Photo by Laia Marín

CHROMATIC reveals chromatin-associated factors contributing to genome topology
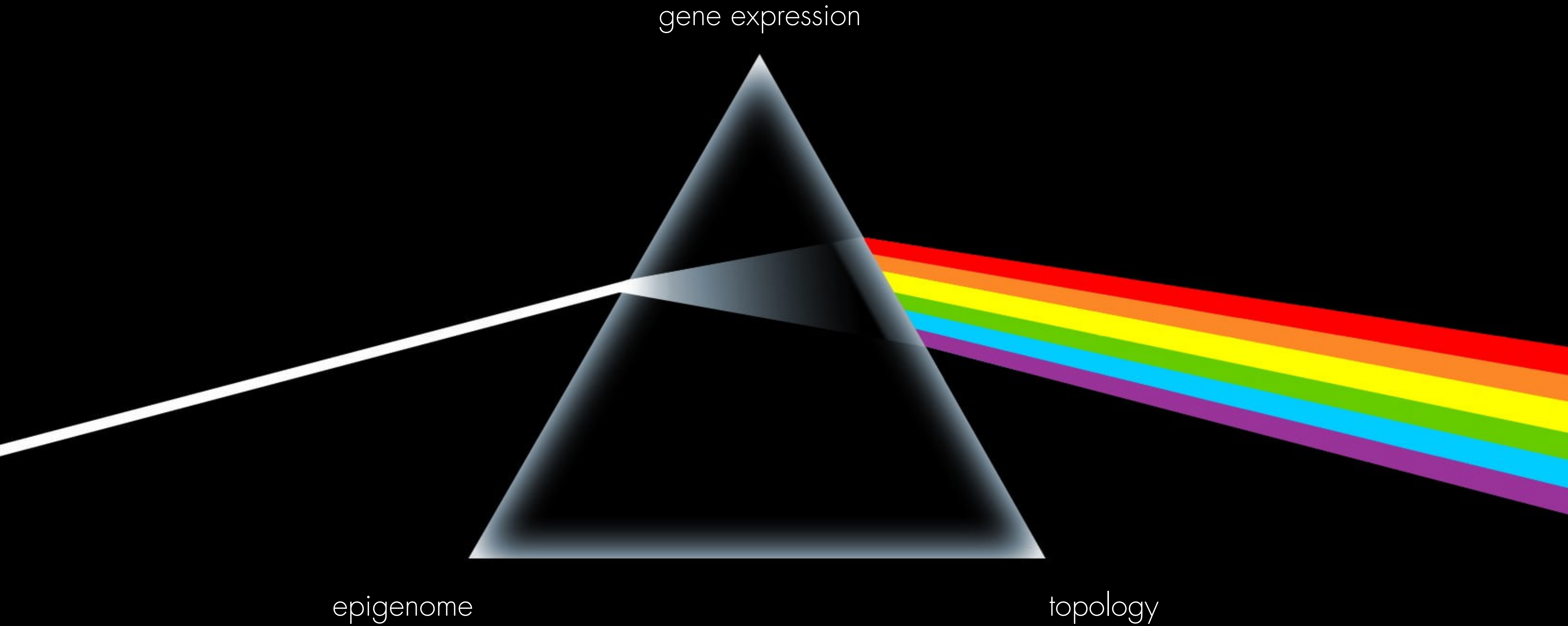
Marc A. Marti-Renom

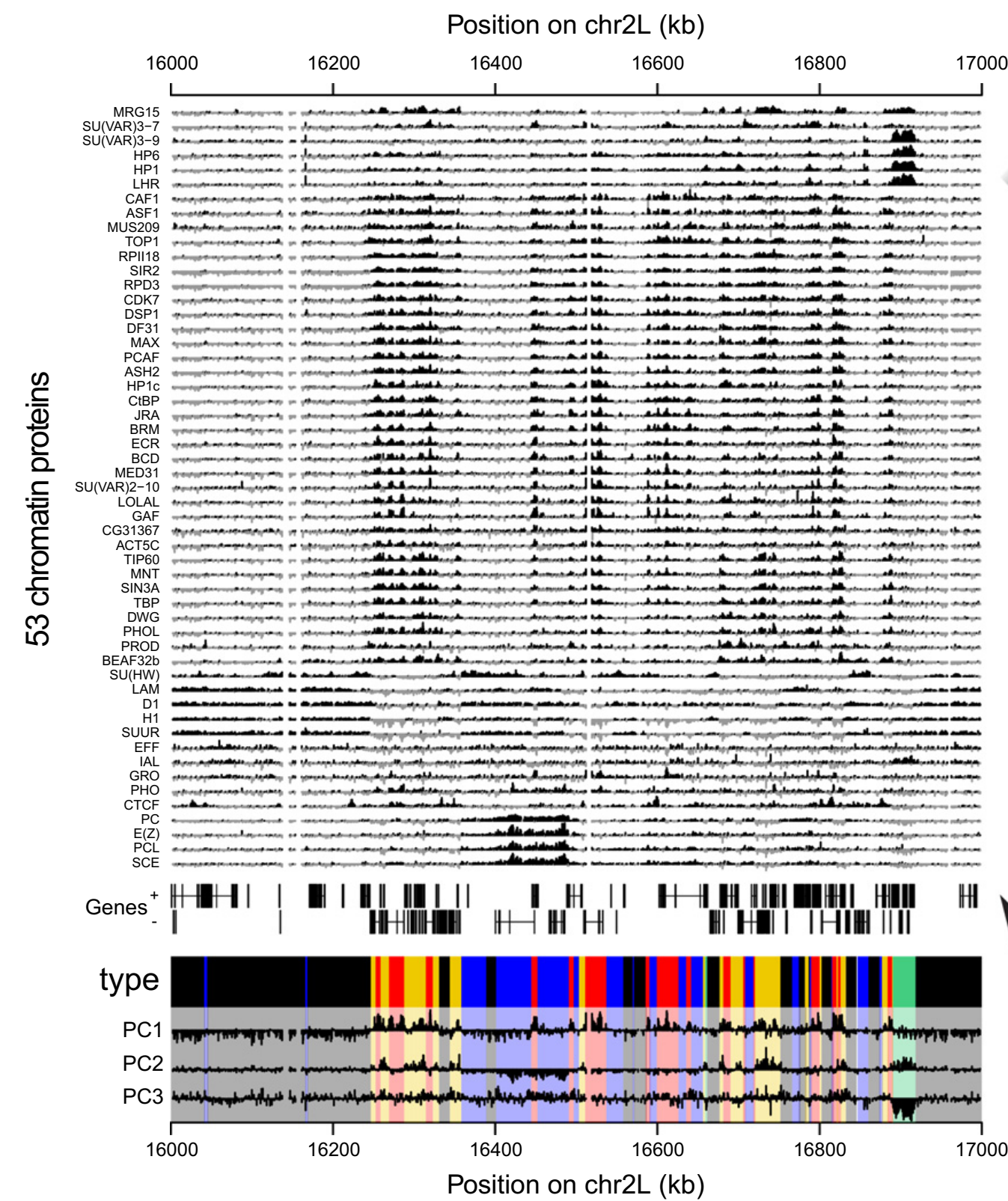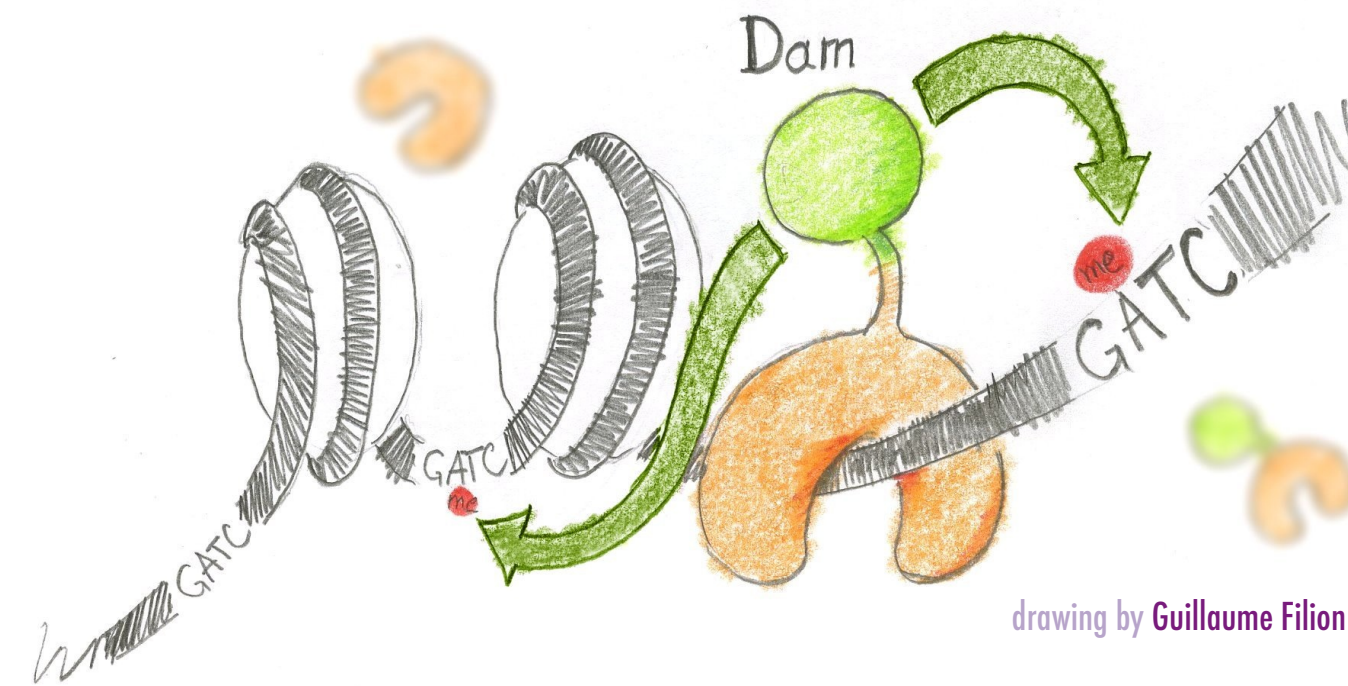CNAG-CRG · ICREA

cnag CRG · ICREA

The genome is a complex system

gene expression

epigenome

topology

Fly Chromatin COLORs

Filion et al. (2010). Cell, 143(2), 212–224.

drawing by Guillaume Filion

Principal component analysis

Position on chr2L (kb)

53 chromatin proteins

Hidden Markov model

Position on chr2L (kb)

GREEN
HP1

YELLOW
Active

RED
Active

BLUE
Polycomb

chromatin proteins

BLACK
Repressive

Nucleus
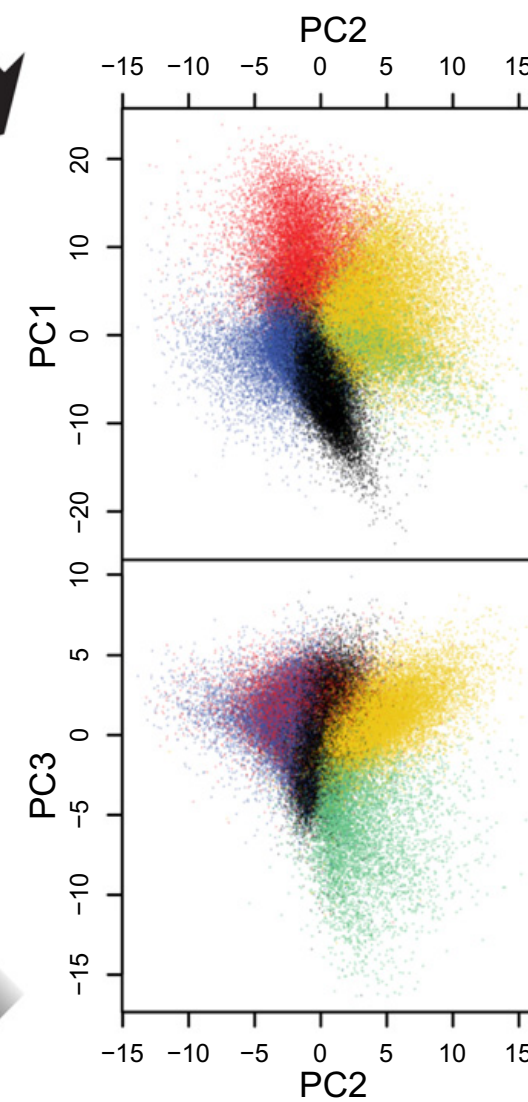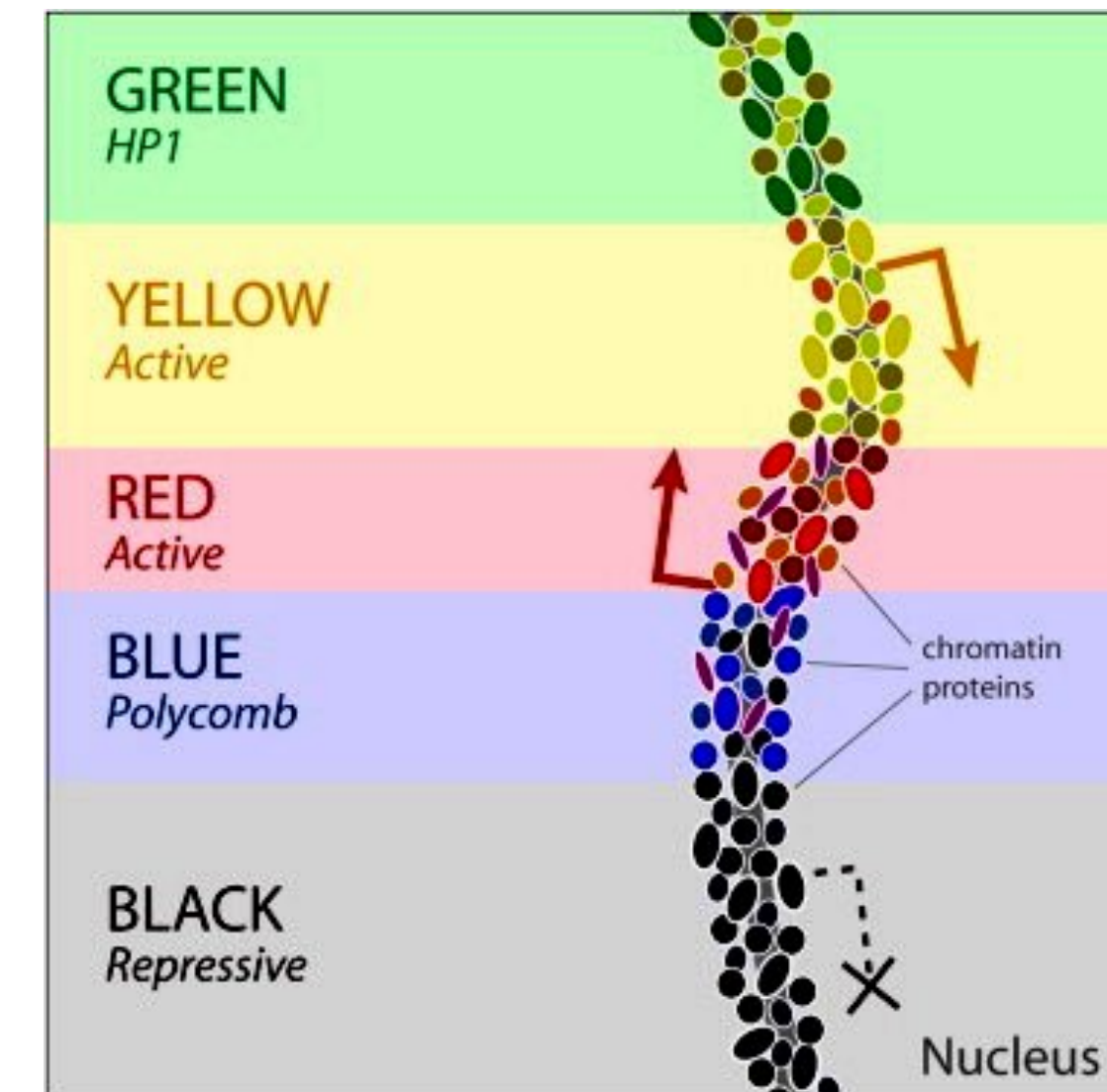
Who "holds" the genome structure?
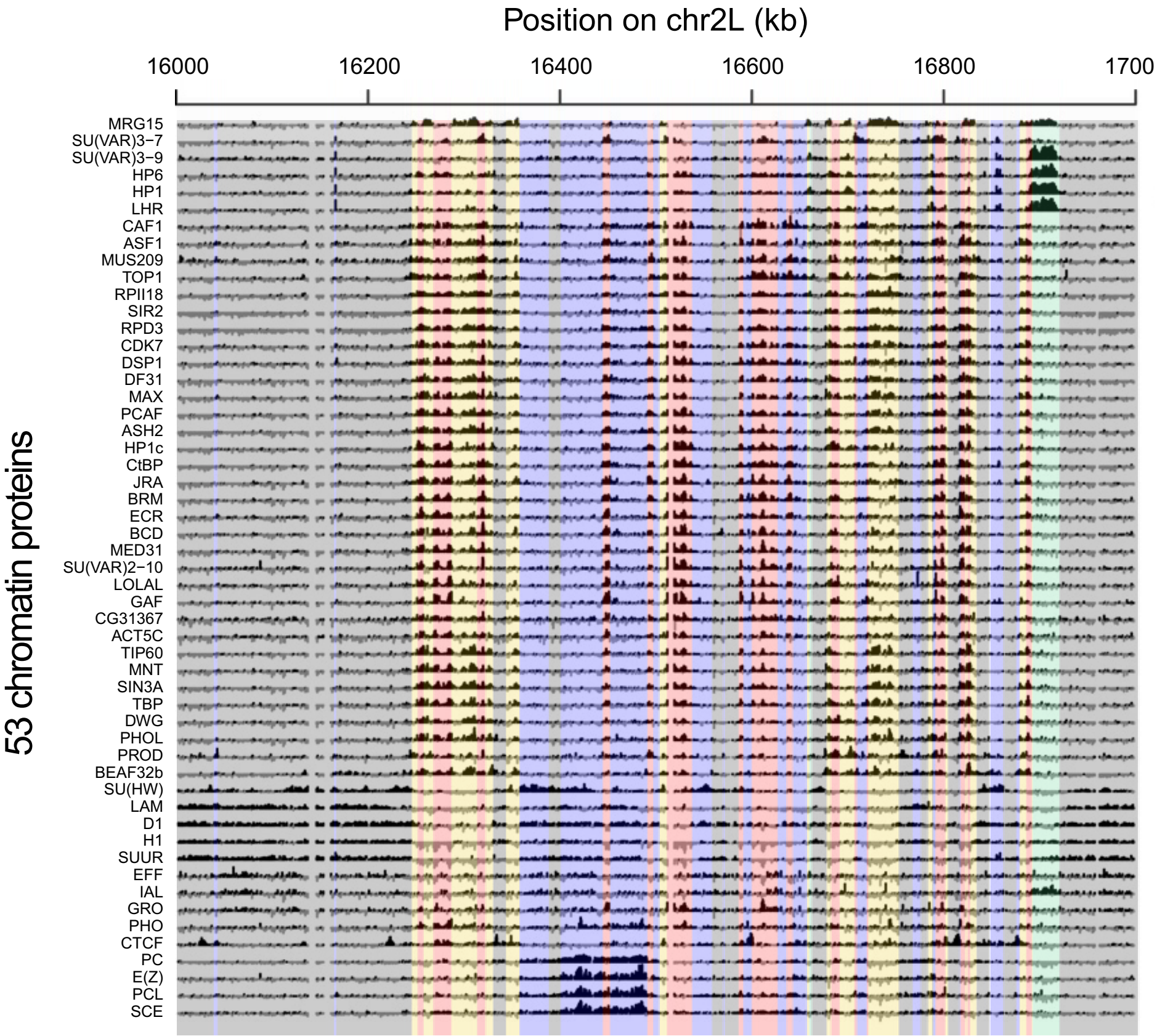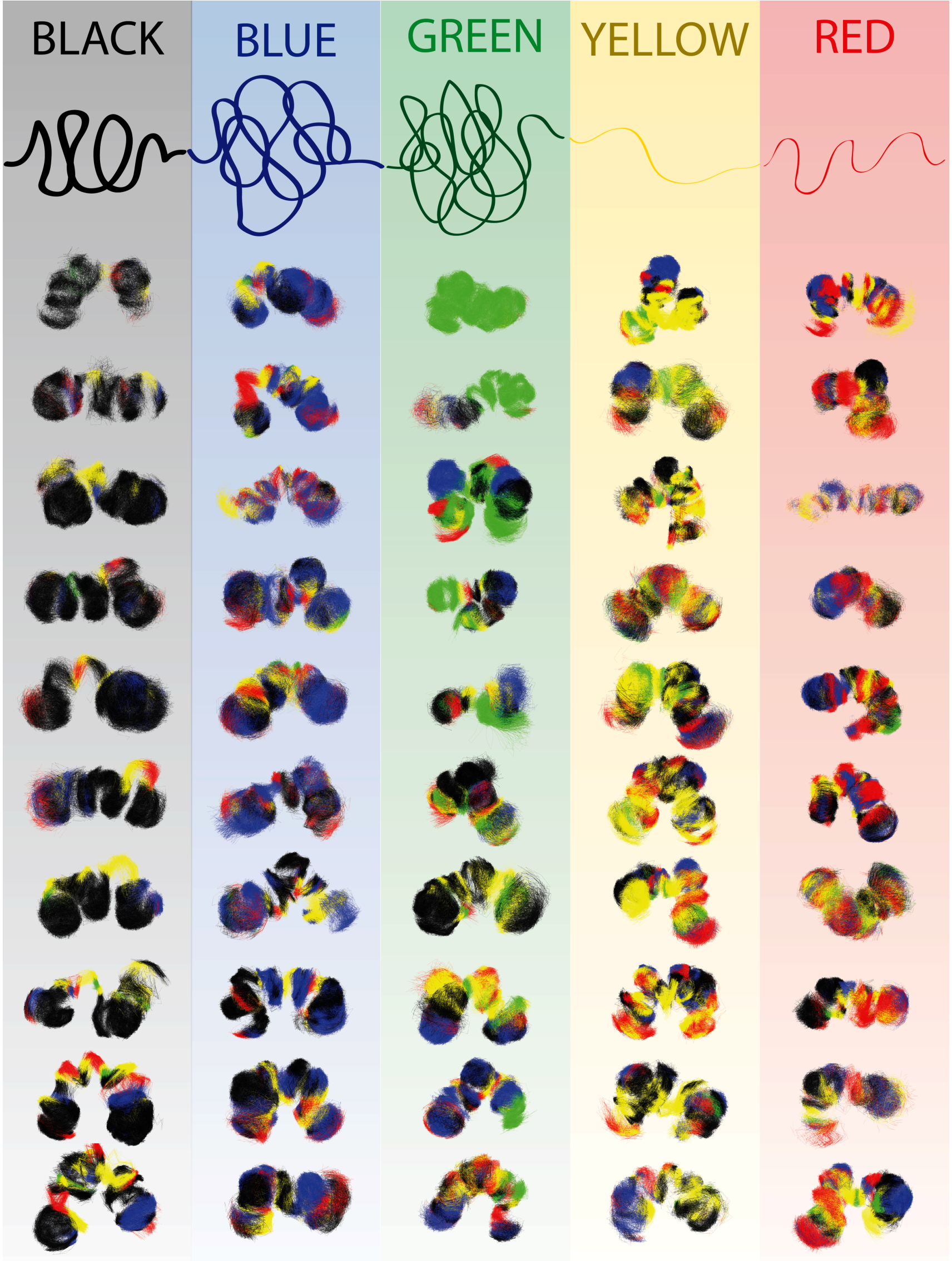
Chromosome Conformation Capture

Dekker, J., Rippe, K., Dekker, M., & Kleckner, N. (2002). Science, 295(5558), 1306–1311.
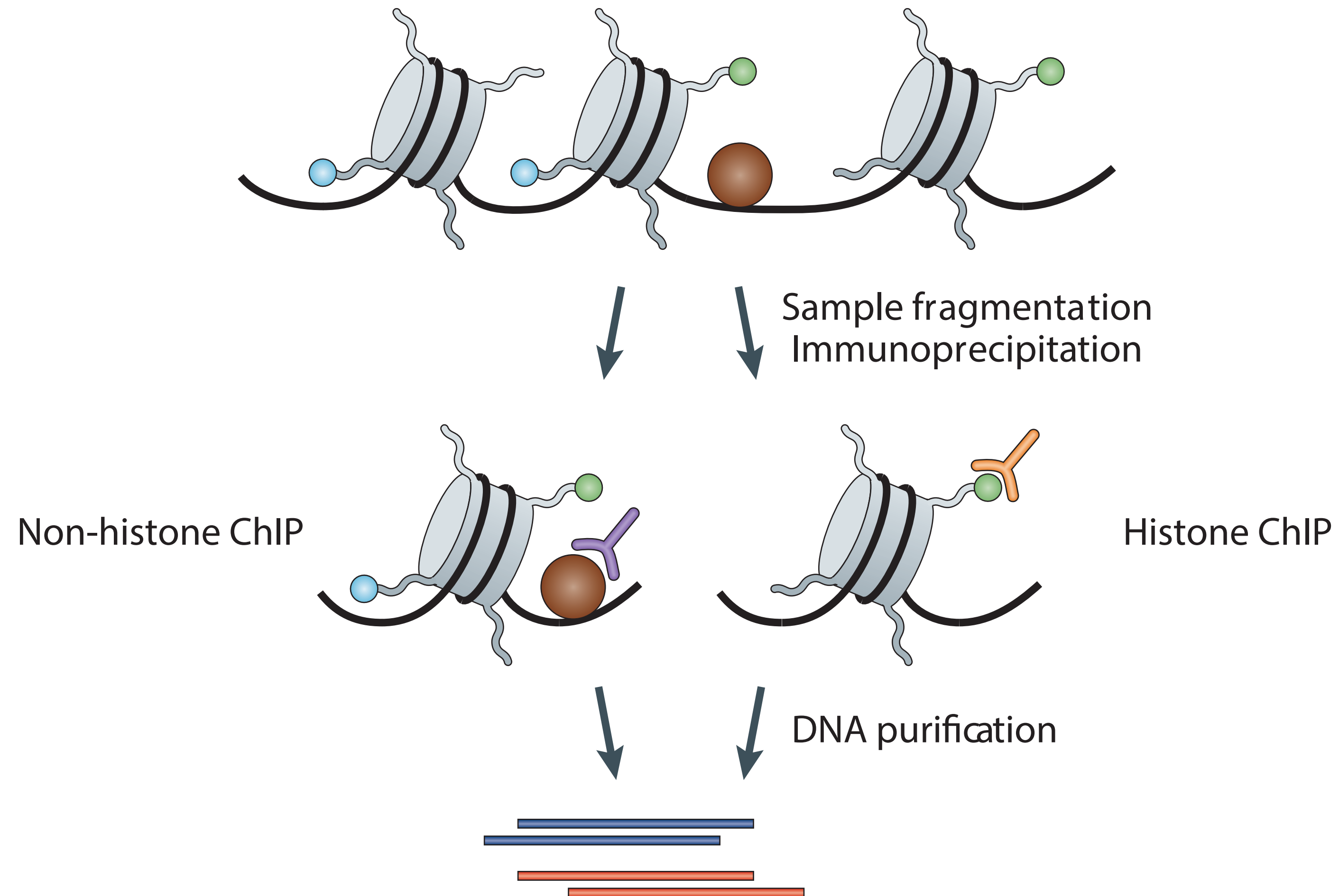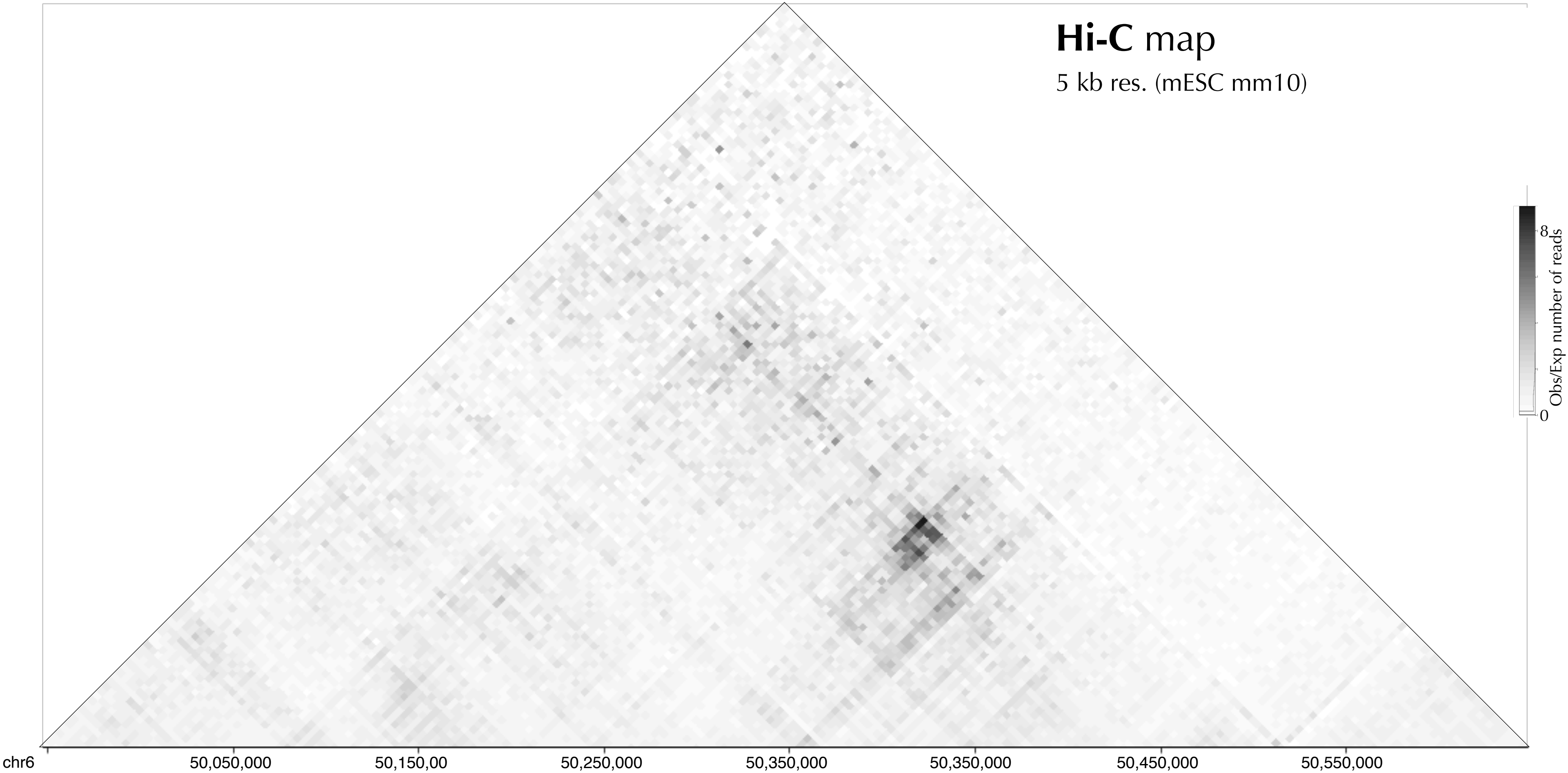Lieberman-Aiden, E., et al. (2009). Science, 326(5950), 289–293.
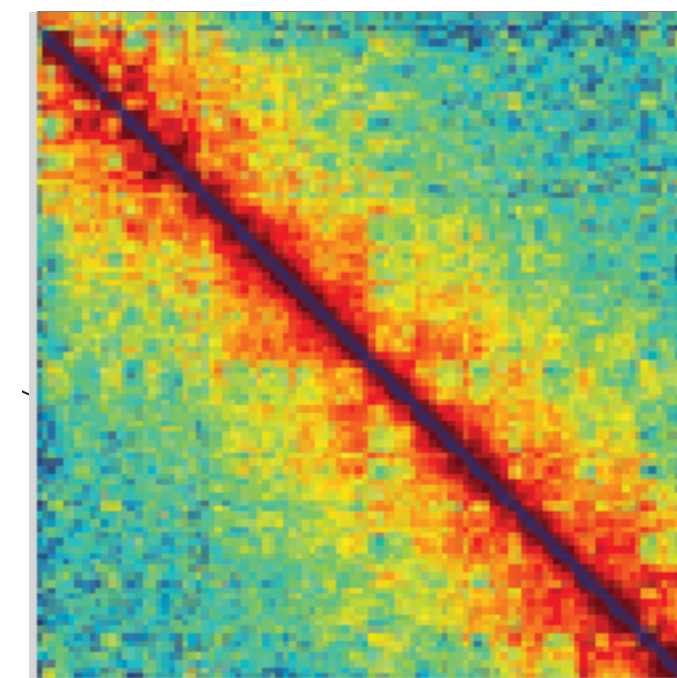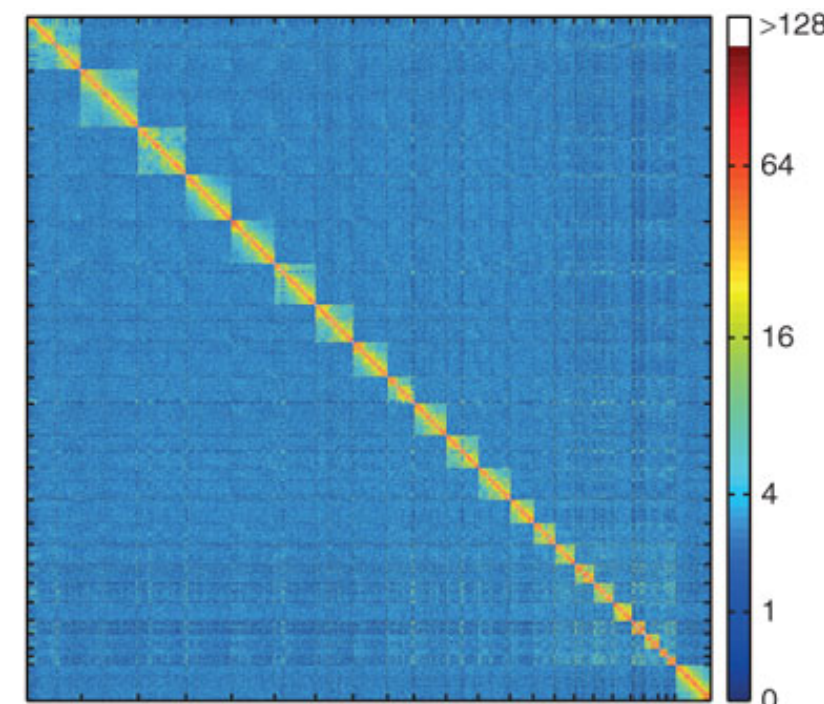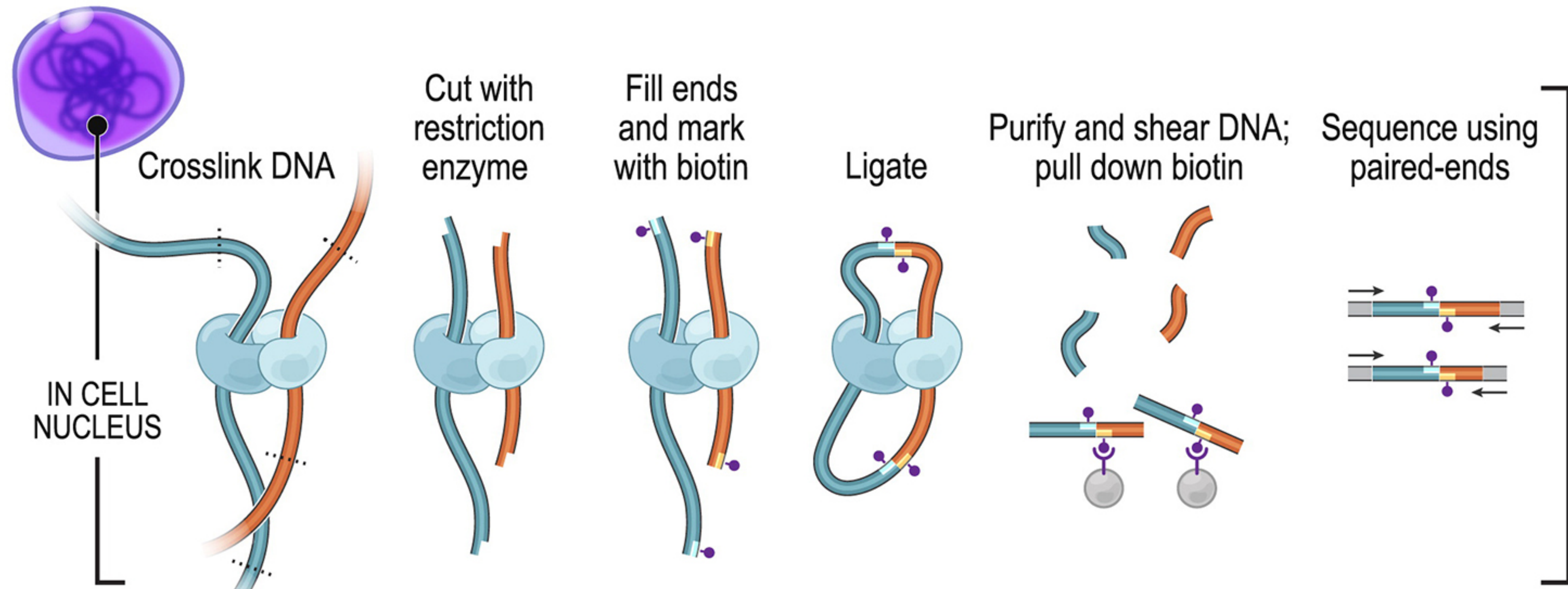
Chromatin Immunoprecipitation (ChIP)

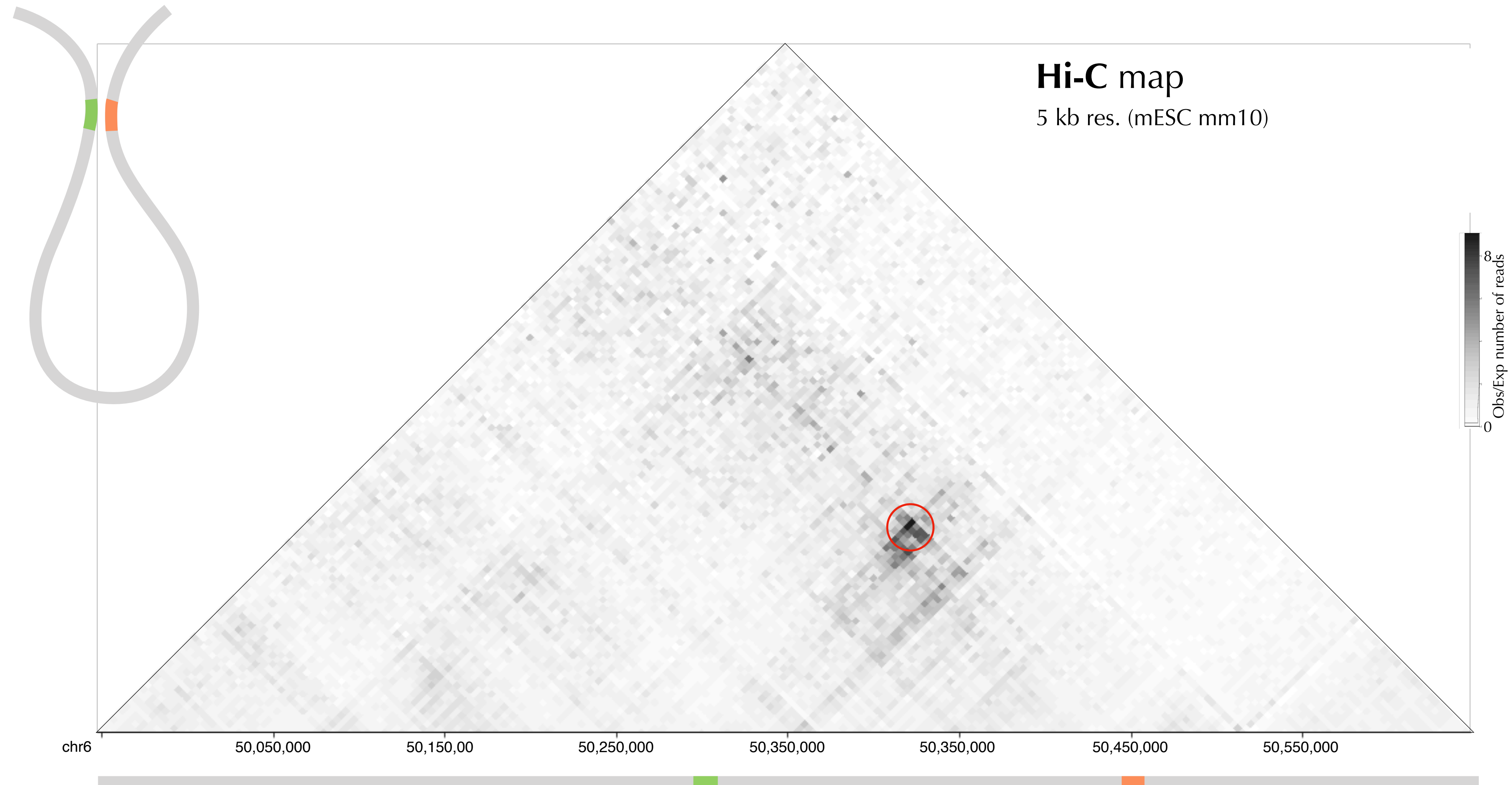Solomon, M. J., Larsen, P. L. & Varshavsky, A. (1988) Cell 53, 937–947.
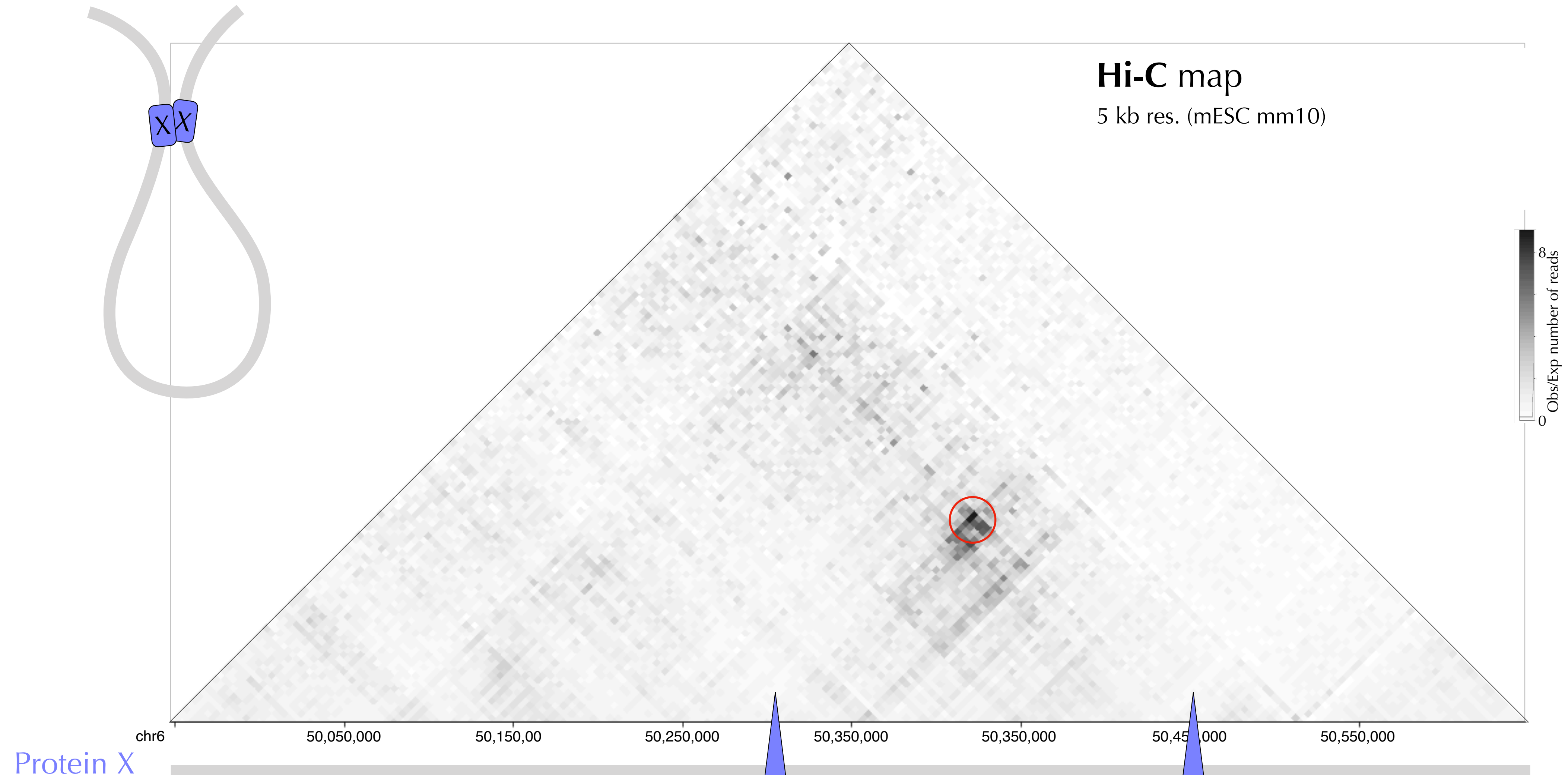Park, P.J. (2009) Nature Reviews Genetics 10, 669–680.

High-throughput Chromosome Conformation Capture (Hi-C)

Hi-C map
5 kb res. (mESC mm10)
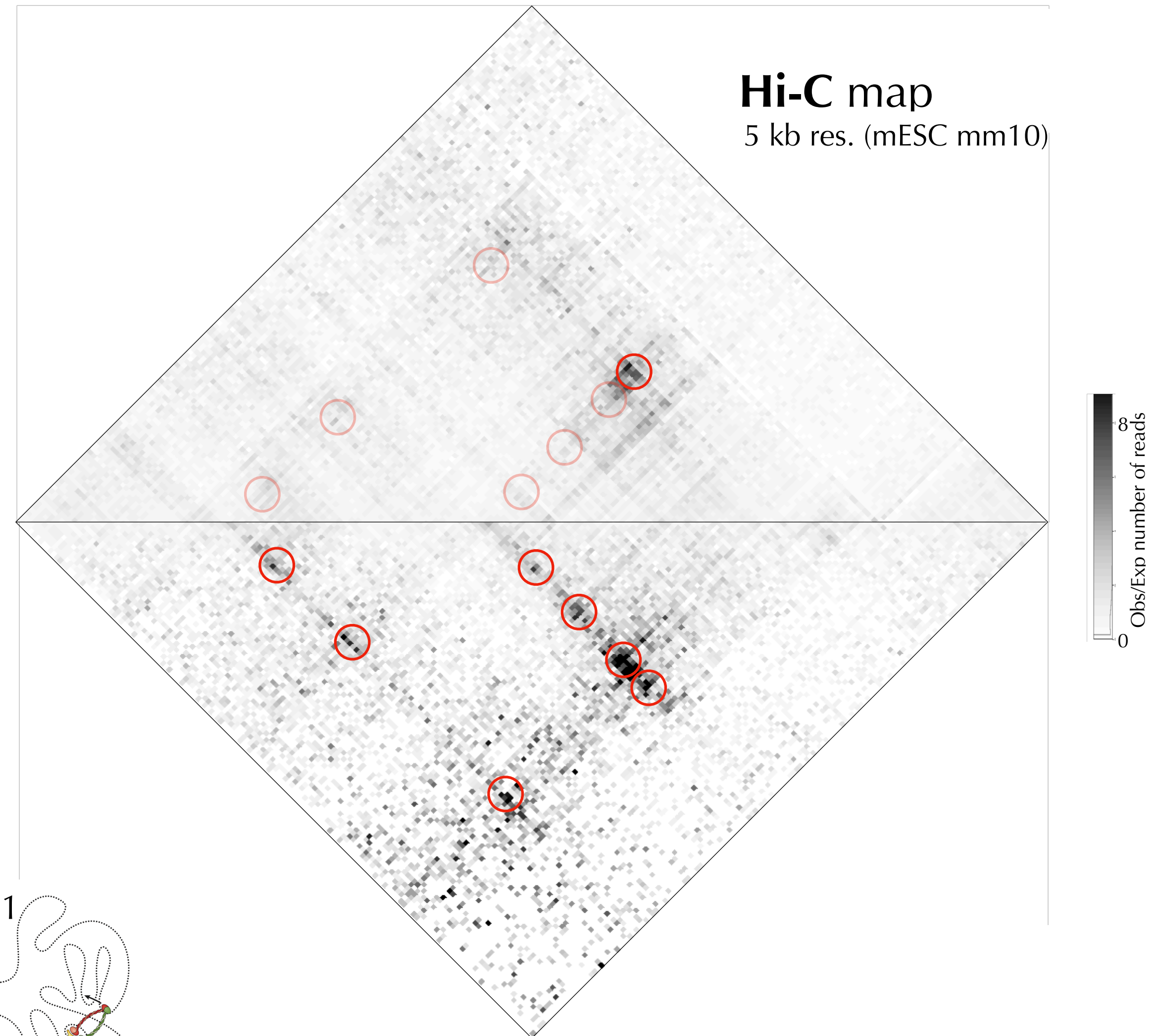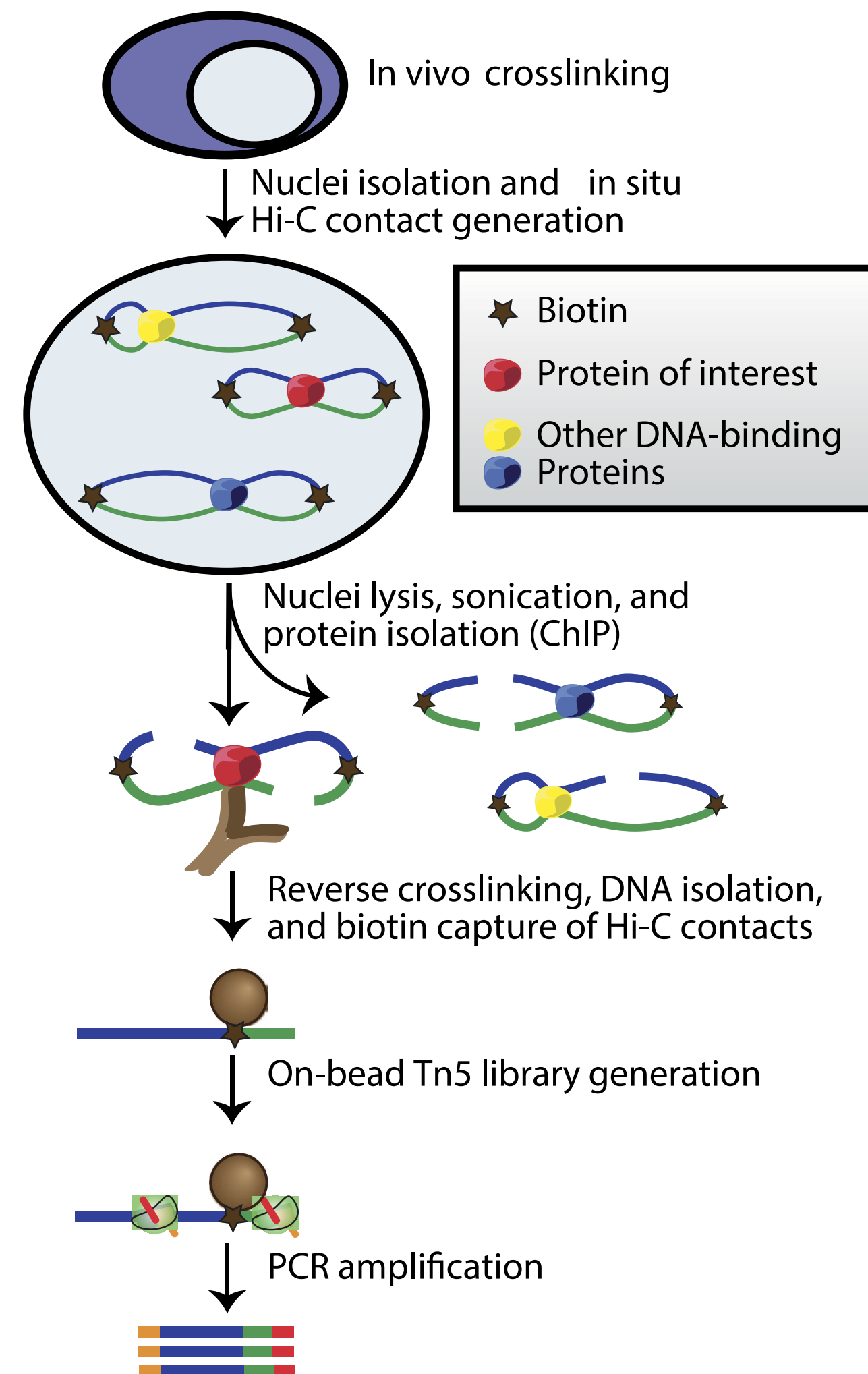
Obs/Exp number of reads

chr6   50,050,000   50,150,00   50,250,000   50,350,000   50,350,000   50,450,000   50,550,000

Chromatin interaction on a Hi-C map

Hi-C map
5 kb res. (mESC mm10)

Obs/Exp number of reads

chr6   50,050,000   50,150,00   50,250,000   50,350,000   50,350,000   50,450,000   50,550,000

Chromatin interaction "mediated" by a protein

Hi-C map
5 kb res. (mESC mm10)

Obs/Exp number of reads
8
0

chr6   50,050,000   50,150,00   50,250,000   50,350,000   50,350,000   50,45   ,000   50,550,000

Protein X

Hi-ChIP: capturing specific protein-mediated interactions
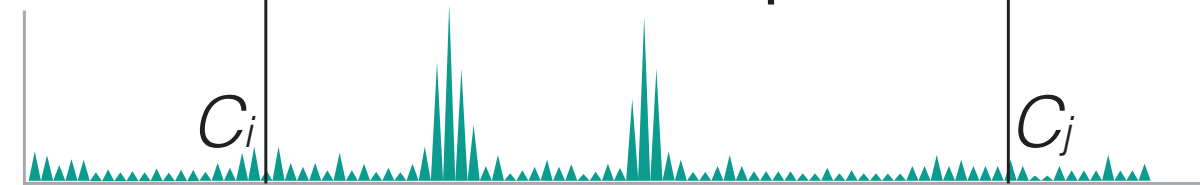
Mumbach, M.R. et al. (2016) Nature Methods 13(11) 919-922.

In vivo crosslinking

Nuclei isolation and in situ Hi-C contact generation

Biotin

Protein of interest

Other DNA-binding Proteins

Nuclei lysis, sonication, and protein isolation (ChIP)

Reverse crosslinking, DNA isolation, and biotin capture of Hi-C contacts

On-bead Tn5 library generation

PCR amplification

Hi-C map
5 kb res. (mESC mm10)

Obs/Exp number of reads

SMC1

SMC1 **Hi-ChIP** map = **Hi-C + ChIP**
5 kb res. (mESC mm10)
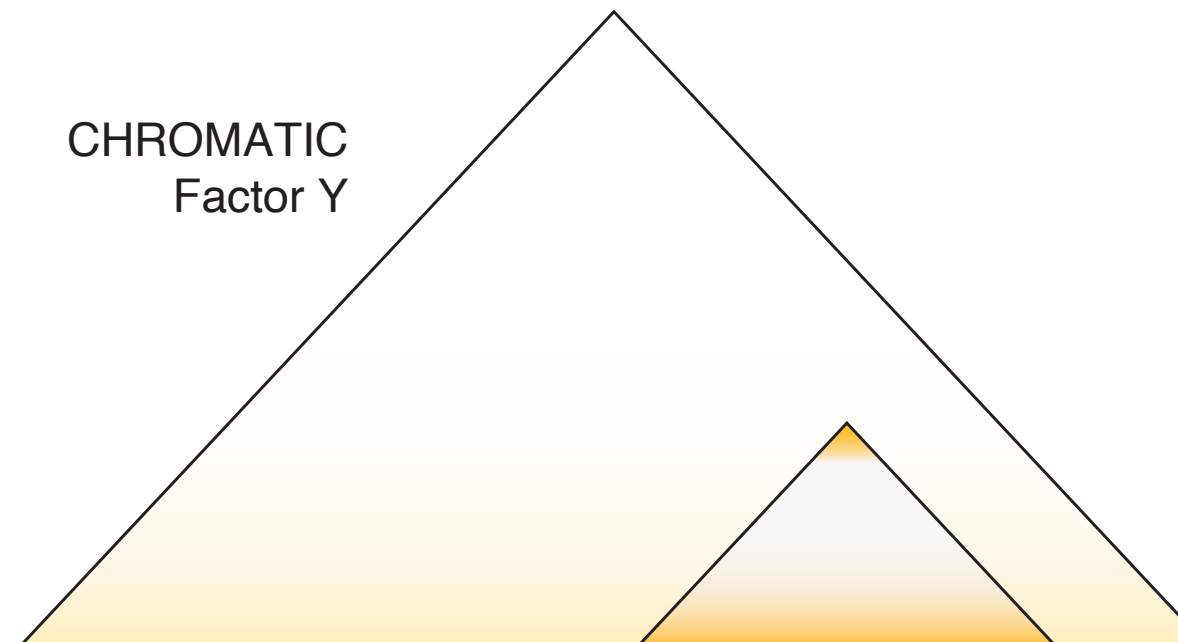
CHROMATIC

Hi-C

H_{ij}

ChIP-seq

Factor X

C_i C_j
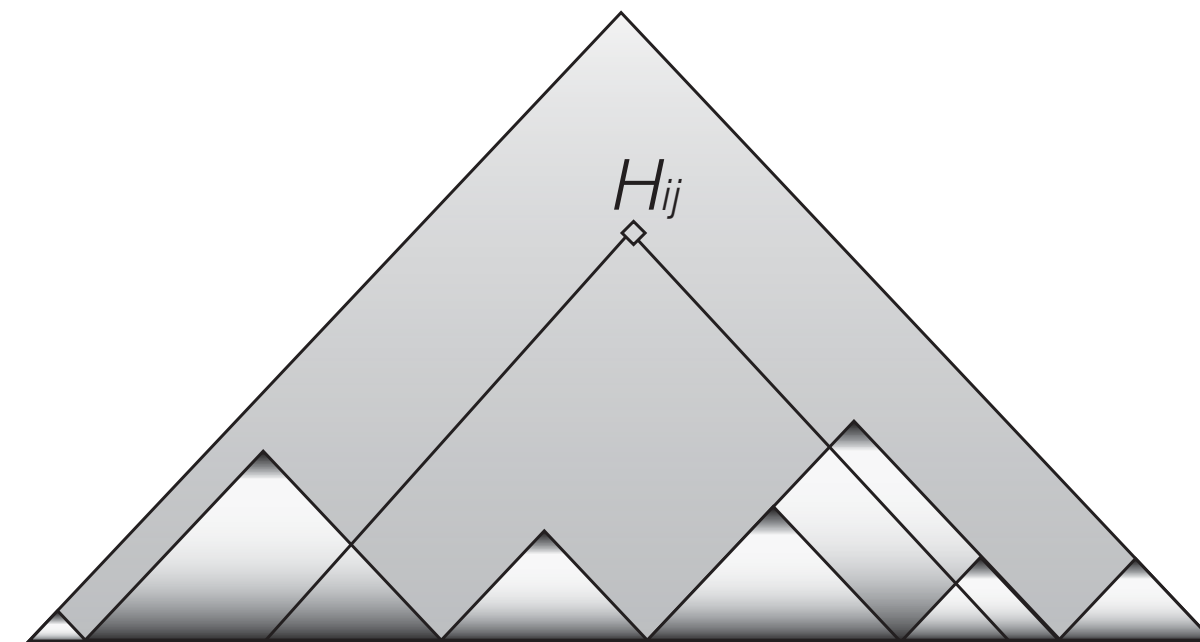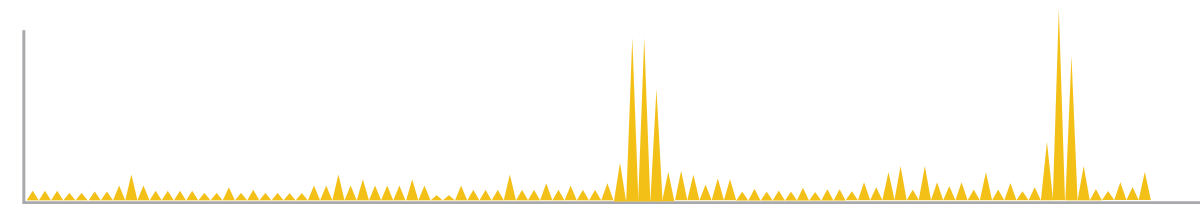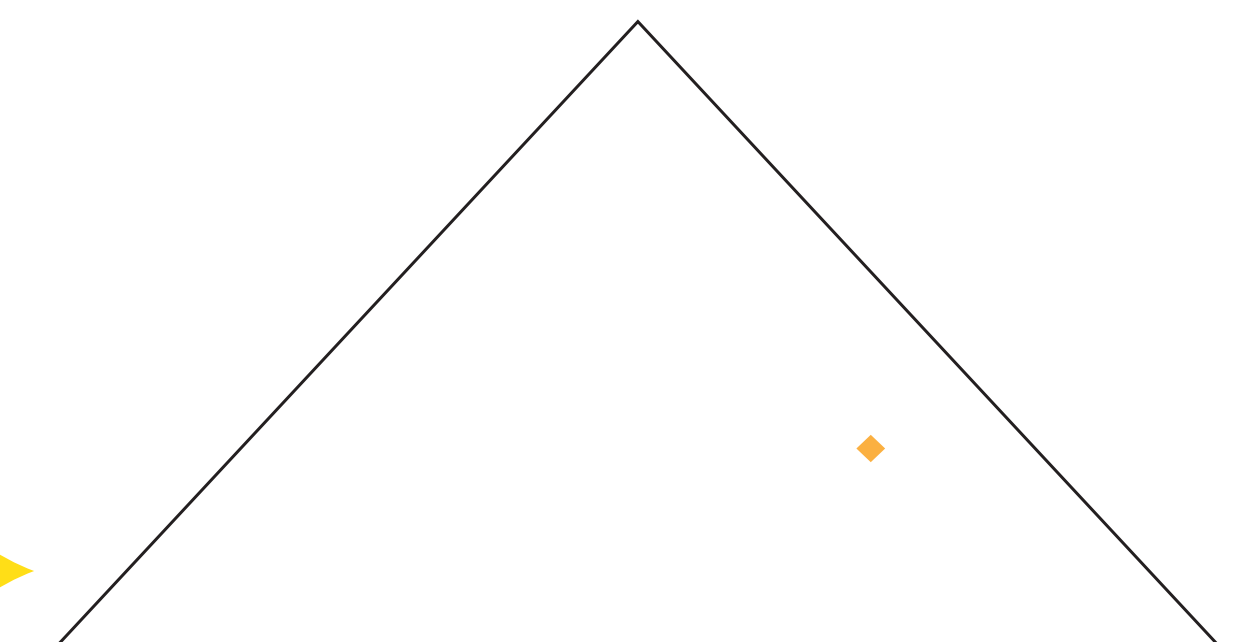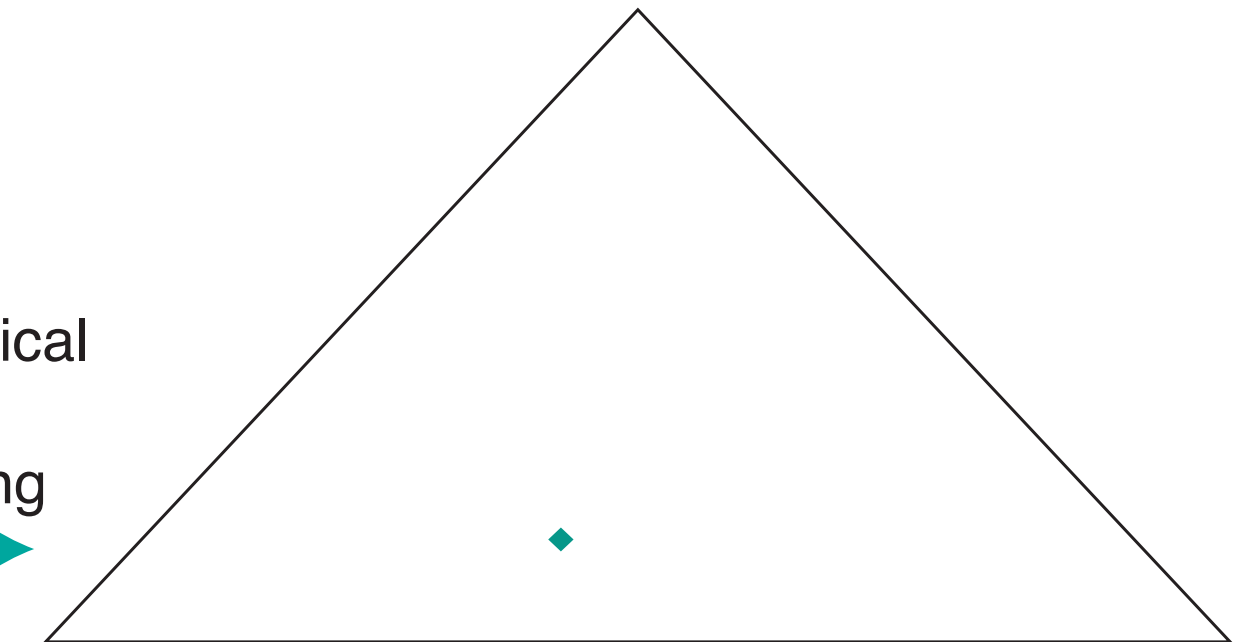
Factor Y

CHROMATIC
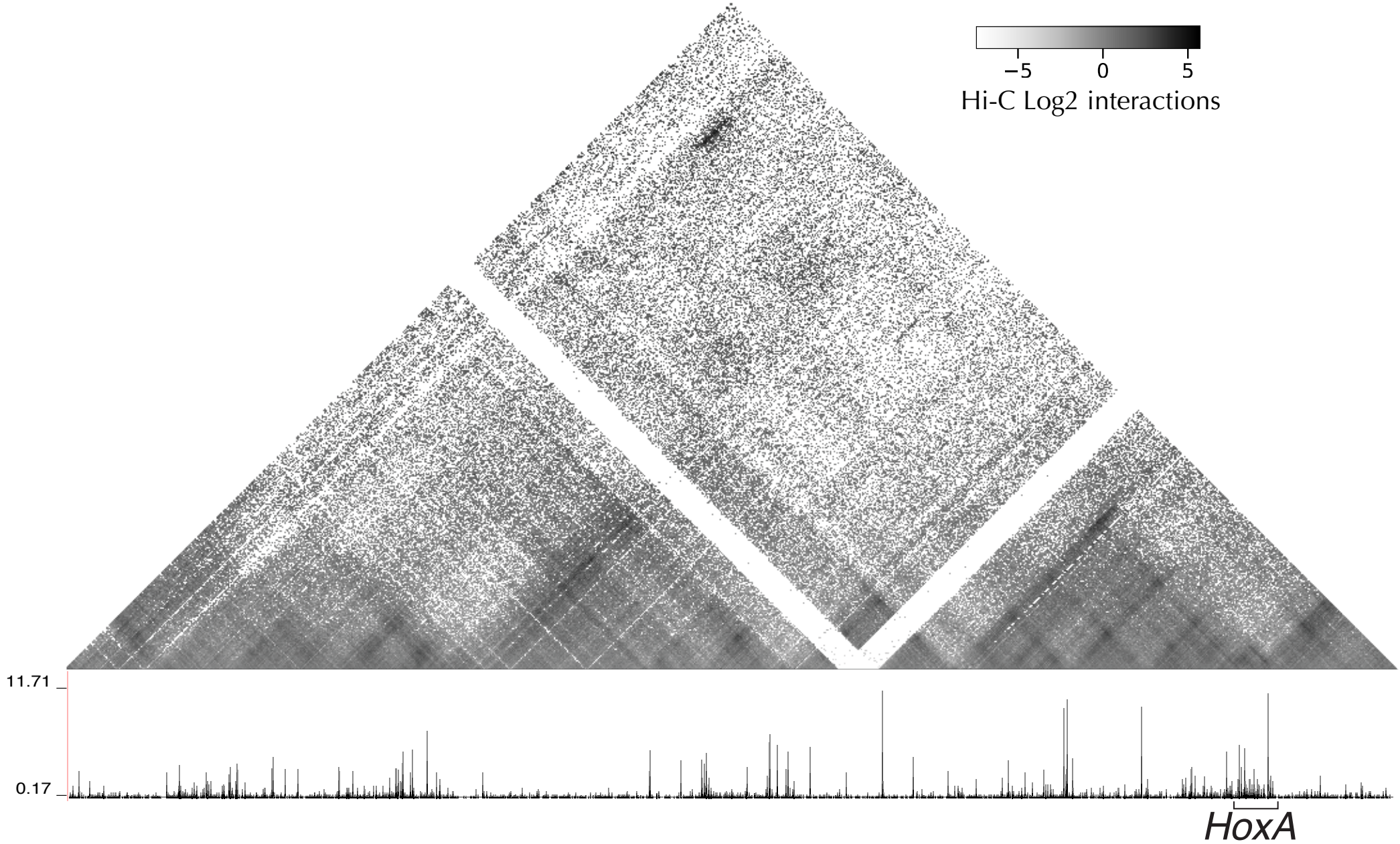
CHROMATIC
Factor X

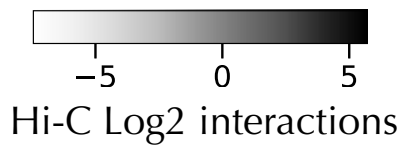$E_{ij}=H_{ij}xC_ixC$
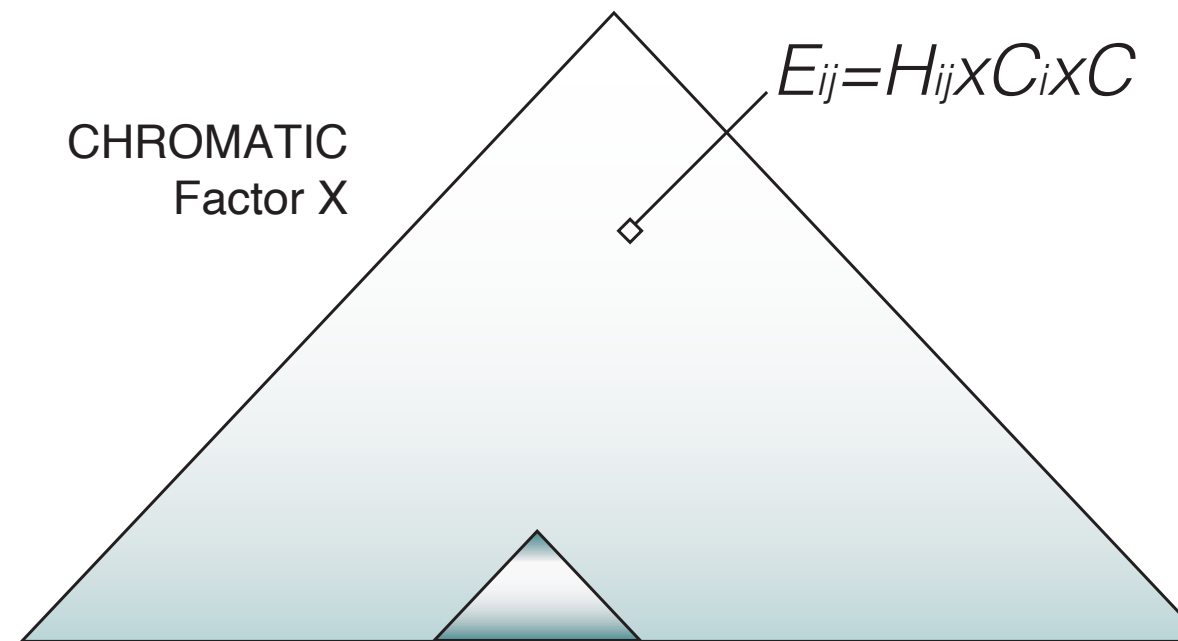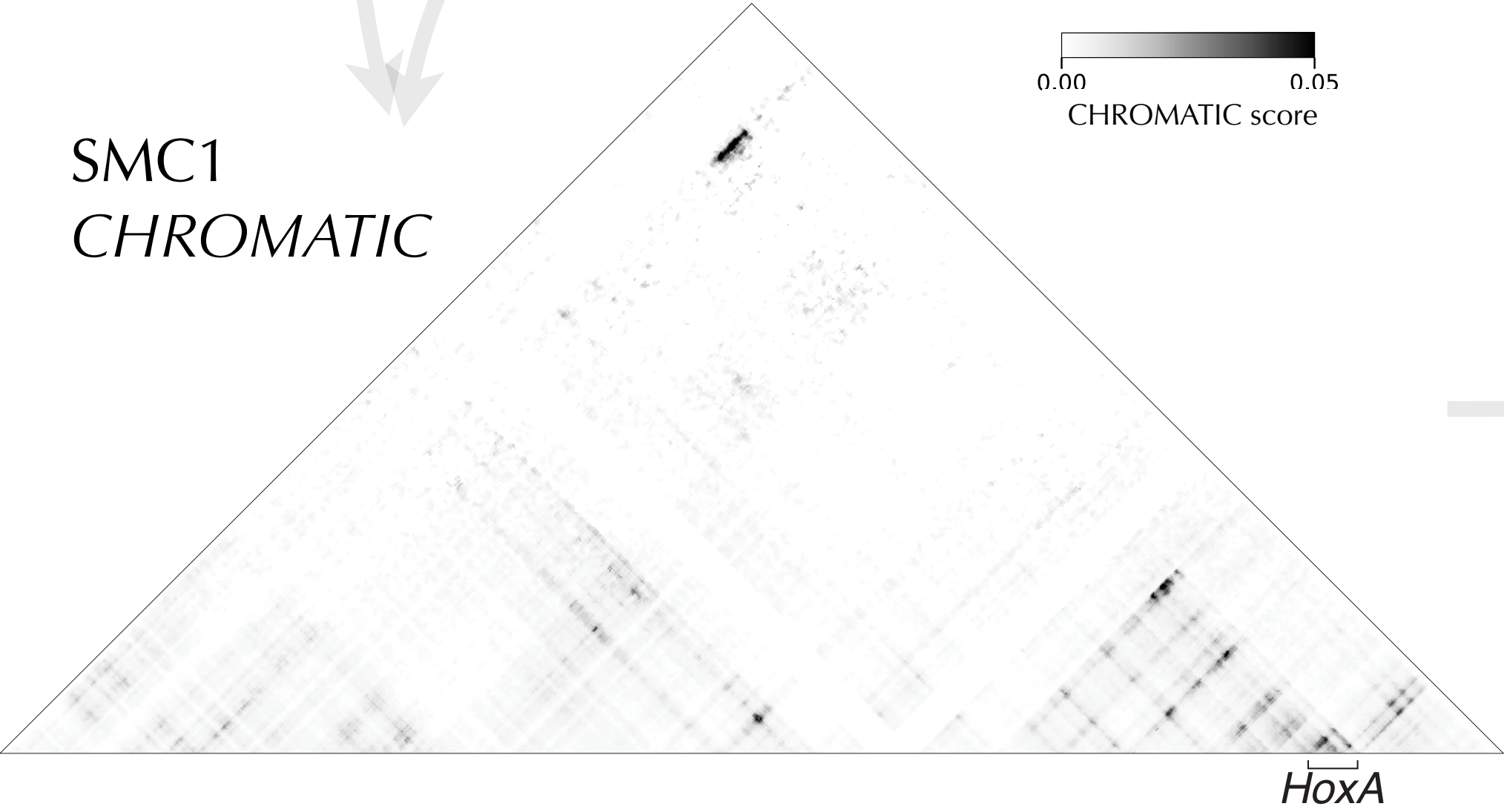
CHROMATIC
Factor Y

Morphological
image
processing

Loops/Hubs

CHROMATIC example

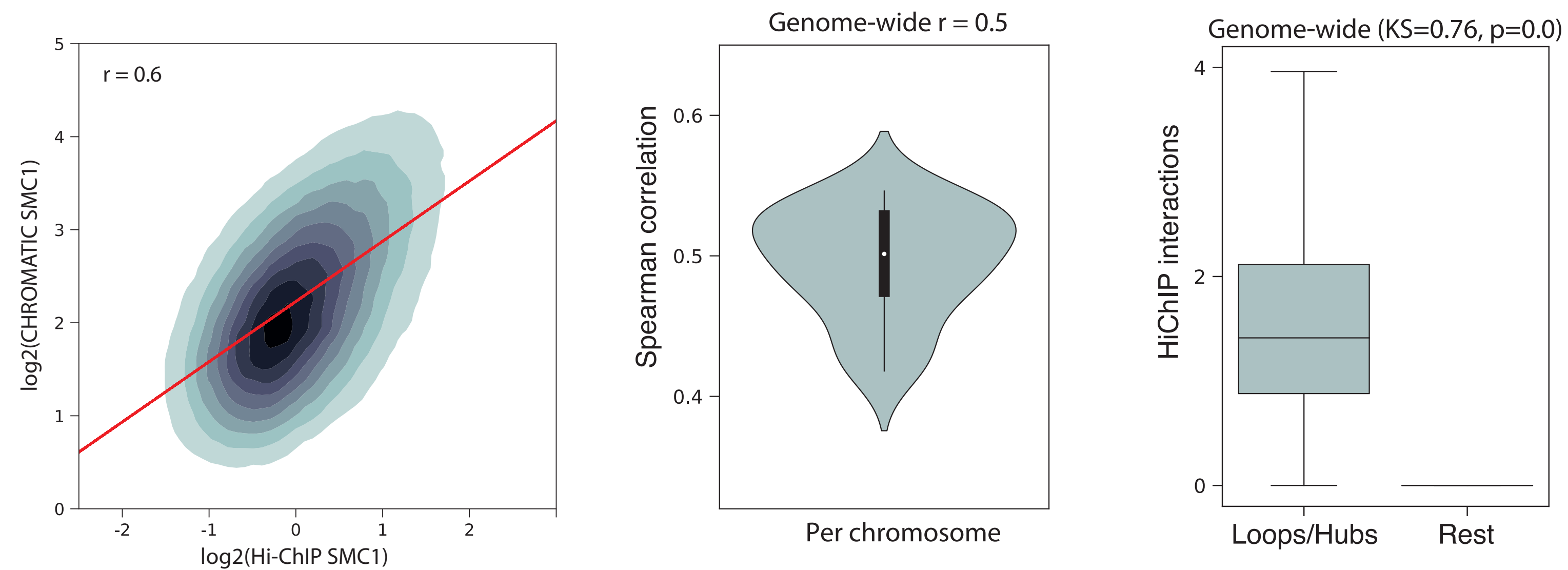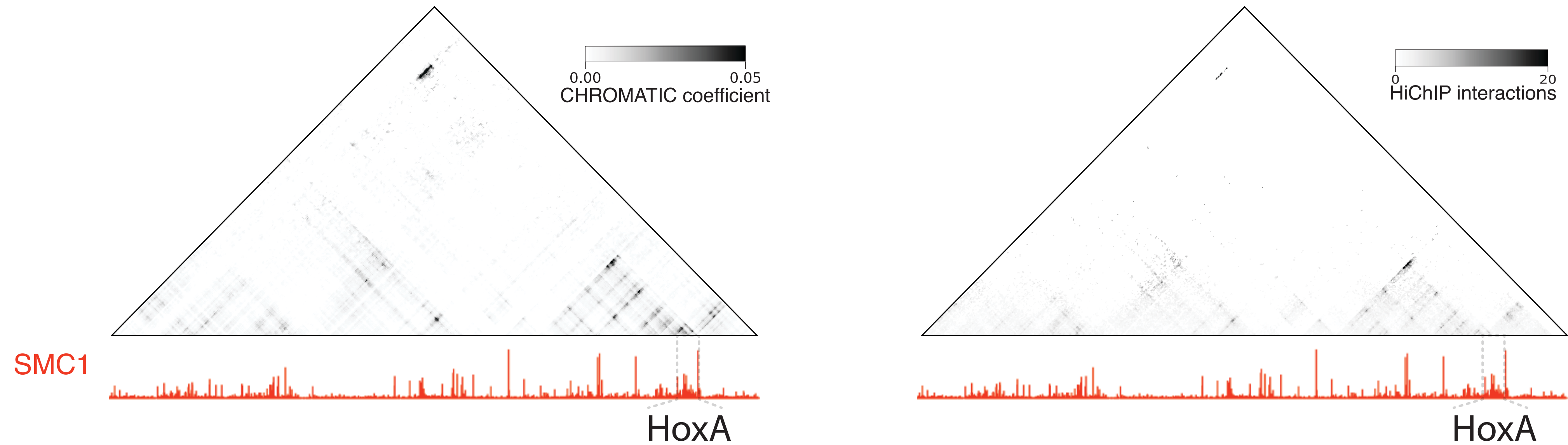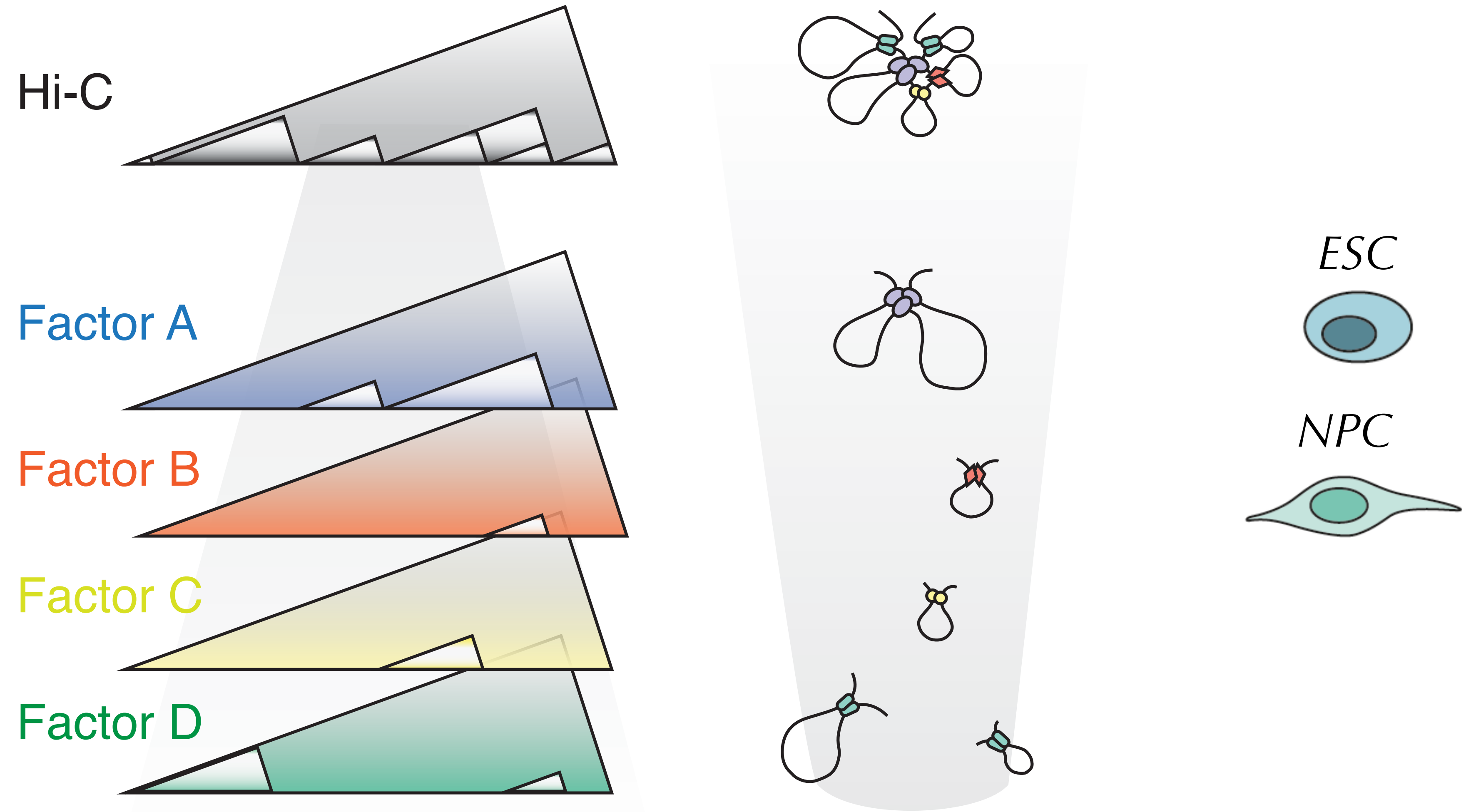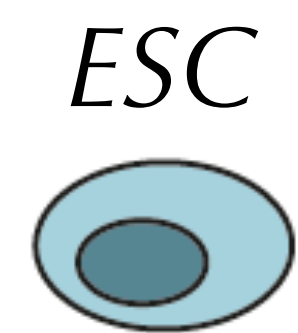mm10   chr6:48,050,001-52,750,006
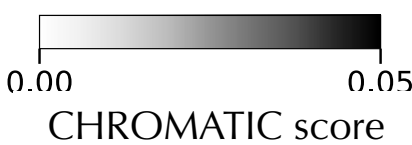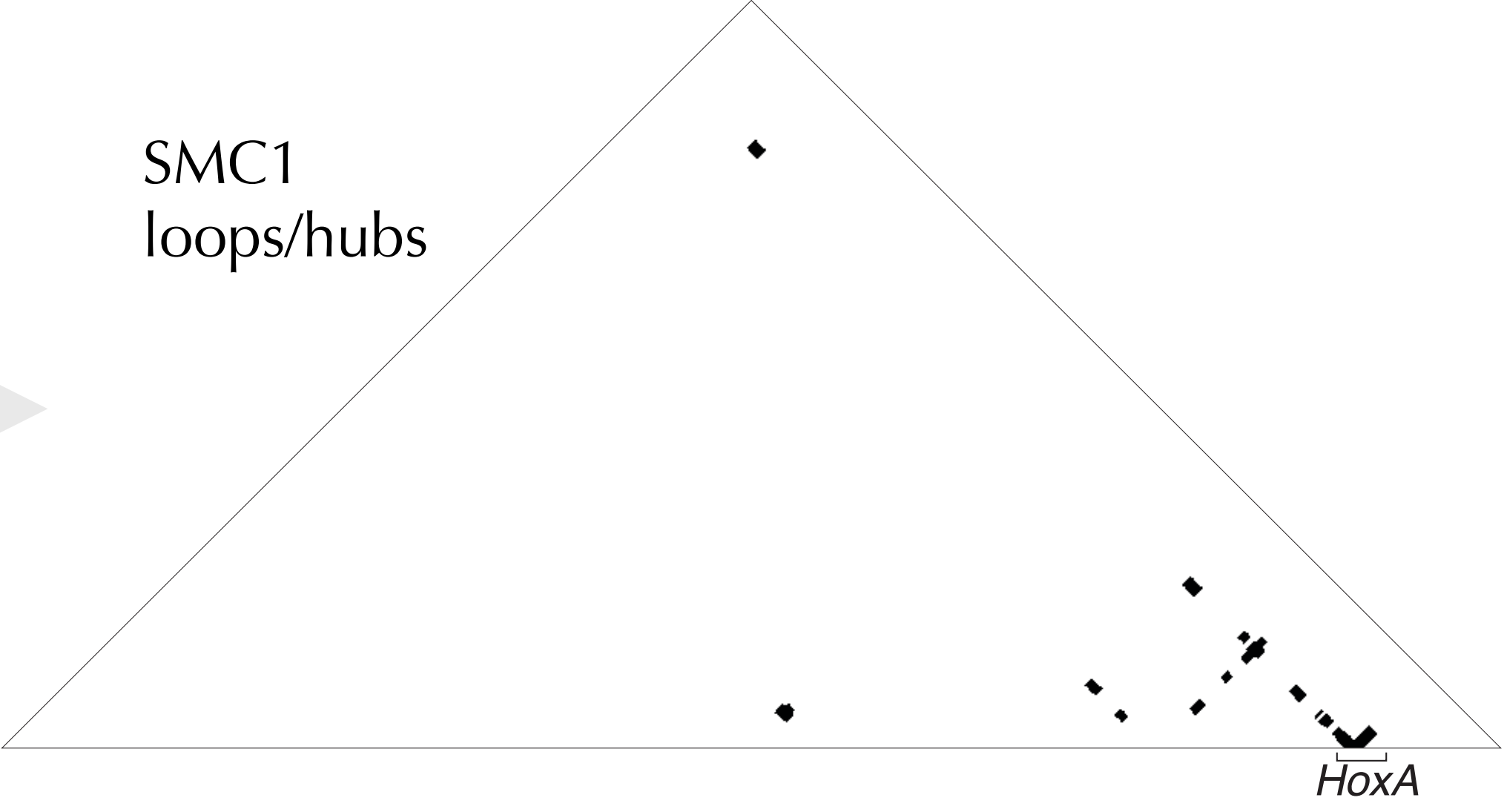
Validation with HiChIP as benchmark
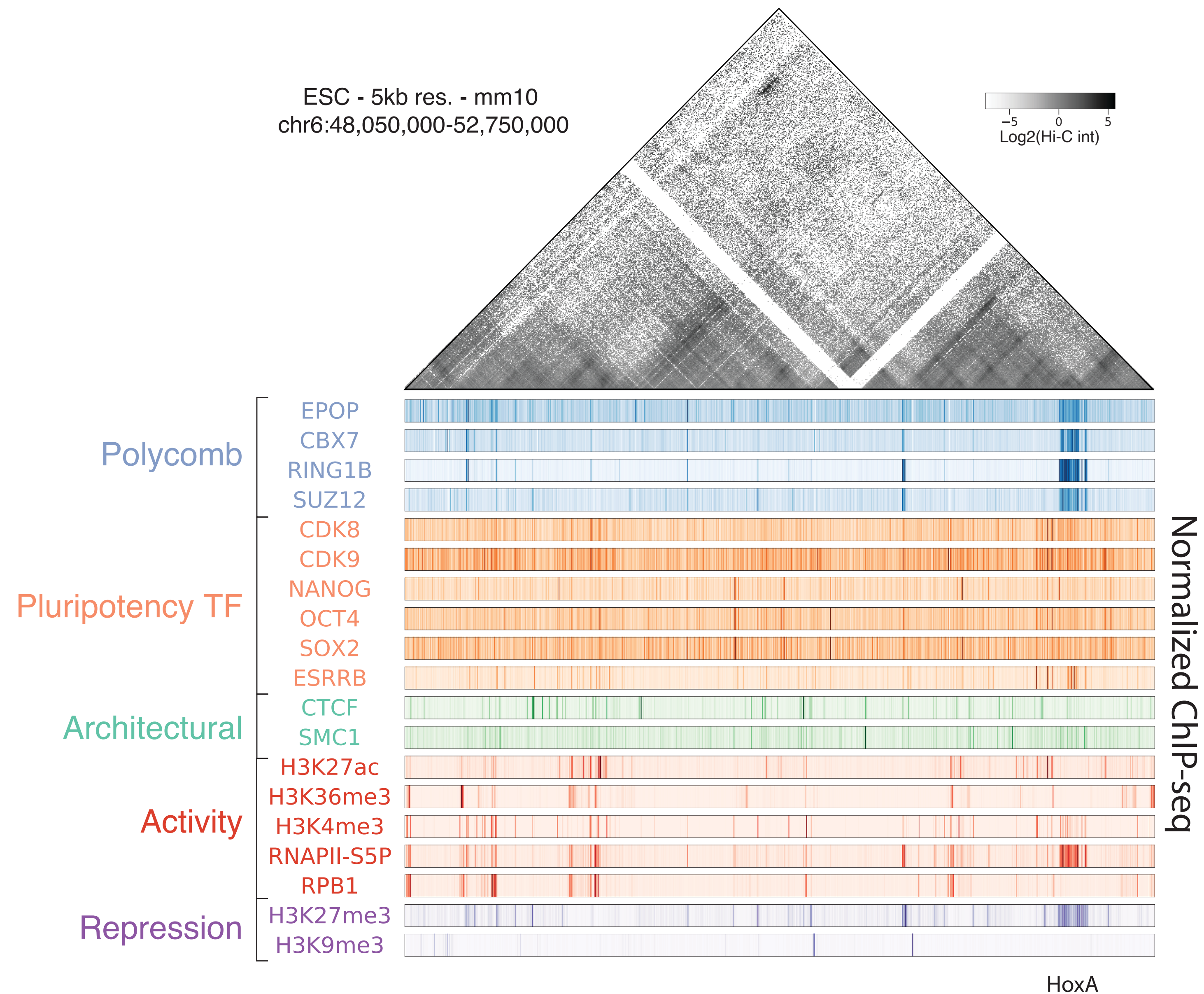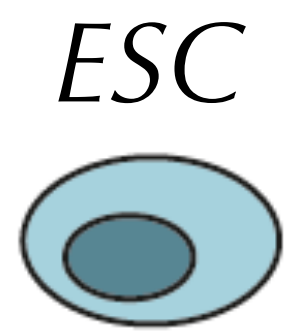
mm10 - mESC - 5kb - chr6:48,050,001-52,750,006
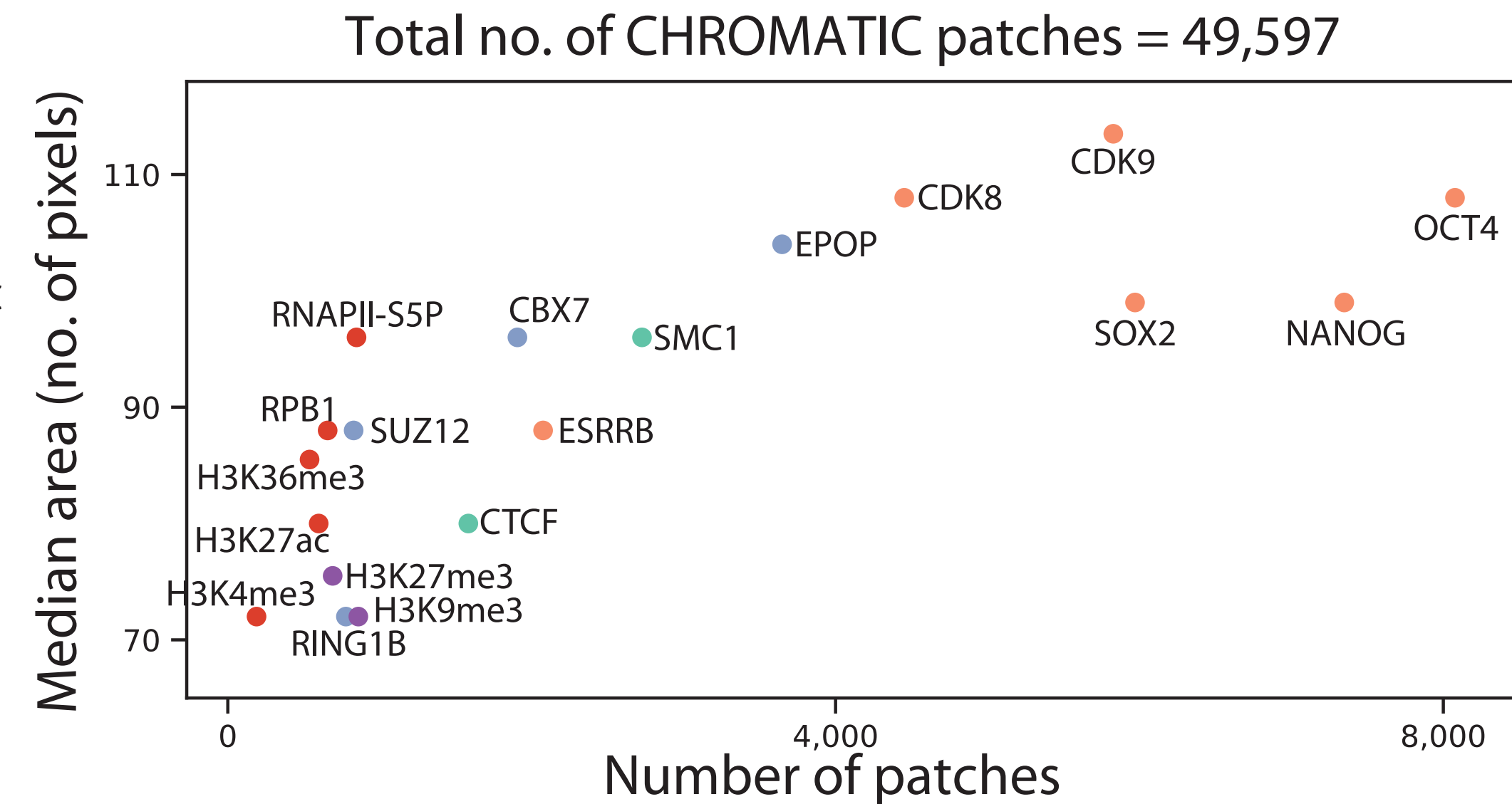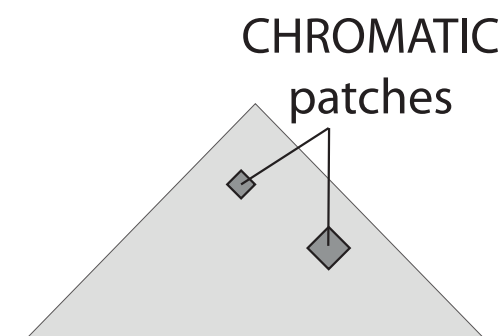
The role of chromatin factors in genome topology

HoxA cluster (example in ESC)

ESC

ESC - 5kb res. - mm10
chr6:48,050,000-52,750,000

Log2(Hi-C int)
−5   0   5

Normalized ChIP-seq

Polycomb
- EPOP
- CBX7
- RING1B
- SUZ12

Pluripotency TF
- CDK8
- CDK9
- NANOG
- OCT4
- SOX2
- ESRRB

Architectural
- CTCF
- SMC1

Activity
- H3K27ac
- H3K36me3
- H3K4me3
- RNAPII-S5P
- RPB1

Repression
- H3K27me3
- H3K9me3

HoxA

3D interactions are mostly associated with pluripotent TFs

Total no. of ChIP-seq peaks = 352,340

Total no. of CHROMATIC patches = 49,597
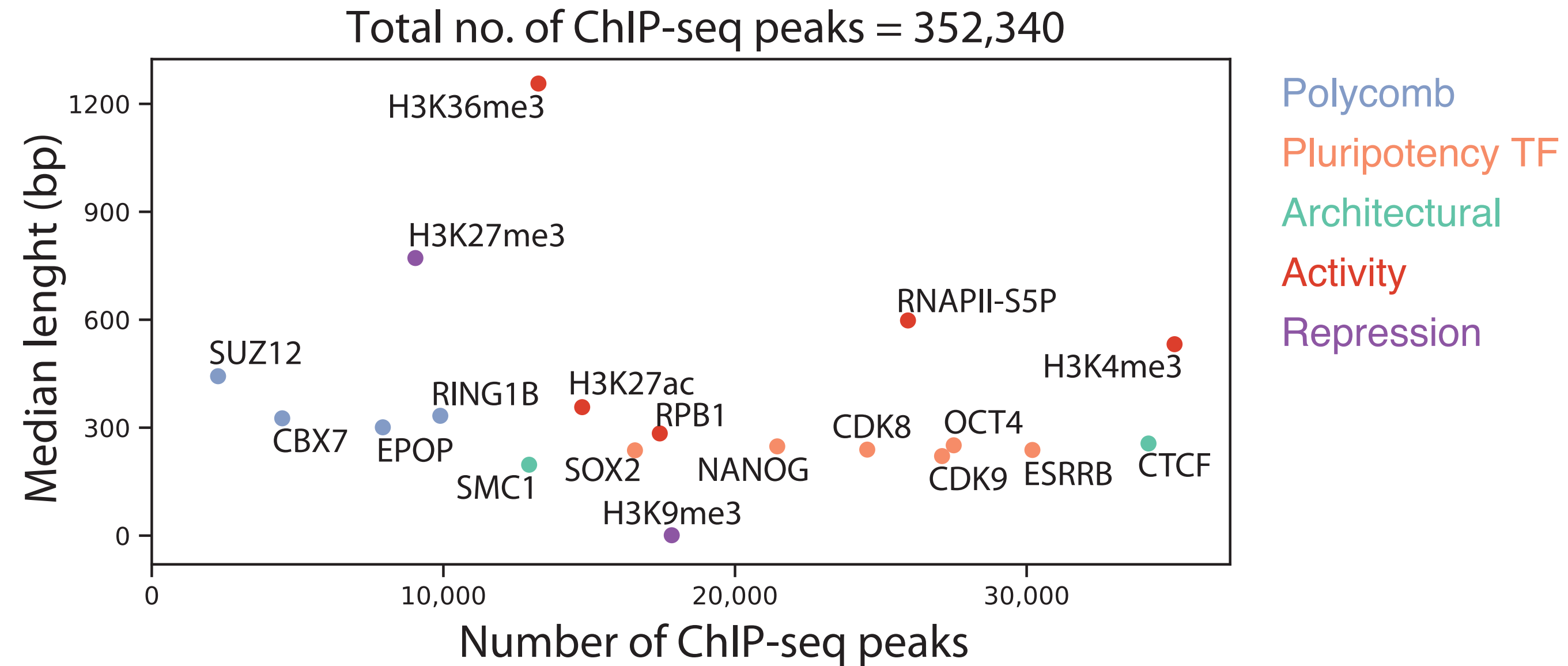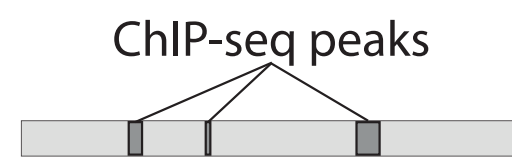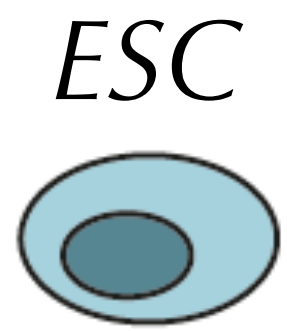
CHROMATIC better represents genome function than ChIP-seq alone
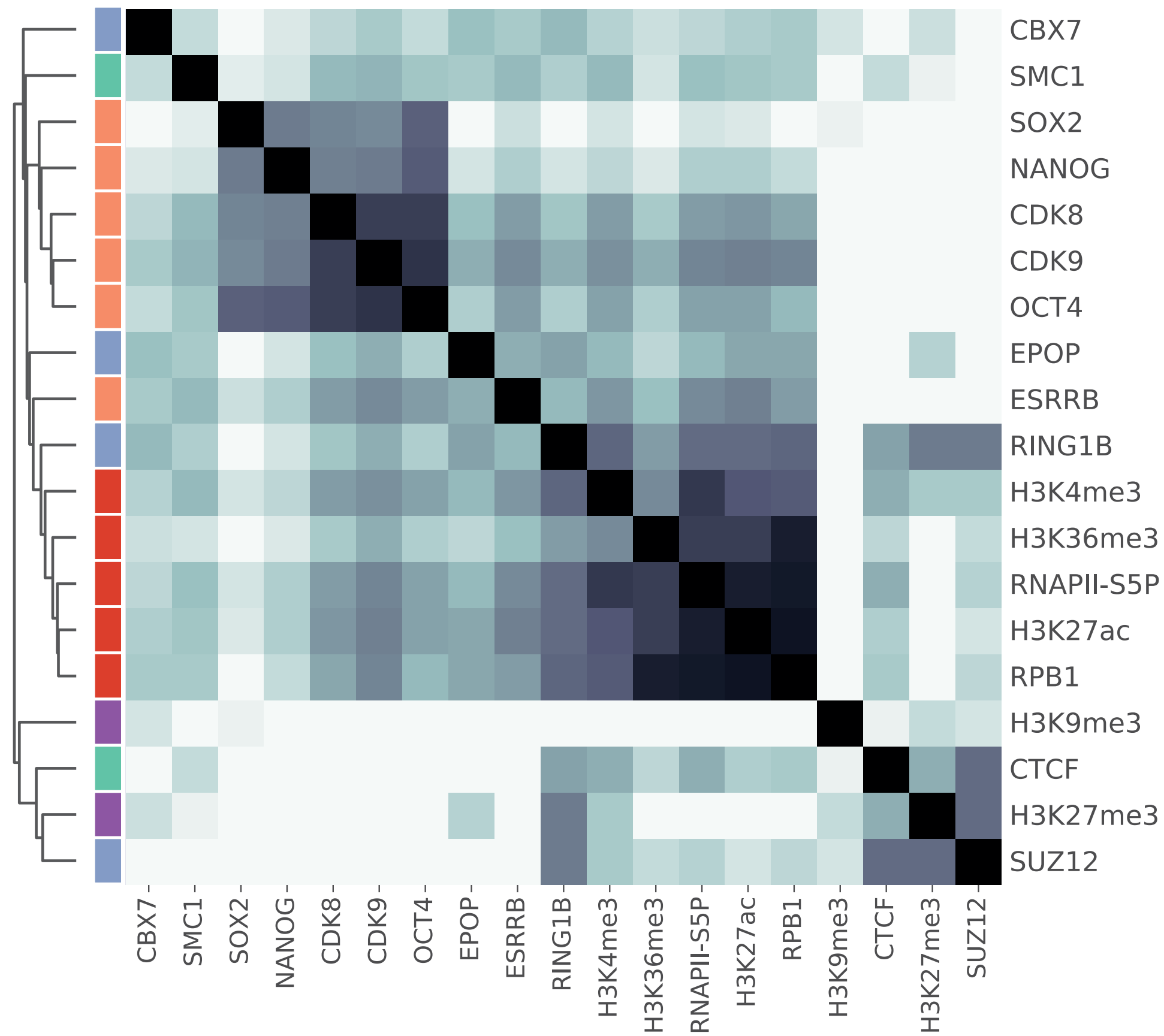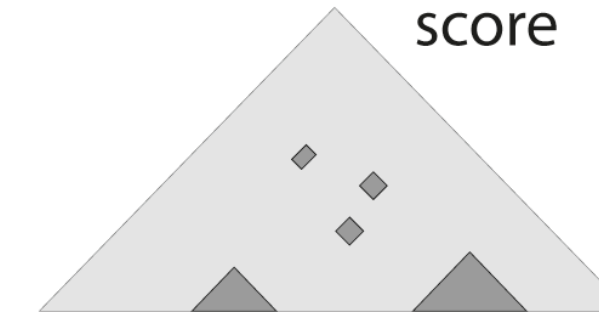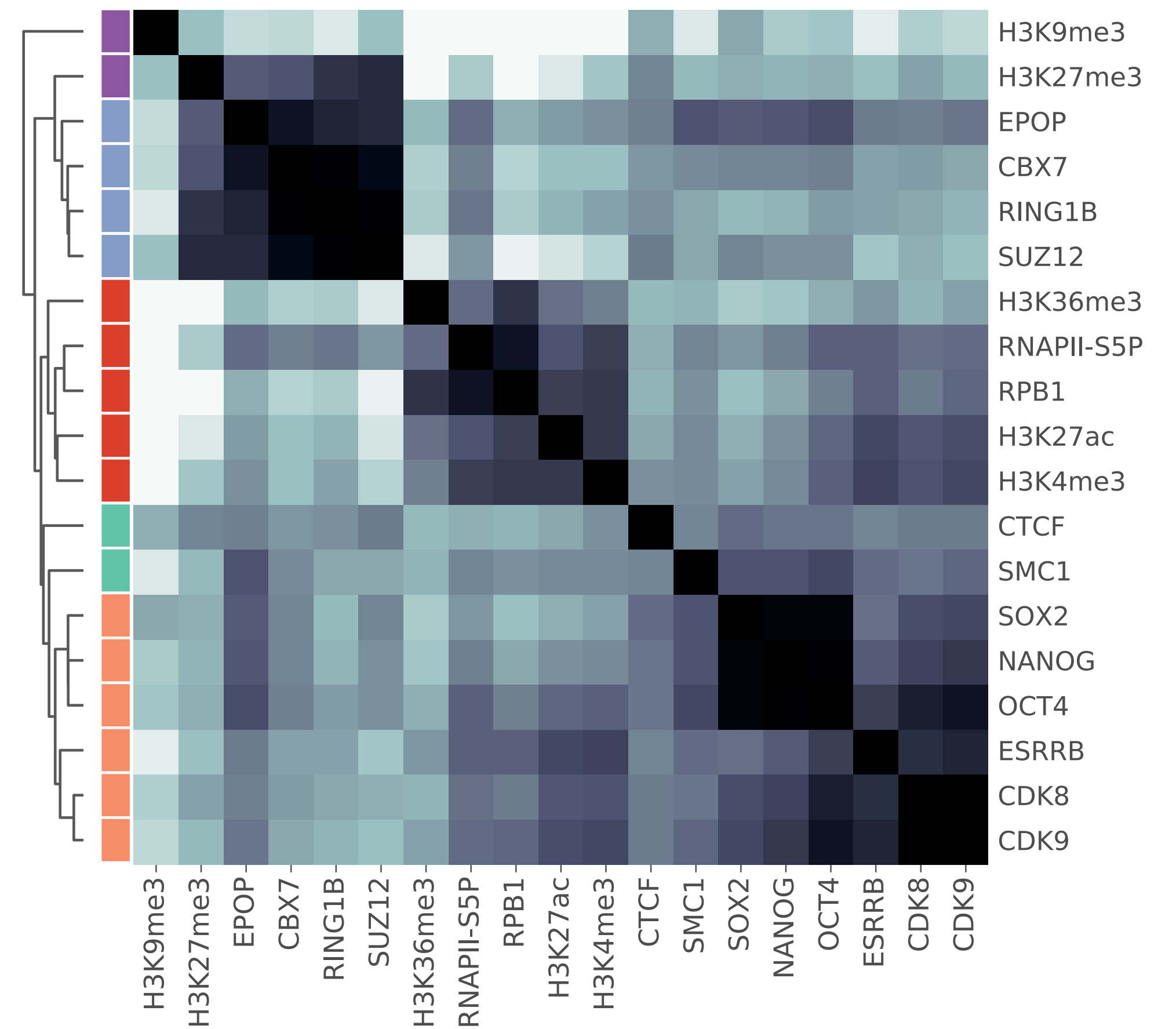
Measure the 3D co-localization of factors

ESC

CHROMATIC interactions

SUZ12

19 factors

...

CTCF

OCT4

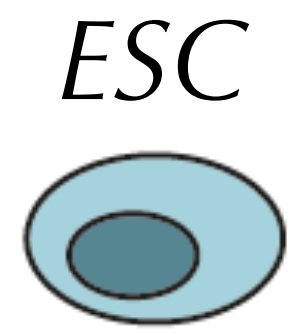chromosome
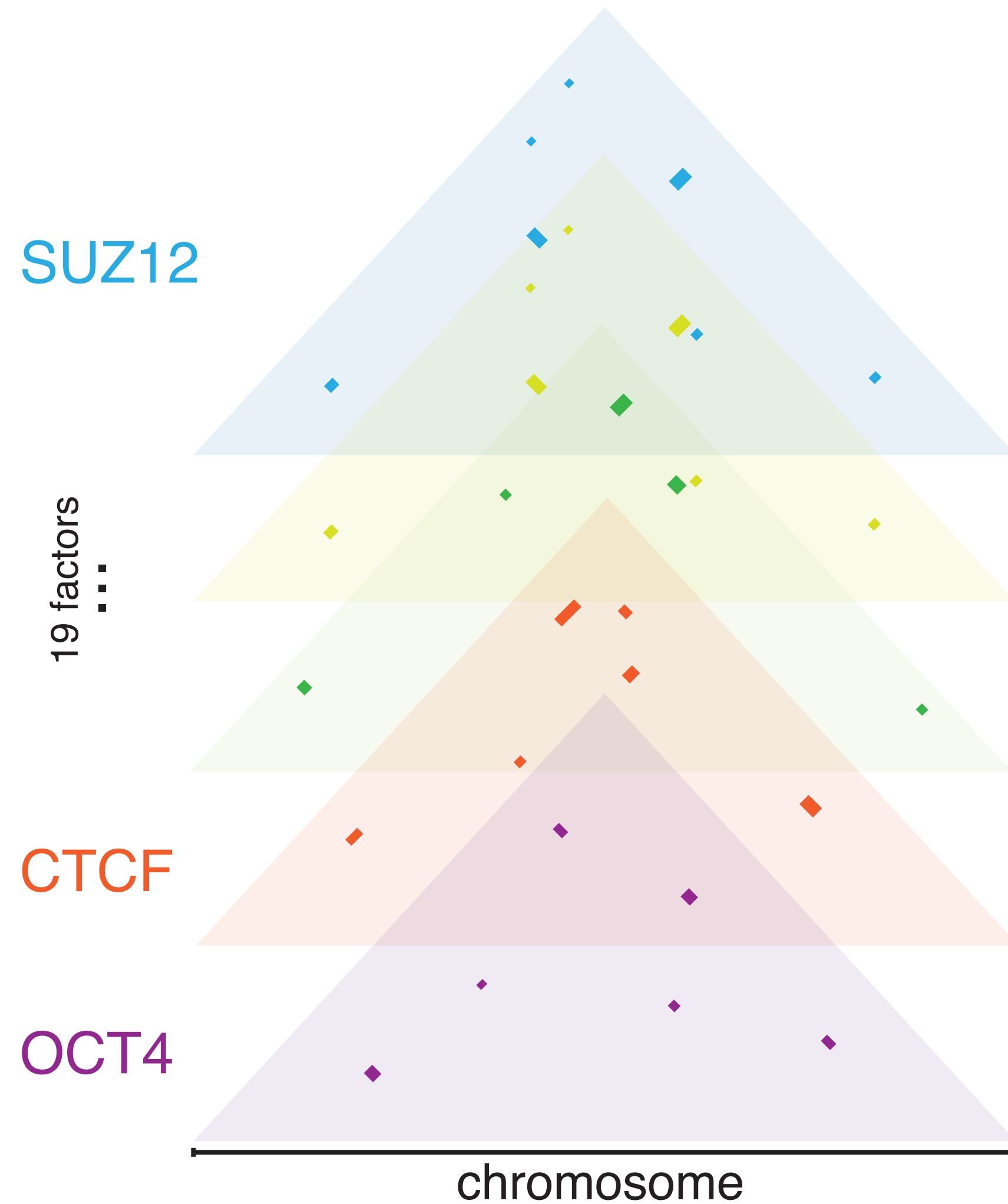
Overlap (19 factors)
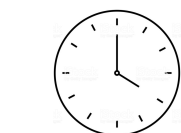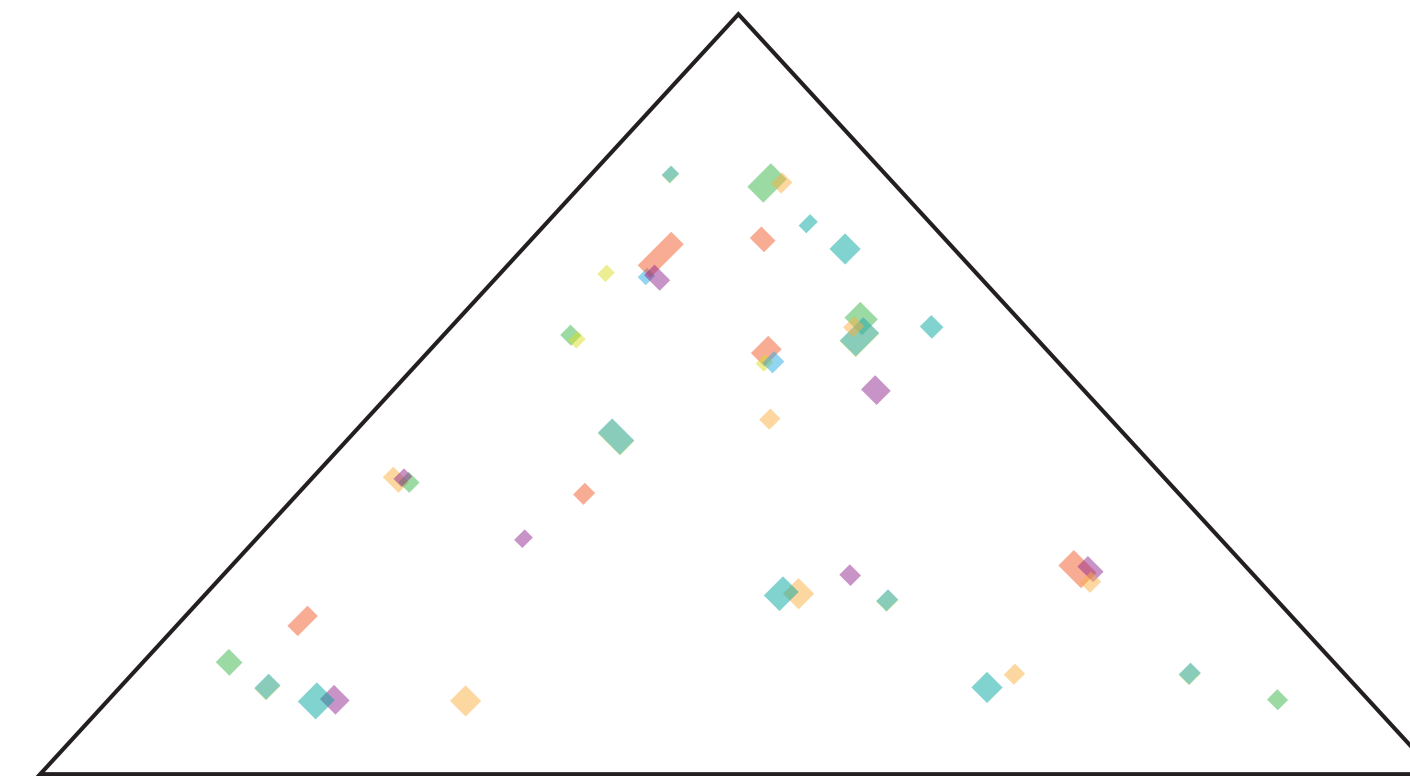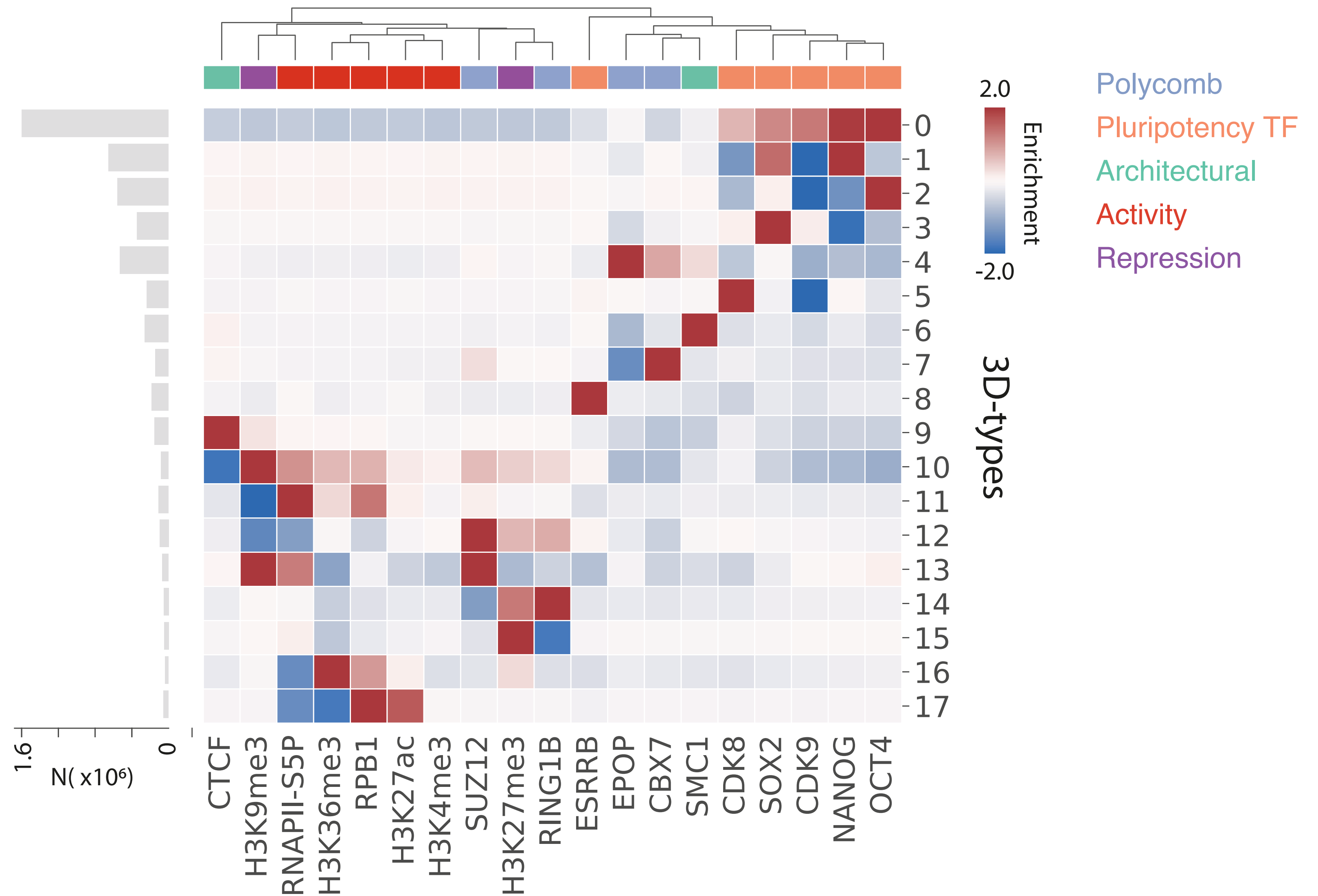
8m 54s
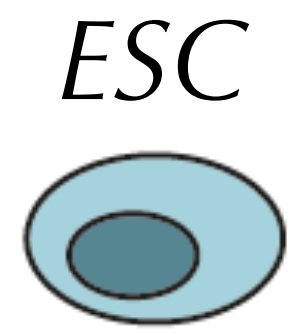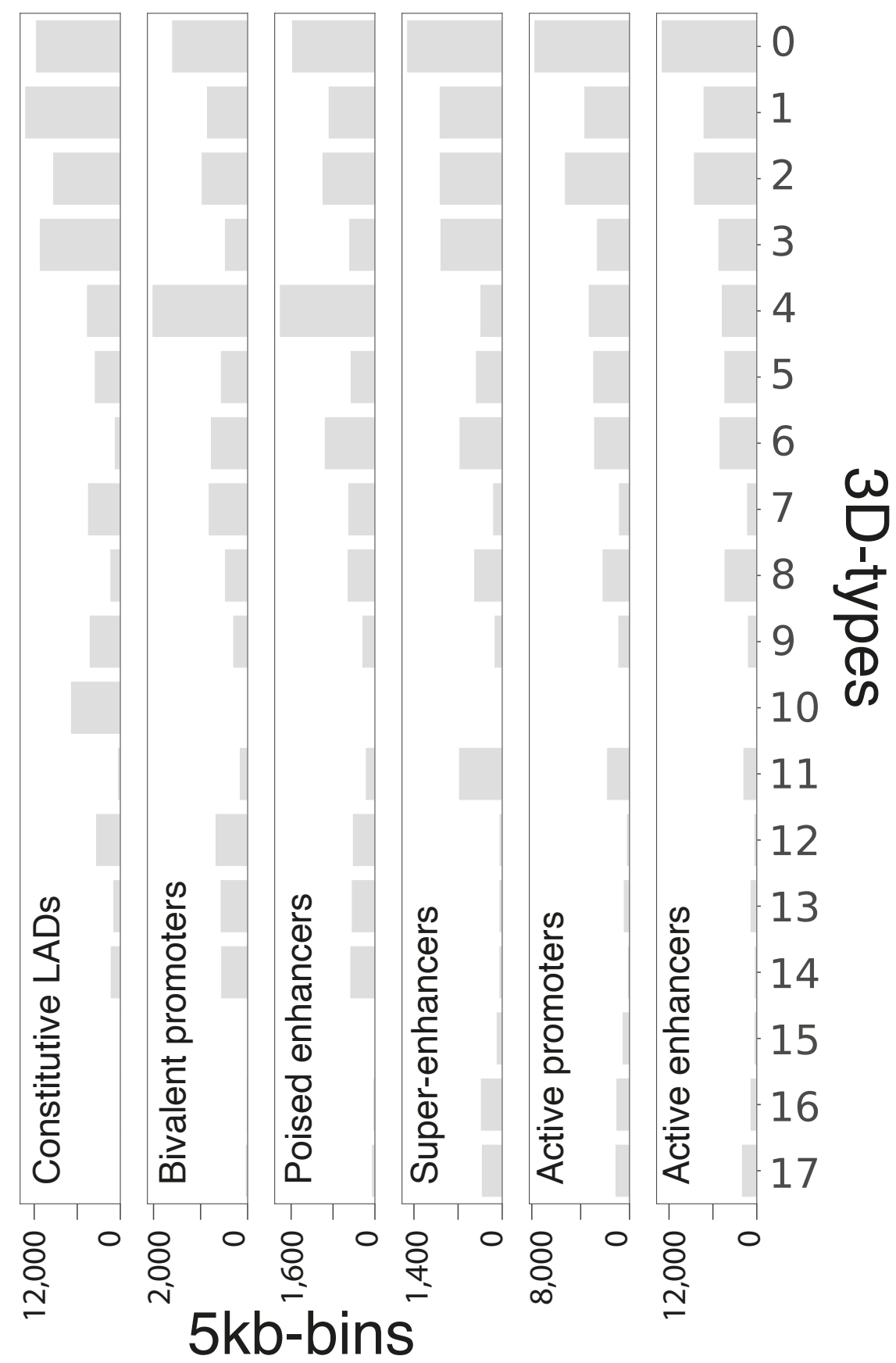
Latent Semantic Analysis

Documents

Doc1: CBX7   NANOG

Doc2: OCT4   CDK8   H3K27ac

Doc3: RING1B

Doc4: H3K36me3   RNAPolIII-S5P

...

...

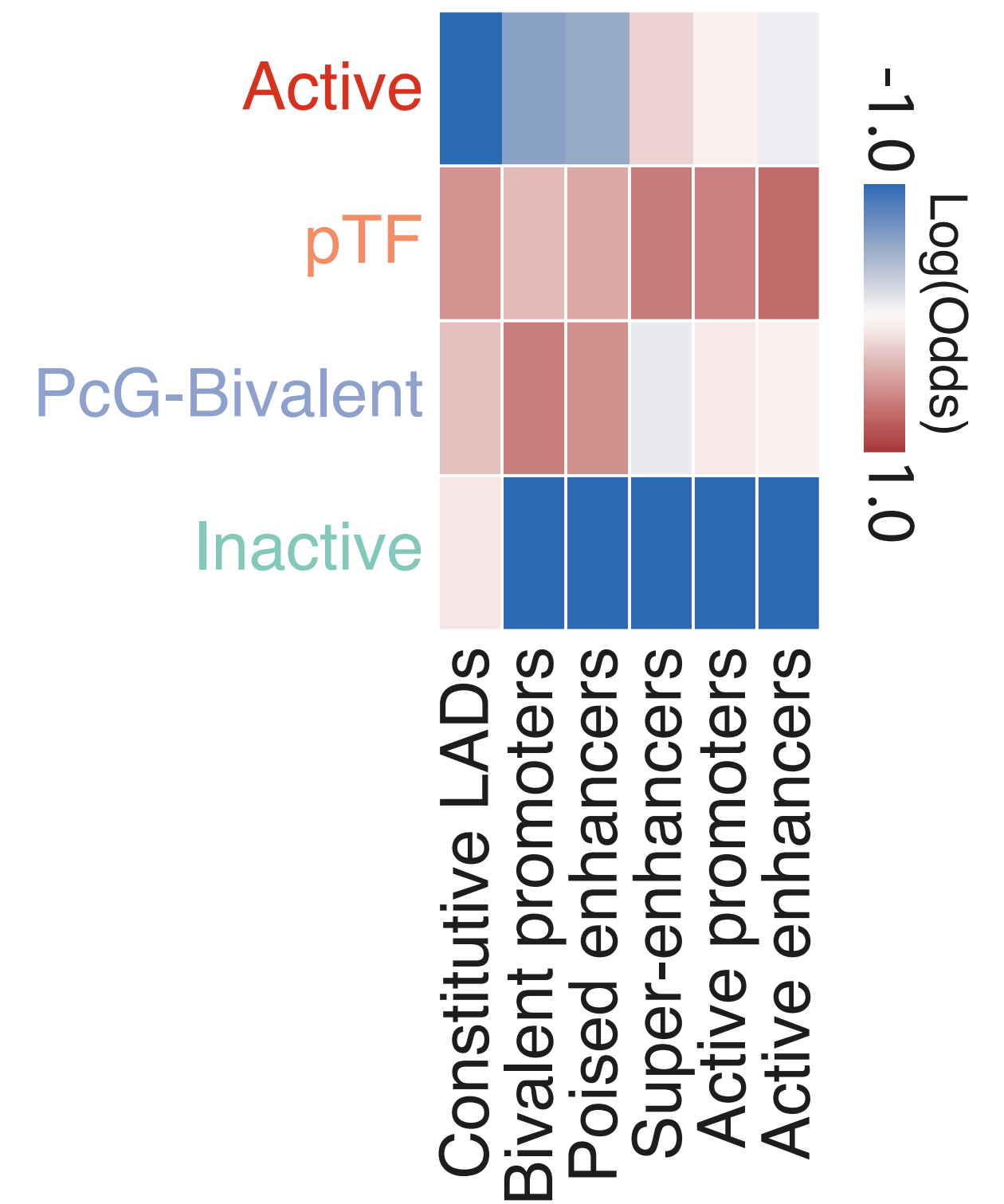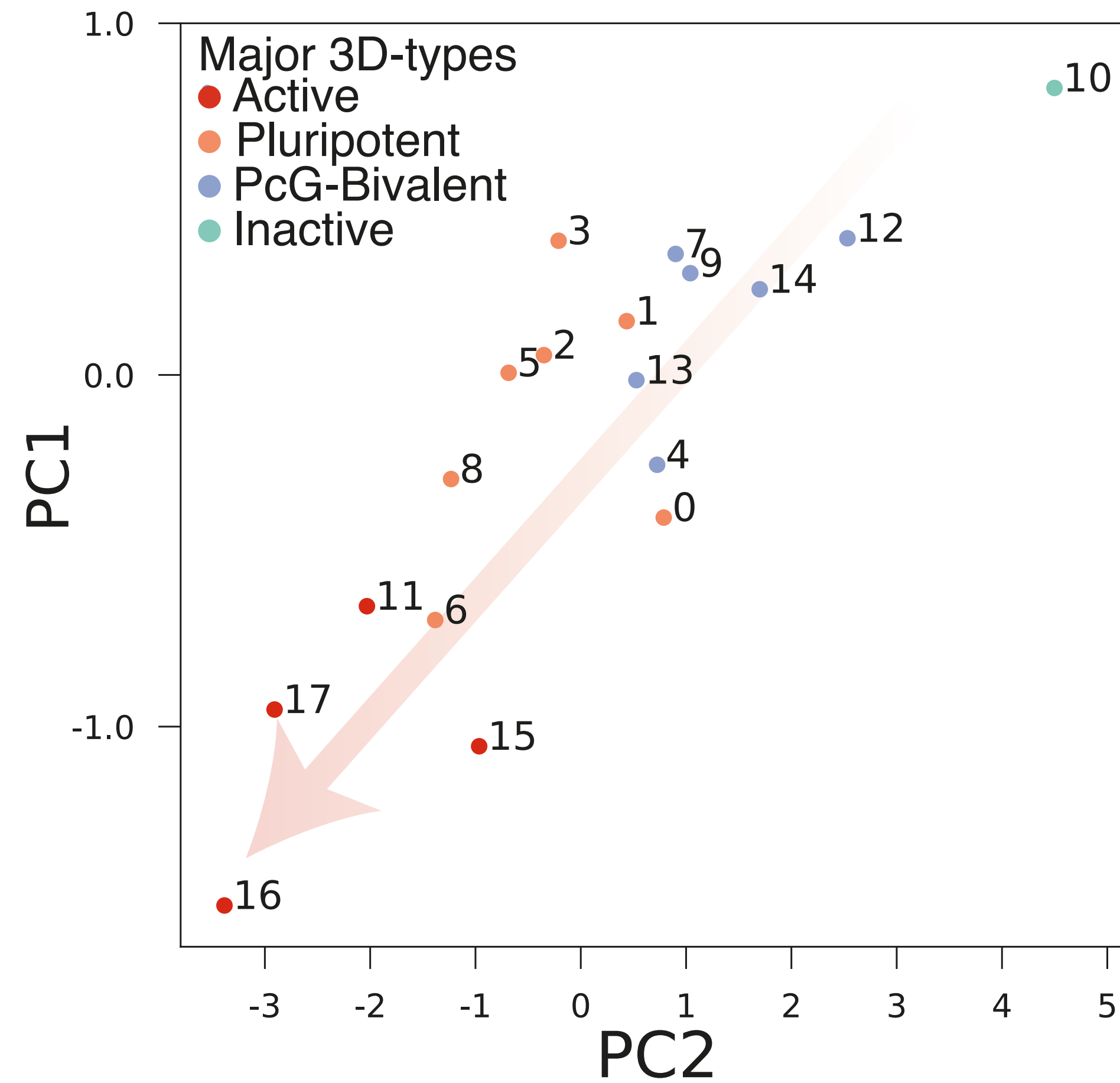# Types of 3D interactions with LSA in ESC
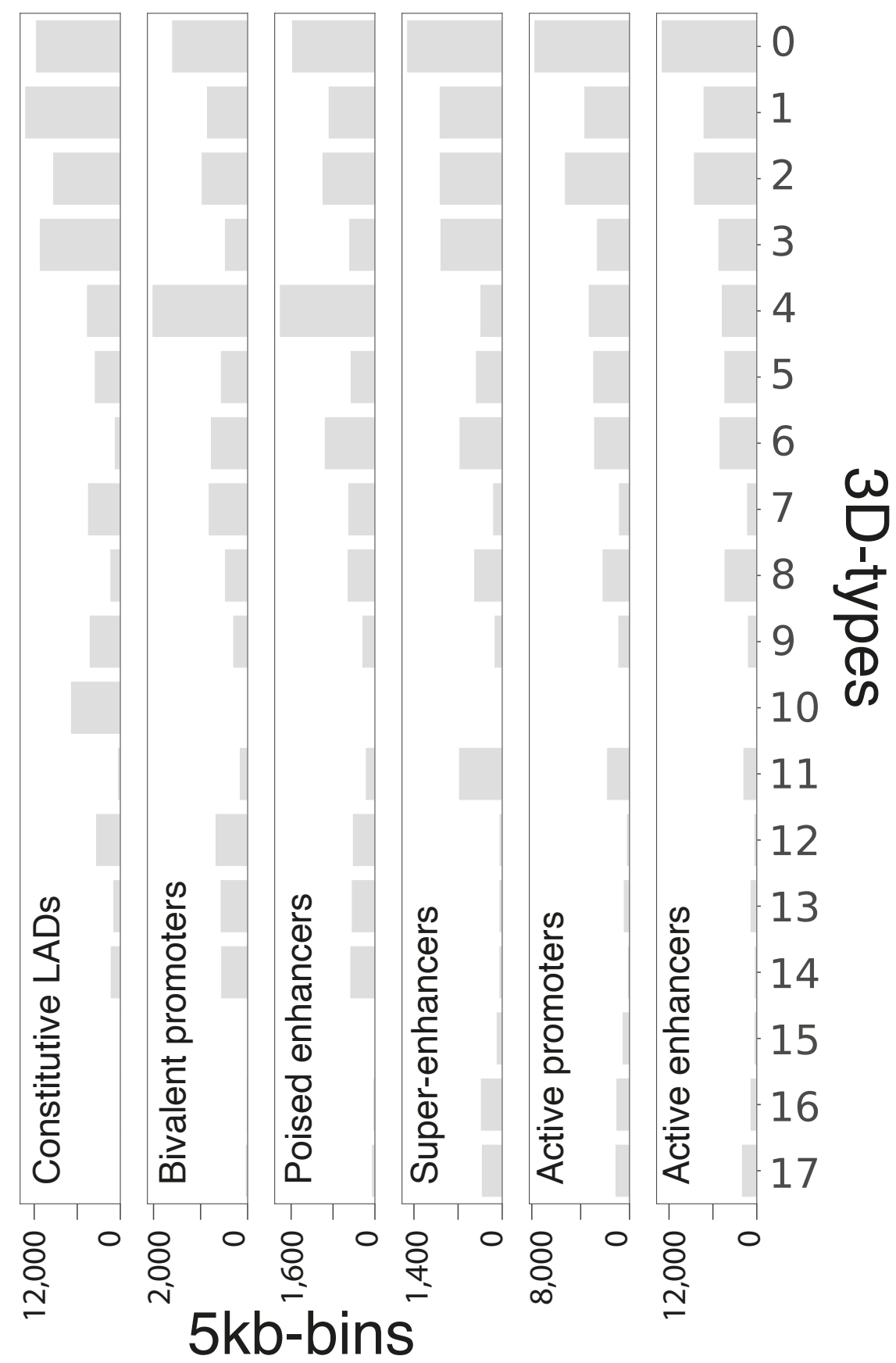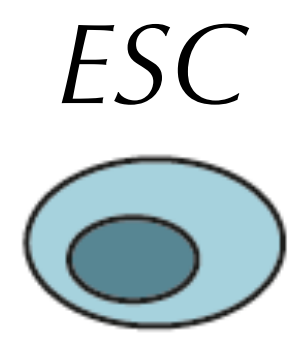
Types of 3D interactions with LSA in ESC

ESC

Annotated using ChIP-Seq and literature

3D-types

5kb-bins

Constitutive LADs
Bivalent promoters
Poised enhancers
Super-enhancers
Active promoters
Active enhancers

Major 3D-types
- Active
- Pluripotent
- PcG-Bivalent
- Inactive

PC1

PC2

Log(Odds)

Active
pTF
PcG-Bivalent
Inactive

Constitutive LADs
Bivalent promoters
Poised enhancers
Super-enhancers
Active promoters
Active enhancers

4 major types of 3D interactions in ESC
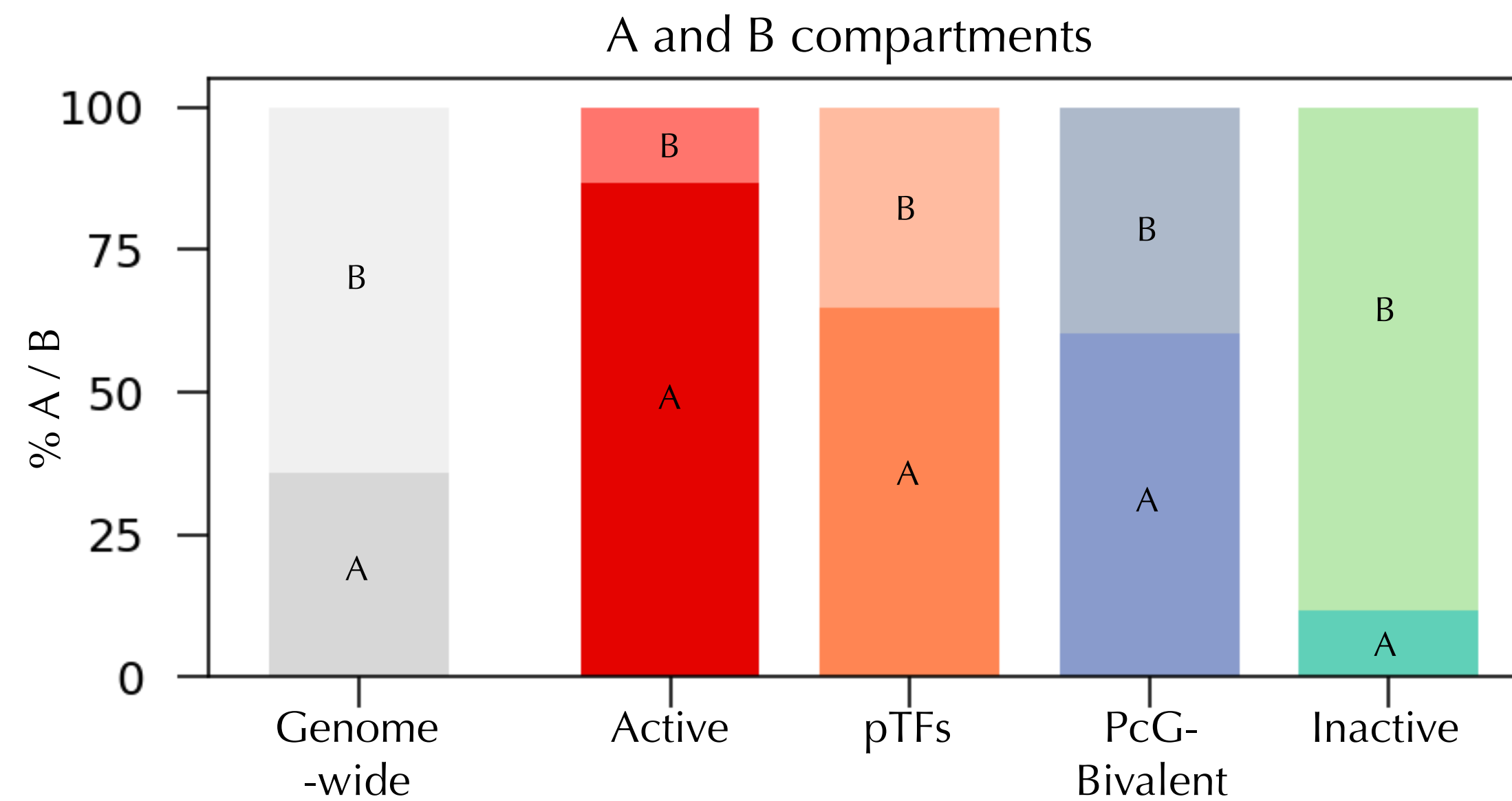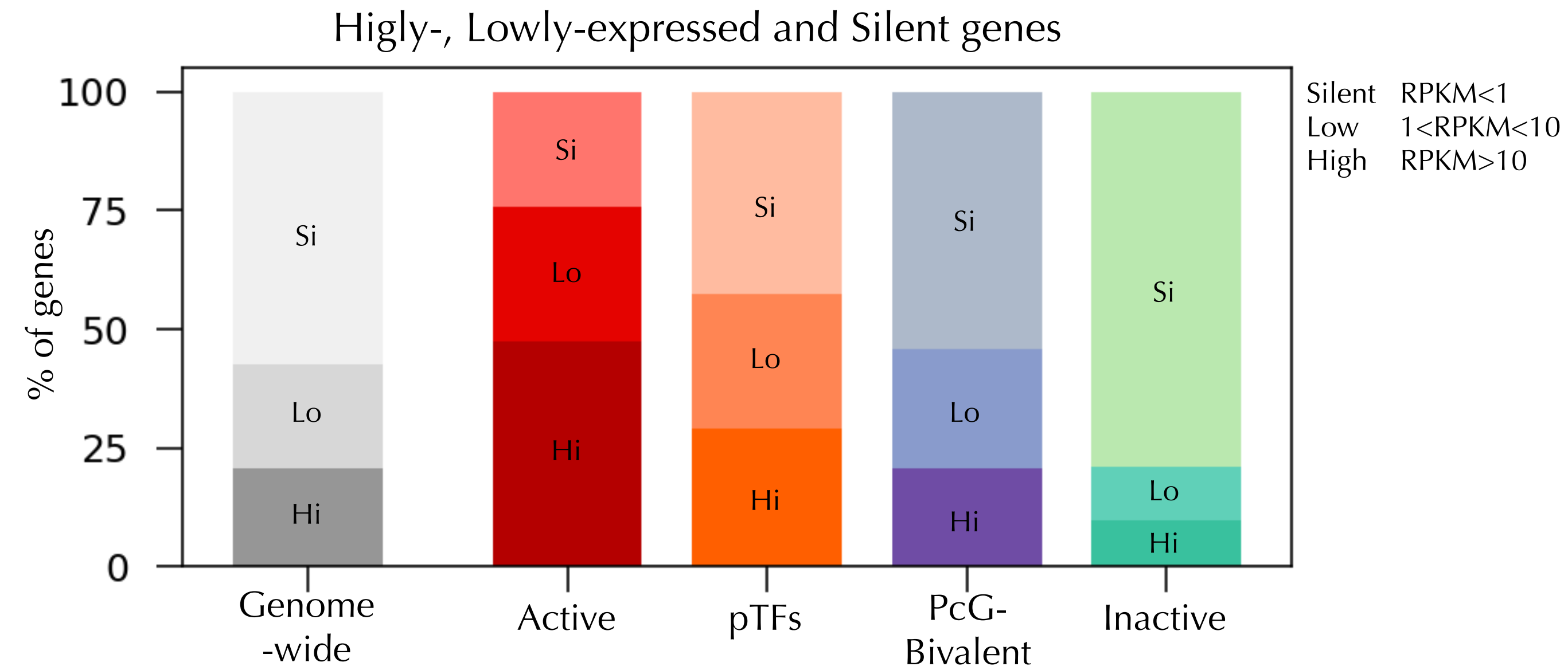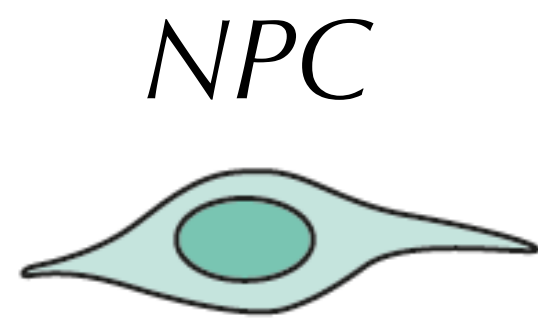
HoxA cluster (example in NPC)

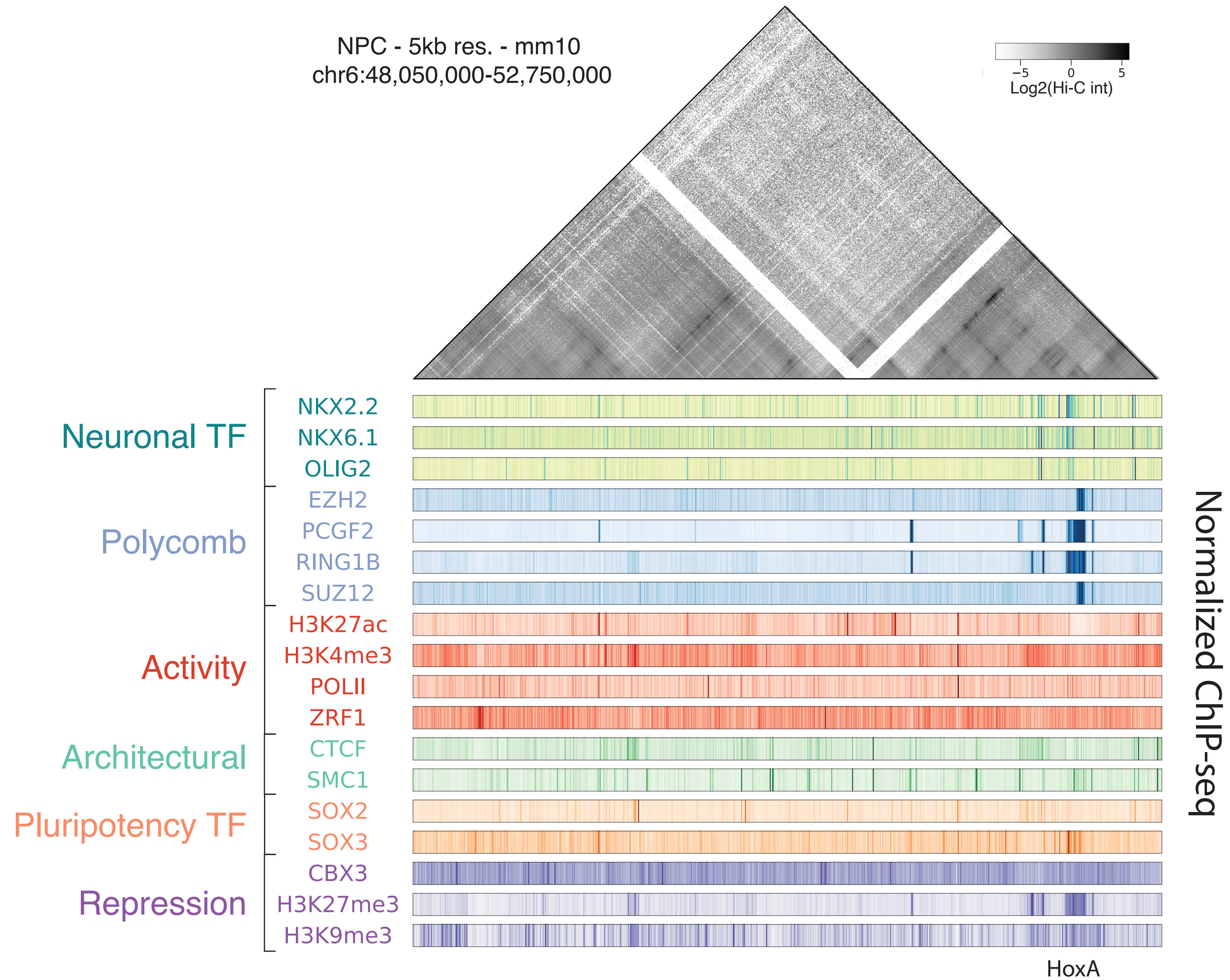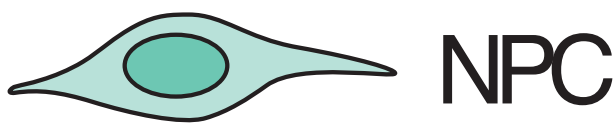# Global 3D interactions rewiring during mouse development
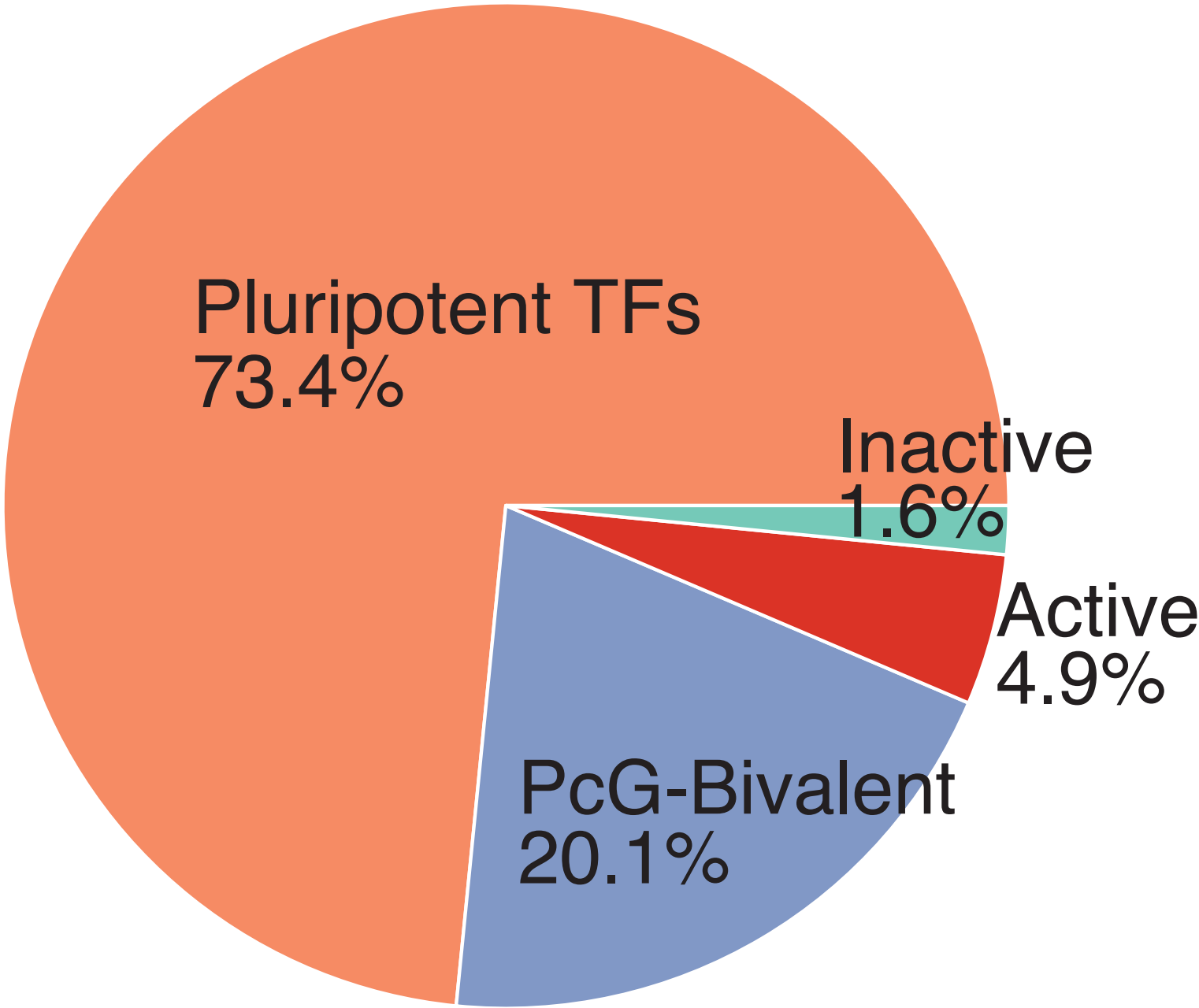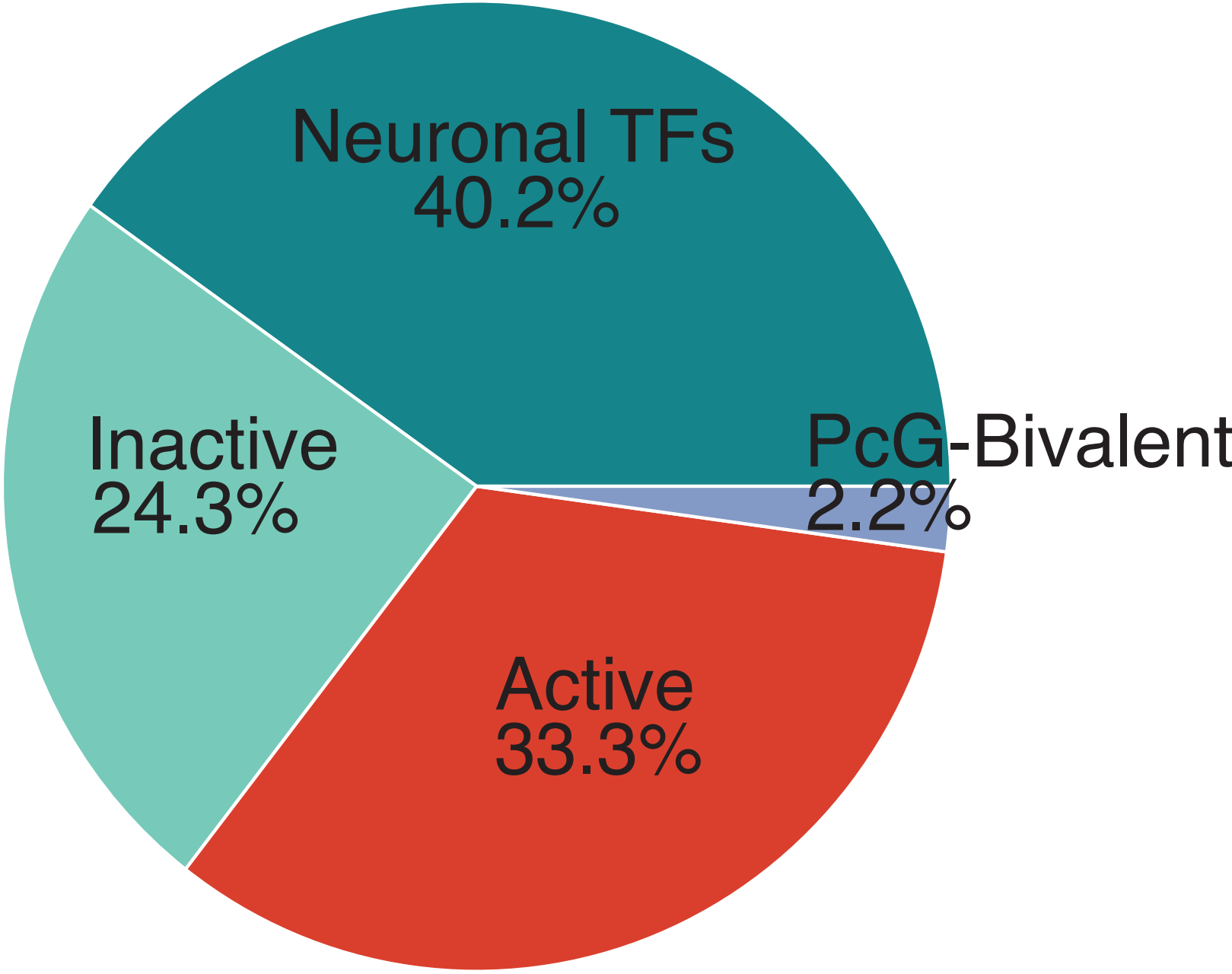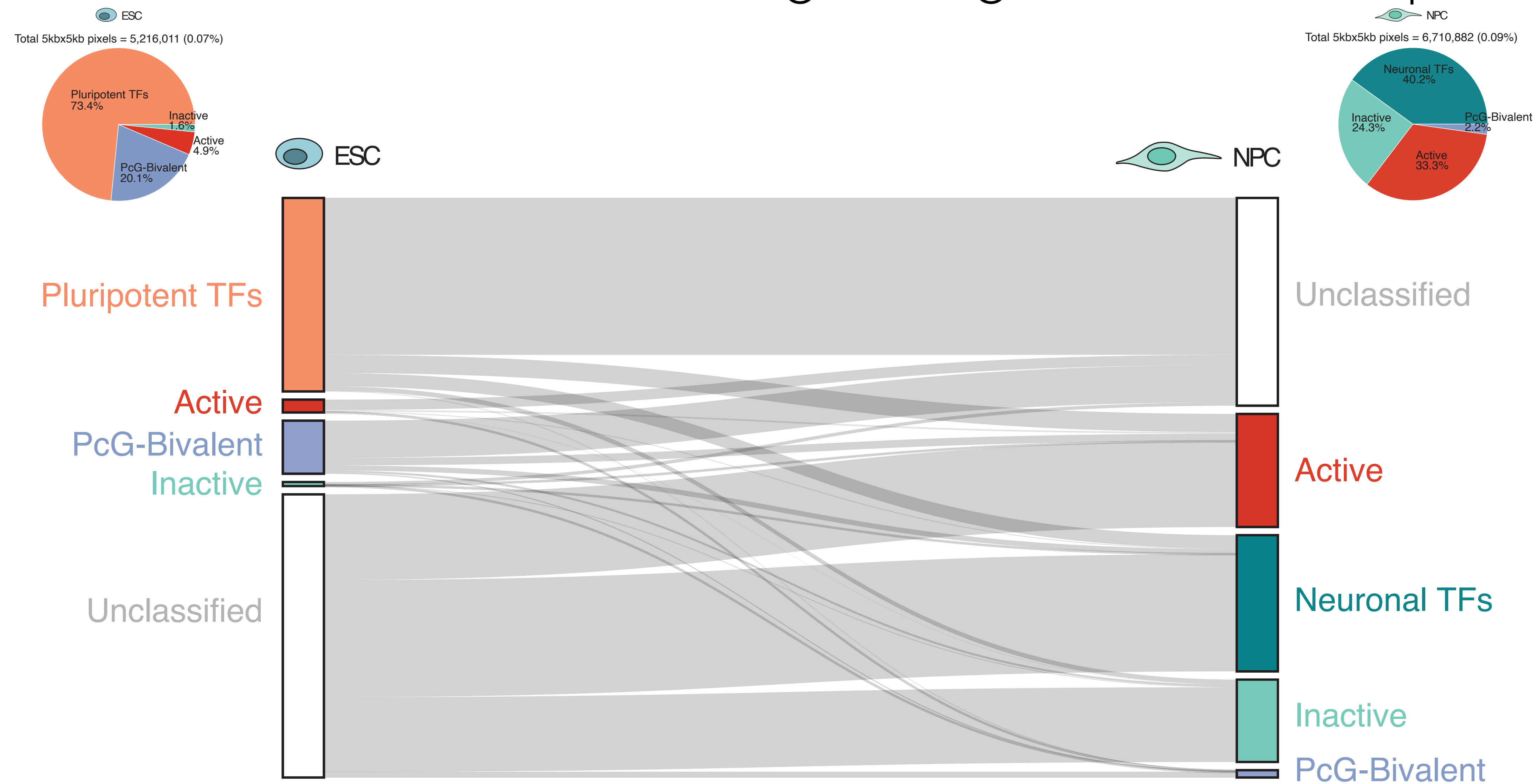
Global 3D interactions rewiring during mouse development

ESC

Total 5kbx5kb pixels = 5,216,011 (0.07%)

Pluripotent TFs
73.4%

Inactive
1.6%

Active
4.9%

PcG-Bivalent
20.1%

NPC

Total 5kbx5kb pixels = 6,710,882 (0.09%)

Neuronal TFs
40.2%

PcG-Bivalent
2.2%

Inactive
24.3%

Active
33.3%

ESC

NPC

Pluripotent TFs

Active

PcG-Bivalent

Inactive

Unclassified

Unclassified

Active

Neuronal TFs

Inactive

PcG-Bivalent

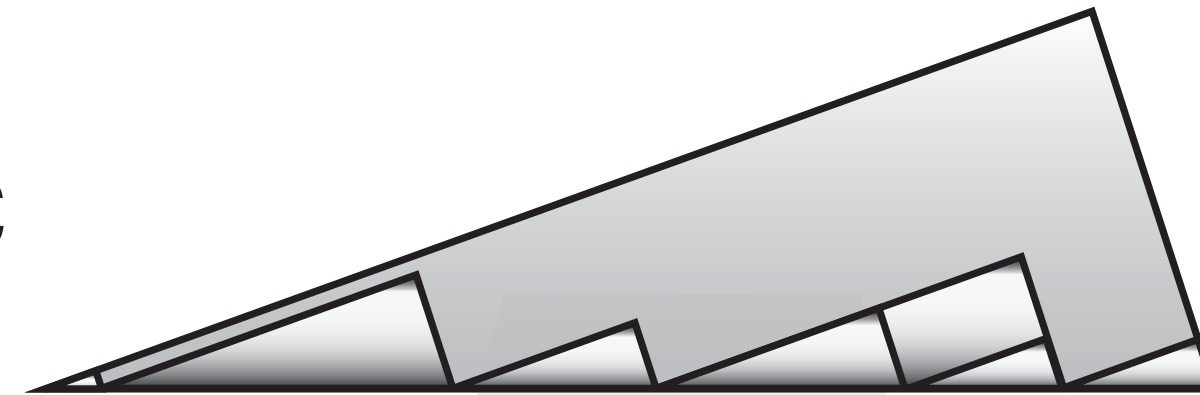# Global 3D interactions rewiring during mouse development
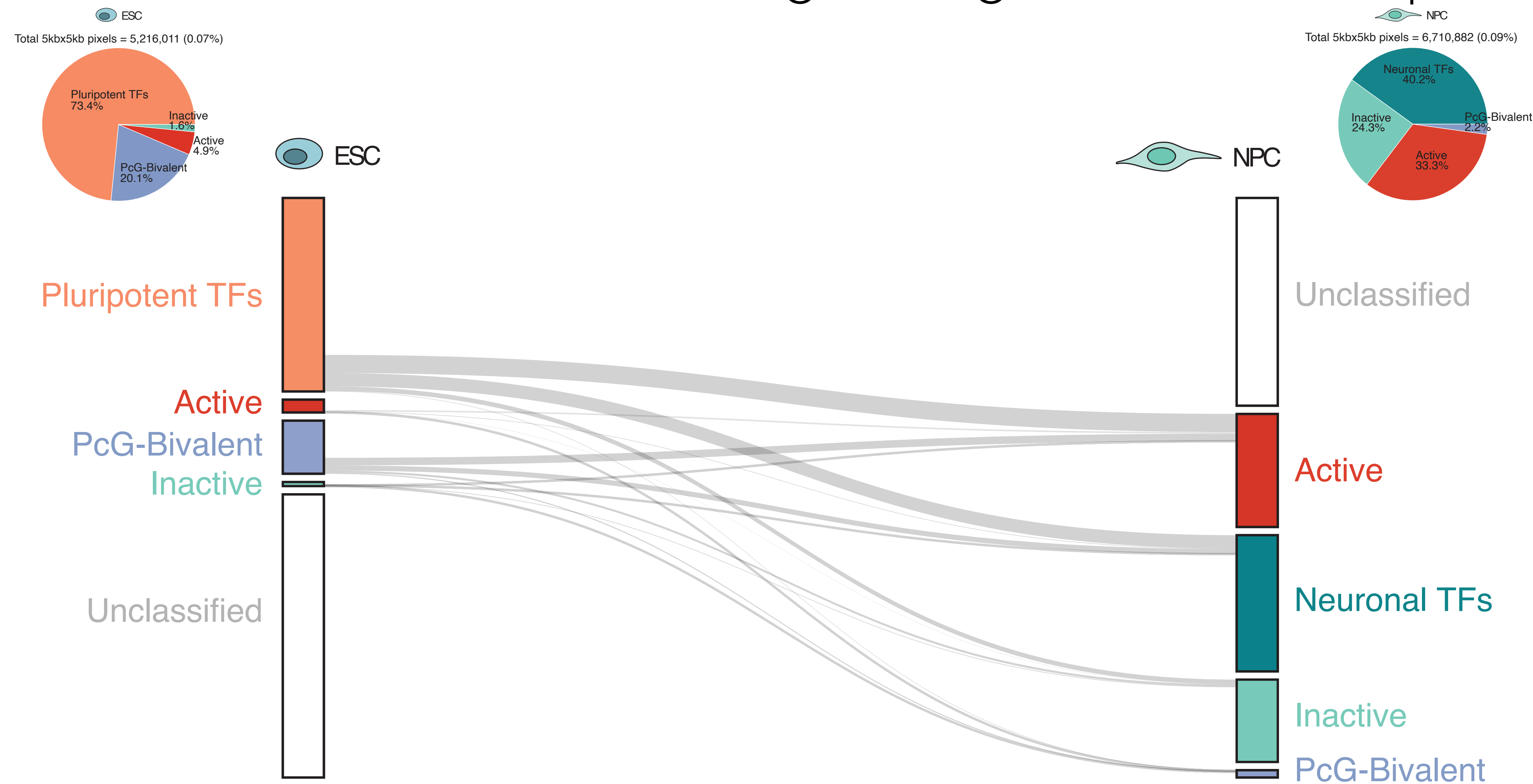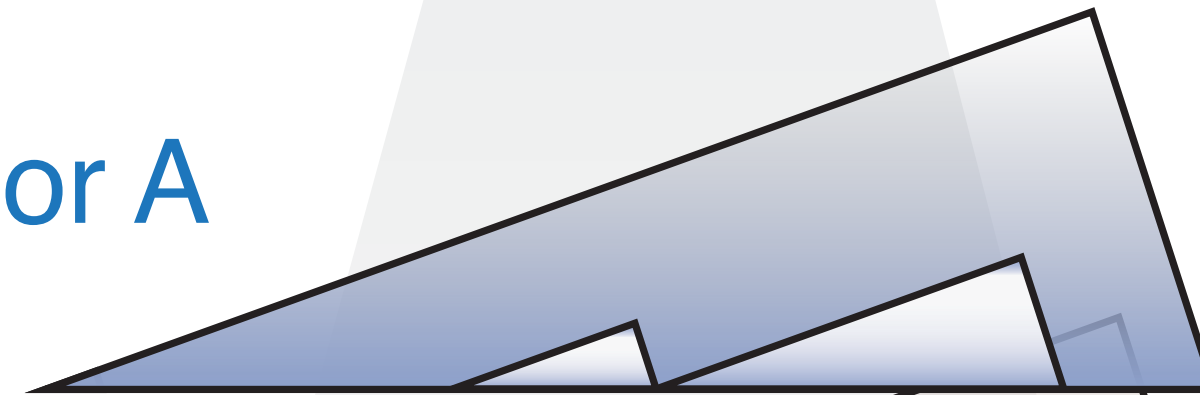
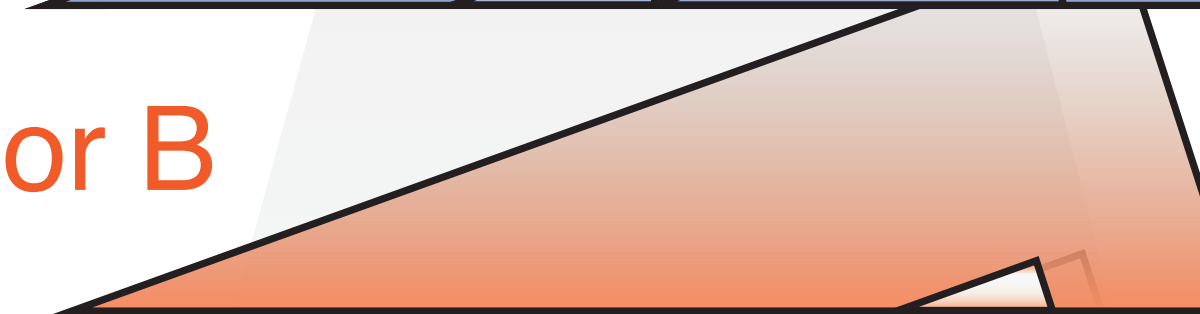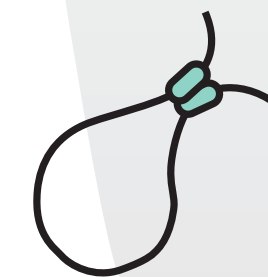# 3D interactions rewiring at Zfp608 locus during mouse neural development
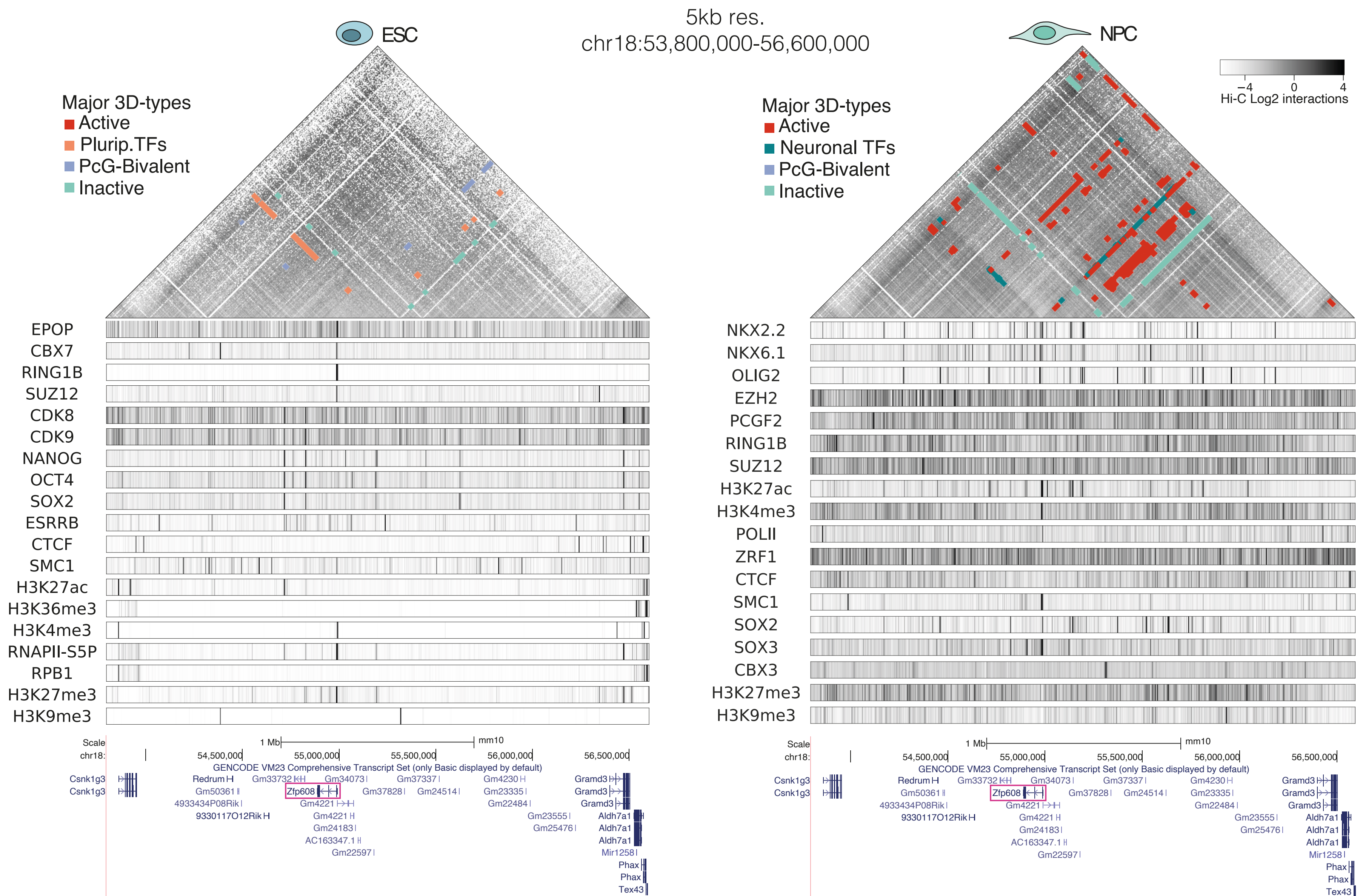
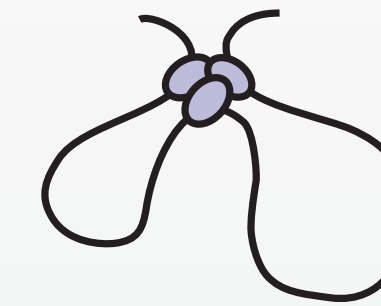# Take home message...

Hi-C

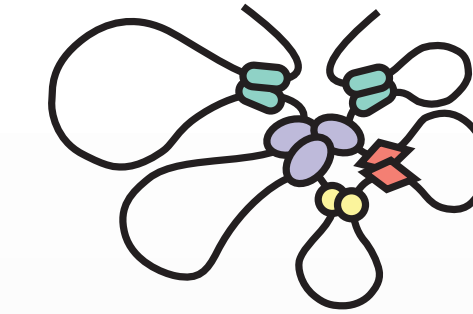Factor A

Factor B

Factor C

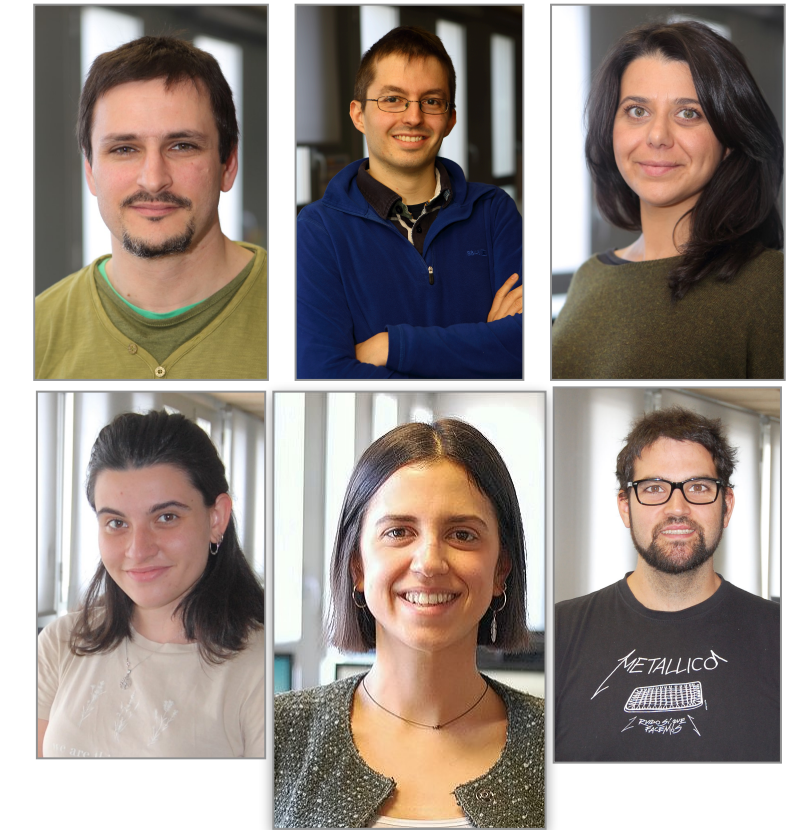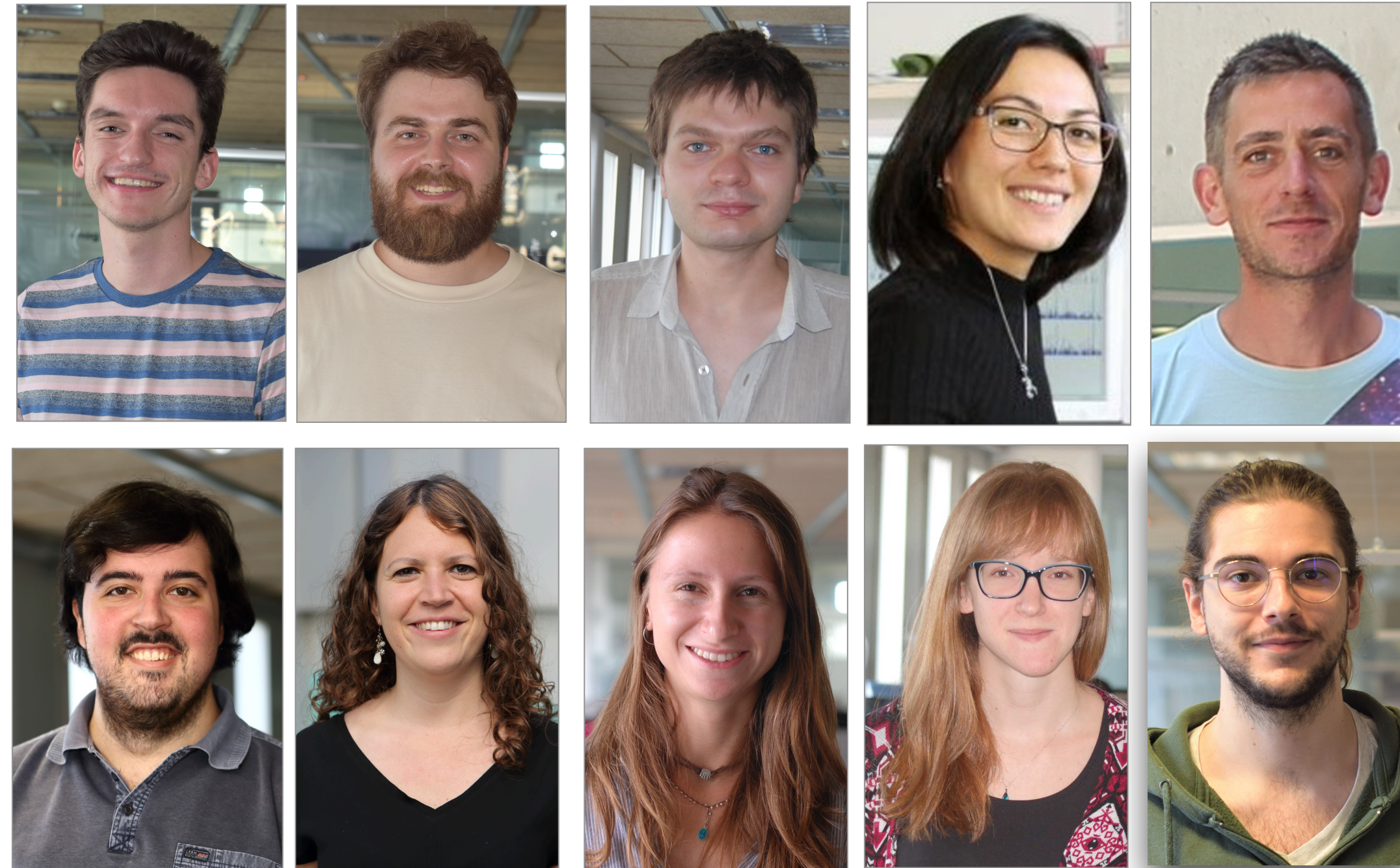Factor D

http://marciuslab.org
http://3DGenomes.org

@marciuslab
@mamartirenom

cnag

CRG Centre for Genomic Regulation

ICREA

Alexander Barclay
Nikolai Bykov
Ronan Duchesne
Iana Kim
François Le Dily
Iago Maceda
Maria Marti-Marimon
Meritxell Novillo
Aleksandra Sparavier
Leo Zuber

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Fundació La Marató de TV3

NIH National Human Genome Research Institute

.: Conflict of Interest Statement :.
Marc A. Marti-Renom serves as a consultant to Acuity Spatial Genomics, Inc., and receives compensation for these services.