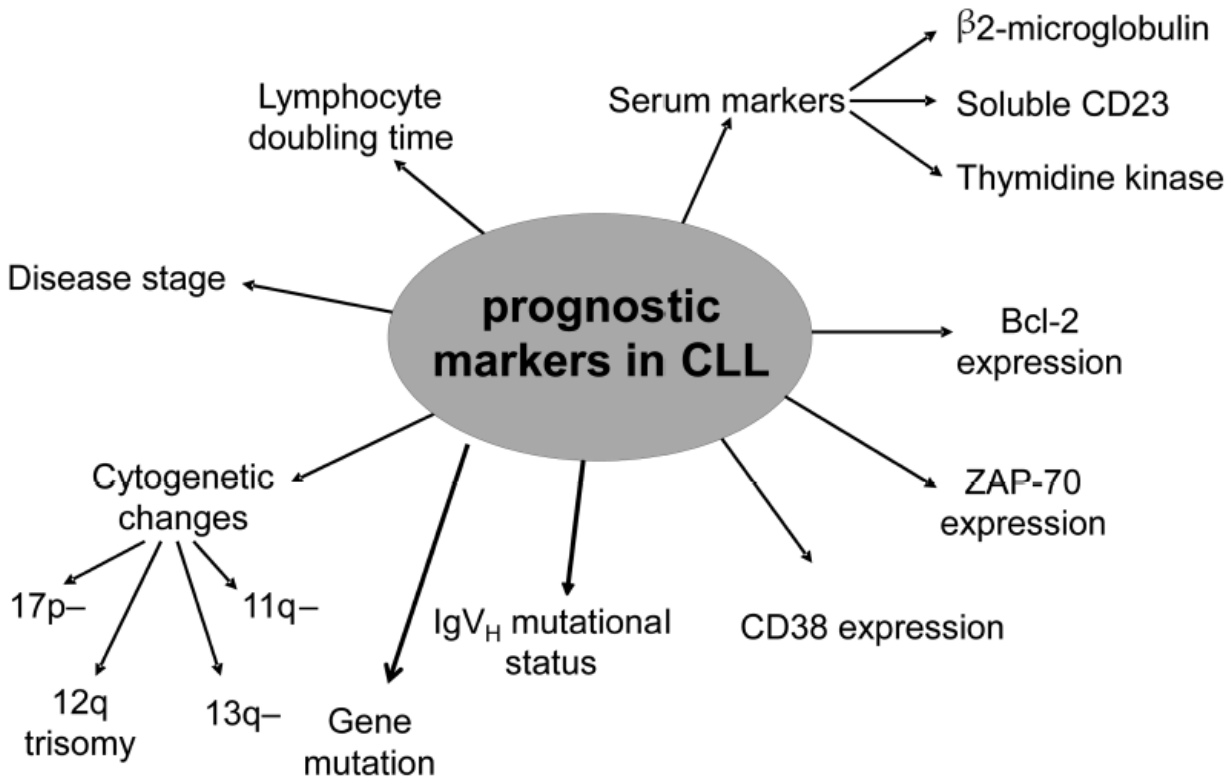
Serum markers		B2M, STK, sCD23, sFLC
Genomic abnormalities	Copy number variation	del 13q, del 11q, p53 loss, gain8q24, +12
	Genomic complexity Chromothripsis	
	Genetic mutations	TP53, ATM, NOTCH1, SF3B1, BIRC3
	Gene SNPs	
DNA methylation	Global arrays	
	Specific genes	ZAP 70
Gene expression	mRNAs	CLLU1, LPL, AID
	miRs	21, 29c,34a, 181b,223
	Protein	CD38, CD49d, CD69, ZAP70, TCL1
	Global assays	Gene expression profiles, proteomics
IGVH genes		Mutational load, VH gene usage, stereotypye
Telomere abnormalities	Telomere length	
	Telomerase activity	
Functional assays	BCR, CD40 signaling	
	P53 function	

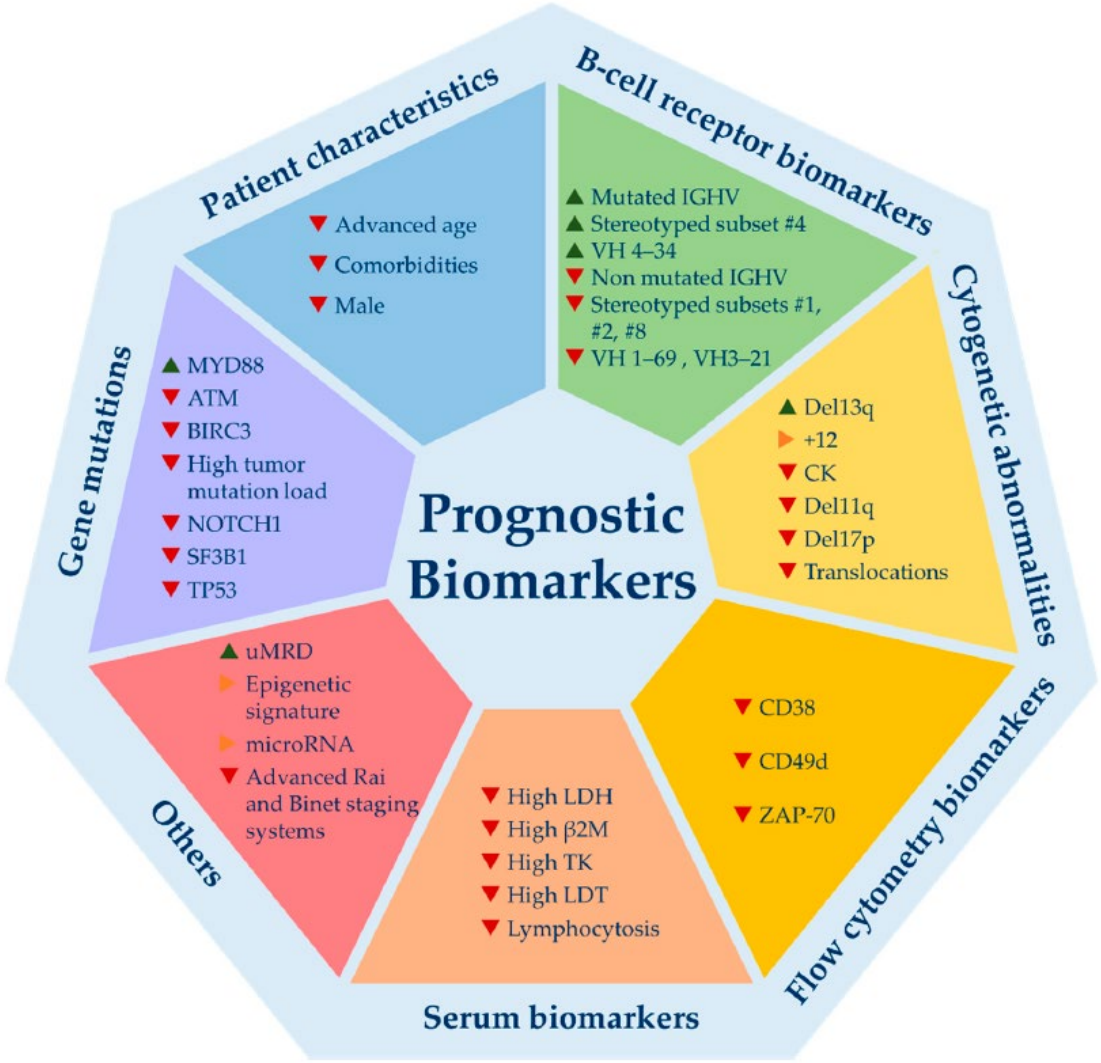
Are we really moving forward?



Yun X, et al. Biomark Res. 2020 Sep 7;8:40.

From 2014 to 2025: are there changes?





J.G. Gribben. EHA. 2014;8:69-74

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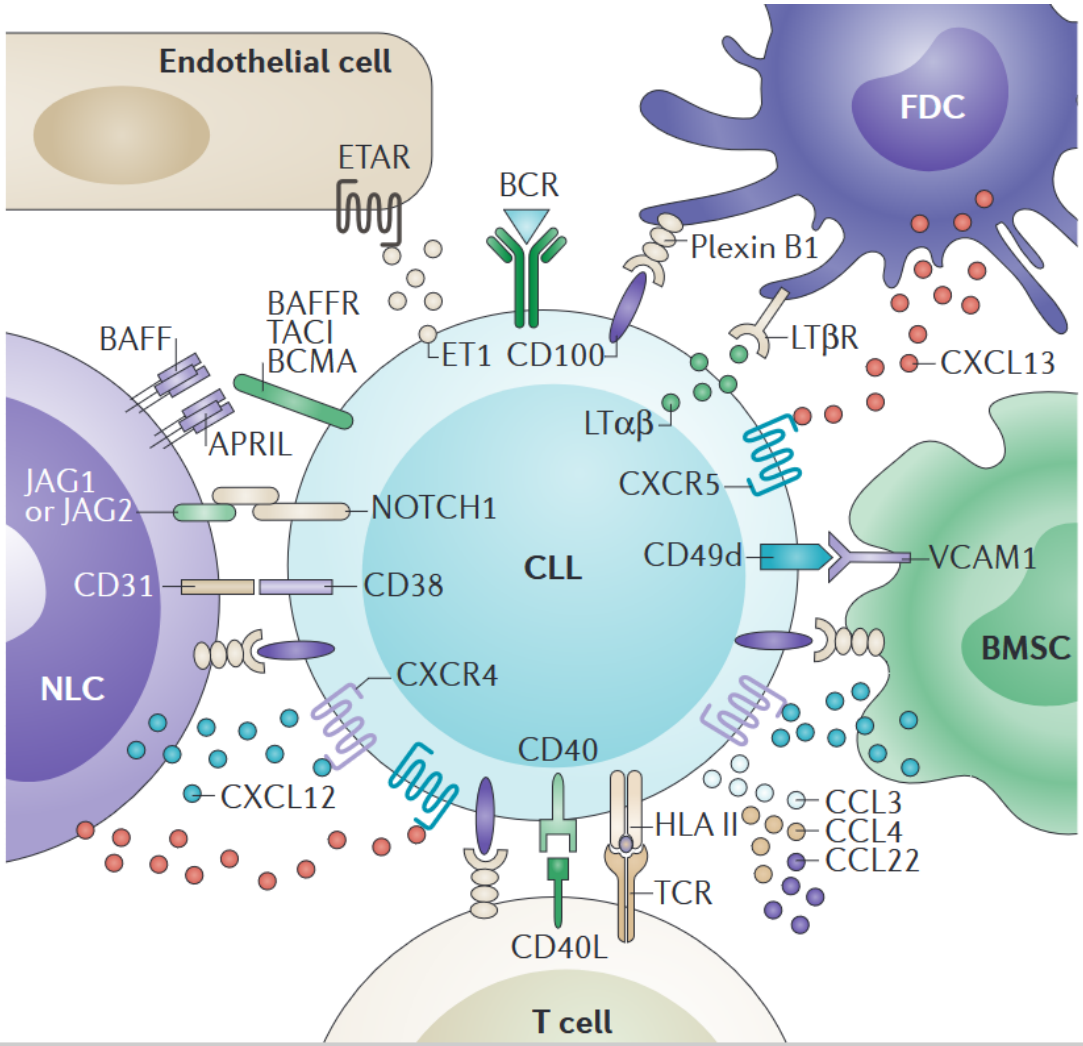
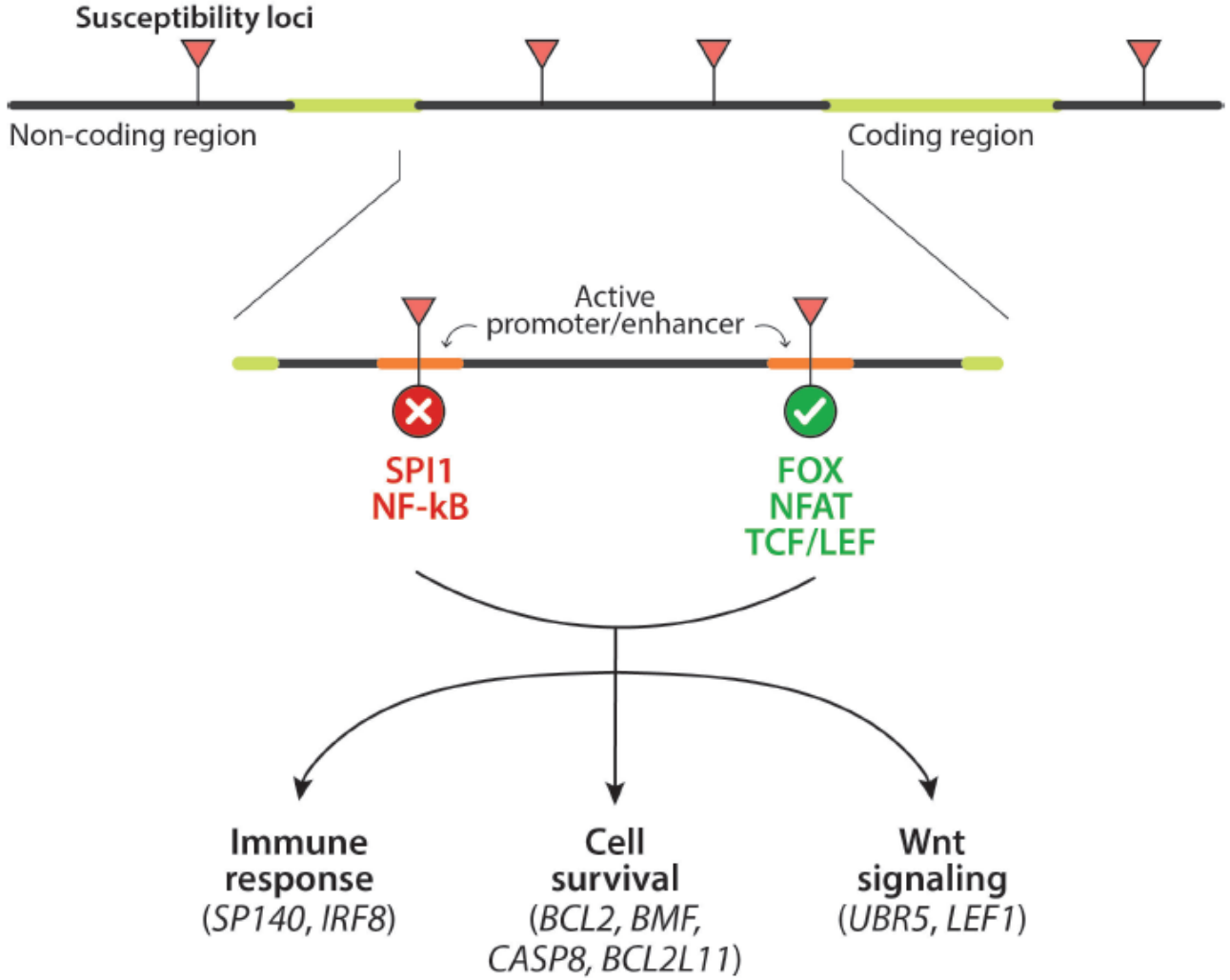
Molecular markers: biological basis



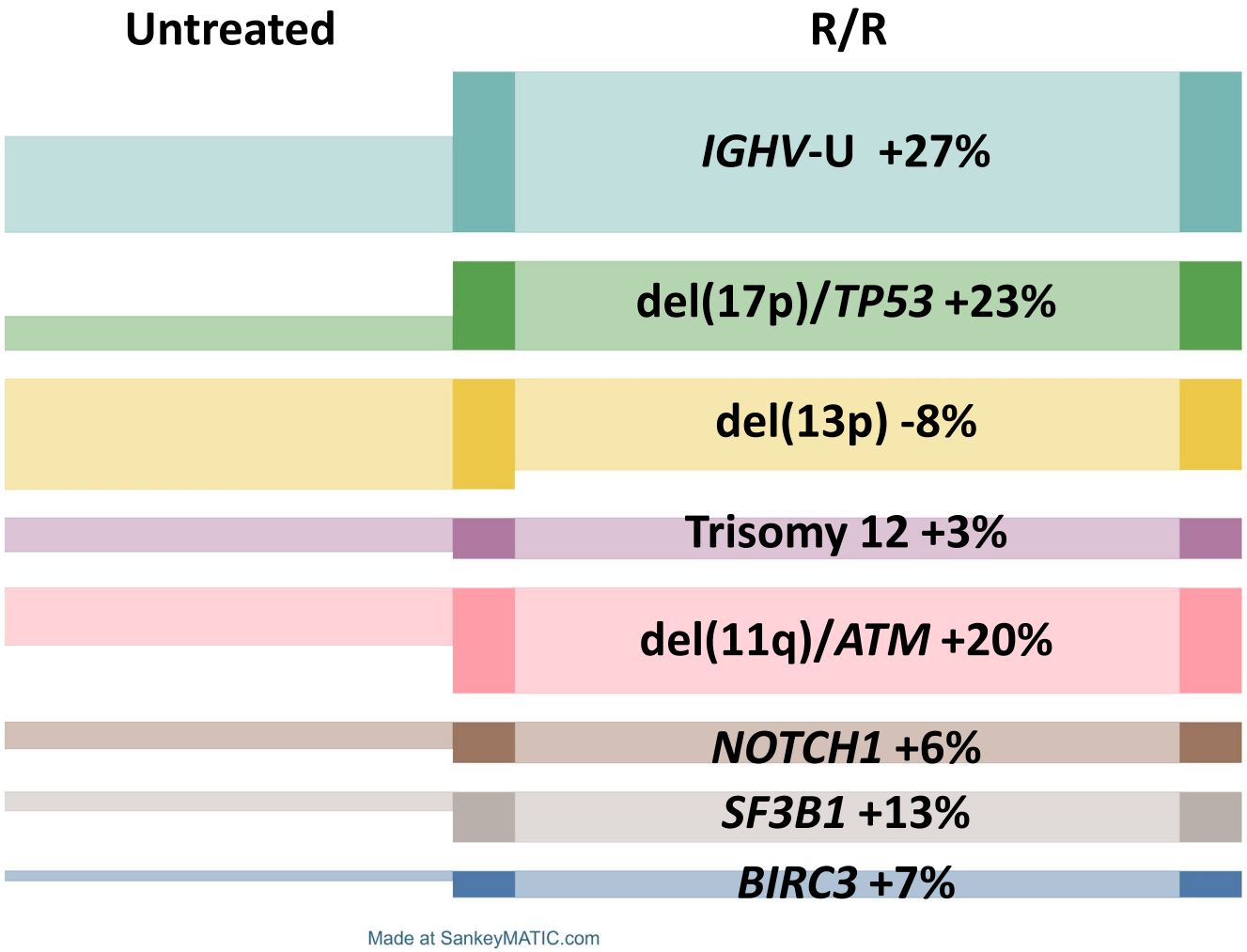


Fabbri G, Dalla-Favera R. Nat Rev Cancer. 2016 Mar;16(3):145-62.

Delgado J, et al. Haematologica. 2020 Sep 1;105(9):2205-2217.

Molecular markers: frequencies and incidence

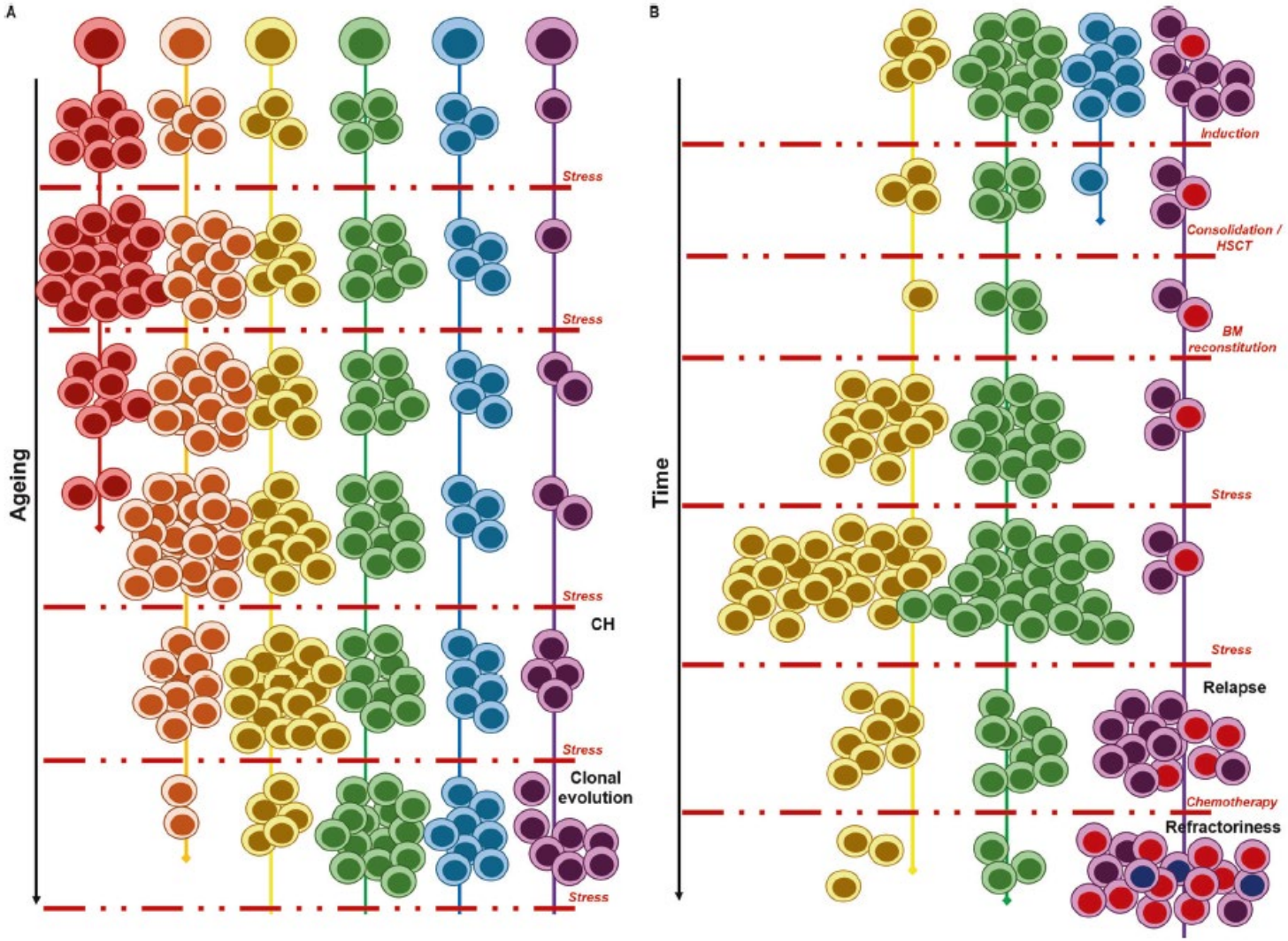
- ***TP53*** mutations, ***IGHV* unmutated** status, **del(17p)**, and **del(11q)**, and **complex karyotype** associated with poor prognosis.
- Trisomy 12 intermediate prognostic factors, whereas del(13q) favorable prognosis.
- Genetic alterations associated with poor prognosis are often **more enriched in patients at advanced stages** or after treatment, such as *TP53* mutations, del(17p), and *IGHV* unmutated status.
- Might reflect the **clonal evolution during natural disease progression** or change in clonal dynamics induced by therapies, especially chemotherapies.



Modified from Lee J, Wang YL. J Mol Diagn. 2020 Sep;22(9):1114-1125.

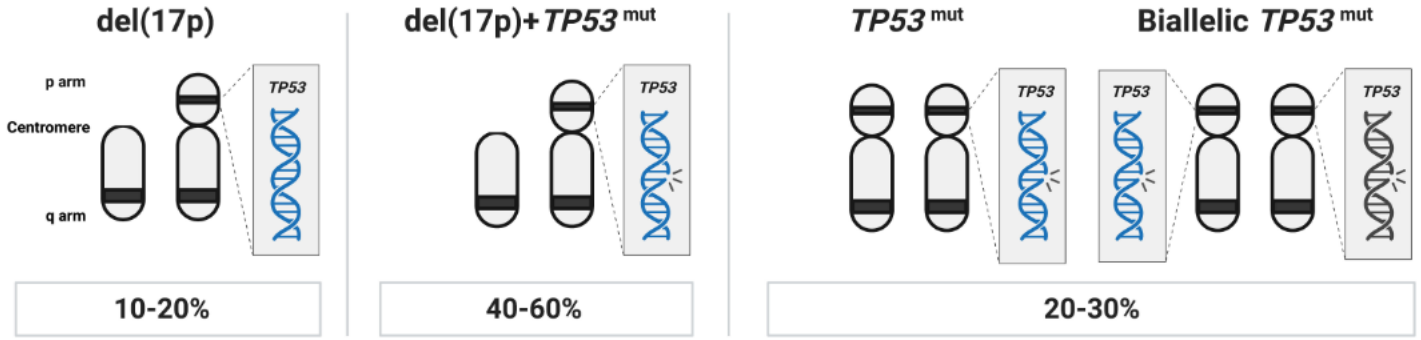
The importance of subclones

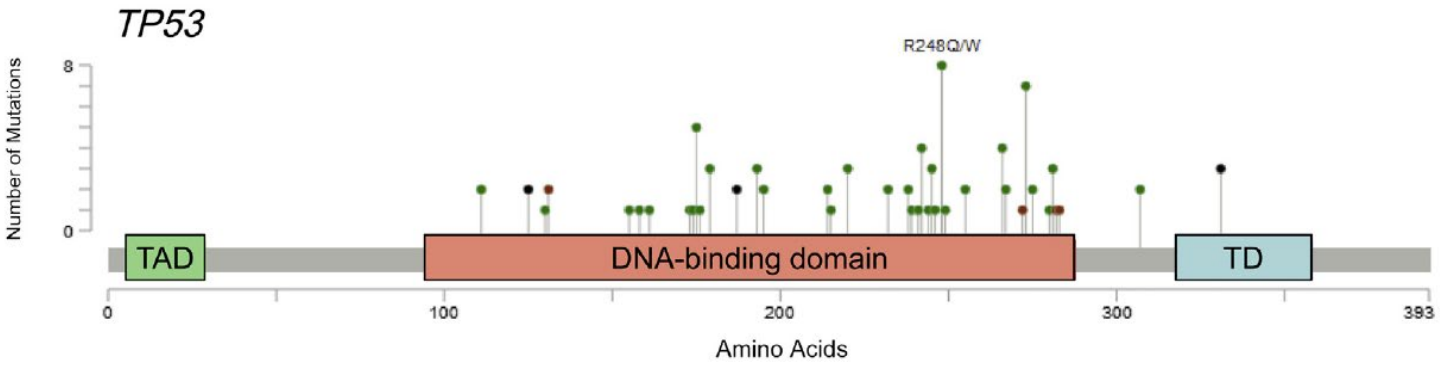
Variable	TTT	OS
TP53		
Clonal	No impact	Shorter OS
Subclonal	No impact	Shorter OS
SF3B1		
Clonal	Shorter TTT	Trend for a shorter OS
Subclonal	No impact	No impact
BIRC3		
Clonal	No impact	Trend for a shorter OS
Subclonal	No impact	No impact
NOTCH1		
Clonal	Shorter TTT	Shorter OS
Subclonal	Shorter TTT	No impact
ATM		
Clonal	Shorter TTT	No impact
Subclonal	*	*



del(17p)/*TP53* mutational status

- Del(17p) and *TP53* mutations are observed in ~14% of untreated CLL, and ~37% of treated CLL.
- Deletion 17p commonly **co-occurs** with *TP53* mutation on the other allele.
- Missense, frame-shift, or splicing mutations, and usually involve the DNA binding domain encoded between exon 4 and exon 8.
- Abolish the function of TP53 as a tumor suppressor.
- Inactivation of a single *TP53* allele is associated with poor survival.
- **Bi-allelic *TP53* inactivation** confers a worse OS in all populations, in both treated and untreated patients.



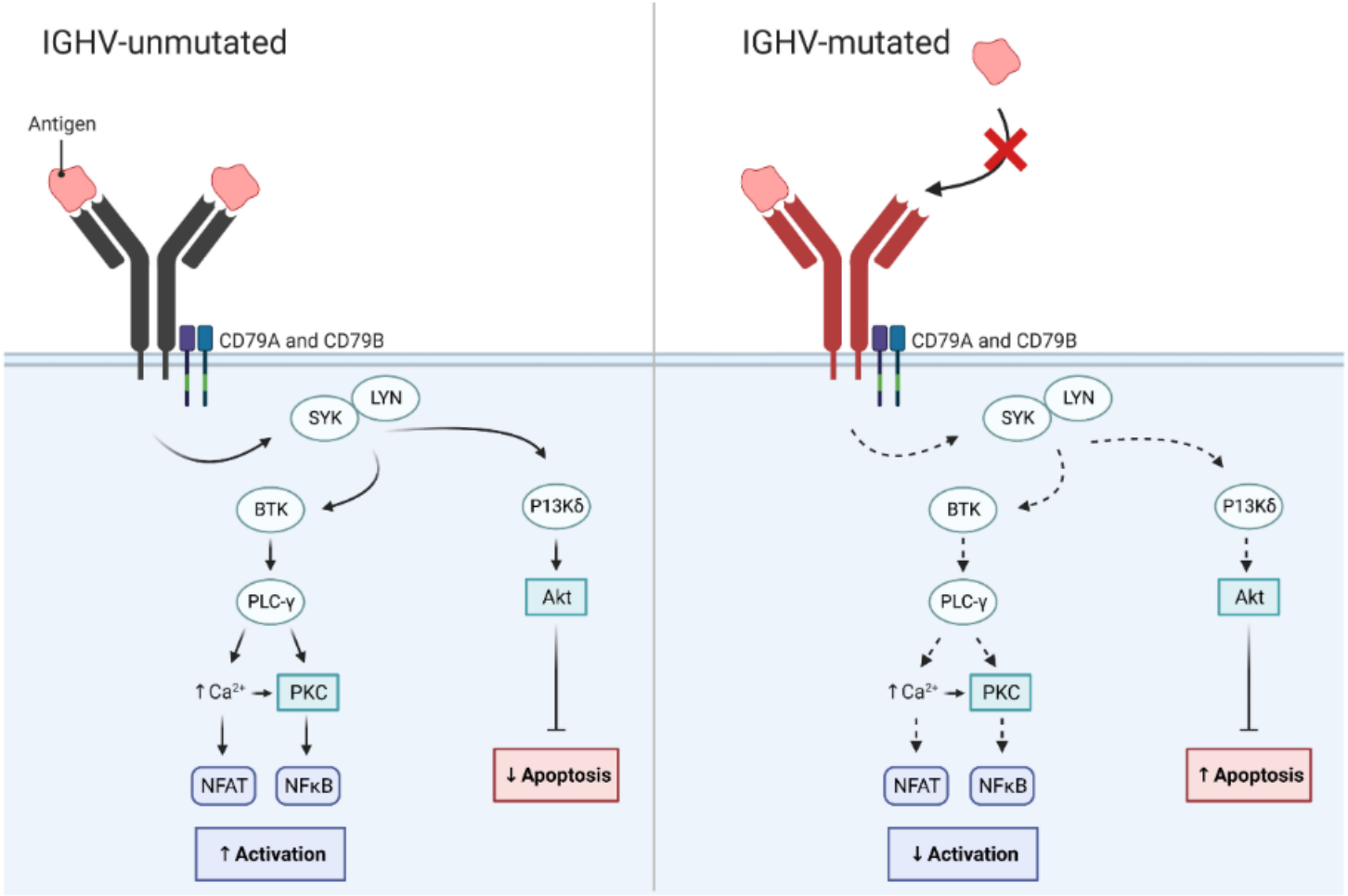


Lee J, Wang YL. J Mol Diagn. 2020 Sep;22(9):1114-1125.

Mollstedt J, et al. Front Oncol. 2023 Mar 21;13:1146486.

IGHV mutational status

- IGHV mutations are generated through somatic hypermutation, a physiological process to generate Ig diversity during normal B-cell maturation.
- Unmutated IGHV is present in **~40%** of untreated cases.
- **Worse prognosis** and poor response to chemoimmunotherapy.
- In the NGS era, IGHV remains one of the strongest independent prognostic marker.
- Unmutated IGHV could increase BCR signaling and malignant cell proliferation.
- **98% homology or 2% mutation** to the germline IGV sequence is interpreted as unmutated and >2% mutation and <98% homology is considered mutated.
- A new category of 97.0% to 97.9% homology. Clinical implication is unclear.



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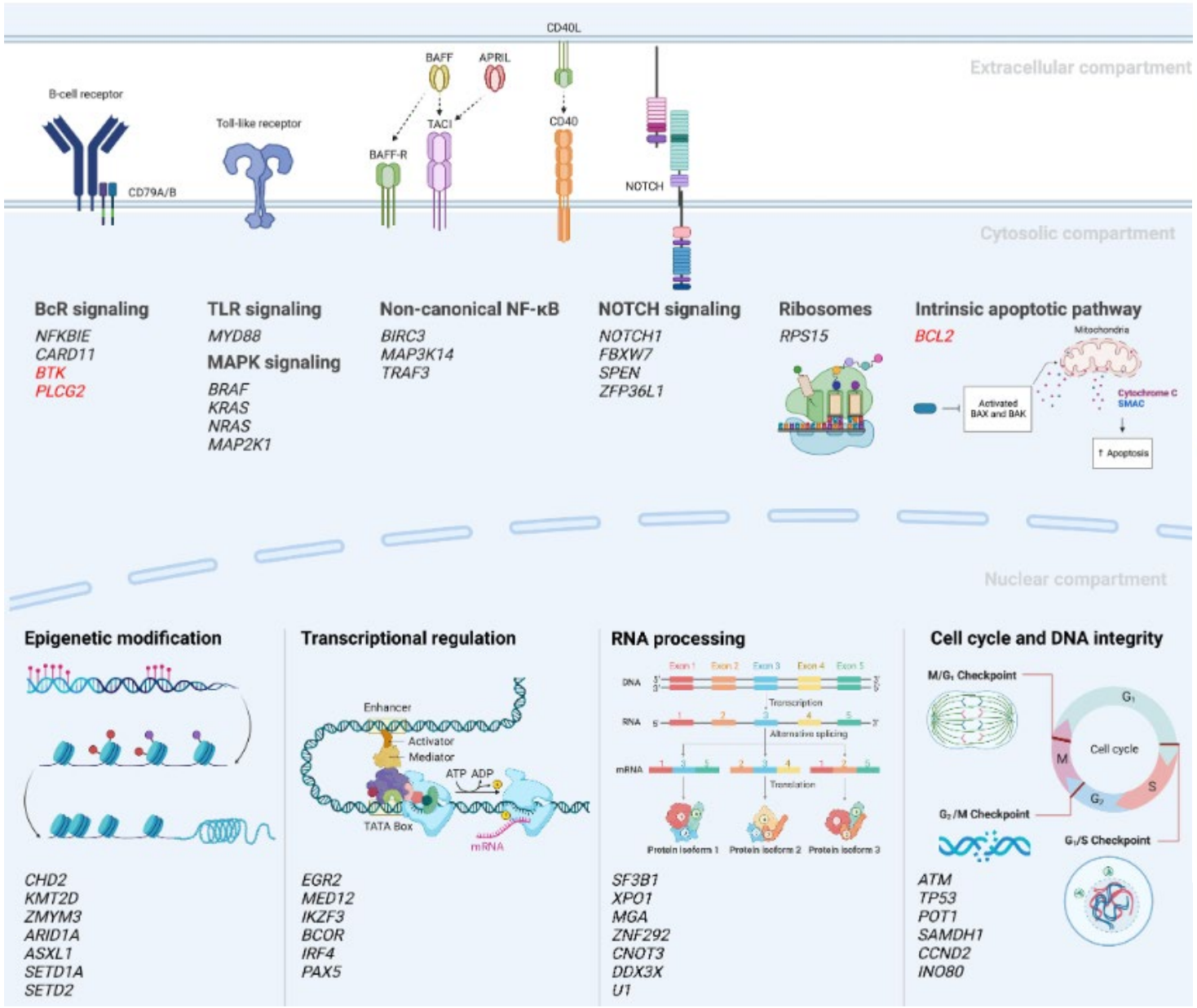


NOTCH1 signaling

NOTCH1
FBXW7

BCR and Toll-like receptor signaling

BCOR *TLR2*
KLHL6 *MYD88*
PAX5 *IRAK1*
IRF4
CARD11

MAPK-ERK pathway

KRAS
NRAS
PTPN11
GNB1
BRAF
MAP2K1

NF-κB signaling

TRAF3
TRAF2
BIRC3
EGR2
NFKBIE
NKAP
NFKB2

Chromatin modifiers

CHD2 *ARID1A*
SETD2 *BAZ2A*
ZMYM3 *HIST1H1B*
KMT2D *HIST1H1E*
SETD1A *SYNE1*
ASXL1

Cell cycle

ATM
TP53
MGA
CCND2
CDKN1B
CDKN2A

DNA damage

ATM
TP53
POT1

RNA splicing and metabolism

SF3B1 *ZNF292*
U1 *NXF1*
DDX3X *MED12*
XPO1 *FUBP1*
RPS15 *CNOT3*

Lee J, Wang YL. J Mol Diagn. 2020 Sep;22(9):1114-1125.

Mollstedt J, et al. Front Oncol. 2023 Mar 21;13:1146486.

Epigenetic changes

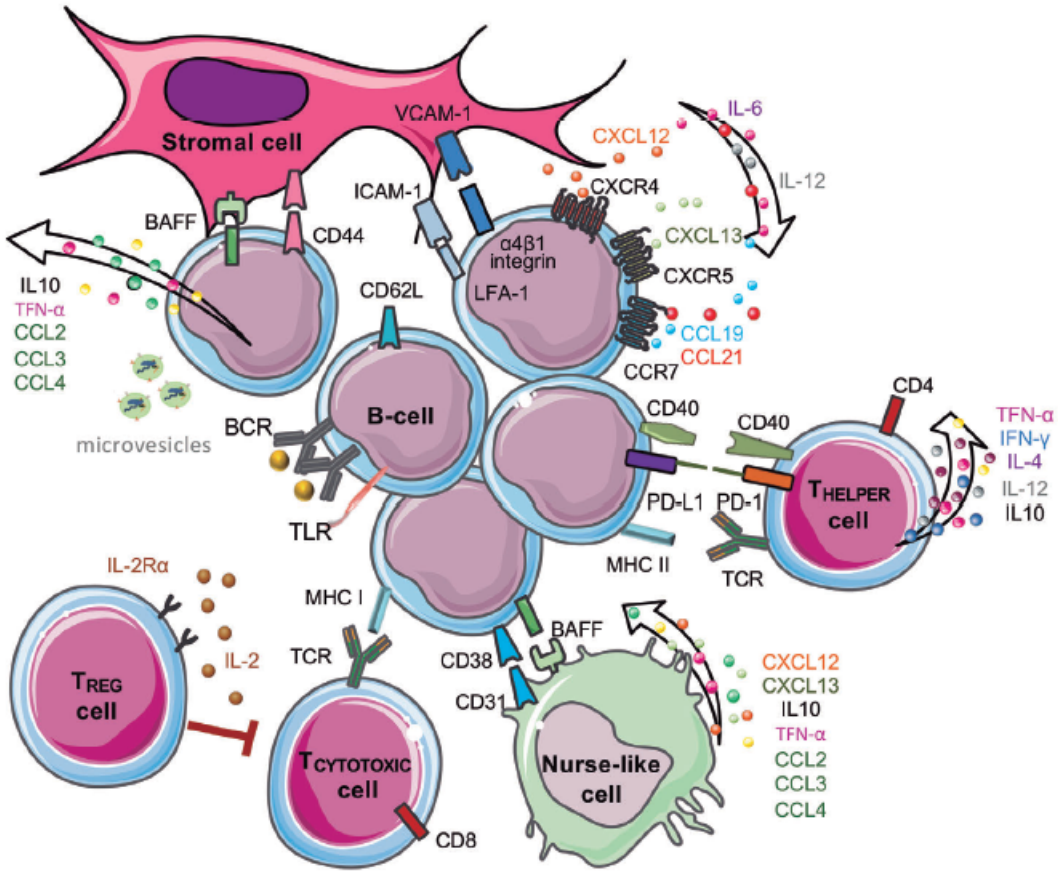
- M-CLL keeps a methylation signature of **germinal center-experienced cells** (memory-like B cells), whereas U-CLL has a **pre-germinal center, naïve-like** methylation signature.
- Major hypomethylation changes occur at transcription factor binding sites such as TCF3, PU.1/SPIB, NFAT and EGR, and enhancers that modulate genes involved in B-cell function, BCR signaling, and NF-κB activation.
- A third subtype with an intermediate profile made of cases with moderate *IGHV* mutation levels.
- Different usage of *IGHV* genes, stereotypes, genomic aberrations and clinical outcome.
- The intermediate epigenetic subtype may be more heterogeneous since it includes most stereotype subset 2 cases with aggressive behavior.

Methylation cell signature	Naïve-like	Intermediate	Memory-like
Typical IG genes	IGHV1, -5, -7 IGHD6-19 IGHJ4 IGKV1-39	IGHV3-21 IGHJ6 IGLV3-21	IGHV4-34 IGHD5-18 IGHJ6 IGKV2-30
Typical stereotype subset	1	2	4
IGHV mutations	Unmutated	Mutated or unmutated (around the 98% cutoff)	Mutated
Mutated drivers	*NOTCH1* *NFKBIE* *TP53*	*SF3B1* del(11q) rarely *TP53*	del(13q)
Clinical outcome[27]	Aggressive; TTFT at 10 years = 97%	Intermediate; TTFT at 10 years = 38%	Indolent; TTFT at 10 years = 24%

Delgado J, et al. Haematologica. 2020 Sep 1;105(9):2205-2217.

Tumor microenvironment

- Environmental or self-antigens and homotypic interactions trigger BCR and TLR signaling, **increasing activation of anti-apoptotic and proliferation pathways**.
- Notch ligands activate cell migration, invasion and angiogenesis.
- BCR and NOTCH1 pathways functionally linked, mutually enhancing their activation.
- MYD88 mutations activate the NF-κB pathway in response to TLR ligands, **increasing cytokine release** involved in recruiting stromal and T cells.
- T- and myeloid-derived cells towards a leukemia-supportive and immunosuppressive microenvironment.
- **CD8+-cell exhaustion** and monocyte M2-like differentiation.



CLL cell niche composition

- B-cells
- T-cells
  T_{HELPER} / $T_{CYTOTOXIC}$ / $T_{REGULATORY}$
- Stromal cells
  MSC / FDC
- Nurse-like cells

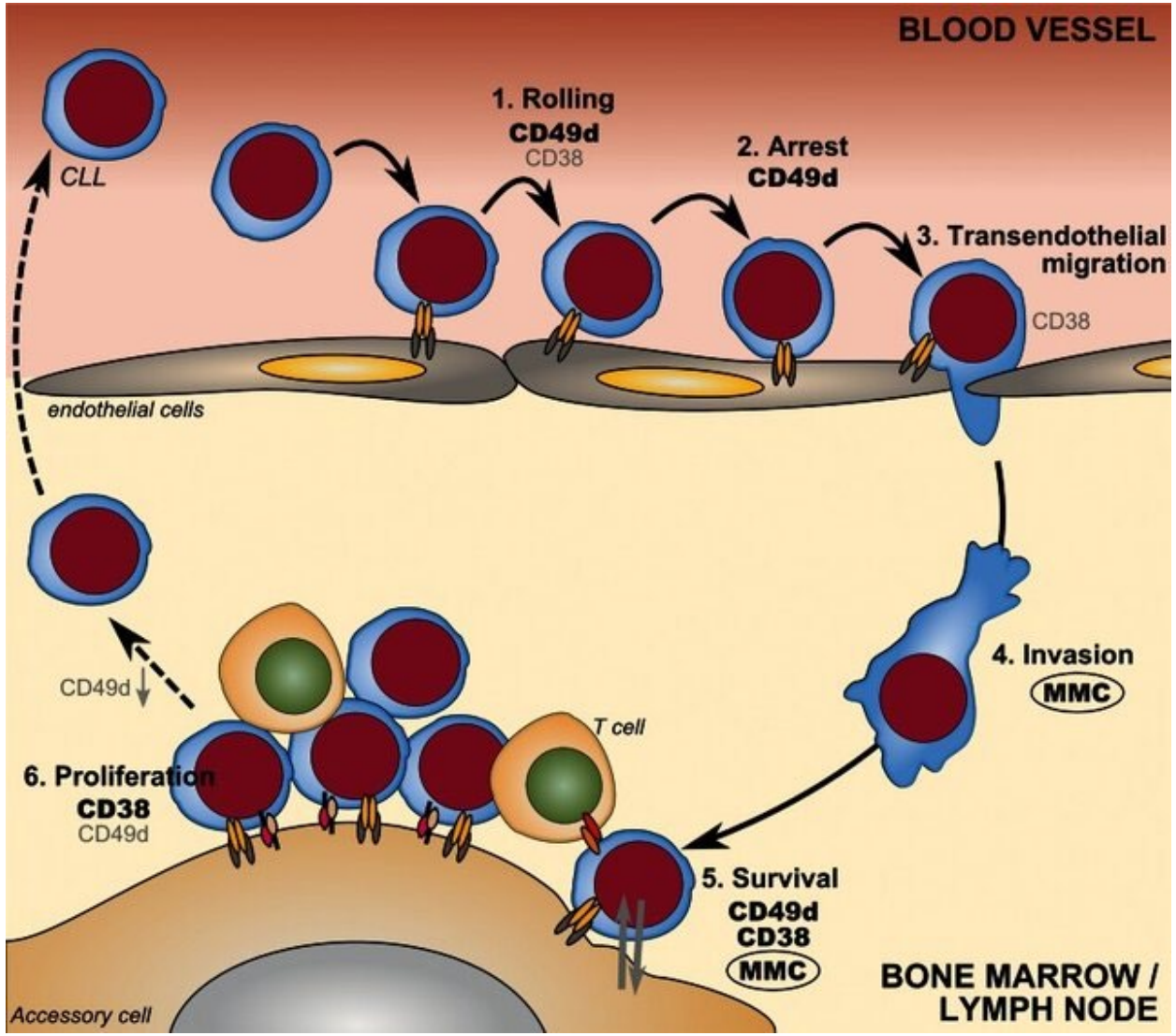
↓

- Survival
- Homing
- Migration
- Drug resistance

Delgado J, et al. Haematologica. 2020 Sep 1;105(9):2205-2217.

CD38/CD49d axis

- CD49d is a key molecule for homing of CLL cells with a major mechanistical role in rolling and arrest of CLL cells on the bone marrow and lymph node endothelia.
- CD38 may contribute to rolling of lymphocytes on the endothelium cells and to transendothelial migration.
- A macromolecular complex (MMC) is relevant for invasion within the lymphoid tissue and survival of CLL cells, with additional individual anti-apoptotic contributions of the molecules.
- CLL proliferation is strongly associated with CD38 expression.
- Mobilization of CLL cells from the lymphoid organs likely requires downregulation of CD49d expression or function.

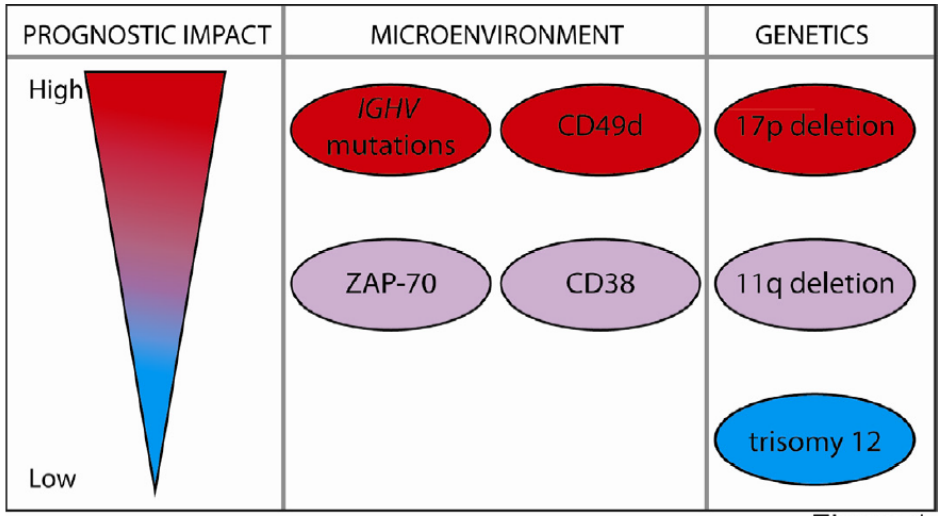


Brachtl G, et al. Ann Hematol. 2014 Mar;93(3):361-74.

	CLL Prognostic Factor
Established Prognostic Factors	**-Clinical Prognostic Models:** 1- Rai and Benet Staging Systems **-Serum Biomarkers:** 1- Lymphocyte doubling time (LDT) 2- Serum LDH 3- Serum β2 microglobulin , and Tyrosine Kinase **-B-Cell Receptor Biomarkers:** 1- IGHV mutational status 2- B-Cell receptor (BCR) stereotype **-Flow Cytometry Biomarkers:** 1- CD38 and CD49d **-Genetic Aberrations:** 1- FISH mutational panel (Del 17p, Del11q, Trisomy 12, Del 13q) 2- Complex Karyotype 3- Tp53
Newer Prognostic Factors	**-Genetic Aberrations:** 1- NOTCH1 2- ATM 3- SF3B1 4- BIRC3
Prognostic Factors of Potential Interest	**-Genetic Aberrations:** 1- MYD88 2- Expression of antiapoptotic genes **-Flow Cytometry Biomarkers:** 1- ZAP70 **-Others:** 1- MicroRNA 2- Serum Cytokines 3- Circulating Micro-vesicles 4- Lipoprotein Lipase A and ADAM29 Expression 5- Telomere Length and Telomerase Activity 6- CLLU1 expression

Braish J, et al. Front Oncol. 2024 May 16;14:1371057.

The science of making risk-stratification systems

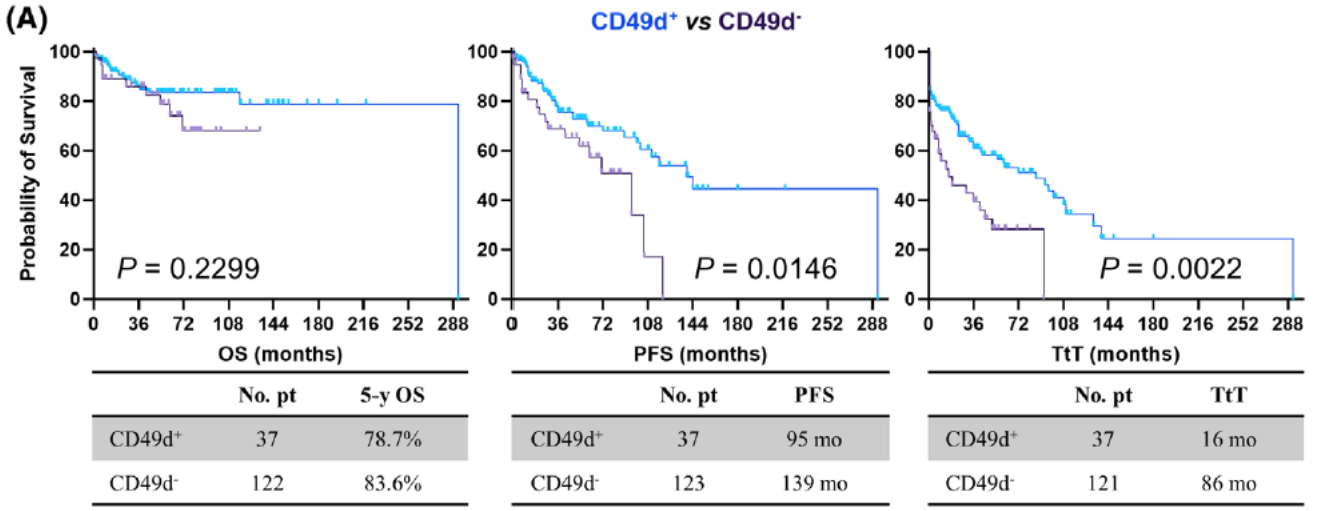


Dal Bo M, et al. Semin Hematol. 2014 Jul;51(3):168-76.

Pérez-Carretero Cet al. Diagnostics (Basel). 2021 May 10;11(5):853.

Why do we still miss flow cytometry markers?

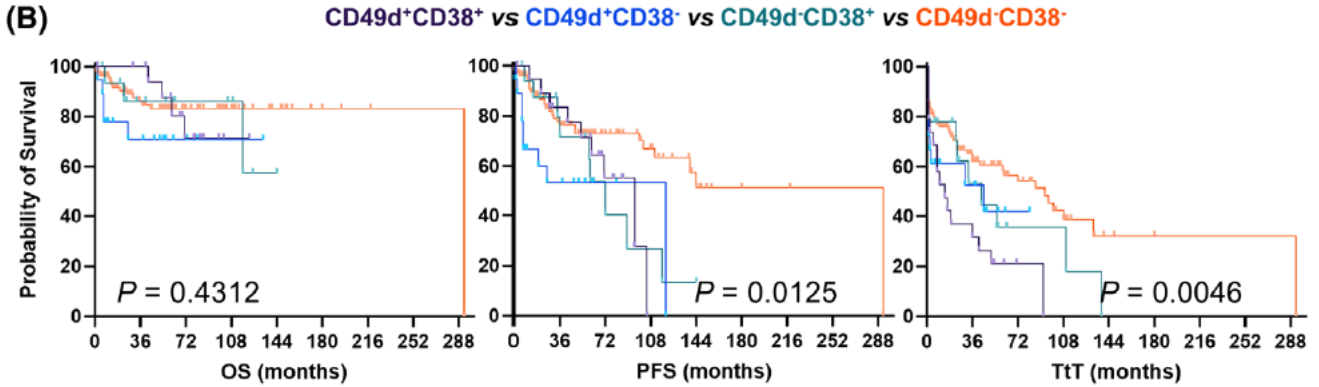
- Patients with **CD49d positivity** show significantly **higher β2-microglobulin** levels.
- Serum levels of β2-microglobulin positively correlated with CD49d expression levels.
- **Unmutated *IGHV*** more frequently found in patients with CD49d.
- **CD49d+** patients have **shorter PFS** (median, 95 months; HR, 1.96; 95%CI, 1.012–3.804), and **shorter TT** (16 months; HR, 2.54; 95%CI, 1.401–4.619).
- Patients with **CD38 positivity** with or without CD49d expression display **the shortest PFS**.
- **Concomitant expression of CD49d and CD38 associated with the shortest TT (14 months).**



	No. pt	5-y OS
CD49d+	37	78.7%
CD49d-	122	83.6%

	No. pt	PFS
CD49d+	37	95 mo
CD49d-	123	139 mo

	No. pt	TtT
CD49d+	37	16 mo
CD49d-	121	86 mo



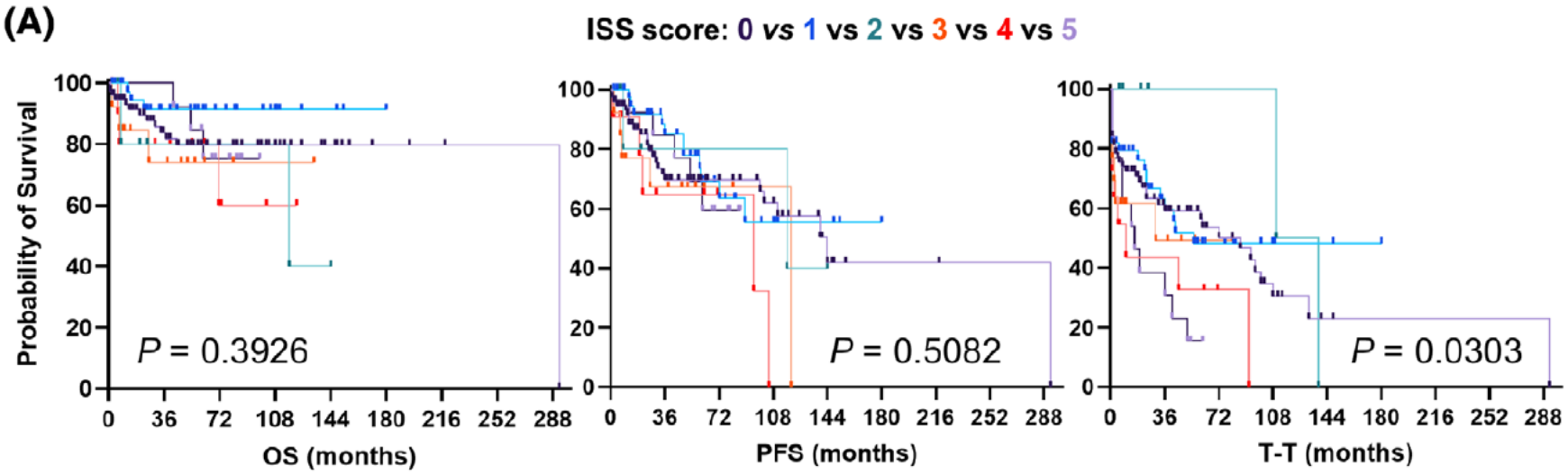
	No. pt	5-y OS
CD49d+CD38+	19	87.5%
CD49d+CD38-	18	70.7%
CD49d-CD38+	18	86.2%
CD49d-CD38-	104	83.1%

	No. pt	PFS
CD49d+CD38+	19	95 mo
CD49d+CD38-	18	120 mo
CD49d-CD38+	18	72 mo
CD49d-CD38-	105	292 mo

	No. pt	TtT
CD49d+CD38+	19	14 mo
CD49d+CD38-	18	45 mo
CD49d-CD38+	18	43 mo
CD49d-CD38-	103	93 mo

Giudice V, et al. Eur J Haematol. 2022 Nov;109(5):483-493.

Why do we still miss flow cytometry markers?



	No. pt	5-y OS
ISS 0	96	79.6%
ISS 1	40	91.3%
ISS 2	8	80%
ISS 3	13	74%
ISS 4	11	80%
ISS 5	13	84.6%

	No. pt	5-y PFS
ISS 0	77	69.7%
ISS 1	40	69.3%
ISS 2	6	80%
ISS 3	13	67.3%
ISS 4	11	64.9%
ISS 5	13	69.2%

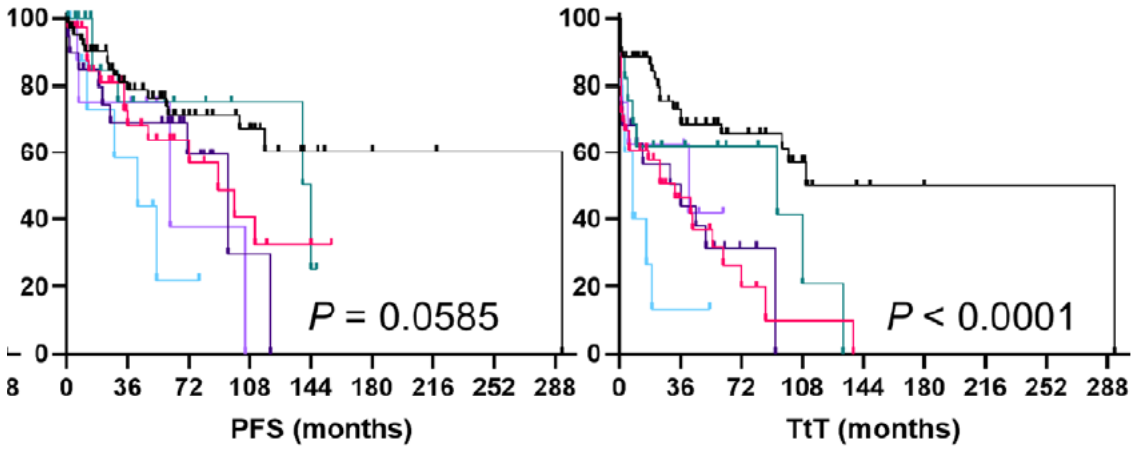
	No. pt	TtT
ISS 0	76	86 mo
ISS 1	39	55 mo
ISS 2	6	124 mo
ISS 3	13	30 mo
ISS 4	11	10 mo
ISS 5	13	16 mo

- Flow cytometry markers alone could identify more aggressive diseases requiring early drug administration.

Giudice V, et al. Eur J Haematol. 2022 Nov;109(5):483-493.

Is it time to incorporate flow cytometry in risk stratification systems?

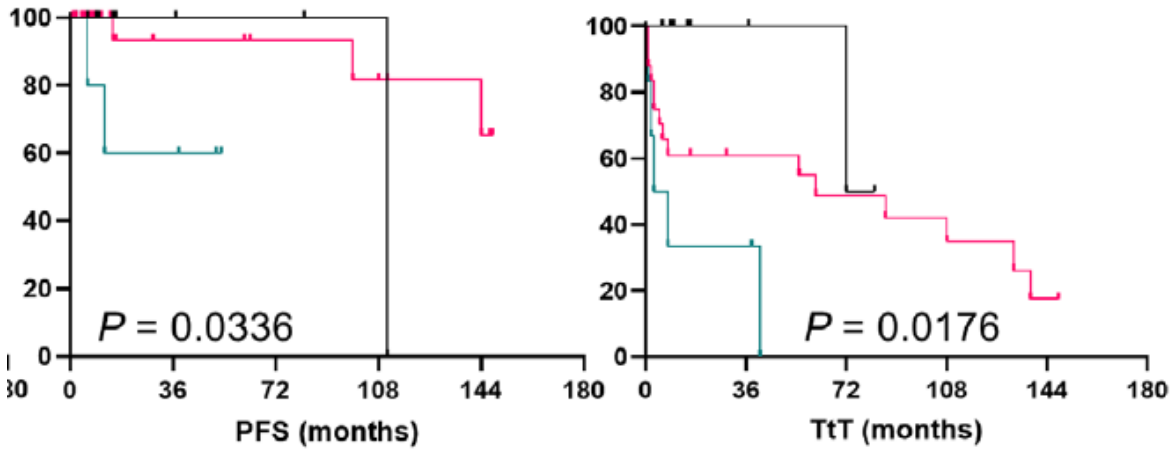
Flow cytometry + CLL-IPI



	No. pt	PFS
Low/low	69	292 mo
Low/Int	36	89 mo
Low/High	18	144 mo
High/Low	20	95 mo
High/Int	8	61 mo
High/High	9	42 mo

	No. pt	TtT
Low/low	69	110 mo
Low/Int	36	33 mo
Low/High	17	93 mo
High/Low	19	36 mo
High/Int	8	41 mo
High/High	10	8 mo

Flow cytometry + IPS-E



	No. pt	5-y PFS
Low/low	8	100%
High/Low	25	93.3%
High/High	6	60%

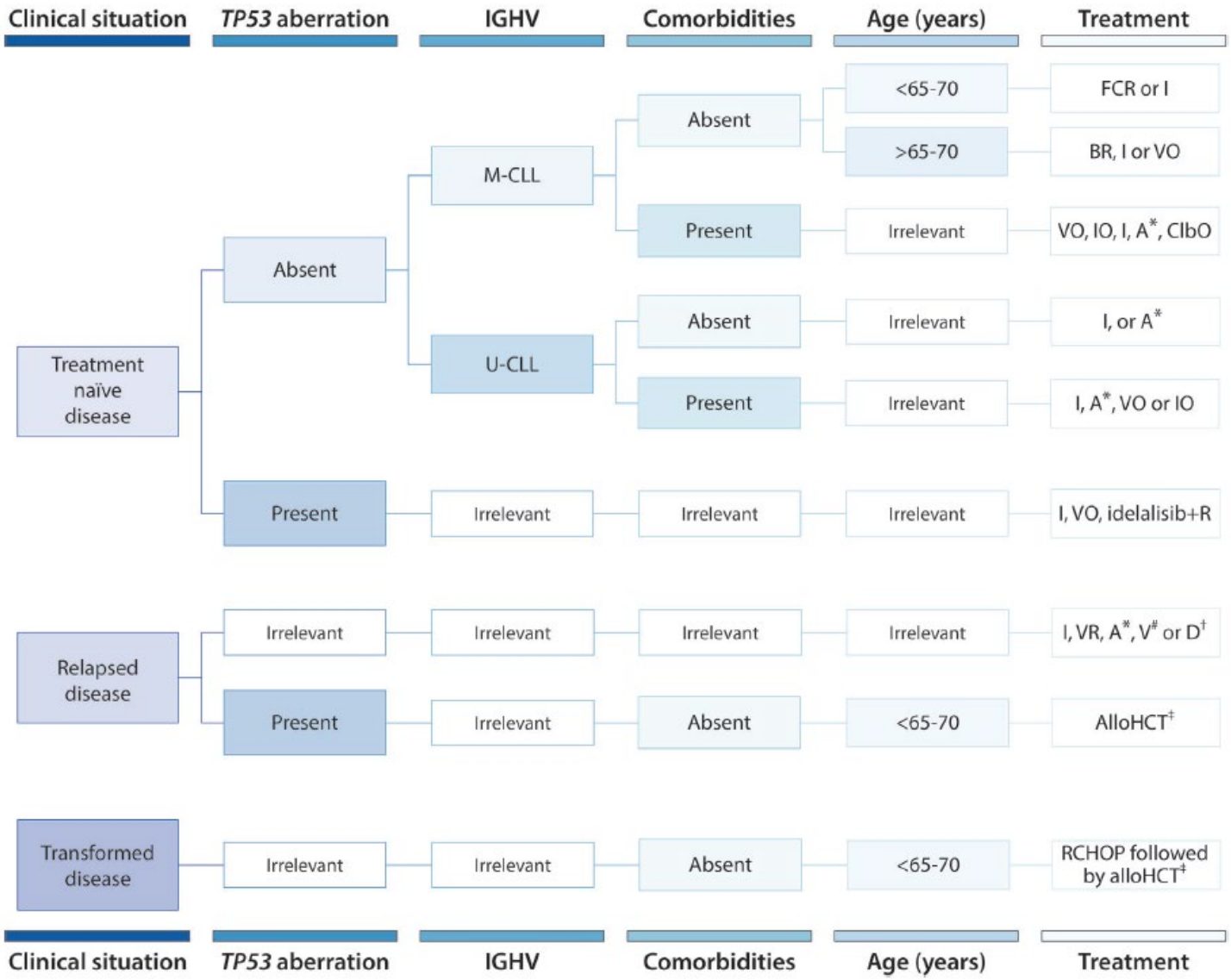
	No. pt	TtT
Low/low	8	77 mo
High/Low	25	61 mo
High/High	6	5.5 mo

- Flow cytometry markers could better stratify CLL patients, especially in intermediate risk.

Giudice V, et al. Eur J Haematol. 2022 Nov;109(5):483-493.

Risk stratification and tailored therapies

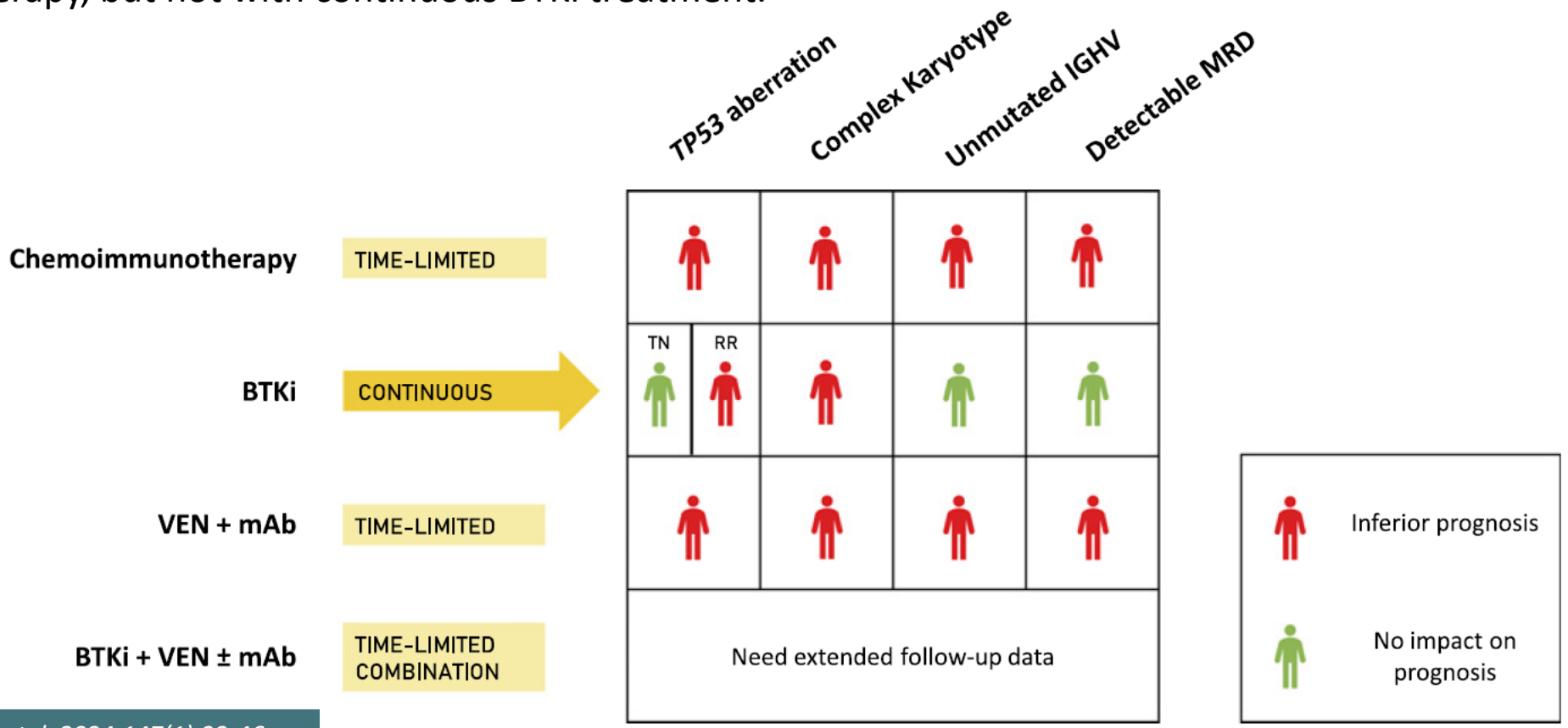
- Treatment for CLL has changed remarkably in the last decade.
- The mainstay of therapy used to be a combination of conventional chemotherapeutic agents plus a monoclonal antibody, such as rituximab or obinutuzumab.
- Novel, targeted agents are now the preferred option, such as the BTK inhibitor ibrutinib, the BCL2 inhibitor venetoclax the PI3K inhibitor idelalisib, and the second-generation BTK inhibitor acalabrutinib and PI3K inhibitor duvelisib.



Delgado J, et al. Haematologica. 2020 Sep 1;105(9):2205-2217.

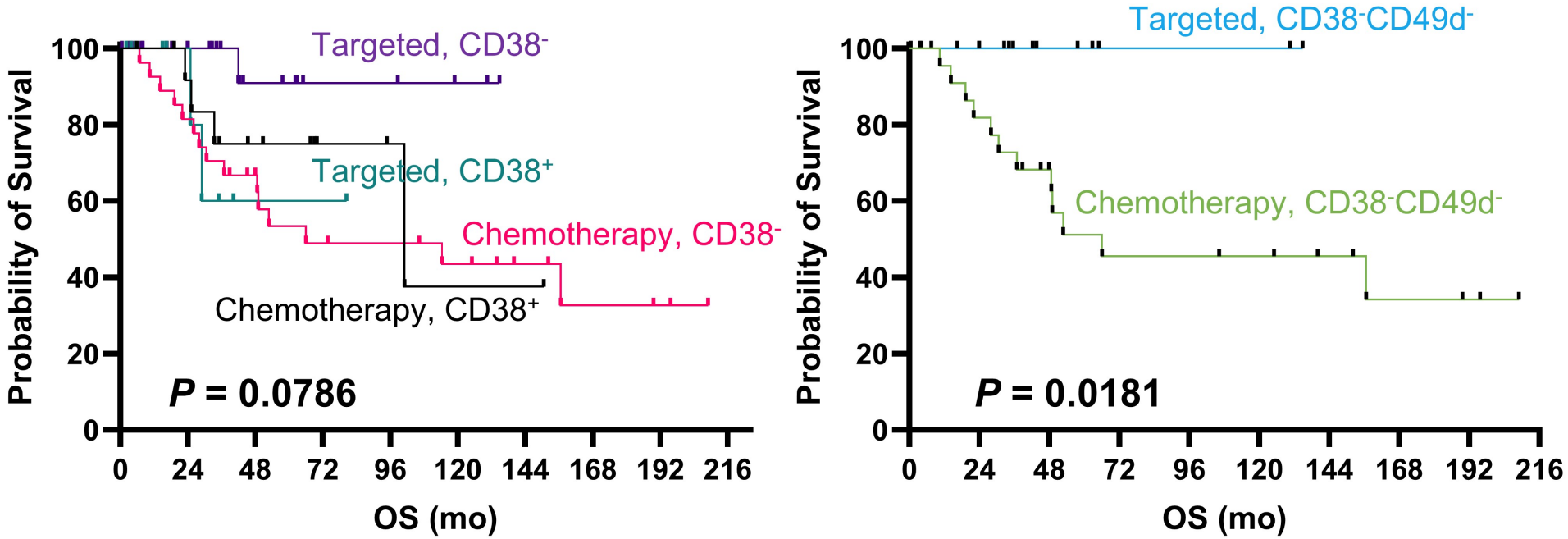
Risk stratification and tailored therapies

- Initial treatment with a BTKi can overcome the poor prognostic value of *TP53* aberration in TN CLL.
- *TP53* aberration continues to be a negative prognostic factor for RR CLL.
- IGHV mutation remains an important prognostic marker predictive of the durability of remission after time-limited targeted therapy, but not with continuous BTKi treatment.



Kang S, Ahn IE. Acta Haematol. 2024;147(1):33-46.

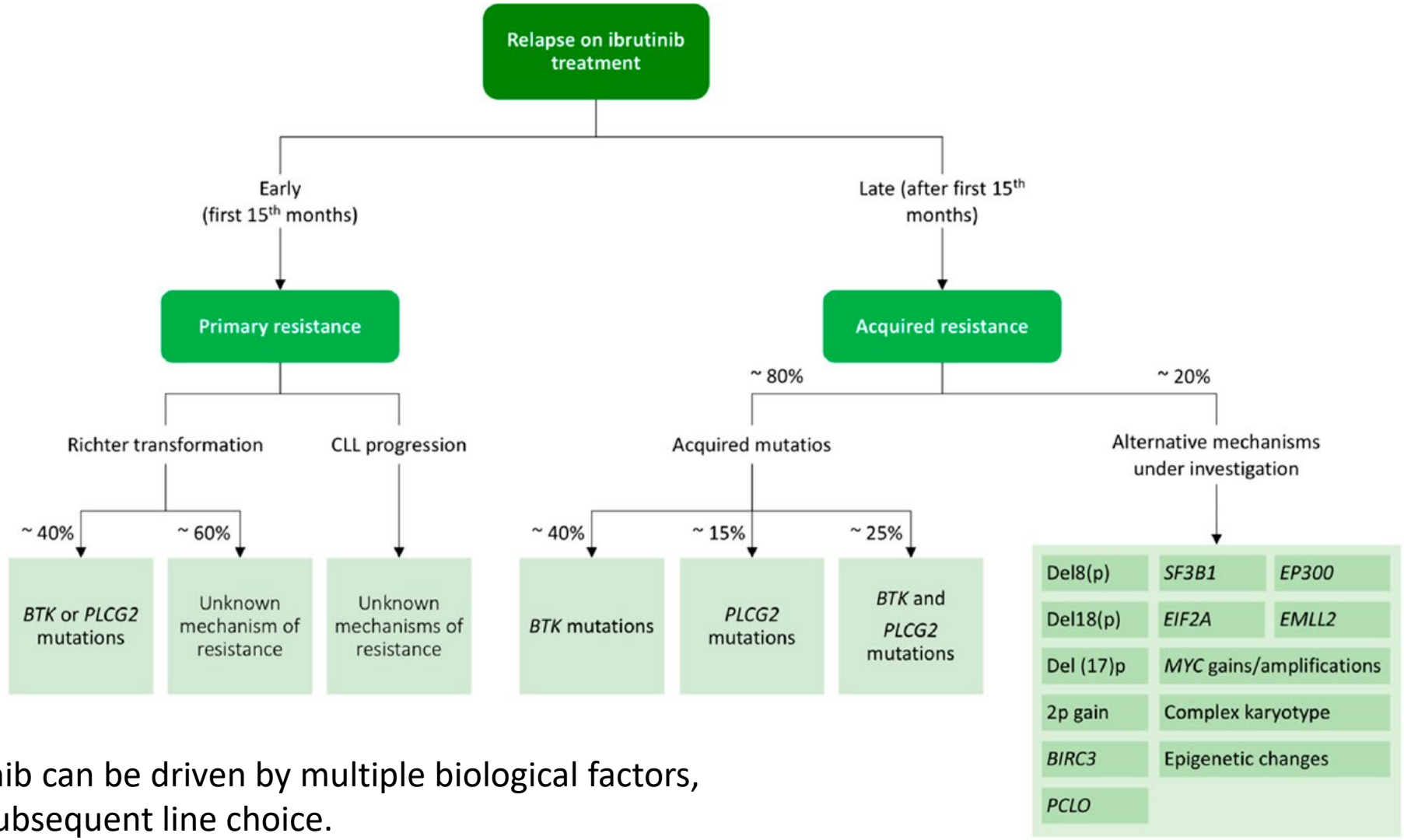
Risk stratification and tailored therapies: we are still missing flow cytometry



- Patients with negativity for CD38 and treated with targeted therapies show the highest benefit, as CD38- subjects treated with standard chemotherapy display the shortest OS (66 months).
- Co-expression of CD38 and CD49d is significantly related to shorter PFS in patients receiving BTKi or BCL2i, while targeted therapies showed an impressive protective effect in CD38-CD49d- CLL/SLL patients (5-year OS, 100% vs 46%; p=0.0181).

Mettivier L, et al. Frontiers in Oncology, under revision.

Treatment response and tailored therapies



- Resistance to ibrutinib can be driven by multiple biological factors, that can influence subsequent line choice.

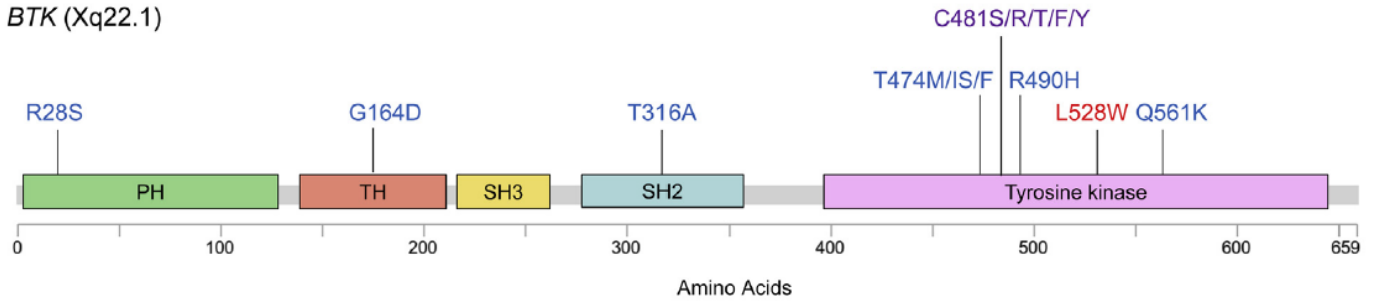
Pérez-Carretero Cet al. Diagnostics (Basel). 2021 May 10;11(5):853.

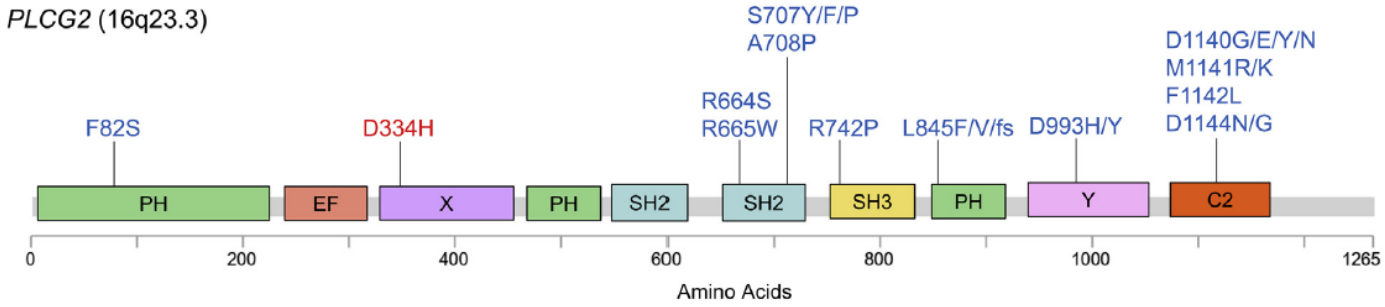
Biomarkers	Clinical Significance in Prognosis
Rai/Binet advance stage	Associated with unfavorable disease course. Not enough to predict disease progression.
β2M high (>3.5 mg/L)	Predicts worse outcome and short-term remission after fludarabine-based CIT. Included in different risk scoring systems.
CD49d expression	Predicts shorter survival and remains valid for predicting treatment-free survival after ibrutinib treatment [1].
IGHV unmutated	Associated with a shorter time to first treatment and poorer response to CIT. Its assessment is highly recommended in pre-treatment evaluation and only once since its status remains stable during disease course.
Del(11q)/*ATM* mutation	Associated with a shorter time to first treatment but better response to BTK inhibitors in the presence of del(11q) [1].
Del(17p)/*TP53* mutation	Confers resistance to CIT and predicts rapid disease progression. Its assessment is mandatory in pre-treatment evaluation.
Complex karyotype	Predicts unfavorable outcome after CIT independently of *TP53* alterations. Its role is controversial after novel targeted agents [1].
NOTCH1 mutation	Refines cytogenetic-risk stratification and is associated with worse outcome and poor response to rituximab treatment [1].
SF3B1 mutation	Refines cytogenetic-risk stratification and has been associated with poor prognostis [1].
BTK/PCLG2 mutation	Confers resistance to BTK inhibitors.
BCL2 mutation	Confers resistance to venetoclax.
MRD positive	Predicts shorter progression free-survival for CIT. Remains valid for venetoclax-based regimens [1].

Pérez-Carretero Cet al. Diagnostics (Basel). 2021 May 10;11(5):853.

Acquired mutations and ibrutinib resistance

- *BTK* C481S mutation prevents the drug from forming a covalent bond with the C481 residue.
- *BTK* T316A mutation functionally confers ibrutinib resistance.
- BTK mutations found in ~70% of CLL patients who progressed on ibrutinib treatment.
- Activating mutations in *PLCG2*, found in ~10% of the.
- PLCG2 mutations mostly missense mutations clustered in the Src homology 2 domain and the calcium-binding C2 domain.
- Complex karyotype, del(17p)/*TP53* mutation, and del(18p) at baseline before ibrutinib treatment are strongly associated with disease relapse.
- Minute CLL clones with BTK or PLCG2 mutations <10% of cancer cell fraction under ibrutinib.





Associated with CLL progression
Associated with Richter transformation
Associated with both CLL and Richter transformation

Time in clinical course	Clinical needs	Predictive markers
Before treatment	To assess risk of disease progression	Complex cytogenetics Del(17p) TP53 Del(18p)
During treatment	To monitor molecular relapse	Small *BTK* or *PLCG2* mutant clones
After relapse	To confirm drug resistance	*BTK* C481 or other mutations *PLCG2* mutations

Lee J, Wang YL. et al. J Mol Diagn. 2020 Sep;22(9):1114-1125.

Acquired mutations and ibrutinib/venetoclax resistance

	Ibrutinib		Venetoclax
Mutation Type	***BTK***	***PLCG2***	***BCL-2***
Prevalence in relapsed patients	57%	13%	47%
Mechanism	Loss of covalent binding of ibrutinib to BTK	Activating BCR signaling independent of BTK	Disruption of the bond of venetoclax to BCL-2
Variants			
More frequent	C481S	Different subclones coexist with low allelic burden	G101V (subclonal)
Others	C481R, C281F, C481Y, R28S, G164D, T316A, T474I/S, R490H, Q516K, L528W, V537I	F82S, P664S, R665W, S707Y, S707P, S707F, L845F, L845V, L845G, L848R, D993Y, D993H, D1140N, M1141K, M1141R, S1192G	D103Y, A103T, A103G, A103V, A113G, A129L, V156A
Median time since drug exposure	34.3 months (14–76.8)	35.1 months (17.4–64.6)	36 months (6.5–73)

Pérez-Carretero Cet al. Diagnostics (Basel). 2021 May 10;11(5):853.

MRD by flow cytometry: where are we

Recommended	Rationale
Measurable residual disease (MRD)	Replaces "minimal" residual disease as a more objective term
Undetectable-MRD (U-MRD)	As a general term, replaces MRD negative or MRD- as a more accurate term in cases where MRD threshold is not specified
MRD4, MRD5, etc.	Specifies upper limit of disease (e.g., MRD4 denotes <0.01%/<10^{-4} disease, MRD5 < 0.001%/<10^{-5} disease, etc) for an individual sample or for a group of patients in clinical trial reporting
Detectable (d) or undetectable (u) within an MRD category	Detectable = residual disease is below the stated threshold but measurable above the next MRD threshold. Undetectable = residual disease is not detectable, but the assay/sample is not suitable for detection of disease at the next threshold MRD4d: < 0.01%/10^{-4} but ≥0.001%/10^{-5} MRD4u: < 0.01%, assay limit of detection does not reach 0.001%/10^{-5}
Always report assay method (e.g., Flow) and analysis technique (e.g., ERIC-FC)	Results may differ by assay method even for assays with identical sensitivity
Always report tissue assayed (e.g., PB, BM)	MRD may differ in different tissues from the same patient/timepoint
In clinical trials, always report MRD rate as percentage U-MRD in ITT population	Avoids confusion with the rate in the MRD-tested population, e.g., MRD4 rate = number of patients with <0.01% MRD as a percentage of the ITT population

BM bone marrow, *CLL* chronic lymphocytic leukemia, *Flow* flow cytometry, *ITT* intention-to-treat, *PB* peripheral blood.

Wierda GW et al. Leukemia 2021

ERIC/ESCCA recommendations

Cytometry Part B (Clinical Cytometry) 94B:121–128 (2018)

Original Article

Reproducible Diagnosis of Chronic Lymphocytic Leukemia by Flow Cytometry: An European Research Initiative on CLL (ERIC) & European Society for Clinical Cell Analysis (ESCCA) Harmonisation Project

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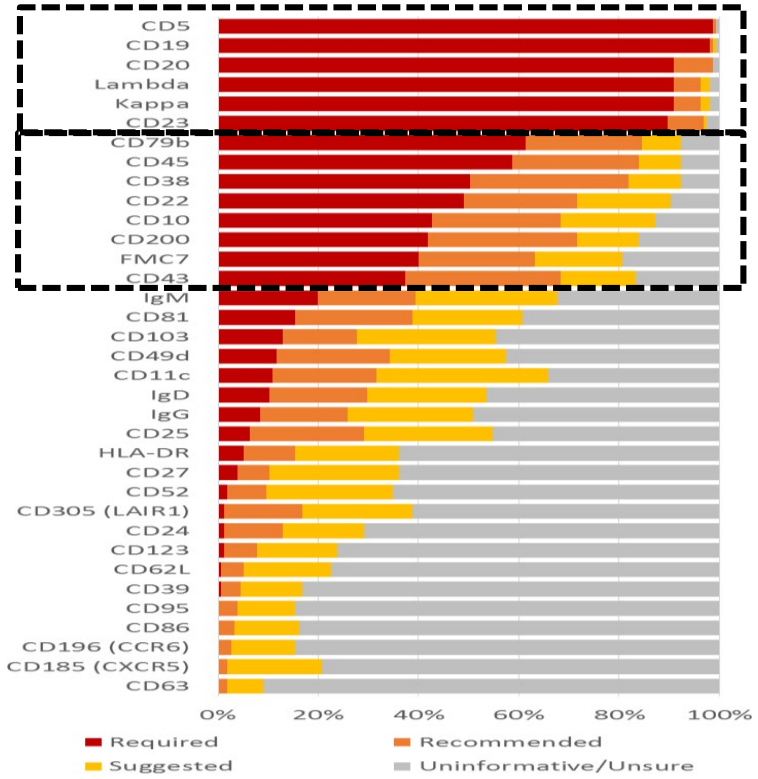


"required": *CD5, CD19, CD20, CD23, κ/λ*
(>75% of respondents)

or "recommended": *CD10, CD43, CD79b, CD200*
(>50% of respondents)

- **ROR1 and CD200 or CD3 are recommended**
  - Facilitate analysis for less experienced operators
  - Permit automated analysis
  - Increase proportion of "atypical" cases that can be monitored
- **Pre-treatment analysis** is required to exclude atypical cases
- **Clonality assessment is recommended**

ESCCA European Society for Clinical Cell Analysis

ERIC european research initiative on CLL

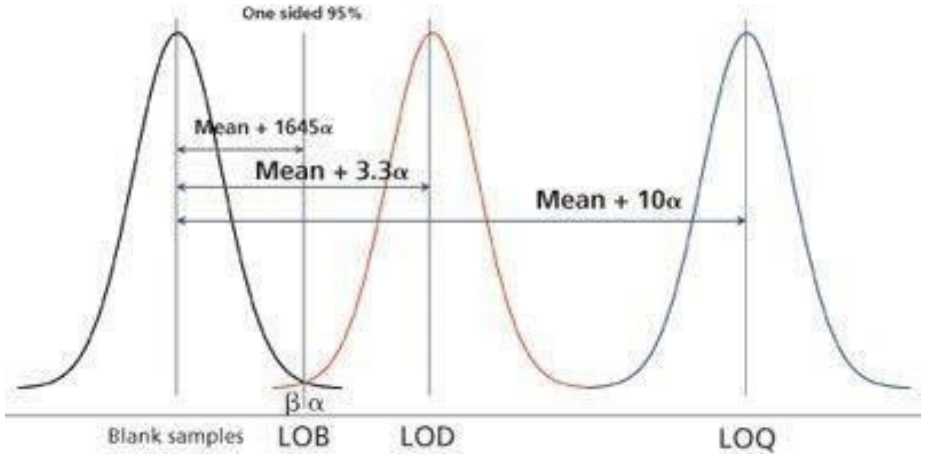
LOB/LOD/LOQ for MRD detection

- **Limit of Blank (LOB)** → highest signal in the absence of measure, calculated as mean (blank) + 1.645 SD
- **Limit of Detection (LOD)** → level at which 95% of samples with low level are detected above the limit of blank, calculated as LOB + 1.645 SD
- **Limit of Quantitation (LOQ)** → lowest level that can be reliably detected and whose total error (bias + imprecision) meets a desired criterion for accuracy (clinical utility)

ERIC specification for the detection of CLL cells in a normal background

The minimum population size for reproducible **detection** of CLL cells in a multiparameter analysis → **20 events**

The minimum population size for reproducible **quantification** of CLL cells in multiparameter analysis → **50 events**



LOQ = 100 * **50** / numero totale di leucociti

LOD = 100 * **20** / numero totale di leucociti

LOB = **< 10** Eventi nei campioni normali
LOD = **> 20** Eventi rilevati nel campione
LOQ = **> 50** Eventi rilevati nel campione
20 e 50 eventi derivano da stime statistiche

Silvia Heltai (IRCCS Ospedale San Raffaele, Milano)

Cell number required for MRD detection

MRD classification	Neoplastic cells / total normal cells	Neoplastic cells % of total cells	Scientific notation	Cell required for flow cytometry	Cells (DNA) required for molecular analysis
MRD3	<1/ thousand	<0.1%	10E-3 (10^{-3})	>20 thousand	>3 thousand (0.02µg DNA)
MRD4	<1/ 10 thousand	<0.01%	10E-4 (10^{-4})	>200 thousand	>30 thousand (0.2µg DNA)
MRD5	<1/ 100 thousand	<0.001%	10E-5 (10^{-5})	>2 million	>300 thousand (2µg DNA)
MRD6	<1/ million	<0.0001%	10E-6 (10^{-6})	>20 million	>3 million (20µg DNA)
MRD7	<1/ 10 million	<0.00001%	10E-7 (10^{-7})	>200 million	>30 million (120µg DNA)

Courtesy of Andy Rawstron

Conclusions and future directions

- CLL is an extremely heterogeneous disease, and risk stratification and prognostication are performed using the CLL-IPI that combines clinical, chromosomal, molecular alterations, and laboratory findings: age; disease stage; β2-microglobulin levels; presence of del(17p); and IGHV mutational status.
- Unmutated CLL has shorter survival and remission duration compared to M-CLL, as well as patients with del(17p)/*TP53* mutations, even though clinical management of these subjects has markedly improved after the introduction of targeted therapies, such as ibrutinib, idelalisib, and venetoclax.
- Point mutations detection could greatly improve clinical management of those patients who develop BTKi resistance, as well as 11q deletion, *TP53* abnormalities, *IGHV* mutational status, complex karyotype and *NOTCH1* mutations.
- However, these systems require integration with other molecular and phenotypical markers, as shown using the IPS-E and ISS systems. Although individual factors can be a very important prognostic tool, reality is more complex, as each patient may harbor several biomarkers with different prognostic value.
- MRD by flow cytometry is still challenging in routinely clinical practice.