

## Data accessibility

### Online session

<http://epigenomegateway.wustl.edu/browser/?genome=mm9&session=bxT0F5m0YY>

Create your own session (optional)

- 1) Goes to <http://epigenomegateway.wustl.edu/browser/>
- 2) Choose mm9
- 3) Choose "Custom tracks"
- 4) Choose "Datahub by URL link"
- 5) Paste [http://wangftp.wustl.edu/~dli/4DN\\_Tcells/datahub\\_online.json](http://wangftp.wustl.edu/~dli/4DN_Tcells/datahub_online.json) to textbox "Datahub file URL" and click "SUBMIT" button. Enjoy!

Data download (~7G)

[http://wangftp.wustl.edu/~dli/4DN\\_Tcells/](http://wangftp.wustl.edu/~dli/4DN_Tcells/) or upon request.

### Data organizations of the online session

Tracks for each development stage from HSPCs to DPs (shown by default)

- 1) Distribution of RNA-Seq read density.
- 2) Distribution of DNase-Seq read density.
- 3) Distribution of A/B compartment score<sup>1</sup>.
- 4) Norm. interaction matrix from replicates (not shown by default to speed loading).

Tracks summarizing change in regulome and 3D nucleome from HSPCs to DPs (shown by default)

- 1) Genomic region showing concordant change in compartment status.
- 2) DHSs exhibiting concordant change in chromatin accessibility.
- 3) TADs that change in intra-TAD connectivity.

Tracks of arc-plots for promoter-enhancer interaction<sup>2</sup> (shown by default)

- 1) pre-commitment (pooled from HSPCs to DN2s).
- 2) Post-commitment (pooled from DN3 to DP).
- 3) Post-commitment (as in 2, but sub-sampled into a size comparable to pre-commitment).

Tracks for public ChIP-Seq data from HSPCs (also HPC7 cell line<sup>3</sup>) to DPs (not shown by default)

- 1) ~100 tracks for histone modifications (H3K4me1, H3K4me3, H3K27me3, H3K27ac, etc).
- 2) ~50 tracks for transcription factors (BCL11B, GATA3, PU.1, etc.).

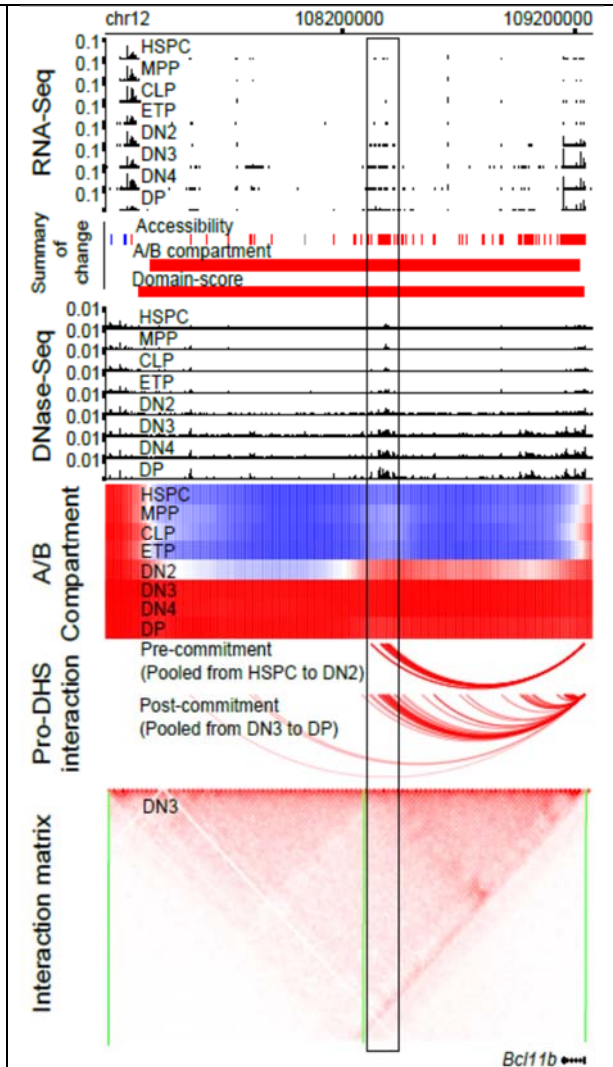
Statistics of Public ChIP-Seqs (As of Oct. 2017)

	H3K4me1	H3K4me2	H3K4me3	H3K27me3	H3K27ac	H2AUBQ	H3AC	H3K79me2	TFs	Total
HSPC	2	3	10	7	2	1	0	1	6	32
MPP	2	1	4	3	2	0	0	0	0	12
CLP	1	1	1	0	1	0	0	0	0	4
ETP	0	3	0	3	0	0	2	0	2	10
DN2	1	1	1	2	0	0	1	0	5	11
DN3	0	7	1	5	0	0	4	0	8	25
DN4	0	0	0	2	0	0	0	0	1	3
DP	2	3	3	6	5	0	2	1	14	36
HPC7	0	0	0	0	0	0	1	0	12	13

## Examples of data integration

### Example 1: The super-enhancer of *Bcl11b*

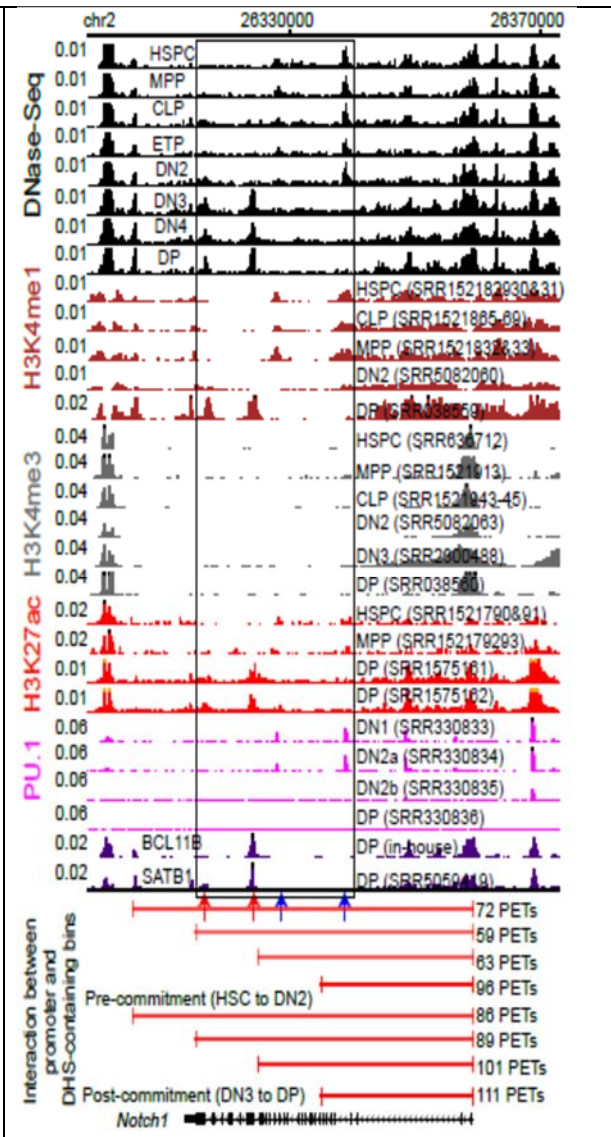
In an earlier study, we discovered a super-enhancer region located about 750K downstream to the *Bcl11b* locus in a previous study (Li et al., 2013). As expected, the expression the RNA-Seq tracks revealed that *Bcl11b* expression was up-regulated from HSPCs to DPs. Interestingly, the RNA-Seq tracks also revealed some expression around the super-enhancer downstream to *Bcl11b*, albeit at low level, corresponding to a lincRNA of *Bcl11b* (Isoda et al., 2017). The DNase-Seq tracks revealed that the chromatin accessibility at the lincRNA region increased dramatically at DN2 and further at DN3. The tracks for compartment scores demonstrated that this region could be attached to periphery in HSCs and relocated to nuclear interior in DN2s. In general, the change in chromatin accessibility at DHSs, the change in compartment status, and the change in intra-TAD connectivity were all coordinated for genomic regions around *Bcl11b*. Additionally, the promoter-enhancer interaction tracks revealed that the lincRNA interacted with *Bcl11b* for a sub-population of the cells before commitment, and the portion increased dramatically post commitment. The interaction between the lincRNA region and *Bcl11b* is also observed by others in in-vitro cultured DN2s (Isoda et al., 2017) and earlier by us in DN3-like leukemic cell line P2C2 (Li et al., 2013). Moreover, Cornelis Murre's lab from UCSF independently discovered the compartment reposition of the lincRNA and elegantly demonstrated that mechanistically the lincRNA transcription activates the looping with the *Bcl11b* promoter and facilitates *Bcl11b* expression (Isoda et al., 2017).



Online WashU genome browser screen shot (edited) showing tracks for RNA-Seq reads, DNase-Seq reads, and A/B compartment score for a genomic region surrounding a super-enhancer of *Bcl11b* (rectangle) from HSPCs to DPs. Summary of change: tracks that summarizes change in accessibility at DHSs, A/B compartment flip, and change in domain score within TADs with red indicating increase in accessibility, B-to-A compartment flip, or increase in domain score, and blue indicating decrease in accessibility, A-to-B compartment flip, or decrease in domain score. Green lines: TAD boundaries; arc plots: interaction between *Bcl11b* promoter and DHSs for stages pooled from HSPC to DN2 and for stages pooled from DN3 to DP. Interaction matrix: shown only for DN2 to save space.

Example2: Enhancer switch within the *Notch1* locus

We noticed that four DHSs within the *Notch1* locus exhibited opposite change in chromatin accessibility from HSPCs to DPs (red arrow heads vs blue arrow heads). The four DHSs exhibited a typical feature of being an enhancer in the presence of H3K4me1 and absence of H3K4me3 in corresponding stages (Heintzman et al., 2007). Two of the enhancers (blue arrow heads) were active pre-commitment, by the presence of H3K27ac, an indicator of active enhancers (Creyghton et al., 2010; Rada-Iglesias et al., 2011). Interestingly, PU.1 bound these sites pre-commitment. As PU.1 binding induces chromatin remodeling (Heinz et al., 2010; Krysinska et al., 2007; Marecki et al., 2004), the observation suggested that the loss of PU.1 binding at the two sites post-commitment is linked to the enhancer decommission. The other two enhancers were newly formed and activated post-commitment (red arrow heads). Remarkably in DPs, the enhancers are characterized by binding of BCL11B and a co-binding of a recruit of chromatin-modulating complexes, SATB1 (Yasui et al., 2002), which physically interacted BCL11B (Kitagawa et al., 2017). Interestingly, the interaction landscape between *Notch1* promoter and DHSs showed no remarkable overall difference pre- and post-commitment, suggesting that the enhancer switch occurred through changing enhancer activity on pre-existing chromatin structure and involving gain and loss of binding of TFs specific to development stages. The enhancer switch within *Notch1* is supplementary to the previously proposed “relay race” model of transcription regulome, in which broadly active gene switches interaction with lineage-specific regulatory elements during development (Kieffer-Kwon et al., 2013).



Online WashU genome browser session showing tracks for DNase-Seq data and representative ChIP-Seq data of H3K4me1, H3K4me2, H3K4me3, H3K27ac, PU.1, BCL11B and SATB1 for stages where the data is available, and interaction between DHS-containing bins and *Notch1* promoter at a resolution of 10K. Red arrow heads: DHSs increasing in accessibility; Blue arrow heads: DHSs decreasing in accessibility; Black rectangle: region of enhancer switch; parenthesis: accession number for public data.

## Notes

<sup>1</sup> Compartment score: PC1 predicted by HOMER (Lin et al., 2012)

<sup>2</sup> As the 3D nucleome was similar within stages pre-commitment and within stages post-commitment, we pooled Hi-C PETs from HSC to DN2 and from DN3 to DP to increase statistical significance. Interactions predicted by FitHiC with ICE normalization: resolution = 10K, q-value < 0.01 (Ay et al., 2014).

<sup>3</sup> HPC7: multipotent hematopoietic progenitor cell line

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