XVIII IWCLL

International Workshop on CLL

20-23 SEPTEMBER 2019 EDINBURGH

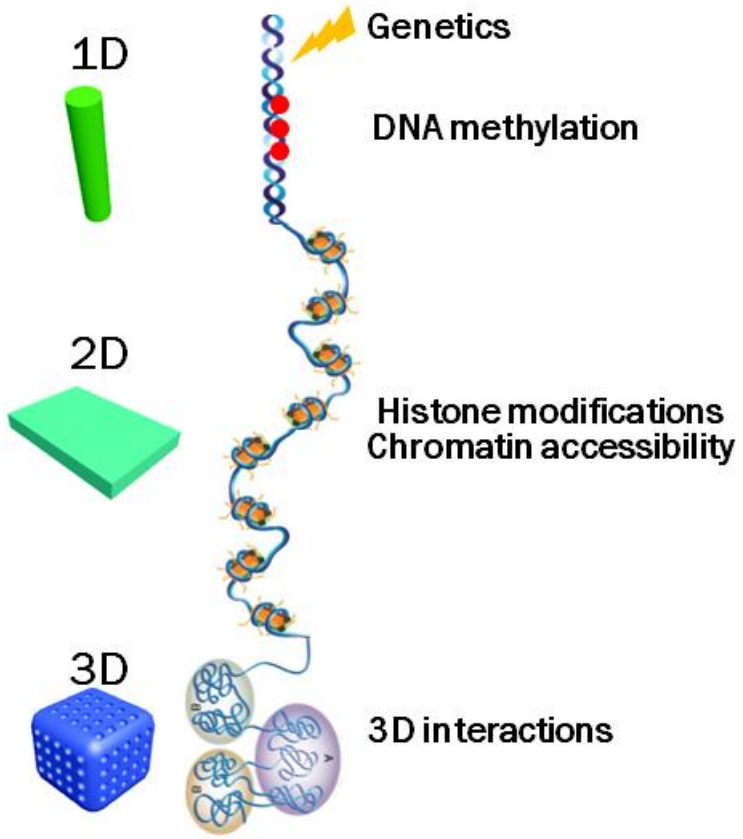
Insights from Tertiary Structure, Chromatin Modifications, and Nuclear Organization

Iñaki Martin-Subero

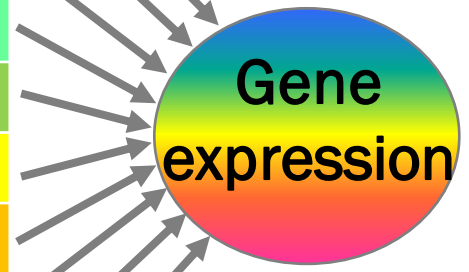
ICREA, IDIBAPS, UB

Barcelona, Spain

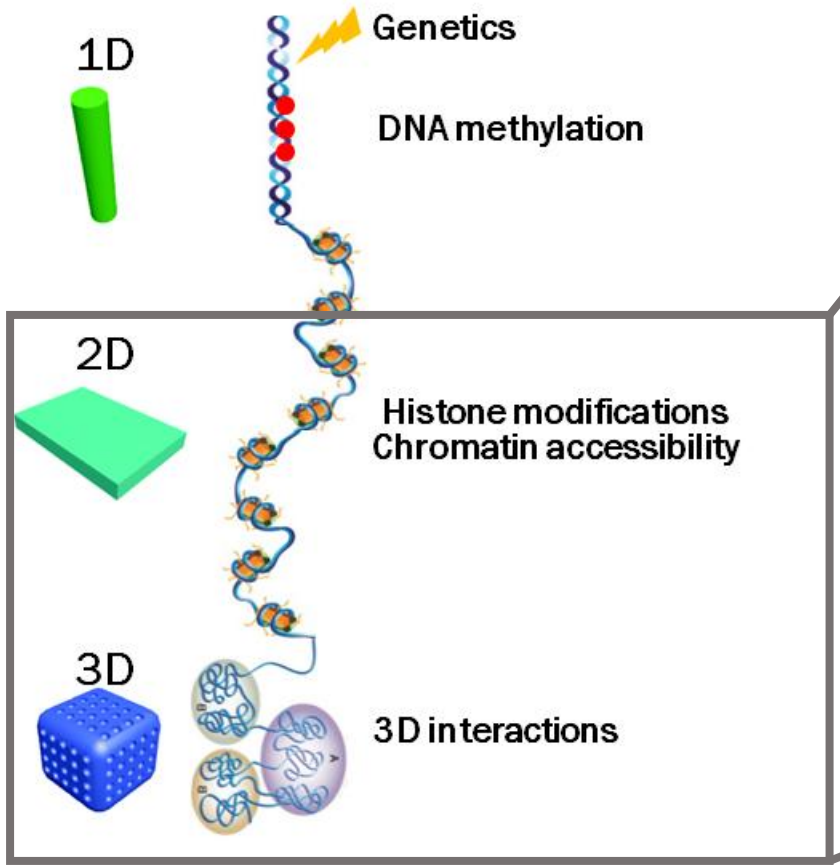
Epigenetic information controls gene expression



LAYER	FUNCTION
Genome	Information storage
DNA methylation	Context dependent
H3K4me3	Promoters
H3K4me1	Enhancers
H3K27ac	Active reg. element
H3K36me3	Txn. Elongation
H3K27me3	Polycomb repression
H3K9me3	Heterochromatin
Chr. accessibility	TF binding
3D interactions	Distant regulation



Three epi & genetic stories

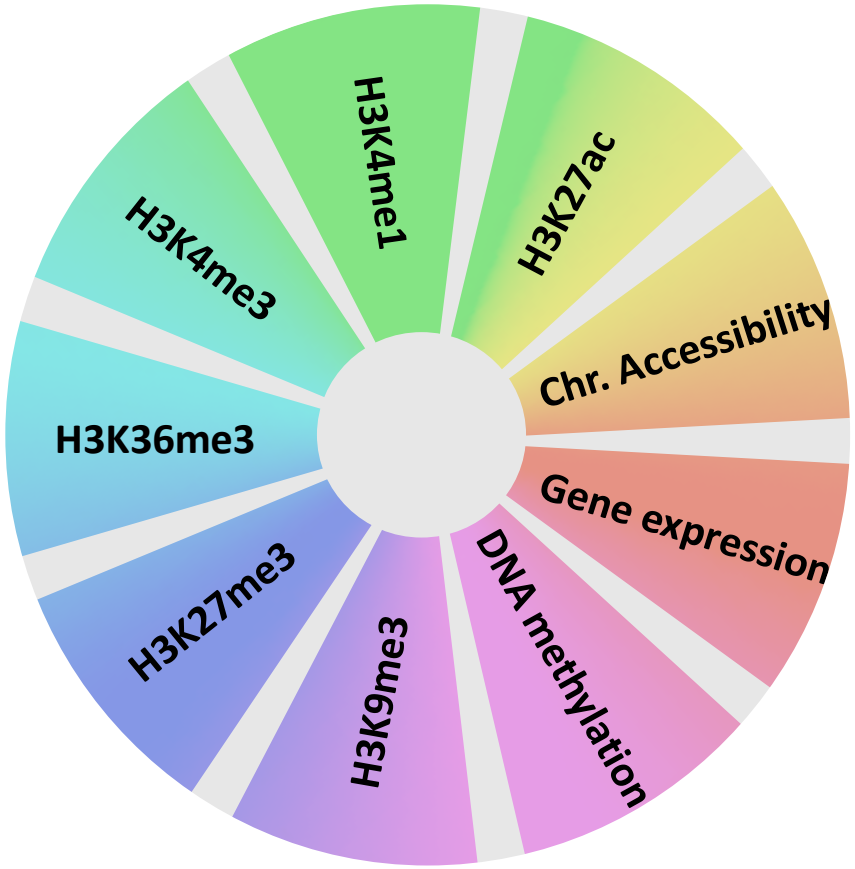


Story 1: Reference epigenomes
Alterations in the chromatin landscape

Story 2: Genetic predisposition and chromatin
Linking risk-loci to chromatin function

Story 3: 3D genome stucture
Broad changes in 3D interactions

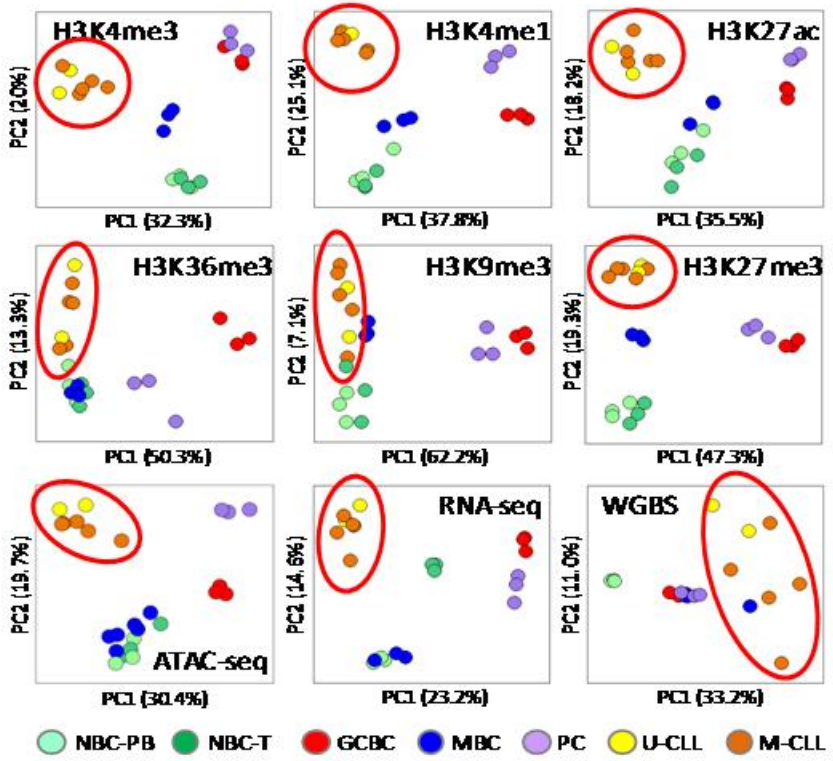
Story 1: Reference epigenomes of CLL

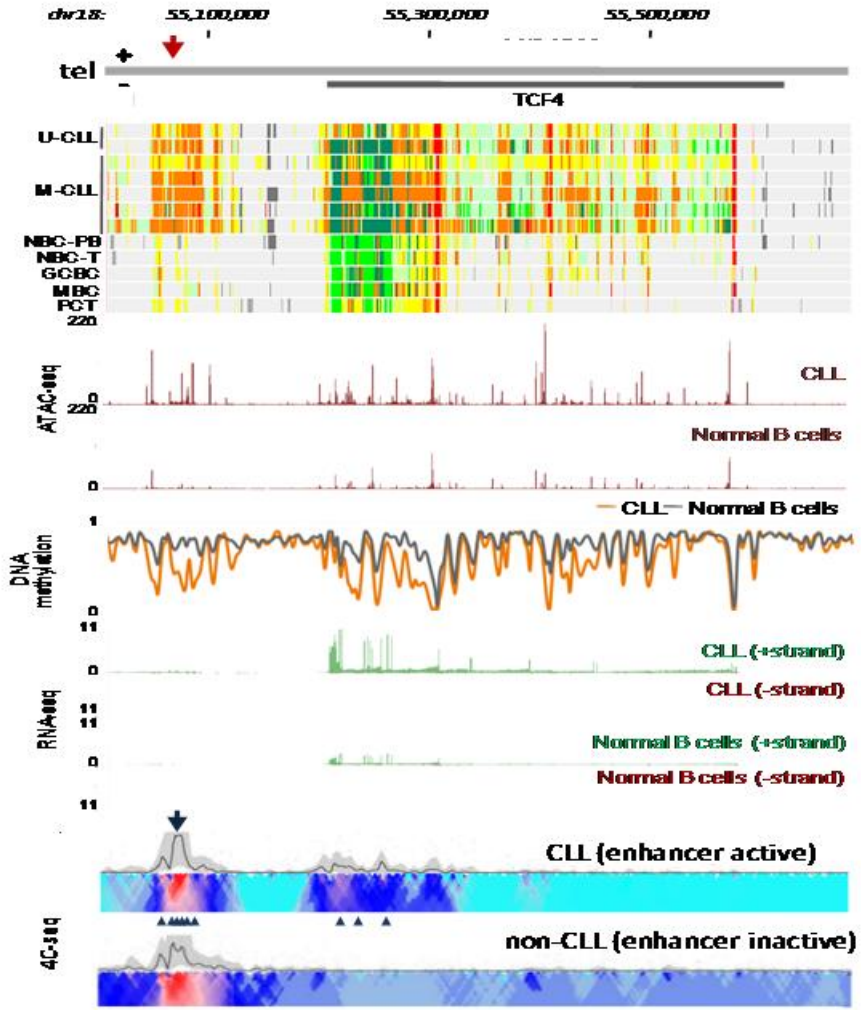


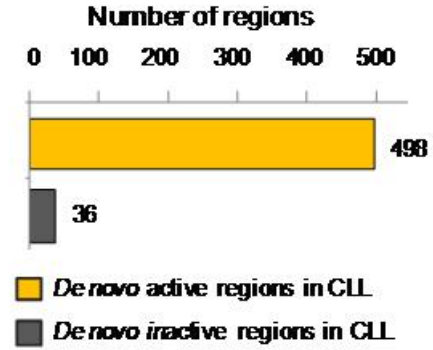
Naive B cells		Germinal center B cells	Memory B cells	Plasma cells	CLL with unmutated IGVH	CLL with mutated IGVH
Reference epigenomes						
3	3	3	3	3	2	5
Extended series ATAC-seq & H3K27ac						
					38	66

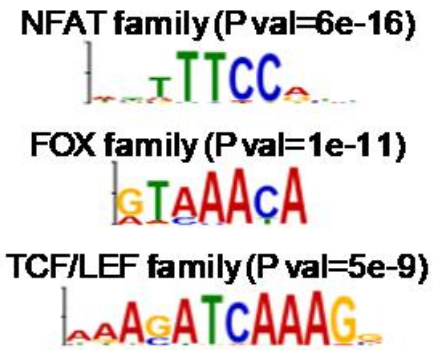
Beekman et al., Nat Med 2018

Story 1: De novo chromatin activation in CLL



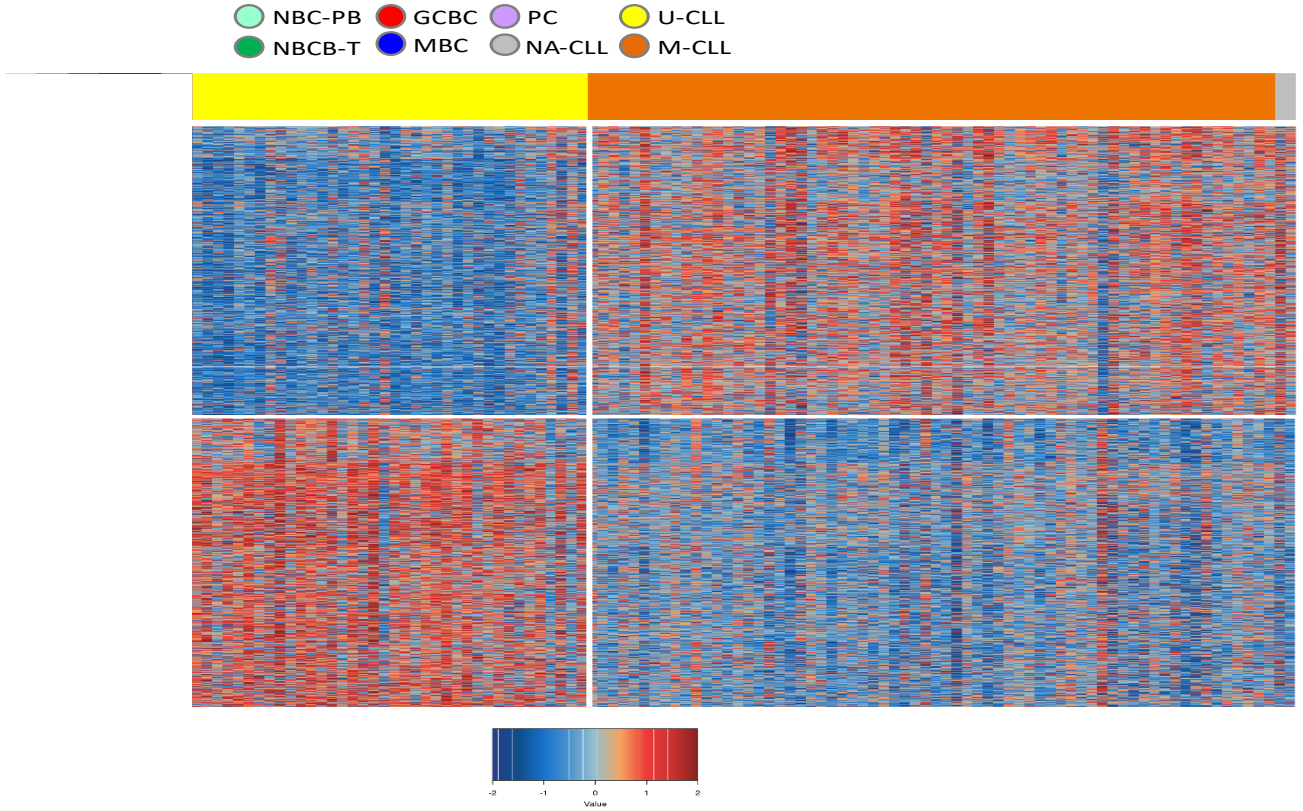






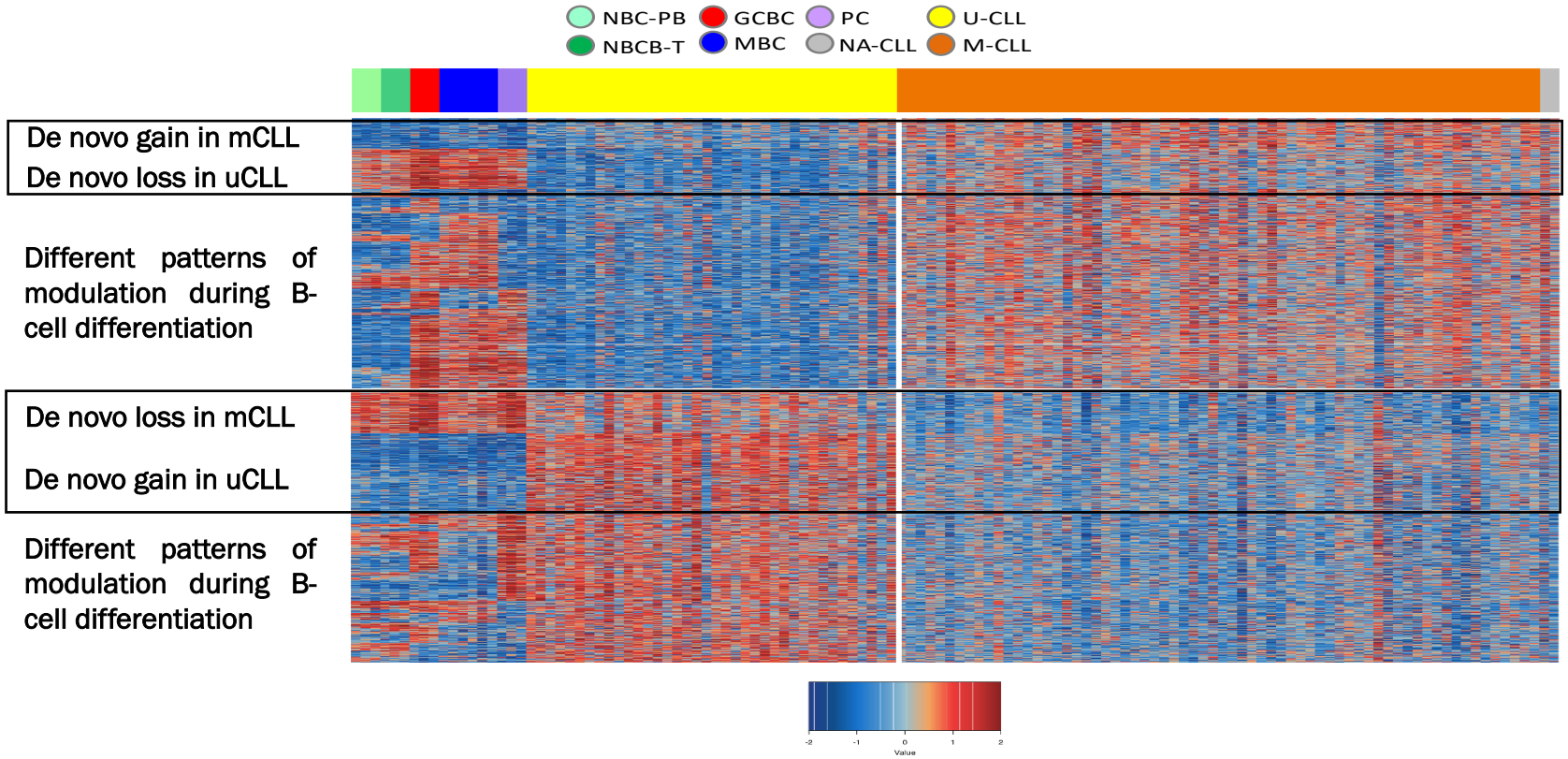
Beekman et al., Nat Med 2018

Story 1: Different chromatin landscapes in uCLL and mCLL



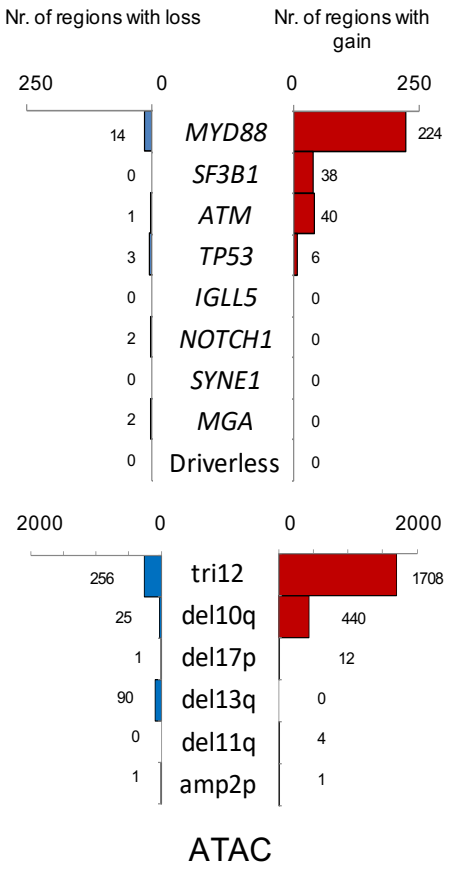
Beekman et al., Nat Med 2018

Story 1: Different chromatin landscapes in uCLL and mCLL

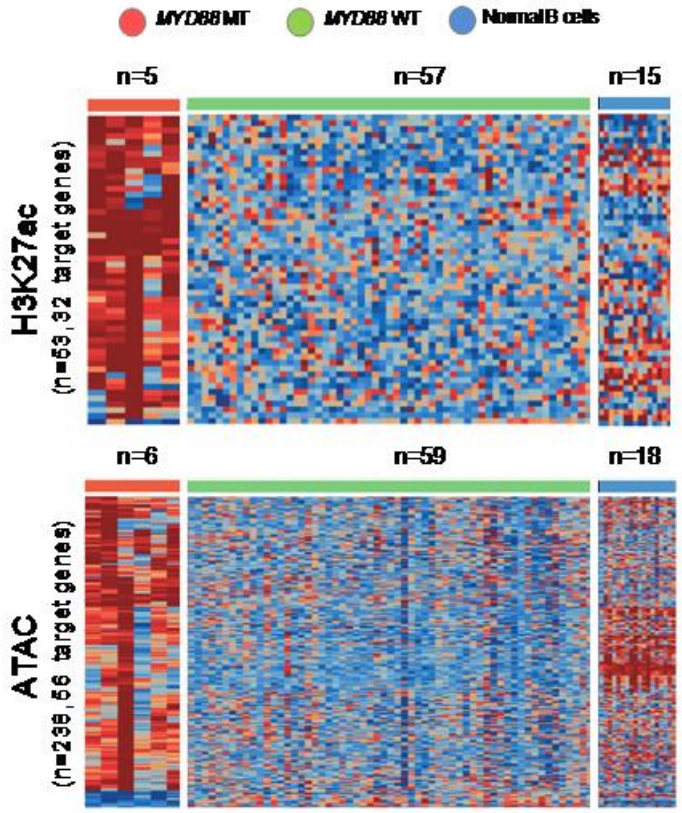


Beekman et al., Nat Med 2018

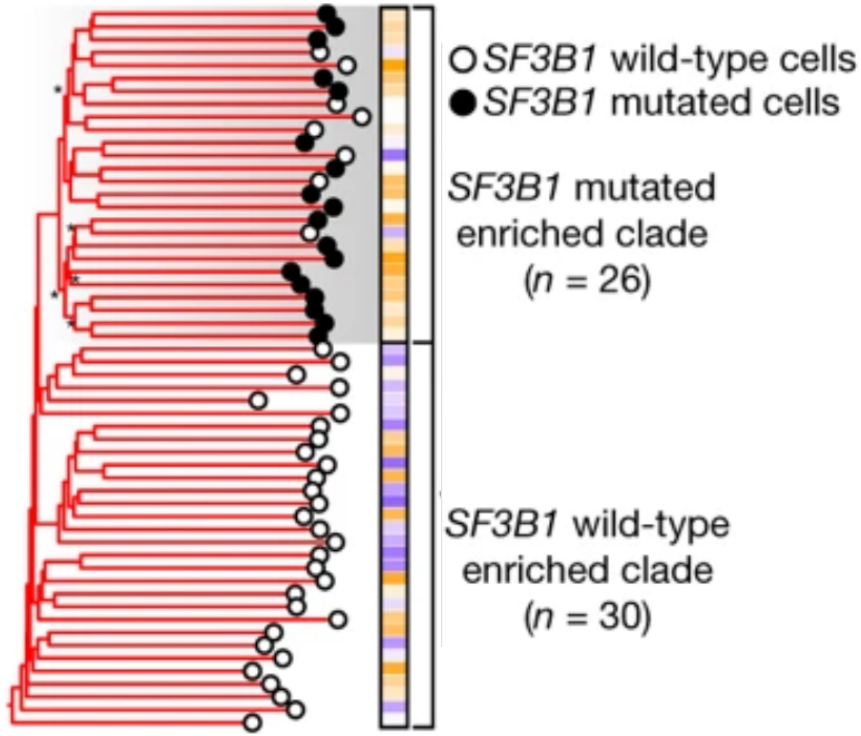
Story 1: Association between genetic changes and chromatin



ATAC



Beekman et al., Nat Med 2018



Gaiti et al., Nature 2019

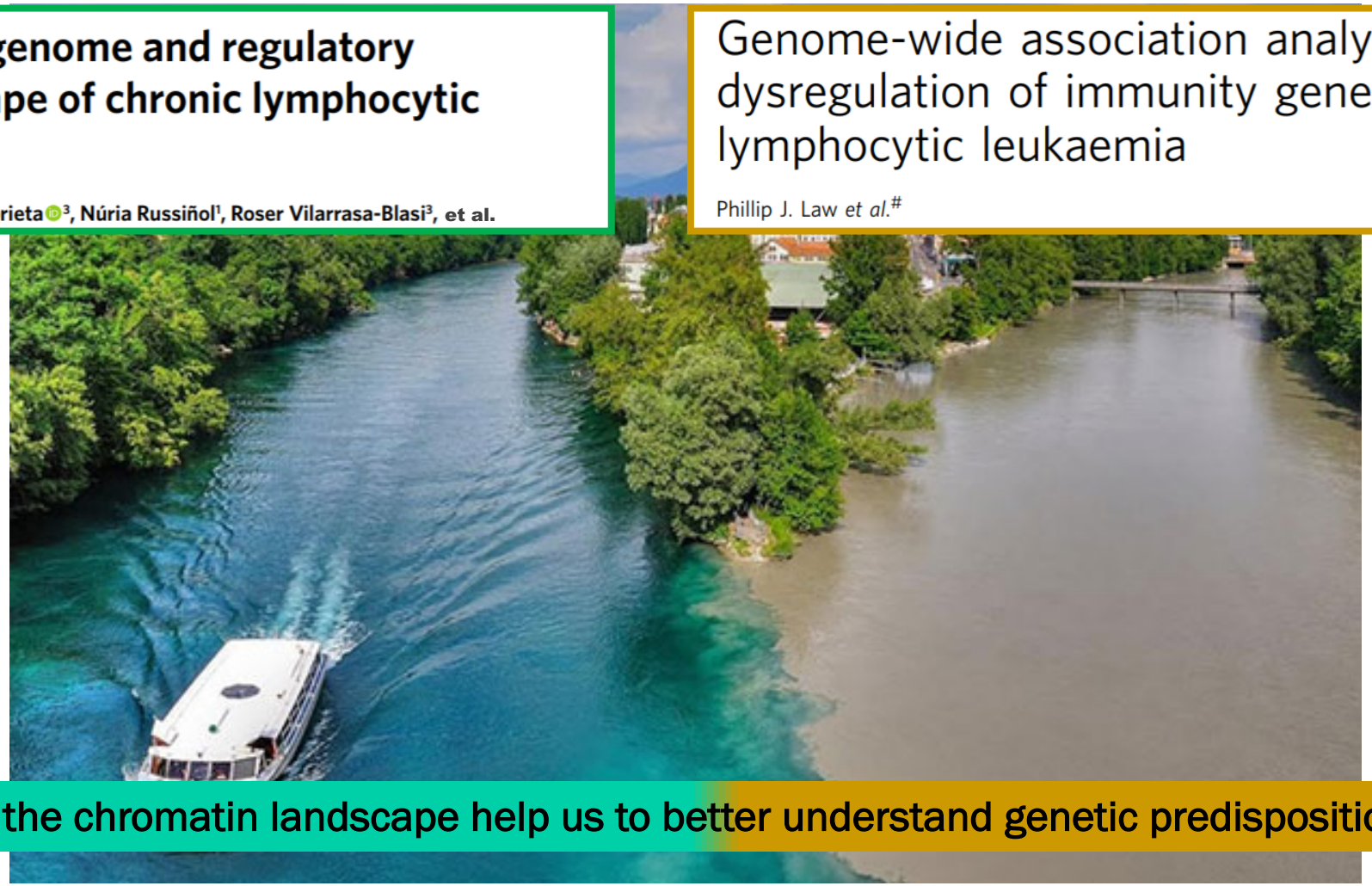
Story 2: Genetic predisposition and chromatin

The reference epigenome and regulatory chromatin landscape of chronic lymphocytic leukemia

Renée Beekman[1,2], Vicente Chapaprieta[3], Núria Russiñol[1], Roser Vilarrasa-Blasi[3], et al.

Genome-wide association analysis implicates dysregulation of immunity genes in chronic lymphocytic leukaemia

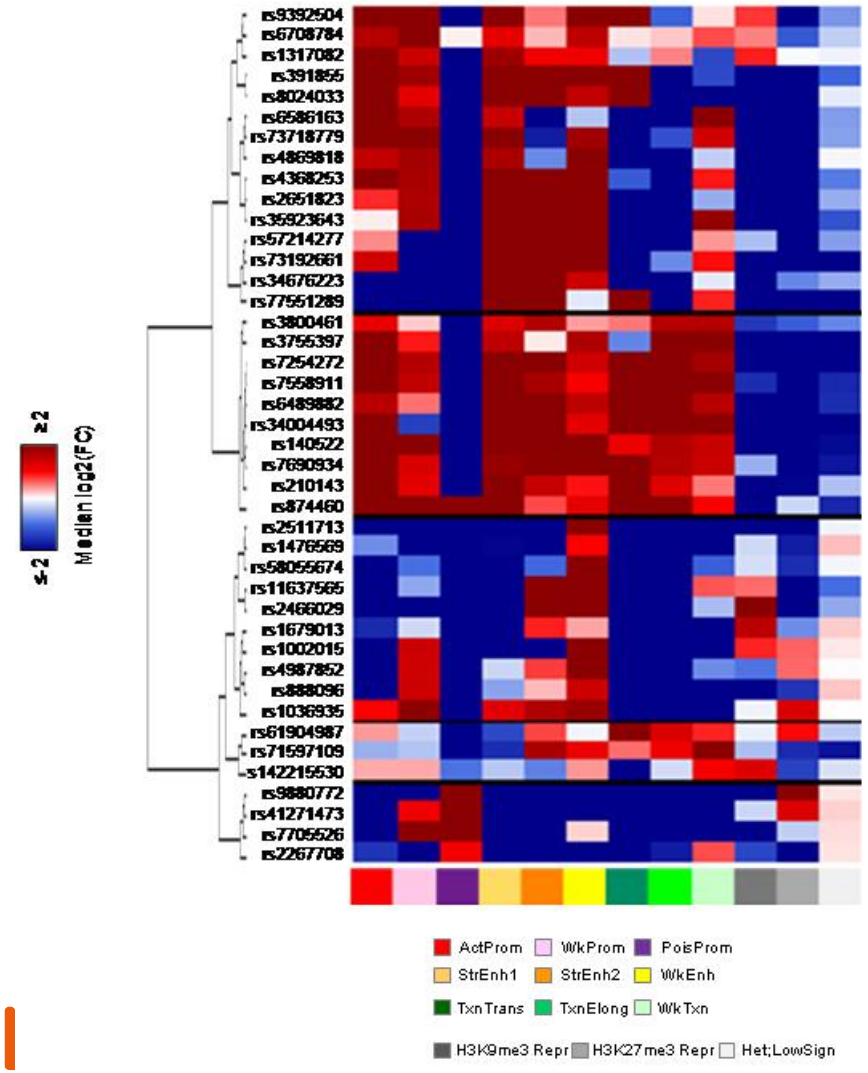
Phillip J. Law *et al.*#

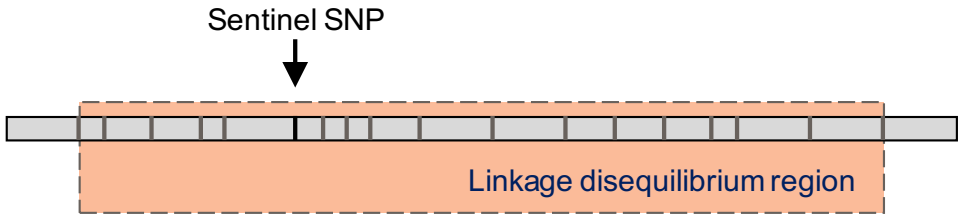


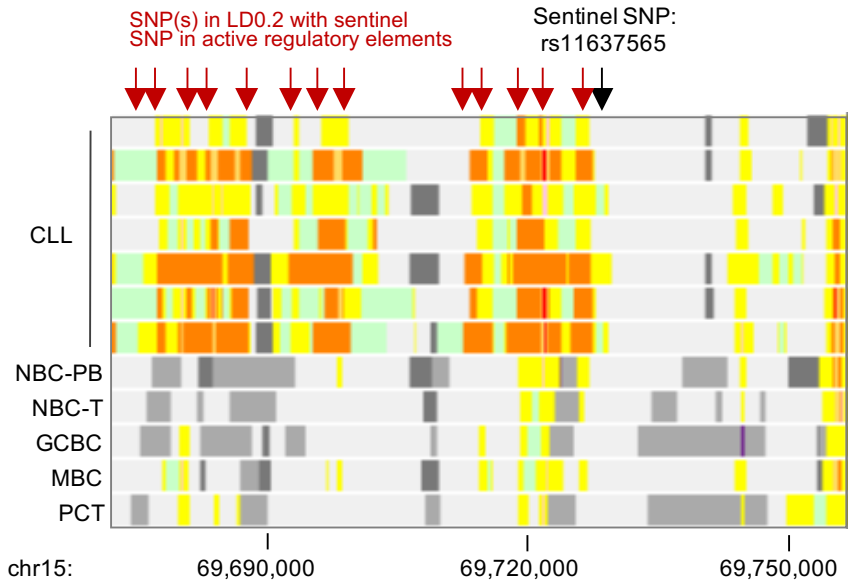
Richard Houlston

Can the chromatin landscape help us to better understand genetic predisposition?

Story 2: Most risk-loci are located in regulatory elements







Speedy & Beekman et al., Nat Comms 2019

Story 2: Target genes & altered TF binding affinity

SNPs (n) — CLL cases (n)

4	Directly genotyped dataset (SNP6.0 array)	502

Quality controls:
14 out of 502 cases failing QC excluded

Recover 33 out of 38 untyped SNPs by imputation:
5 out of 38 SNPs with info score < 0.85 excluded

37	Directly genotyped and imputed dataset (SNP6.0 array)	488

Use WGS data of 146 out of 488 cases to check fidelity of imputation:
1 case with discordance > 10% excluded, >99% concordance across all other genotypes

Complete missing genotypes for remaining 145 out of 487 cases with WGS data:
Including genotypes of 5 SNPs with imputation info score < 0.85

42	Final dataset for QTL analysis	487

ATAC-seq	H3K27ac	DNA methylation	Gene expression
99 cases	97 cases	486 cases	452 cases
30 QTLs	19 QTLs	61 QTLs	36 QTLs
14 risk loci	9 risk loci	28 risk loci	23 risk loci

QTL analysis

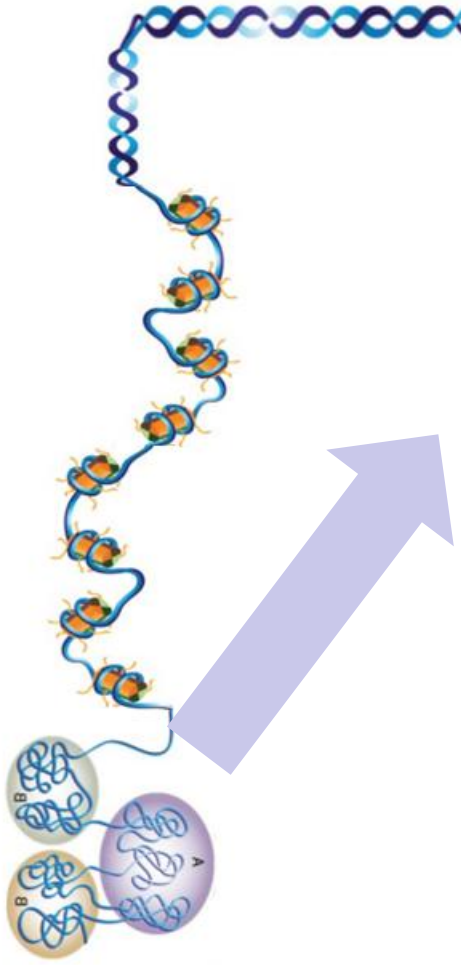
SNP	Position	ATAC-seq	H3K27ac	DNA methylation	Gene expression	Candidate target genes
rs35923643	chr11:123484683	■	■	■	■	*GRAMD1B*
rs6489882	chr12:112943571	■	■	■	■	*OAS1, OAS3*
rs11637565	chr15:69728186	■	■	■	■	*TLE3*
rs2511713	chr8:102565637	■	■		■	*UBR5*
rs7558911	chr2:201159226	■		■	■	*CASP8,CFLAR, PPIL3*
rs7690934	chr4:108104709	■		■	■	*LEF1*
rs1679013	chr9:22206988	■		■	■	*DMRTA1*
rs8024033	chr15:40111456	■		■	■	*BMF*
rs391855	chr16:85895015	■		■	■	*IRF8*
rs3755397	chr2:241355498		■	■	■	*SEPT2, ING5, MTERF4*
rs140522	chr22:50532837		■	■	■	*CHKB-CPT1B, CPT1B, ODF3B, TYMP*
rs2466029	chr8:127188726	■	■			
rs1002015	chr2:110859042	■		■		
rs210143	chr6:33579153	■		■		
rs142215530	chr15:56485493	■		■		
rs1036935	chr18:50317164		■	■		
rs41271473	chr1:228744549			■	■	*CCSAP*
rs6708784	chr2:111169802			■	■	*BCL2L11*
rs1317082	chr3:169779797			■	■	*MYNN*
rs3800461	chr6:34648545			■	■	*C6orf106, SNRPC, TAF11*
rs4869818	chr6:154150090			■	■	*IPCEF1*
rs6586163	chr10:88992261			■	■	*ACTA2, FAS*
rs2651823	chr11:2300420			■	■	*C11orf21, TSPAN32*
rs7254272	chr19:4069121			■	■	*TBXA2R*
rs57214277	chr4:184333619	■				
rs71597109	chr4:101819845		■			
rs9880772	chr3:27736288			■		
rs7705526	chr5:1285859			■		
rs9392504	chr6:412802			■		
rs73718779	chr6:2969044			■		
rs2267708	chr7:124752458			■		
rs4368253	chr18:59955055			■		
rs34004493	chr2:230289297				■	*SP140, SP140L*
rs73192661	chr3:188411006				■	*LPP*
rs77551289	chr18:63121512				■	*VPS4B*
rs874460	chr19:46673495				■	*FKRP, PRKD2*

Potential functional SNP (sentinel SNP)	Transcription factor	Risk allele	Binding affinity risk allele	Motif
rs210142 (rs210143)	SPI1	C	Decreased	
rs12531150 (rs142215530)	PAX5	T	Decreased	
rs4767033 (rs6489882)	MEF2A	T	Decreased	
rs919054 (rs11637585)	TCF3	G	Increased	
rs7701411 (rs4869818)	NFATC1	T	Increased	
rs9383367 (rs4368253)	FOXH1	G	Increased	

Speedy & Beekman et al., Nat Comms 2019

Story 3: From 1D to 3D genomes in CLL

Huge impact on our understanding and the clinical management of hematological malignancies



Genetic changes

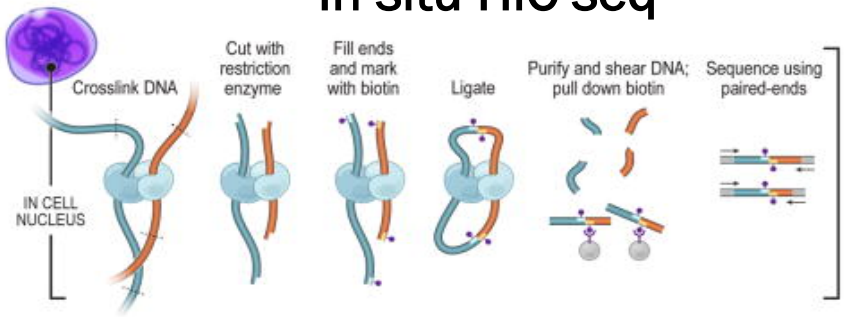
Gene expression changes

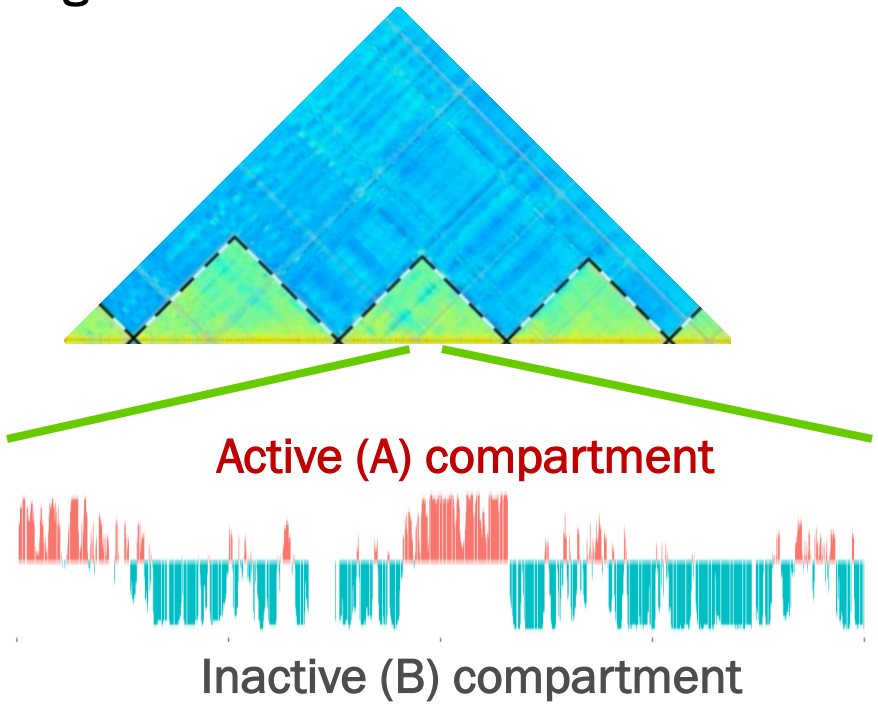
DNA methylation changes

Active (A) compartment

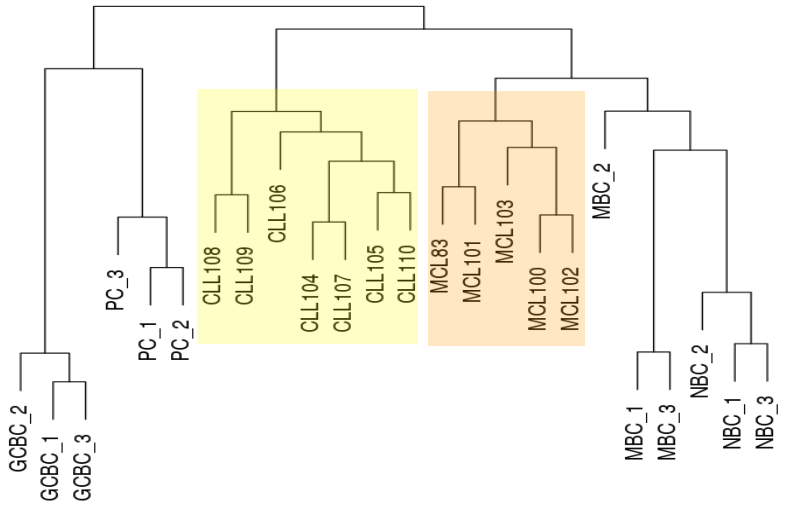
Inactive (B) compartment

In situ HiC-seq





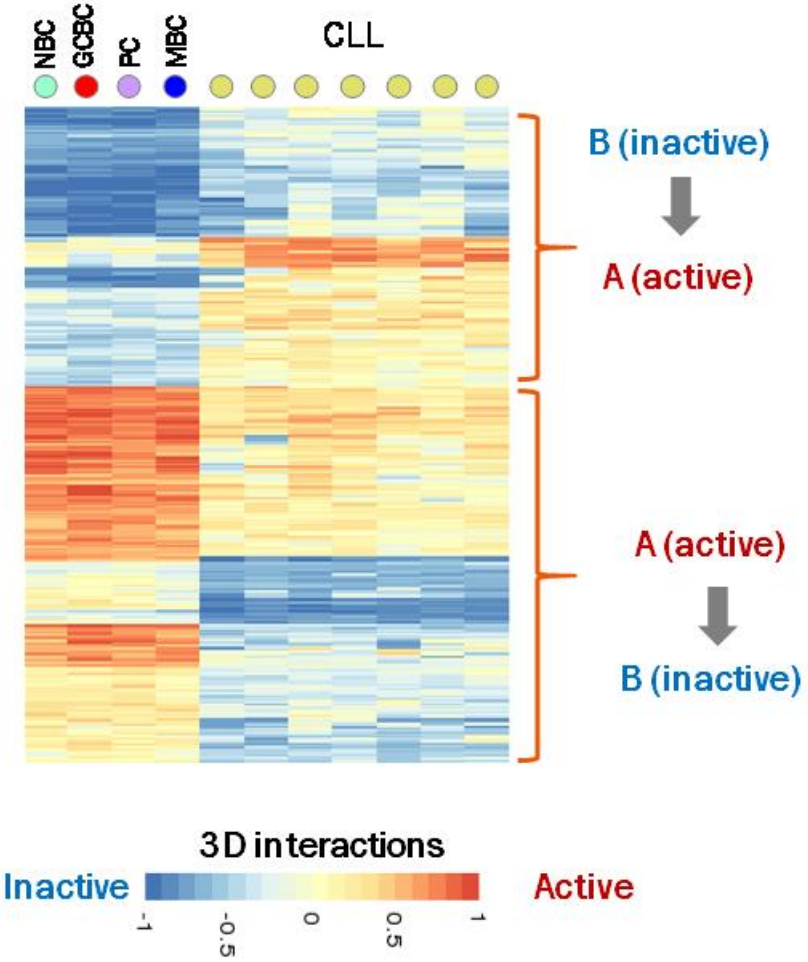
Story 3: Changes in the 3D genome structure



- The 3D genome structure changes during cell differentiation and is maturation stage-specific
- CLLs and MCLs cluster are in the same cluster as NBCs and MBCs
- CLLs and MCL show differential 3D structures

28% of the 3D genome changes during B-cell differentiation

72% is stable in normal B cells

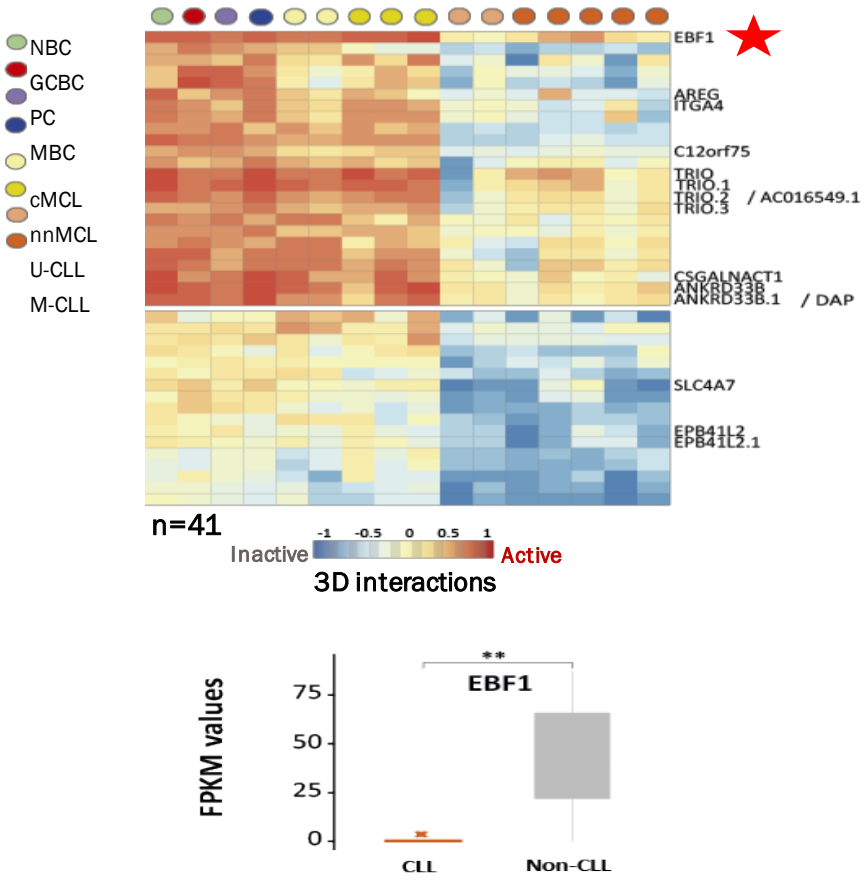


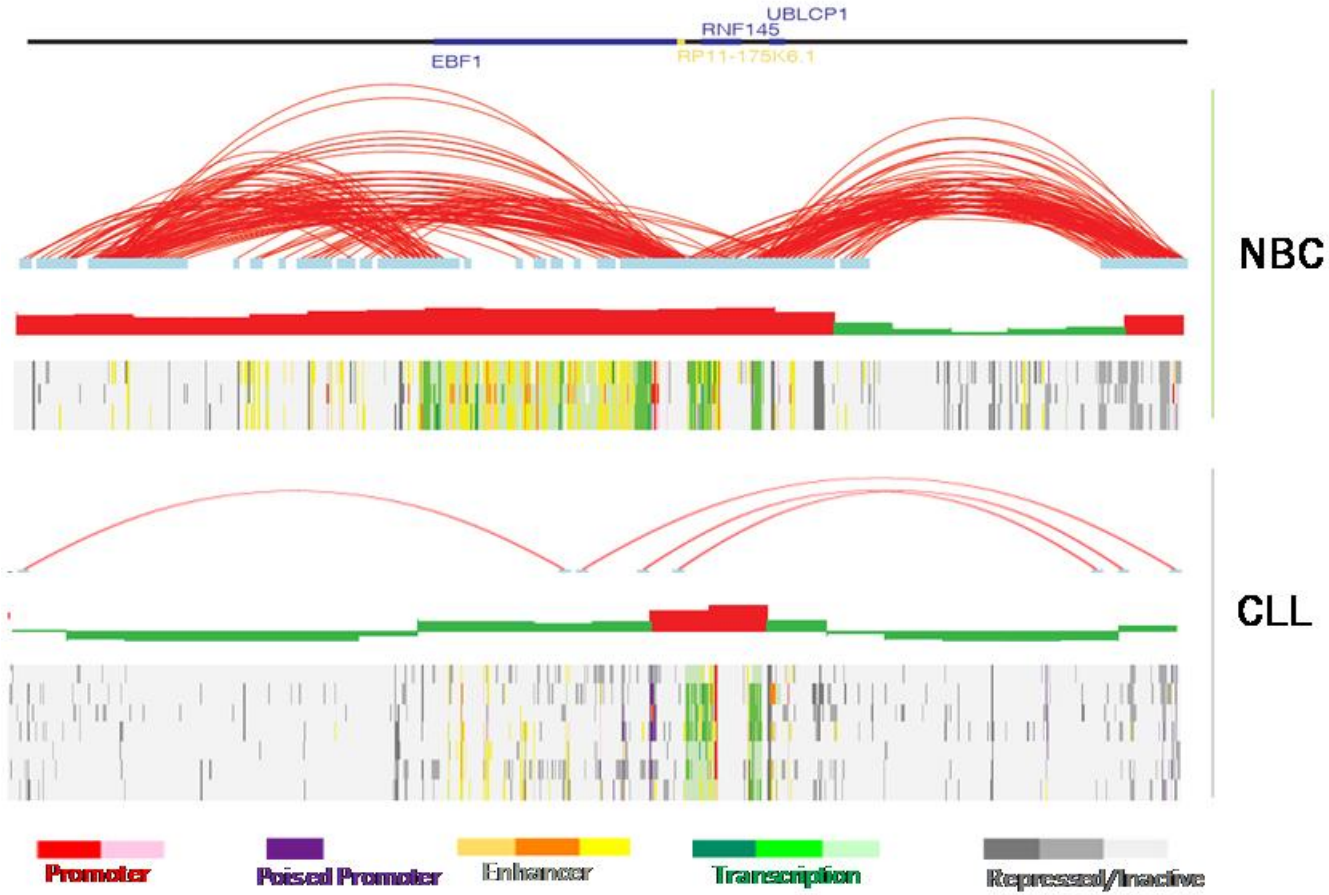
Can we detect changes in the 3D genome structure of CLL?

XVIII IWCLL
International Workshop on CLL
20-23 SEPTEMBER 2019 EDINBURGH

Vilarrasa-Blasi & Soler-Vila et al., submitted
https://www.biorxiv.org/content/10.1101/764910v2

Story 3: CLL-specific loss of 3D interactions





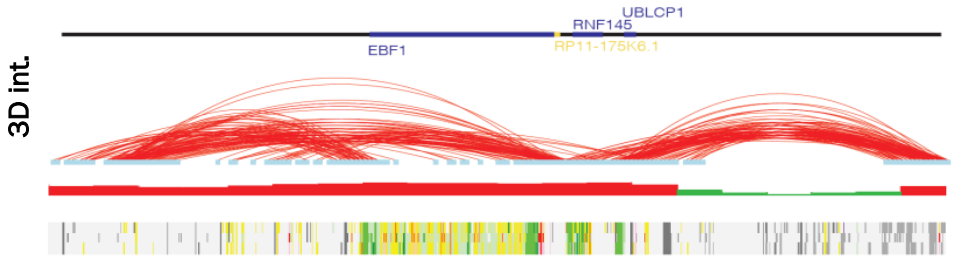
How is *EBF1* structurally organized into the nucleus?

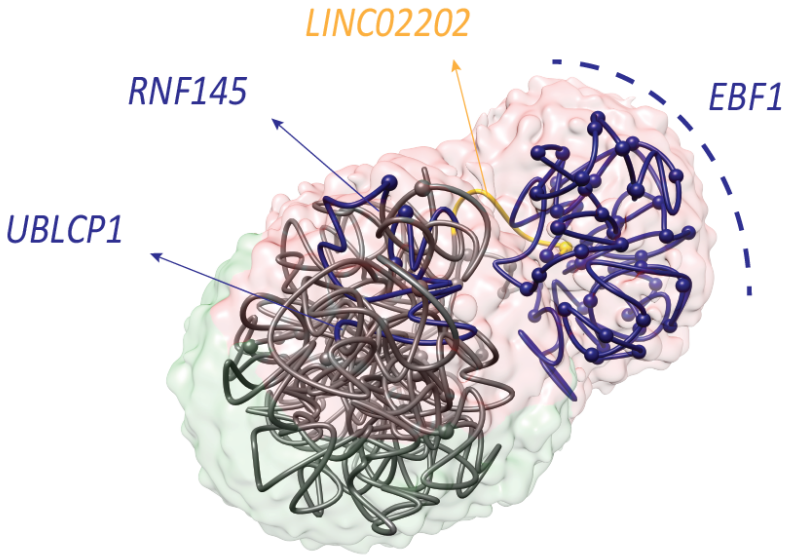
XVIII IWCLL
International Workshop on CLL
20-23 SEPTEMBER 2019 EDINBURGH

Vilarrasa-Blasi & Soler-Vila et al., submitted
https://www.biorxiv.org/content/10.1101/764910v2

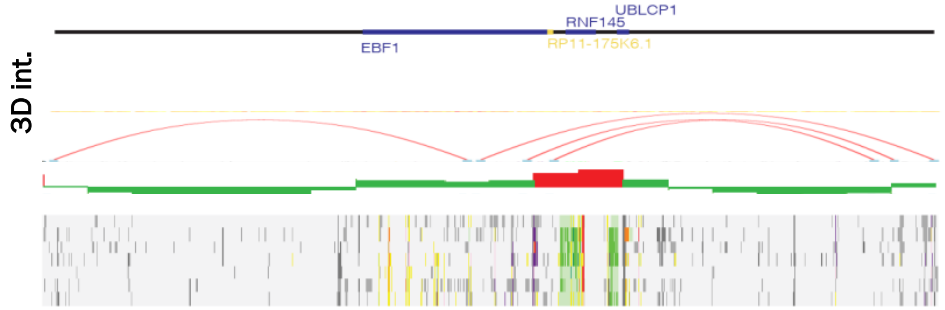
Story 3: Structural model of EBF1 in normal B cells and CLL

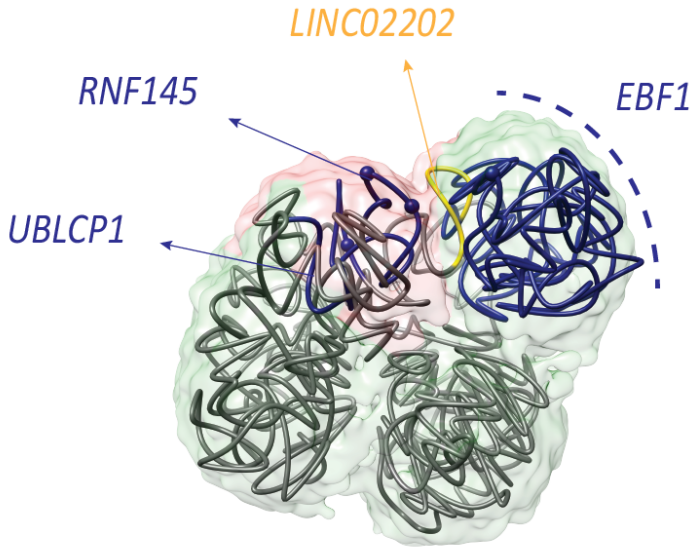
NBC

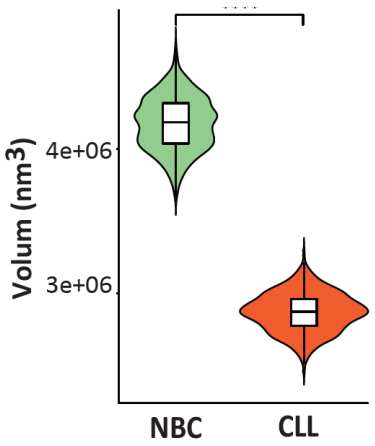




CLL







XVIII INCLL
International Workshop on CLL
20-23 SEPTEMBER 2019 EDINBURGH

Vilarrasa-Blasi & Soler-Vila et al., submitted
https://www.biorxiv.org/content/10.1101/764910v2

Take-home messages

- The regulatory chromatin landscape is extensively altered in CLL as a whole and in IGHV-based subtypes, and seems to be mediated by the action of few TF families.
- The great majority of the SNPs associated with CLL risk are located in regions enriched in regulatory elements.
- CLL shows an altered 3D genome structure and DNA blocks changing their level of 3D interactions contain genes related to disease pathogenesis.

Acknowledgements

IDIBAPS, Hospital Cinic and Universitat de Barcelona, Spain

Renee Beekman
Roser Vilarrasa-Blasi
Vicente Chapaprieta
Núria Russiñol
Núria Verdaguer-Dot
Marta Kulis
Martí Duran-Ferrer
Guillem Clot
Sílvia Beà
Dolors Colomer
Elías Campo

Radboud University Nijmegen, The Netherlands

Joost H.A. Martens
Hendrik G. Stunnenberg

Euopean Bioinformatics Institute, Hinxton, UK

Avik Datta
Ernesto Lowy
Paul Flicek

University of Navarra, Spain

Xabier Agirre
Felipe Prosper

Centro Nacional de Análisis Genómico (CNAG), Barcelona, Spain

Paula Soler-Vila
Marco Di Stefano
Irene Farabella,
Marc Marti-Renom

The Institute of Cancer Research (ICR), London, UK

Helen Speedy
Giulia Orlando
Philip Law
Dan Catovsky
Richard Houlston

Instituto de Salud Carlos III

GOBIERNO DE ESPAÑA · MINISTERIO DE ECONOMÍA Y COMPETITIVIDAD · SECRETARÍA DE ESTADO DE INVESTIGACIÓN, DESARROLLO E INNOVACIÓN

European Commission

worldwide cancer research

aecc Contra el Cáncer

MARIE CURIE ACTIONS

EHA

XVIII IWCLL
International Workshop on CLL
20-23 SEPTEMBER 2019 EDINBURGH