

SUPPLEMENTAL METHODS

Generation of Hi-C libraries

Hi-C analysis was performed as described previously (Belton et al. 2012), with the minor modifications described below. A total of 3-5 million cells were pelleted and resuspended in 10 ml of 1× PBS (10 mM phosphate buffer, 2.7 mM KCl 137 mM NaCl, pH7.4) and fixed with 1% formaldehyde (Sigma) for 10 min at room temperature. The reaction was stopped by adding 2 M glycine to a final concentration of 125 mM. The cell suspension was incubated on ice for 5 min, and the cells were pelleted (10 min at 1000 x g in a pre-chilled (4°C) centrifuge). The supernatant was removed, and the cell pellet was washed with 500 µl of 1× PBS, frozen and, if necessary, stored in a liquid nitrogen. After thawing, the cells were lysed for 15 min in 1.5 ml of ice-cold lysis buffer (50 mM Tris-HCl (pH8.0), 150 mM NaCl, 0.5% (v/v) NP-40, 1% (v/v) Triton-X100, 1× Protease Inhibitor Cocktail (CALBIOCHEM #539137 of Thermo Scientific #78430)). The nuclei were harvested at 5000 x g for 7 min in a pre-chilled centrifuge, washed twice with 100 µl of 1.2× restriction NEBuffer 2 (NEB) and resuspended in 200 µl of 1.2× NEBuffer 2. Then, 20% SDS was added to a final concentration of 0.3%, and the nuclei were incubated at 37°C for 1 h with constant shaking. After incubation, 330 µl of 1.2× NEBuffer 2 was added, 20% Triton X-100 was added to a final concentration of 1.8%, and the samples were incubated for 1 h under the same conditions. DNA was digested overnight with 600-800 U of 100 U/µl HindIII-HF restriction endonuclease (NEB) at 37°C with shaking. The next morning, 200 U of HindIII-HF was added, and the samples were incubated for 2 h under the same conditions. The nuclei were harvested for 10 min at 5000 x g and 25°C and resuspended in 50 µl of the collected supernatant. Cohesive DNA ends were biotinylated by adding 7.6 µl of the biotin fill-in mixture prepared in 1× NEBuffer 2 (0.025 mM dATP (Thermo Scientific), 0.025 mM dGTP (Thermo Scientific), 0.025 mM dTTP (Thermo Scientific), 0.025 mM biotin-14-dCTP (Invitrogen), 0.8 U/µl Klenow enzyme (NEB)). The samples were incubated at 37°C for 75 min with slow shaking, and 20% SDS was added to a final concentration of 1.8%; the samples were heated at 65°C for 20 min to inactivate the Klenow enzyme and residual HindIII-HF. The samples were transferred into 10-ml tubes with 3 ml of 1× T4 DNA ligase reaction buffer (Thermo Scientific) supplied with 1.8% Triton X-100, incubated at 37°C for 1 h and then cooled on ice. Next, 100 U of T4 DNA ligase (Thermo Scientific) was added, and the DNA was ligated for 6 h at 22°C with slow agitation. The cross-links were reversed by overnight incubation at 65°C in the presence of proteinase K (100 µg/ml). After cross-link reversal, the DNA was purified by single phenol-chloroform extraction followed by ethanol precipitation (tRNA (Sigma) at a concentration of 7.5 µg/ml and glycogen (Thermo Scientific) at a concentration of 20 µg/ml were used as co-precipitators due to the low amount of DNA in the samples). After

precipitation, the pellets were dissolved in 500 μ l 10 mM Tris-HCl pH 8.0. To remove residual salts and DTT, the DNA was additionally purified using AMICON Ultra Centrifugal Filter Units (0.5 ml, 30 K, Millipore #UFC5030BK) by washing with 10 mM Tris-HCl pH 8.0 (after two washing steps, the final volume of the DNA solution was 40-50 μ l). Biotinylated nucleotides from the non-ligated DNA ends were removed by incubating the Hi-C libraries in the presence of 1.5 U of T4 DNA polymerase (NEB) in NEBuffer 2 supplied with 0.025 mM dATP and 0.025 mM dGTP at 20°C for 4 h. Next, the DNA was purified by phenol-chloroform extraction followed by ethanol precipitation. The DNA pellets were dissolved in 500 μ l of sonication buffer (50 mM Tris-HCl (pH 8.0), 10 mM EDTA, 0.1% SDS) and treated with 50 μ g of RNase A (Thermo Scientific) for 45 min at 37°C; the DNA was then sheared to a size of approximately 100-1000 bp using a VirSonic 100 (VerTis). The samples were concentrated (and simultaneously purified) using AMICON Ultra Centrifugal Filter Units. The fraction of 100-500-bp DNA fragments was captured using Agencourt AMPure XP beads (Beckman Coulter), and the fragments were eluted with 50 μ l of 10 mM Tris-HCl (pH 8.0). The DNA ends were repaired by adding 62.5 μ l MQ water, 14 μ l of 10 \times T4 DNA ligase reaction buffer, 3.5 μ l of 10 mM dNTP mix, 5 μ l of 3 U/ μ l T4 DNA polymerase (NEB), 5 μ l of 10 U/ μ l T4 polynucleotide kinase (NEB), and 1 μ l of 5 U/ μ l Klenow DNA polymerase (NEB) and incubating at 20°C for 30 min. The DNA was purified with Agencourt AMPure XP beads and eluted with 50 μ l of 10 mM Tris-HCl (pH 8.0). To perform an A-tailing reaction, the DNA samples were supplemented with 6 μ l 10 \times NEBuffer 2, 1.2 μ l of 10 mM dATP, 1 μ l of MQ water and 3.6 μ l of 5 U/ μ l Klenow (exo-) (NEB). The reactions were carried out for 30 min at 37°C in a PCR machine, and the enzyme was then heat-inactivated by incubation at 65°C for 20 min. The DNA was purified using Agencourt AMPure XP beads and eluted with 100 μ l of 10 mM Tris-HCl (pH 8.0). Biotin pull-down of the ligation junctions was performed as described previously, with minor modifications. Briefly, 4 μ l of MyOne Dynabeads Streptavidin C1 (Invitrogen) beads were used to capture the biotinylated DNA, and the volumes of all buffers were decreased by 4-fold. The washed beads with captured ligation junctions were resuspended in 50 μ l of adapter ligation mixture composed of 41.5 μ l MQ water, 5 μ l 10 \times T4 DNA ligase reaction buffer (Thermo Scientific), 2.5 μ l of Illumina TruSeq adapters and 1 μ l of 5 U/ μ l T4 DNA ligase (Thermo Scientific). Adapter ligation was performed at 22°C for 2.5 h, and the beads were sequentially washed twice with 100 μ l of TWB (5 mM Tris-HCl (pH8.0), 0.5 mM EDTA, 1 M NaCl, 0.05% Tween-20), once with 100 μ l of 1 \times binding buffer (10 mM Tris-HCl (pH8.0), 1 mM EDTA, 2 M NaCl) and once with 100 μ l of CWB (10 mM Tris-HCl (pH 8.0) and 50 mM NaCl) and then resuspended in 20 μ l of MQ water. Test PCR reactions containing 4 μ l of the streptavidin-bound Hi-C library were performed to determine the optimal number of PCR cycles needed to generate enough PCR

products for sequencing. The PCR reactions (volume of each reaction is 25 μ l) were performed using KAPA High Fidelity DNA Polymerase (KAPA) and Illumina PE1.0 and PE2.0 PCR primers (10 pmol each). The temperature profile was 5 min at 98°C, followed by 6, 9, 12, 15 and 18 cycles of 20 s at 98°C, 15 s at 65°C, and 20 s at 72°C. The PCR reactions were separated on a 2% agarose gel supplied with ethidium bromide, and the number of PCR cycles necessary to obtain a sufficient amount of DNA was determined based on the visual inspection of gels (typically 12-15 cycles). Four preparative PCR reactions were performed for each sample. The PCR mixtures were combined, and the products were purified with QIAGEN PCR Purification Kit. The DNA was eluted with 50 μ l of 10 mM Tris-HCl (pH 8.0) and separated on a 1.8% agarose gel supplied with ethidium bromide; 200-600-bp fragments were excised from the gel, purified using QIAGEN Gel Extraction Kit and sequenced with an Illumina HiSeq 2000 by paired-end 101-nt reads.

SUPPLEMENTAL FIGURE LEGENDS

Supplemental Figure S1. (A) Heatmaps of biological replicates of Hi-C experiments (chromosome 3L is shown). Resolution of heatmaps is 20 kb. (B) Graph of contact probability as a function of genomic distance.

Supplemental Figure S2. Hi-C interaction maps (heatmaps) of the four studied cell lines at 20-kb resolution.

Supplemental Figure S3. Visual annotation of long-range interarm (A) and interchromosomal (B) contacts of Polycomb-occupied TADs in BG3 cell line (highlighted by red circles; white circles mark interactions between Polycomb-occupied TADs located on the same chromosomal arm). Resolution of the heatmaps is 50 kb, and resolution of TAD images under the maps is 20 kb.

Supplemental Figure S4. Distributions of TAD lengths (excluding boundary bins) and inter-TAD lengths (including TAD boundaries) in the four studied cell lines.

Supplemental Figure S5. (A) Distribution of chromatin colors near TAD boundaries. Number of bins in each group is shown above the diagram. All designations are as in Fig. 2B. (B) Diagram showing the inverse dependence between the proportion of active chromatin colors within a genomic bin and γ_t (the minimal value of the scaling parameter gamma required to annotate a bin as a TAD boundary or an inter-TAD). Numbers of bins in each group are shown above the diagram (for example, 240 bins were annotated as inter-TADs at gamma values of 0–0.1 (excluding $\gamma = 0.1$), 17 bins were annotated as inter-TADs at gamma values 0.1–0.2 (excluding $\gamma = 0.2$)). Bins that have not been annotated as TAD boundaries or inter-TADs at gamma values from 0 to 10 (excluding $\gamma = 10$) are highlighted in red. All other designations are as in Fig. 3B. P-values (two-sided Mann-Whitney test) are presented in Supplemental Table S6.

Supplemental Figure S6. Boxplots showing distributions of individual chromatin marks and binding proteins near TAD boundaries, some of them were used for the construction of panel (C) in Figure 2. P-values (two-sided Mann-Whitney test) are presented in Supplemental Table S6.

Supplemental Figure S7. The observed representation of dCTCF at TAD boundaries and inter-TADs does not depend on the gamma value used for TAD annotation. The distribution of dCTCF, H3K4me3 and Su(Hw) around TAD boundaries at different values of the scaling parameter gamma used for TAD annotation is shown. Curves smoothed with LOESS show the median Z-transformed values. Thick rectangles show TAD boundary bins.

Supplemental Figure S8. Principal components analysis (A) and cluster analysis (B) of biological replicates of poly(A)+ RNA-seq of the cell lines. (C) The level of polyA(+) transcripts around TAD boundaries in the Kc167, OSC and S2 cells (extension of Fig. 3A). (D) Boxplots showing inverse dependence between the transcription level within a genomic bin and γ in the S2 and Kc167 cells (extension of Fig. 3D).

Supplemental Figure S9. Scatter plots demonstrating transcription level and the proportion of active chromatin colors in individual TADs excluding boundary bins (orange spots), and inter-TADs including boundary bins (blue spots), in BG3, S2 and Kc167 cells. Horizontal lines separate inter-TADs by the 20% quintile according to the transcription level; TADs were also separated according to this level. Vertical lines separate inter-TADs (left panels) and TADs (right panels) by a threshold of 30% of active chromatin within inter-TADs and TADs, respectively (sum of 1, 2 and 3 colors for the BG3 and S2 cells, and RED and YELLOW for the Kc167 cells). Other designations are as in Fig. 3B.

Supplemental Figure S10. Pie charts showing the distribution of differentially or uniformly transcribed bins (A) and bins with different proportions of active chromatin (B) in the four groups, as defined by a pairwise comparison of the cell lines (extension of Fig. 4B).

Supplemental Figure S11. Scatter plots illustrating independent variations in proportions of active or repressed chromatin within the interior and boundaries of conserved TADs, as observed in a pairwise comparison of BG3 and S2 cell lines.

Supplemental Figure S12. Scatter plots demonstrating the proportion of interband-specific CYAN and BLUE chromatin colors (Zhimulev et al. 2014) in TADs and inter-TADs of different sizes in the BG3 and S2 cells.

Supplemental Figure S13. Results of 12 individual realizations of our polymer model, similar to Fig 6. Distances are measured in numbers of monomer units (nucleosomes).

Supplemental Figure S14. Predicted spatial configuration of a polymer composed of equal blocks of acetylated (green) and non-acetylated (black) nucleosomes. Each block consists of 500 nucleosomes. (B) Spatial proximity map (distance heatmap) of the polymer configuration presented in (A). (C) Simulated Hi-C map constructed at resolution of 4 kb (20 beads) for three consecutive TADs. The starting conformation of a polymer chain was a random walk trajectory in the $64 \times 64 \times 64$ simulation box.

Supplemental Figure S15. (A) Scatter plot demonstrating negative correlation between the number of contacts within simulated TADs and their radius of gyration (measured in diameters of polymer beads). This plot demonstrates that the total sum of Hi-C interactions within a TAD may be interpreted as a measure of density of a TAD. For each of the 19 TADs in each of the 12 independent simulations, we calculated the total number of contacts between regions more than 4 kb (20 nucleosomes) apart. We also calculated radius of gyration, which is the square root of the mean squared distance to the center of mass. The two show strong negative correlation. (B) Graph of contact probability as a function of distance along the polymer chain estimated for beads located within simulated TAD (red) or between TADs (blue). All pairs of regions not belonging to the same TAD, independent of their status (inter-TADs or different TADs) contributed to the between-TAD plot. We note that comparing this plot to experimental Hi-C data is not feasible, because this plot ends at 100 kb, a typical TAD size in *Drosophila*, while our Hi-C maps start at 20 kb, leaving less than an order of magnitude overlap.

Supplemental Figure S16. Spatial configuration (A) and heatmap (B) of the model polymer obtained when volume interactions instead of saturating interactions were used. All designations are as in Figures 6 and S13.

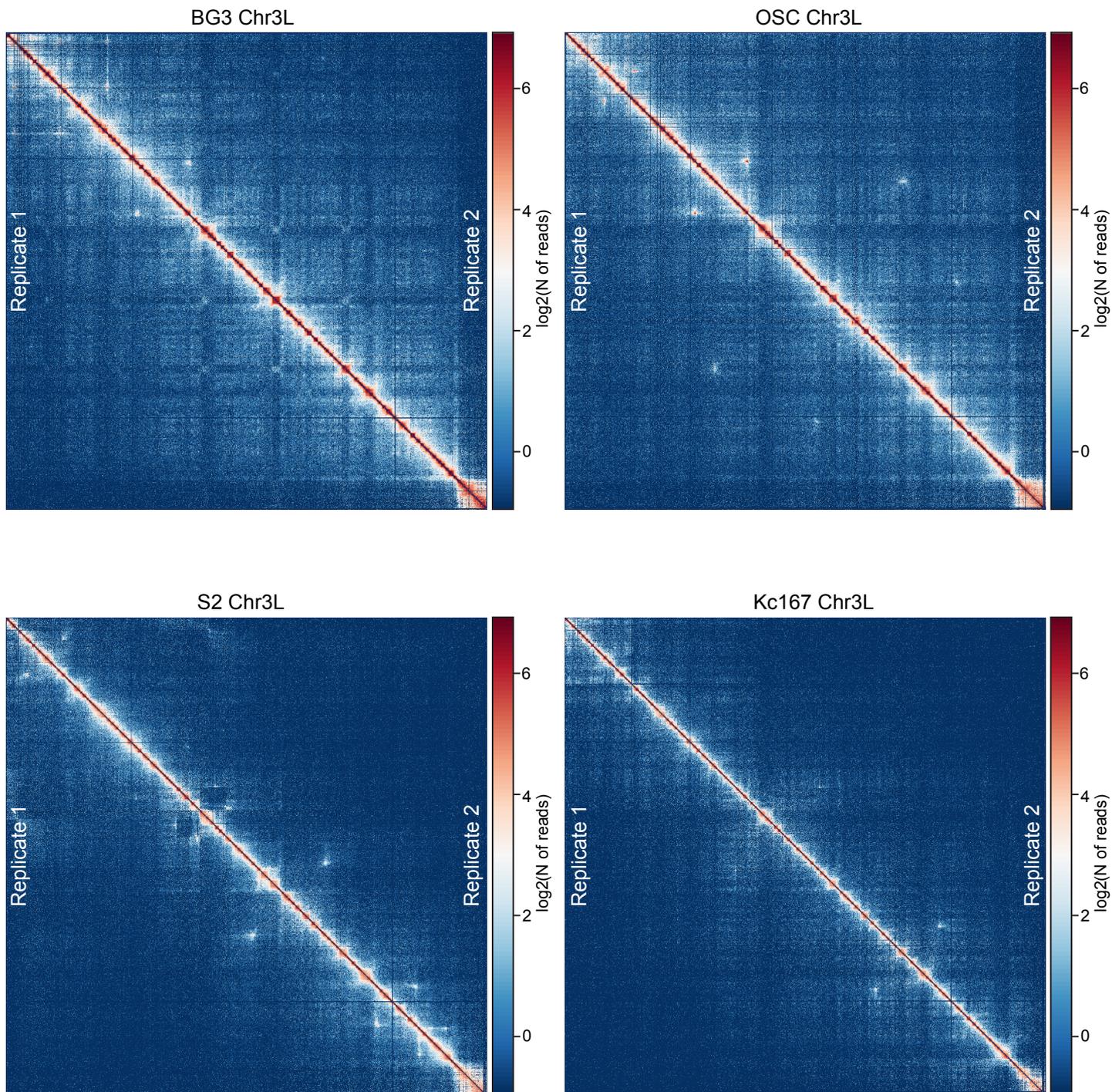
Supplemental Figure S17. The TAD profiles of X chromosome are the same in the male and female cell lines. (A) Heatmap of a 2.8-Mb region of the X chromosome in studied cell lines. (B) Venn diagram showing the numbers of TADs located on the X chromosome and common (with both boundaries located at the same genomic bin or at adjacent bins) for all studied cell lines.

Supplemental Figure S18. Different methods of analysis alter the shape of the distribution of chromatin marks near the TAD boundary. (A) The height of each peak of chromatin mark was multiplied by the proportion of the bin intersecting with the peak, and the results for all peaks in the bin were summed and Z-transformed, as in Fig. 2C. Smoothed curves

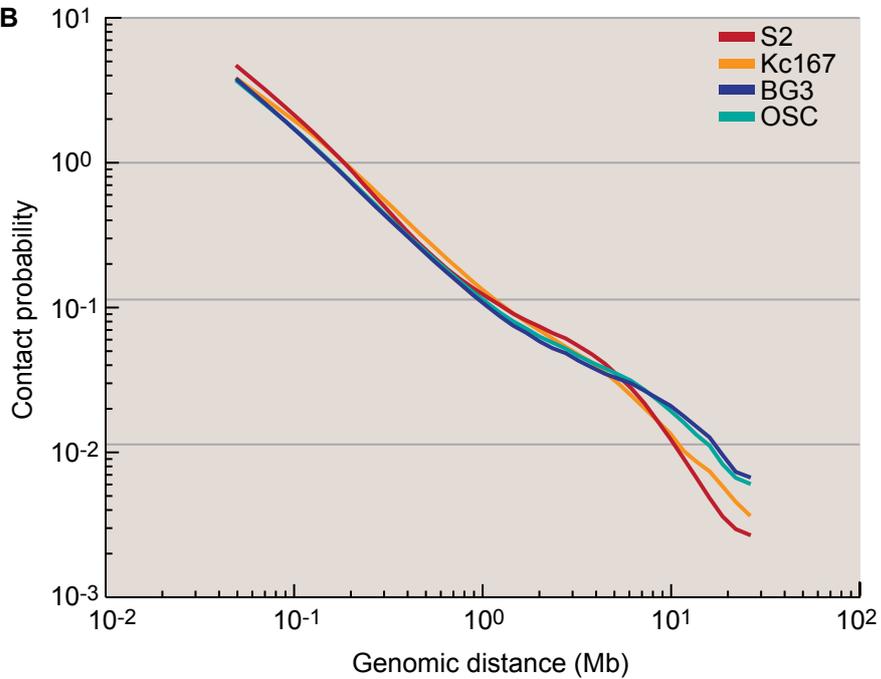
show the median values. (B) Z-transformed proportion of chromatin mark peaks in a bin. The height of peaks was not taken into account. Smoothed curves show the median values. (C) Z-transformed number of peaks of a chromatin mark per bin. The height and the length of peaks were not taken into account. Smoothed curves show the median values. (D) Number of peaks of a chromatin mark per bin. The height and the length of peaks were not taken into account. Smoothed curves show average values. Data files containing called peaks were used in this analysis. (E) Analysis of dCTCF, Su(Hw), Chriz and RNA polymerase II binding within TAD boundaries identified by Hou et al. (2012) in Kc167 cells. TAD boundary positions were rounded to 1 kb. Calculation method as in (A) and Fig. 2C.

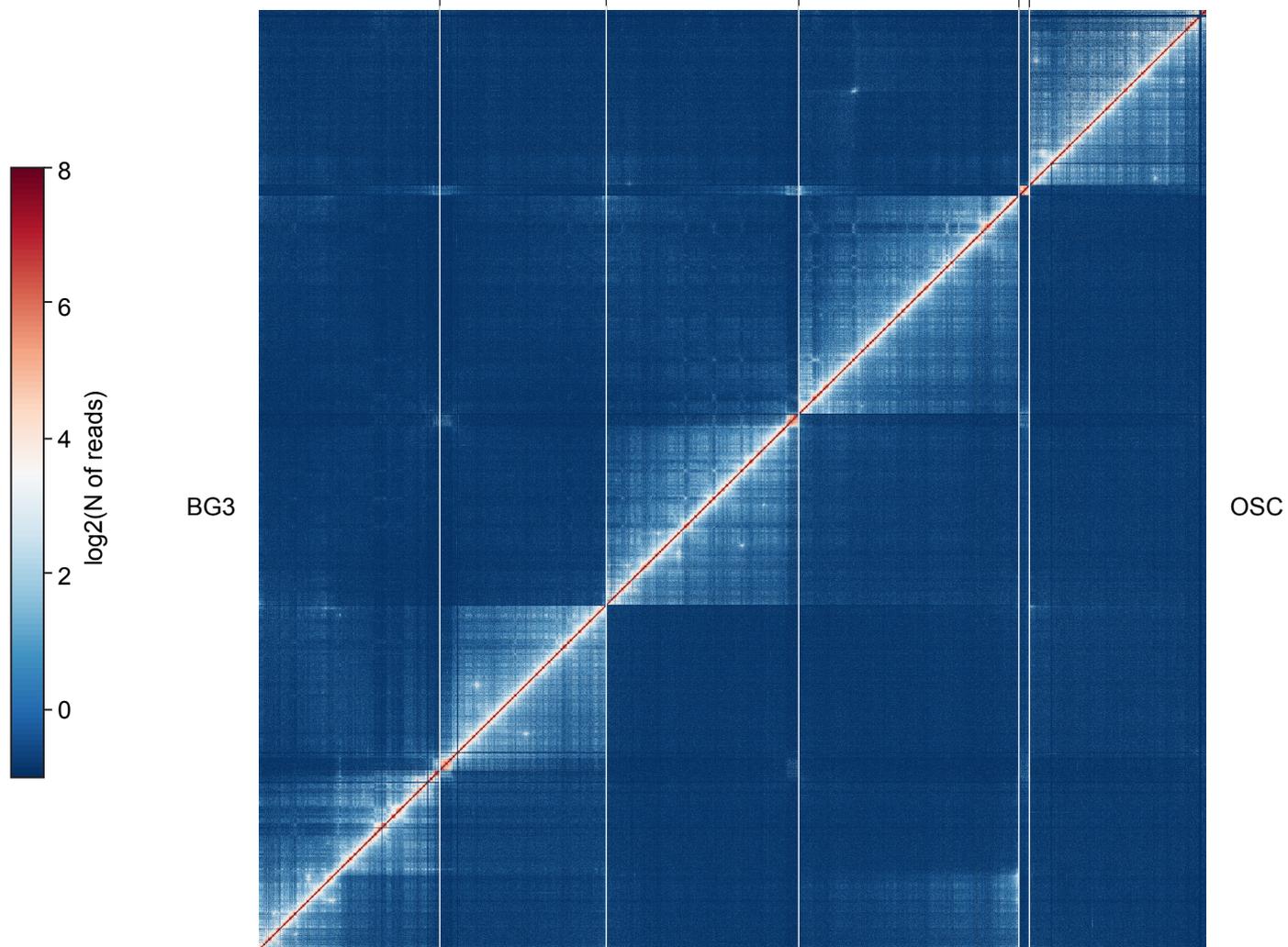
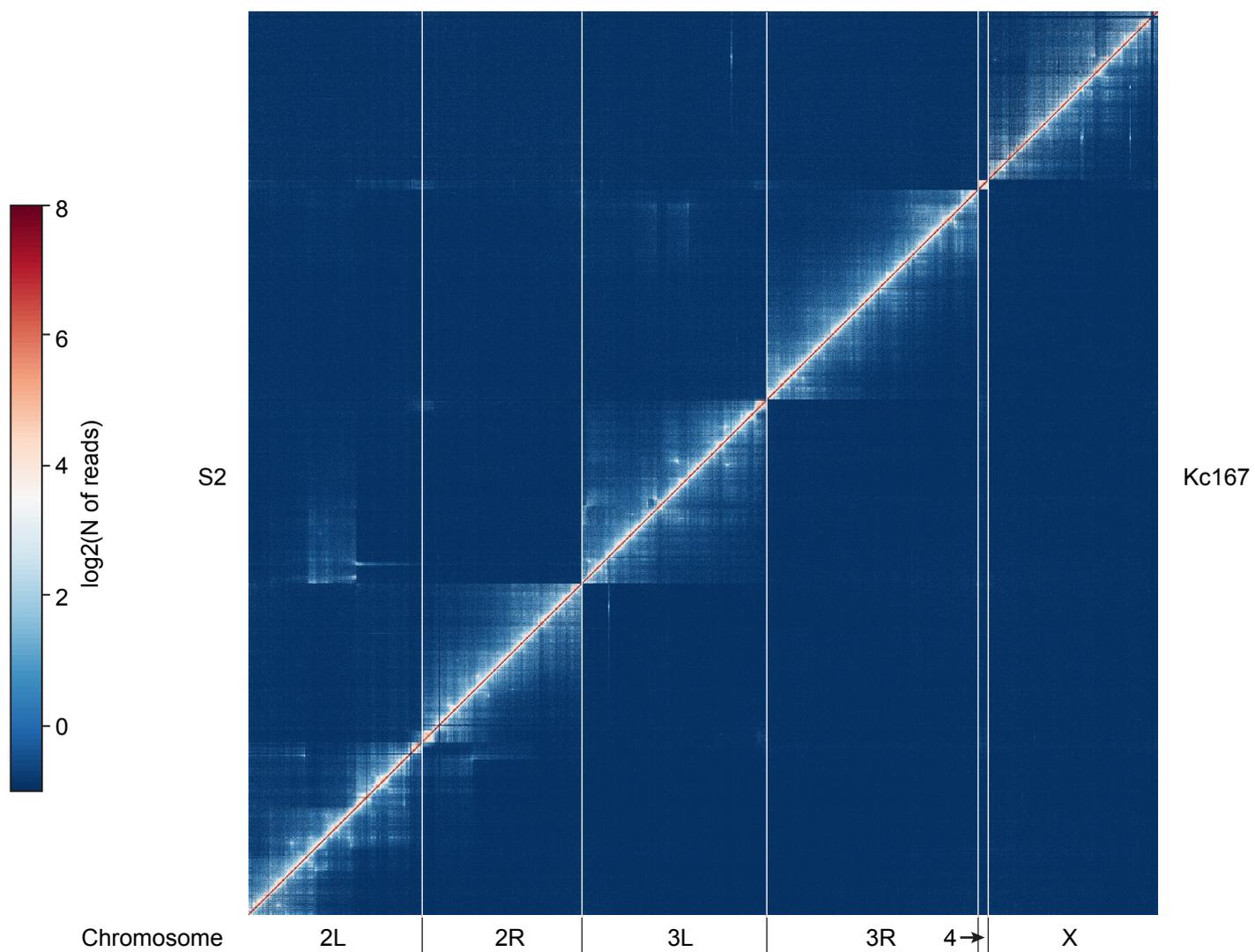
Supplemental Video S1. 3D-display of one of the predicted spatial configurations of the model polymer (a snapshot from this video is presented in Fig. 6A, and the heatmap of this configuration is shown in Fig. 6B).

A

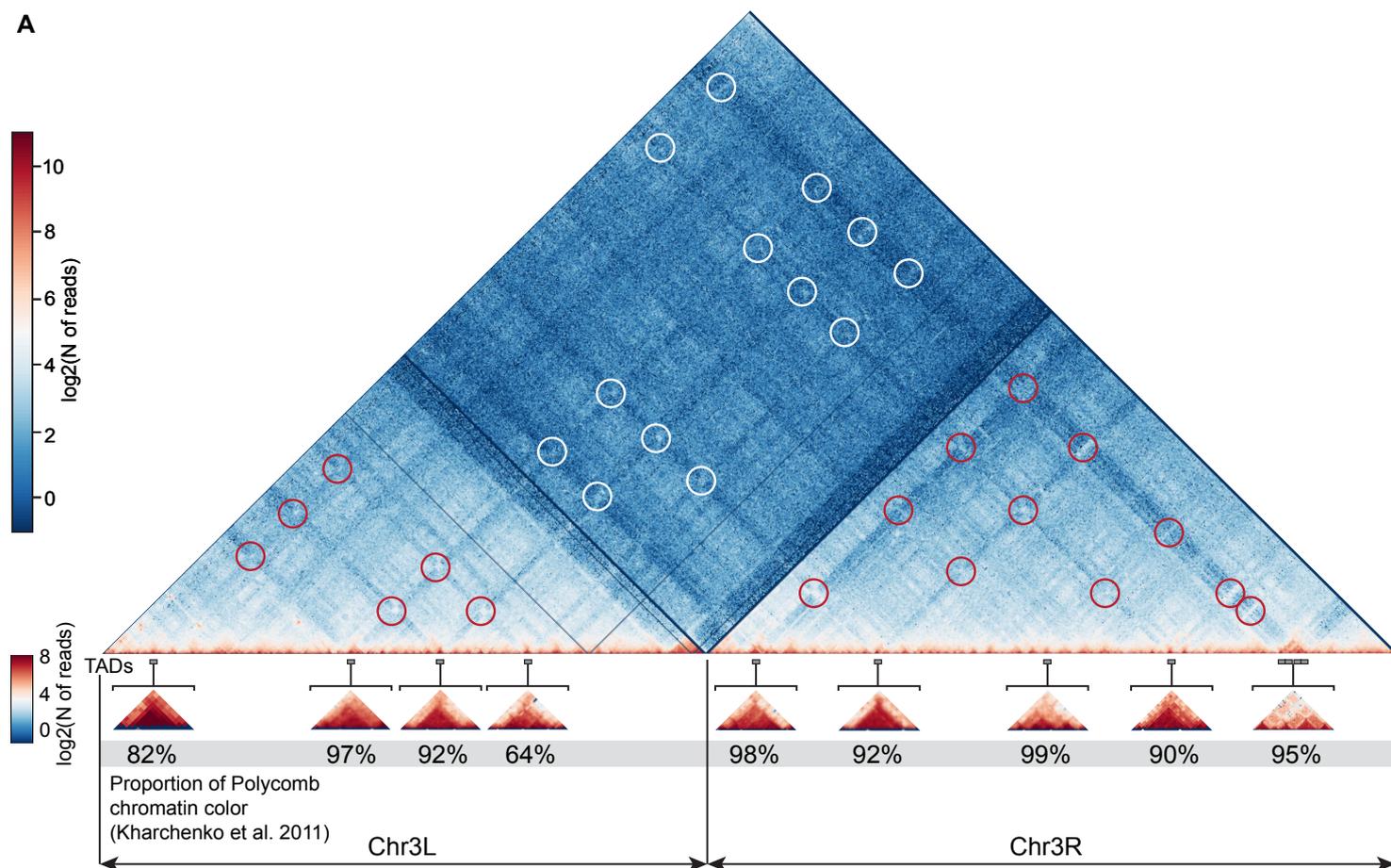


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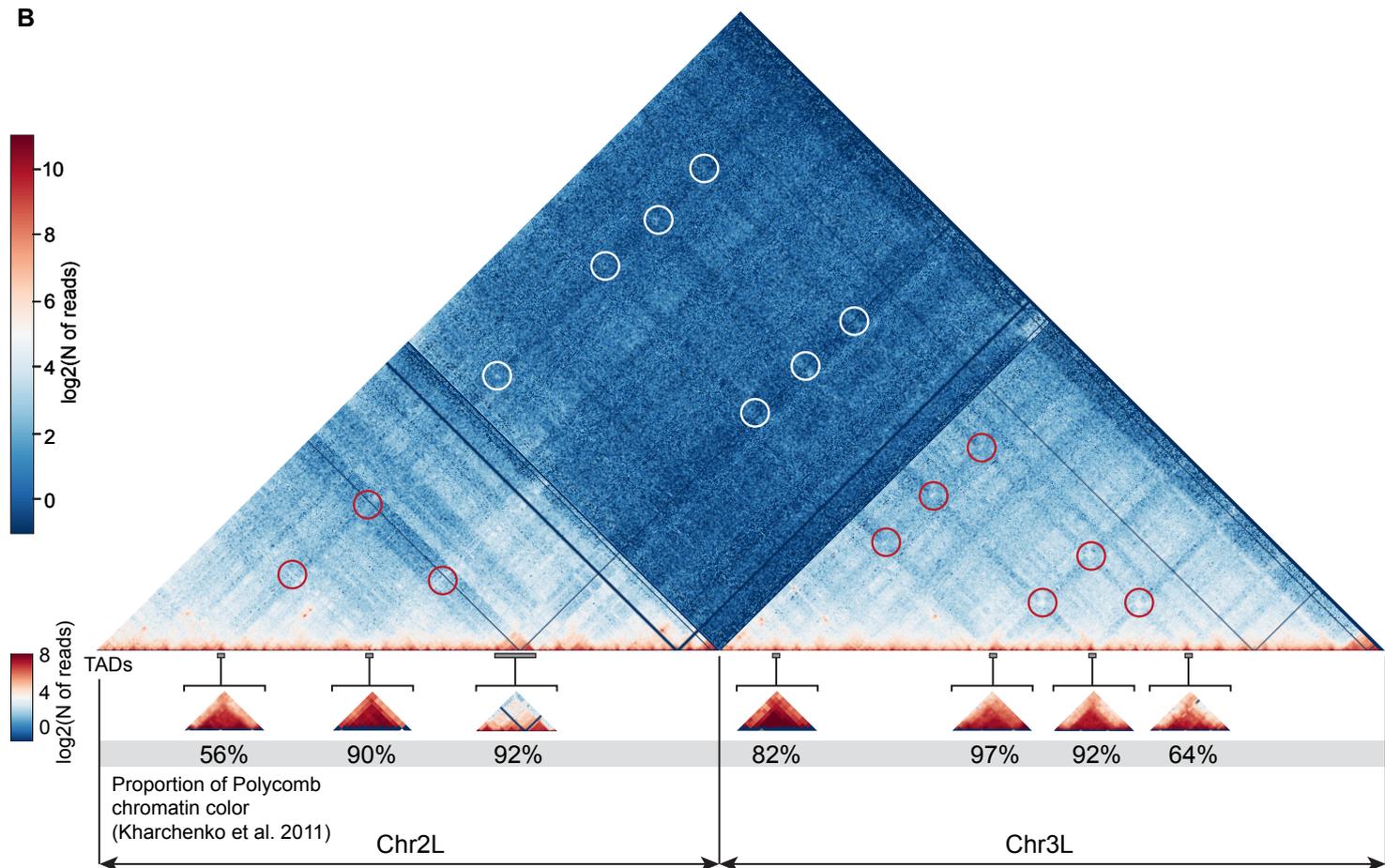




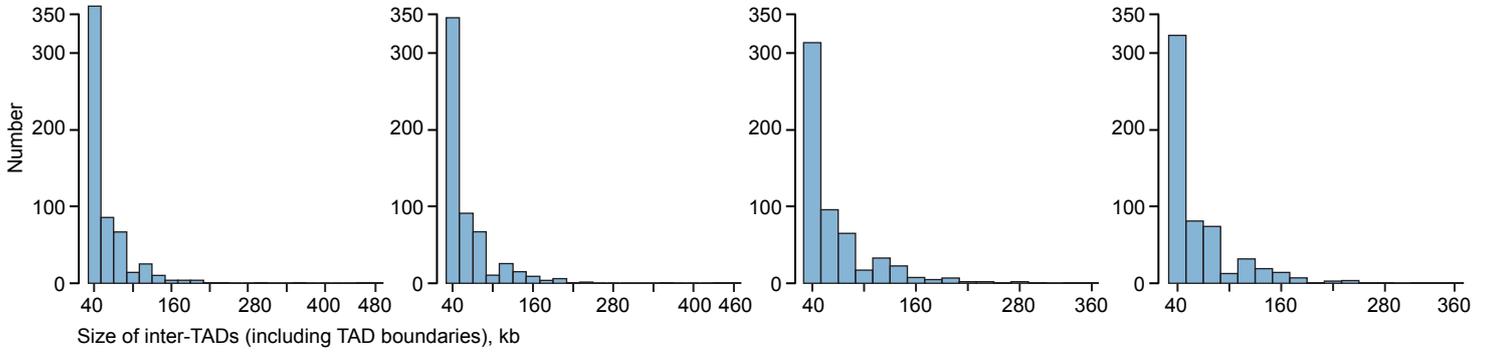
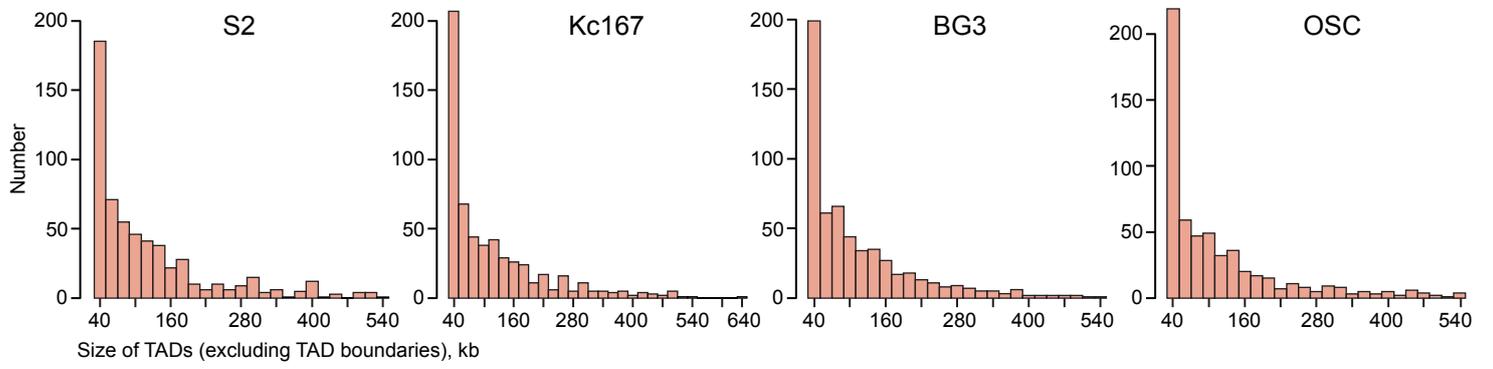
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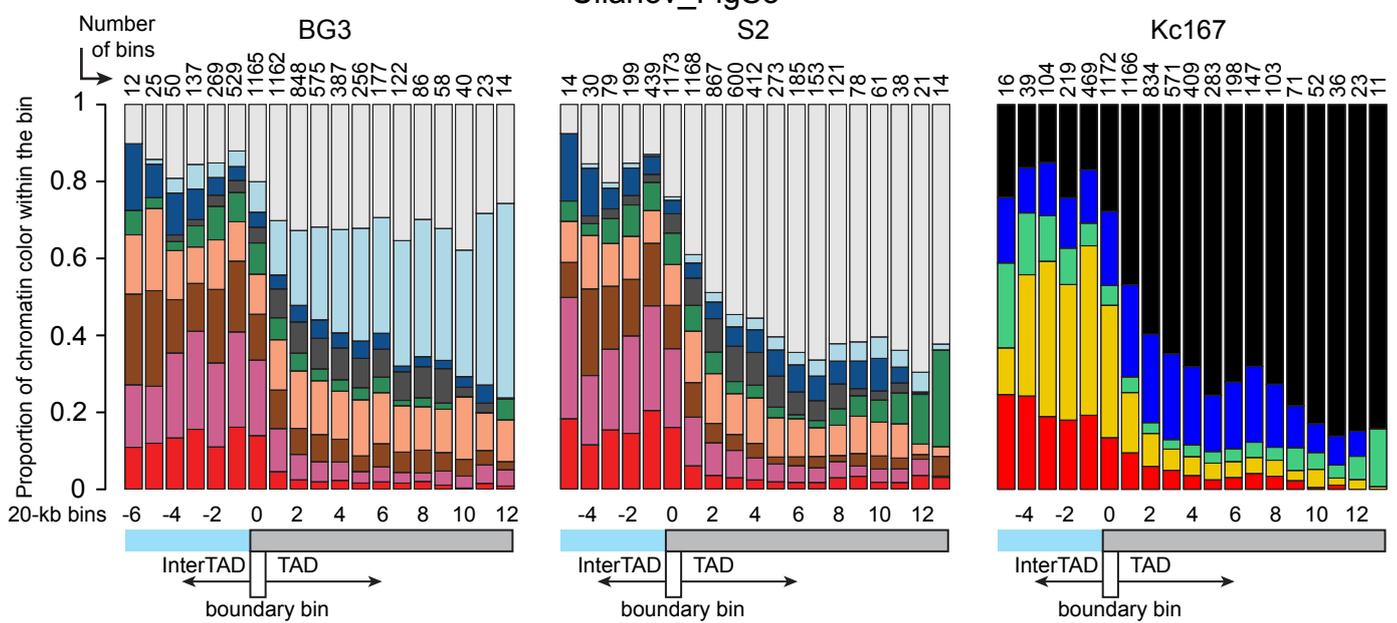
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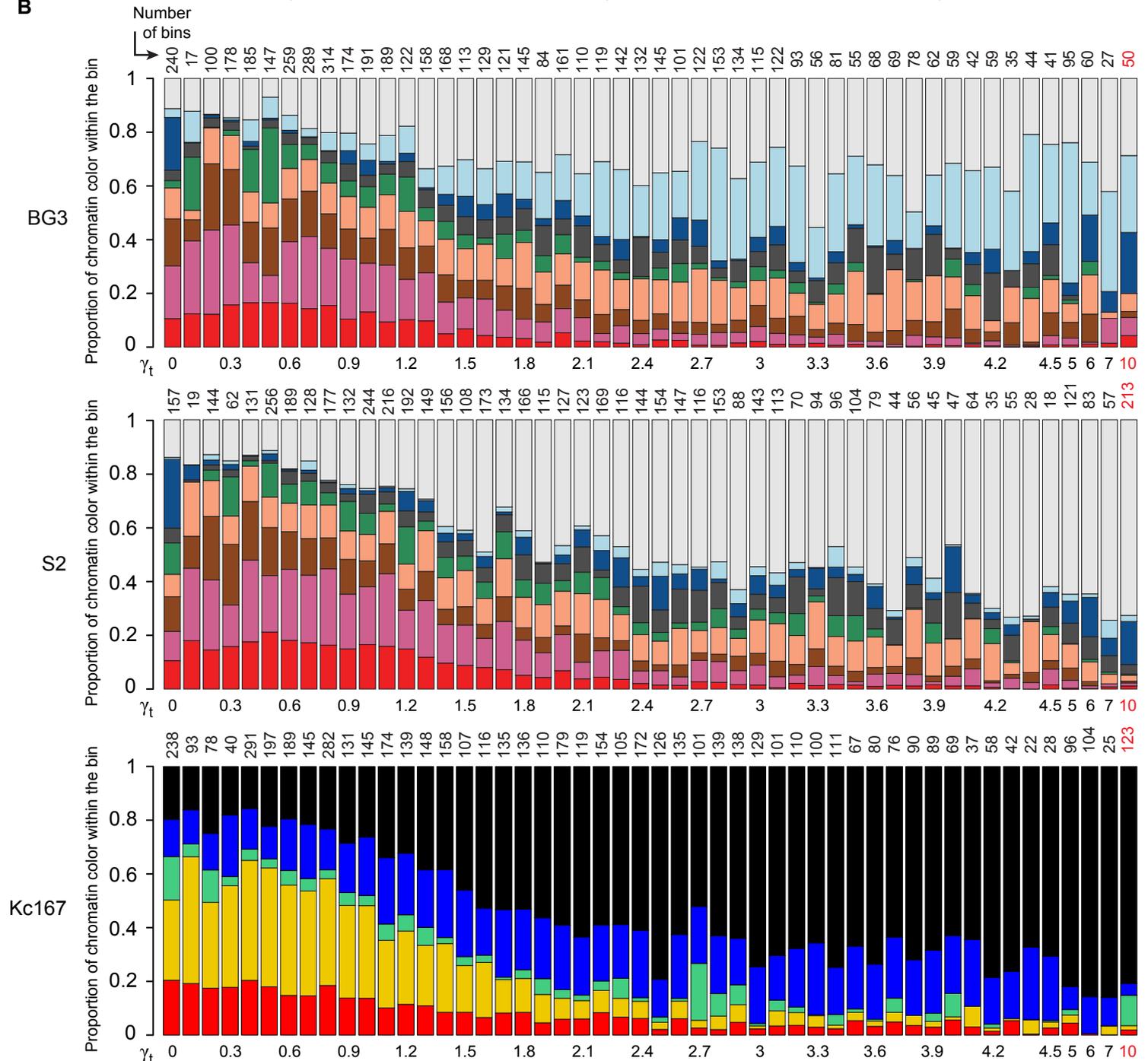
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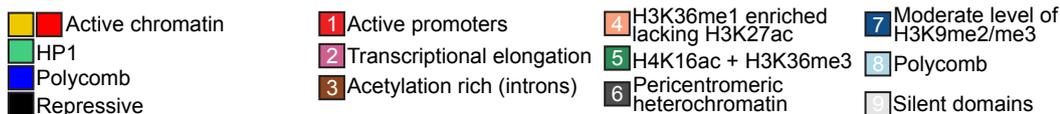
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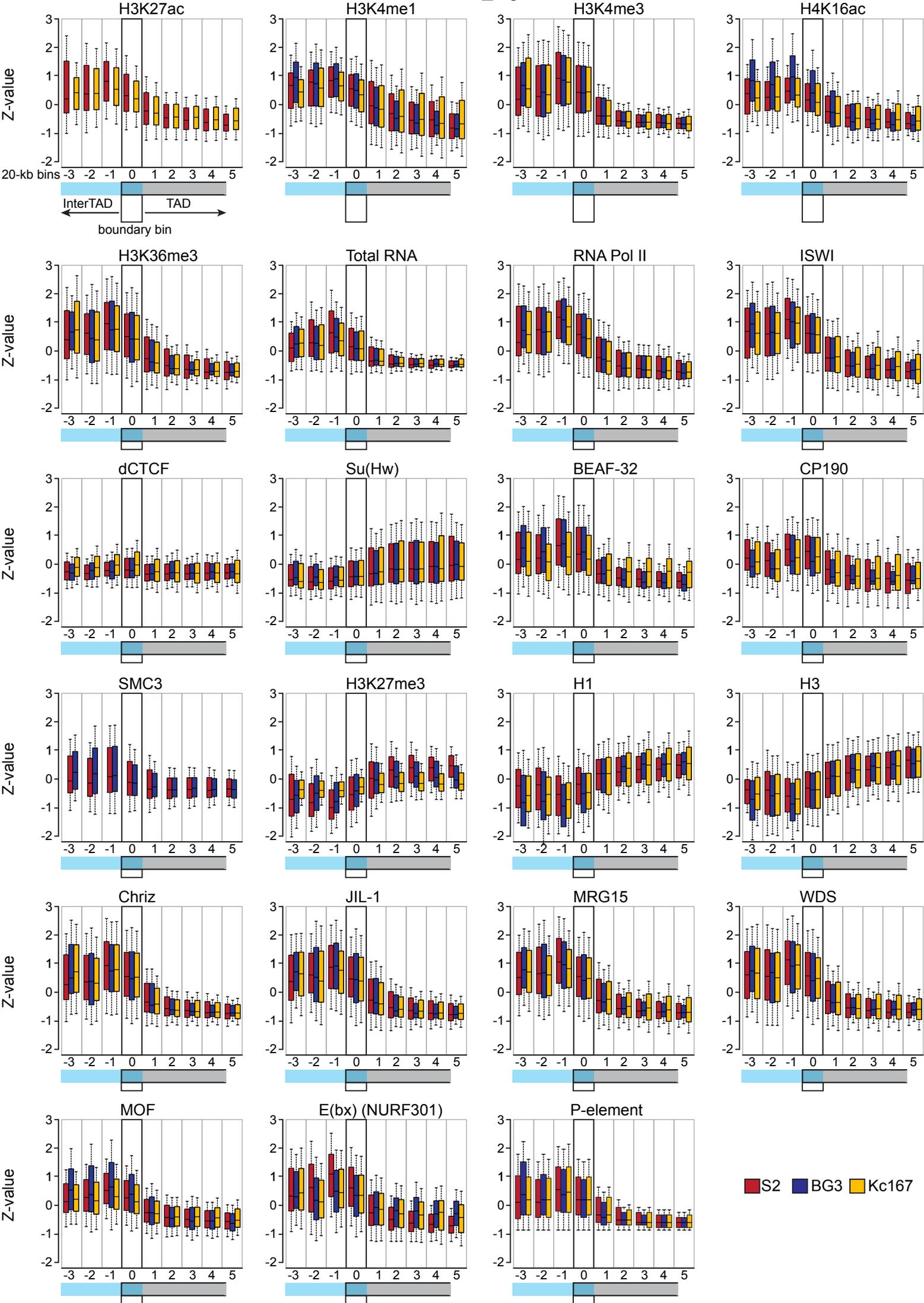


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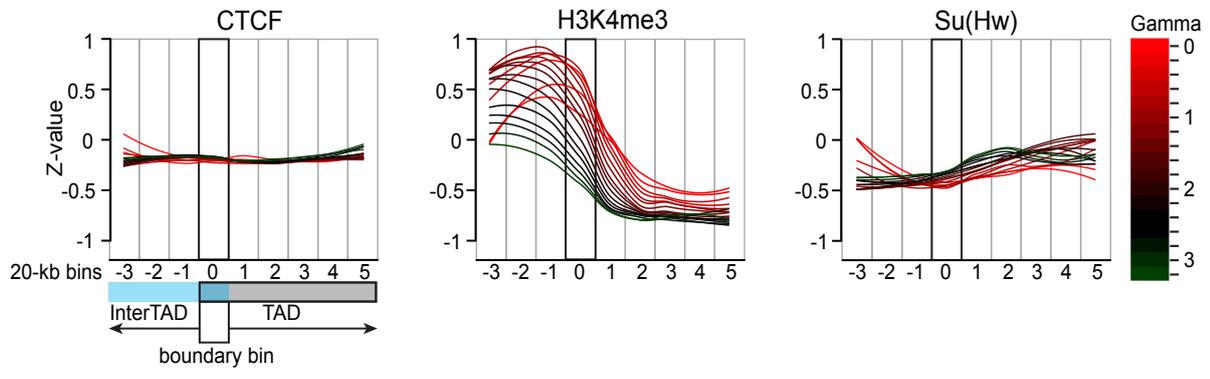


Chromatin colors in Kc167: Chromatin colors in S2 and BG3:

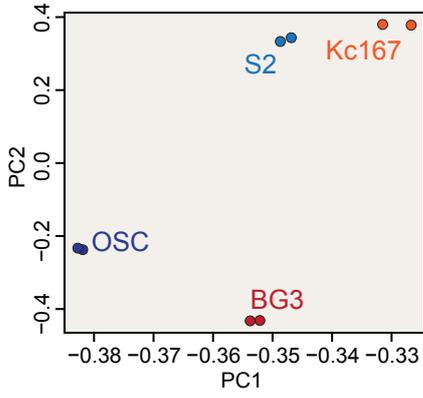




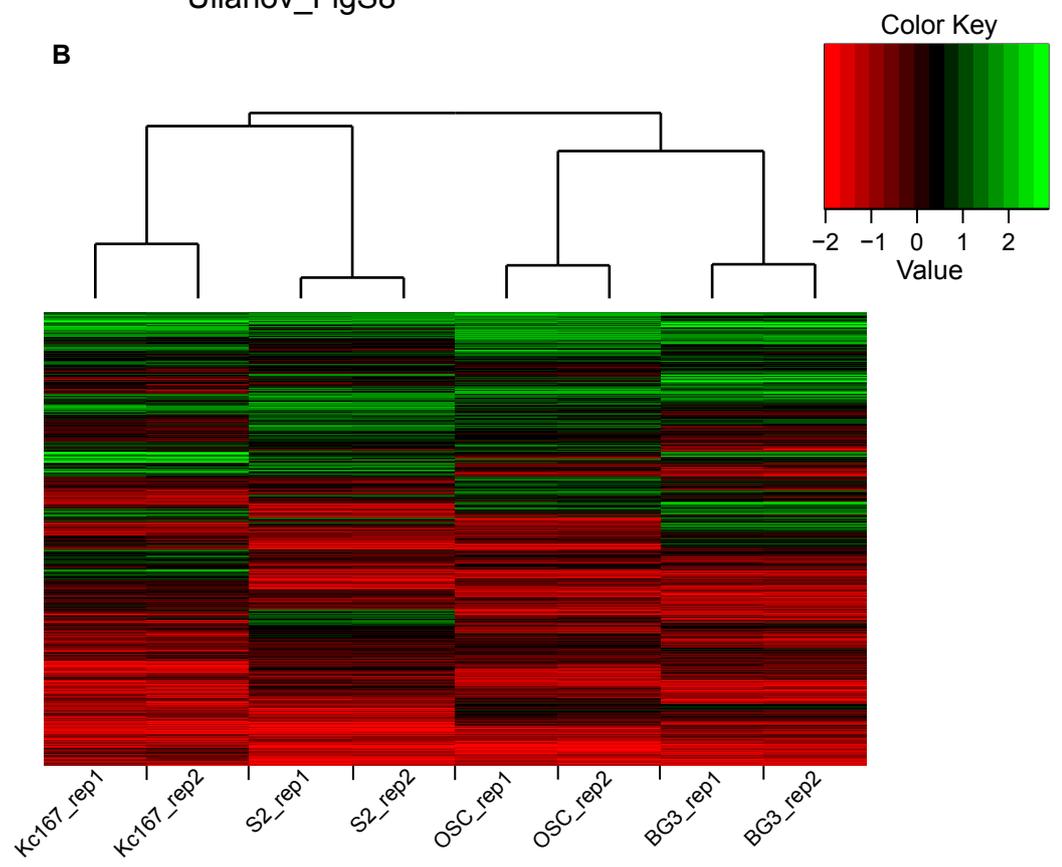
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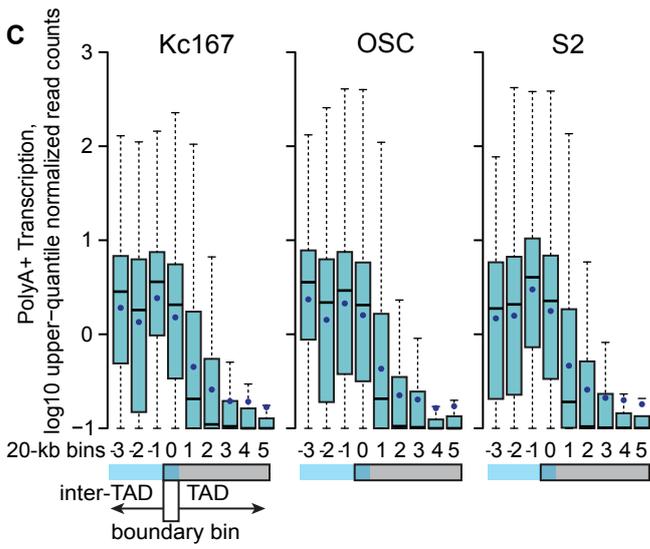
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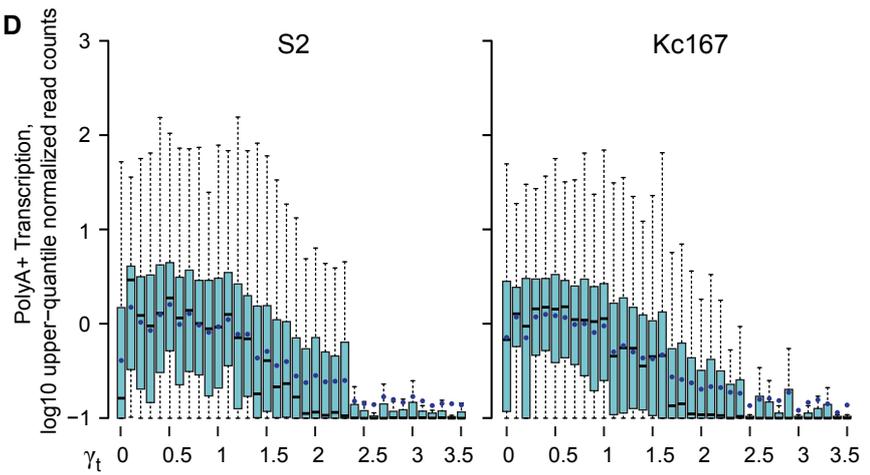
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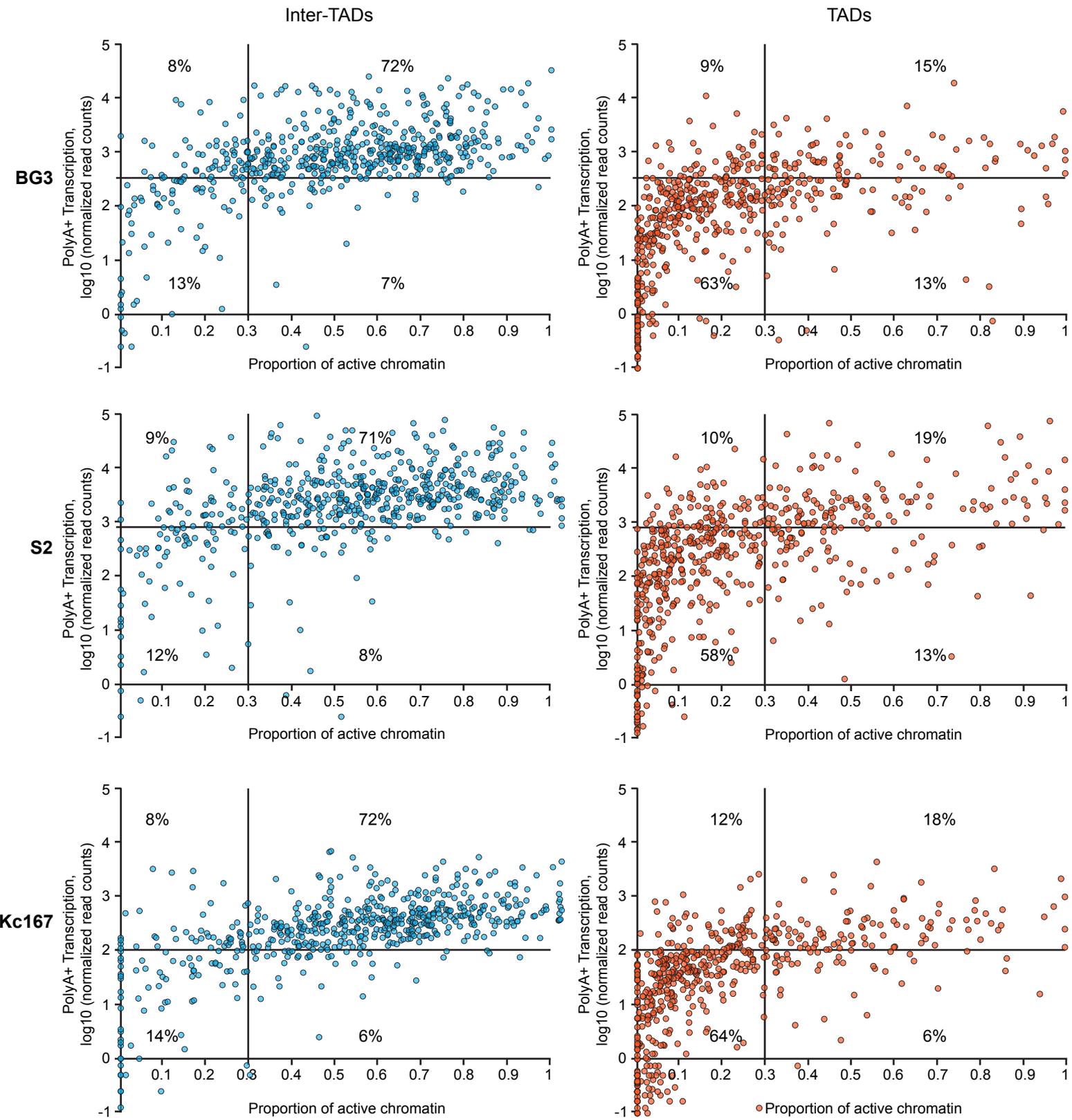


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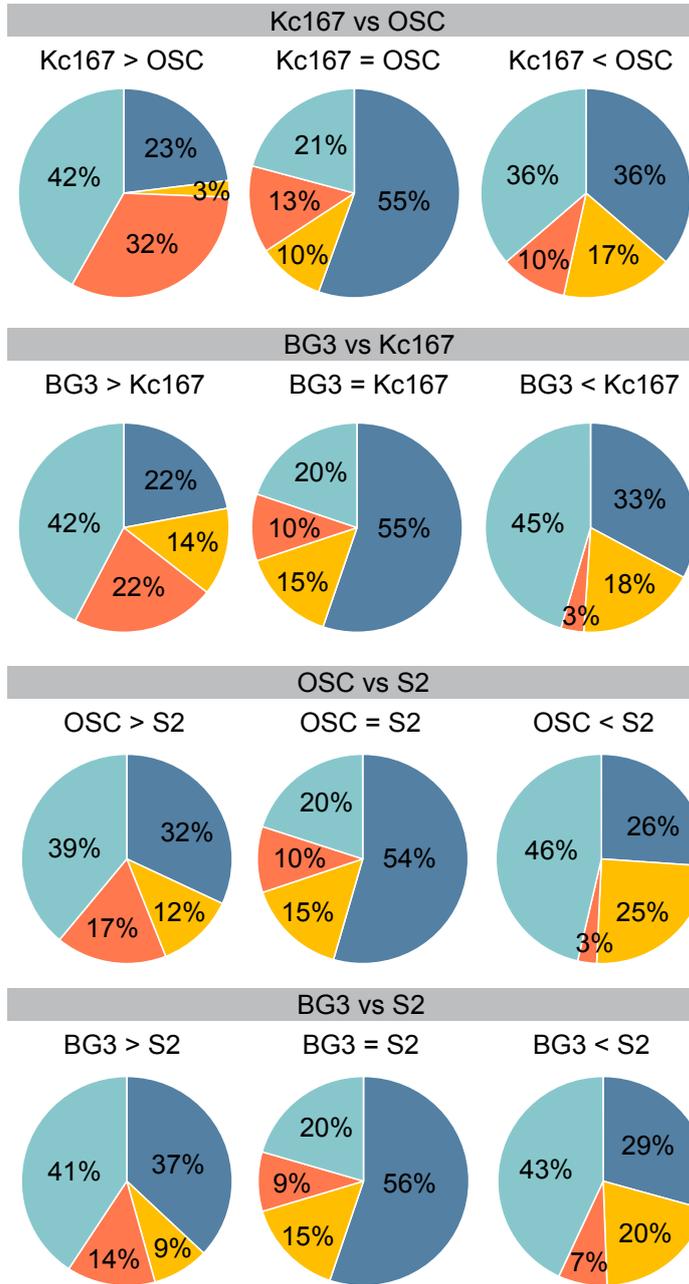
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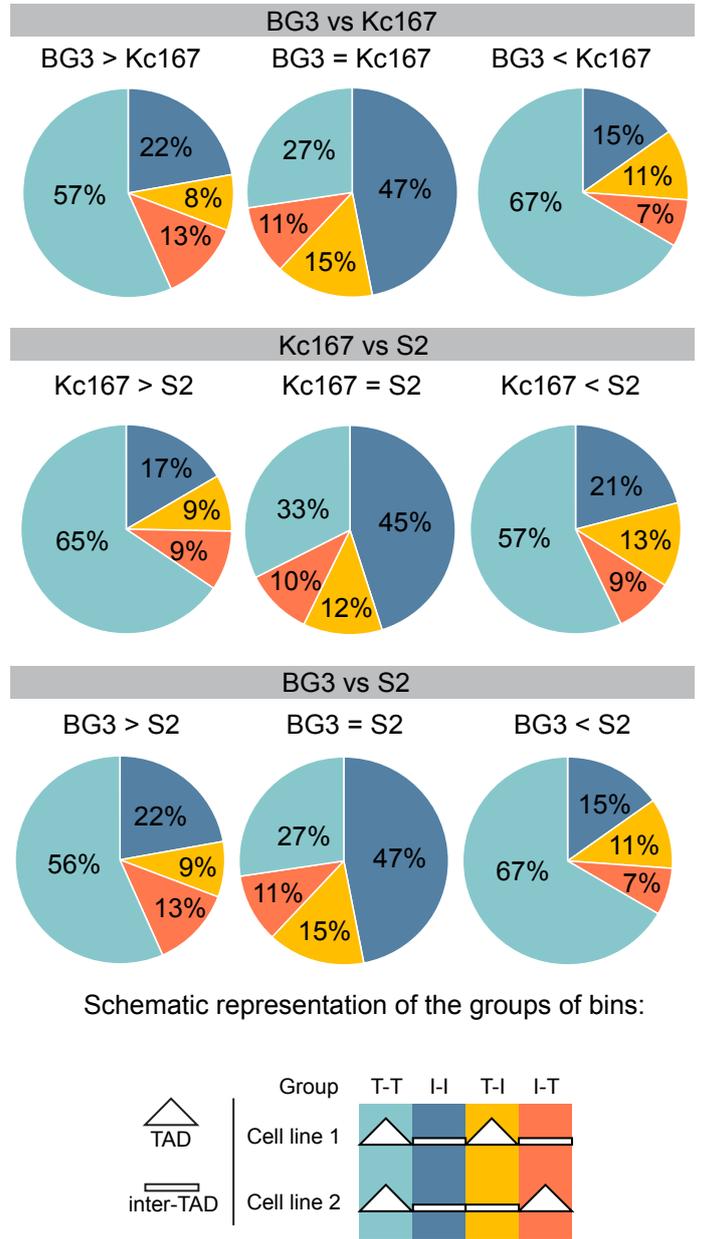


Active chromatin = RED + YELLOW chromatin colors in Kc167,
 1 (RED) + 2 (MAGENTA) + 3 (BROWN) chromatin types in BG3 and S2

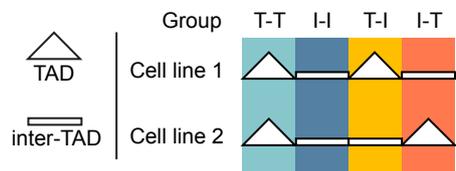
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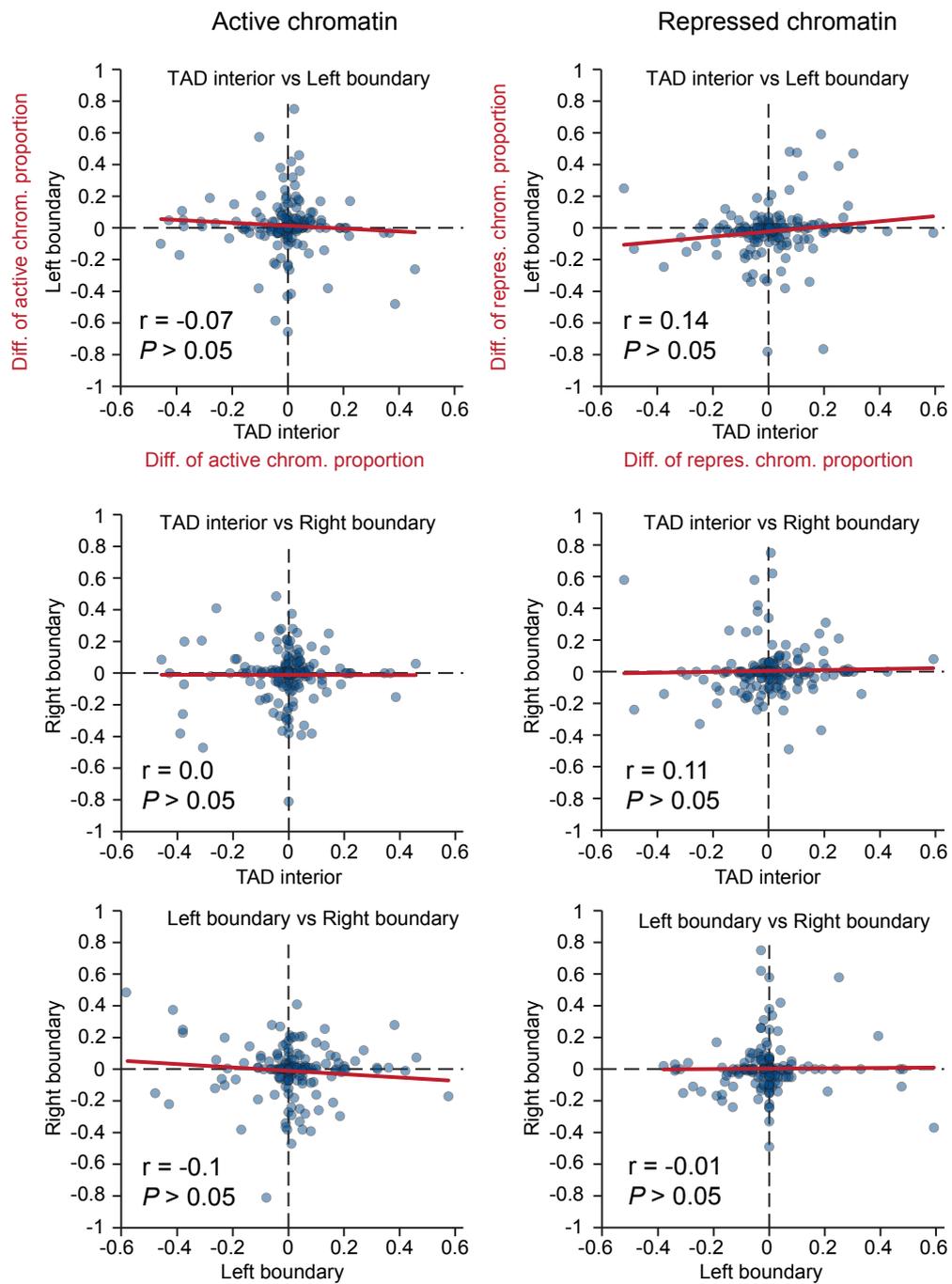


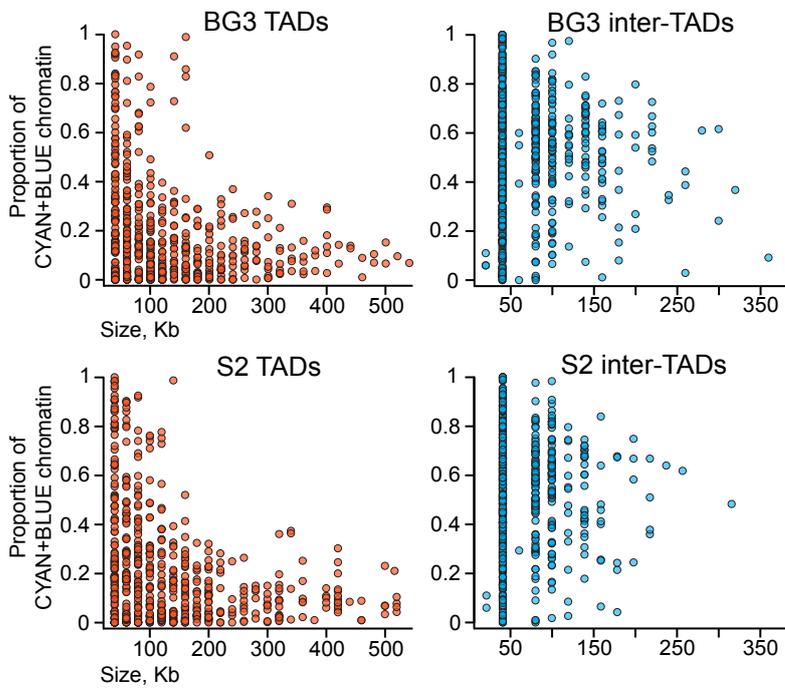
B Active chromatin within the genomic bin (FC of 3)

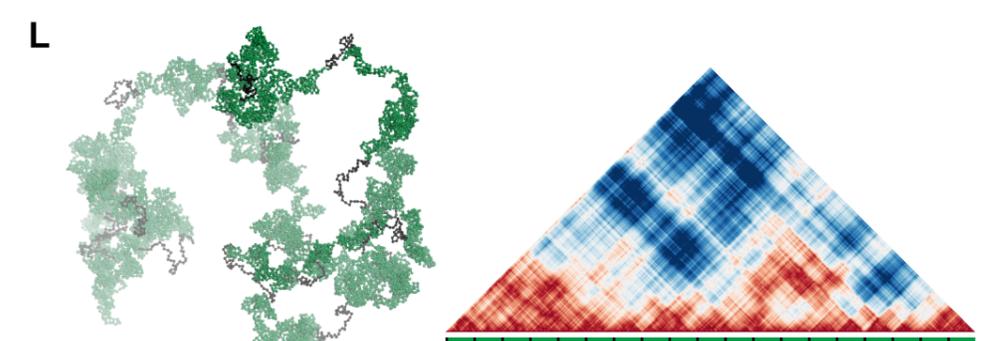
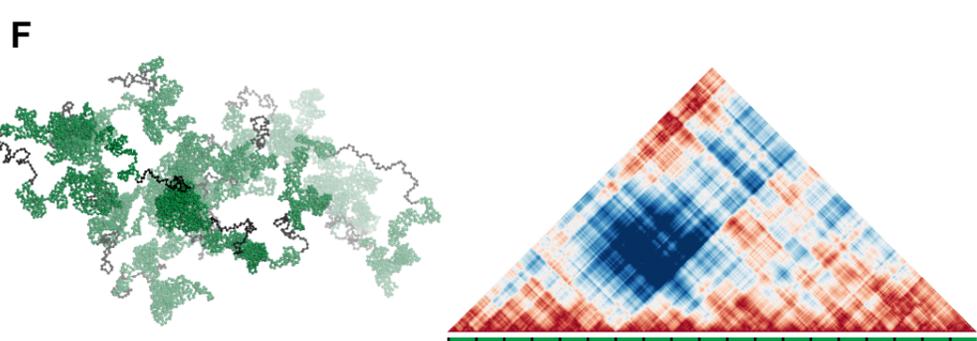
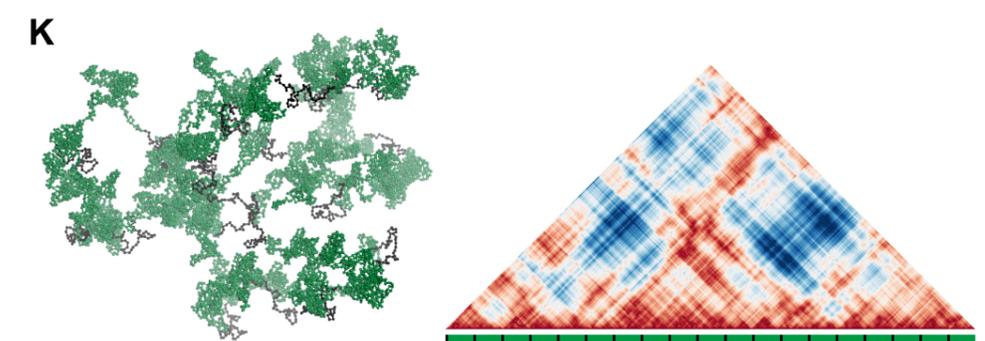
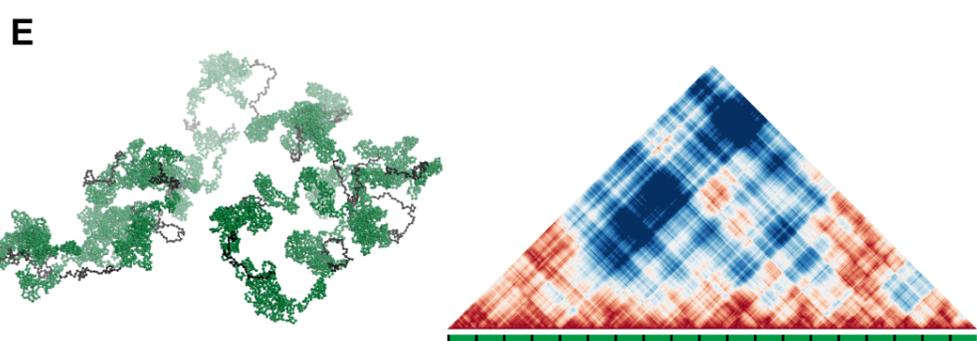
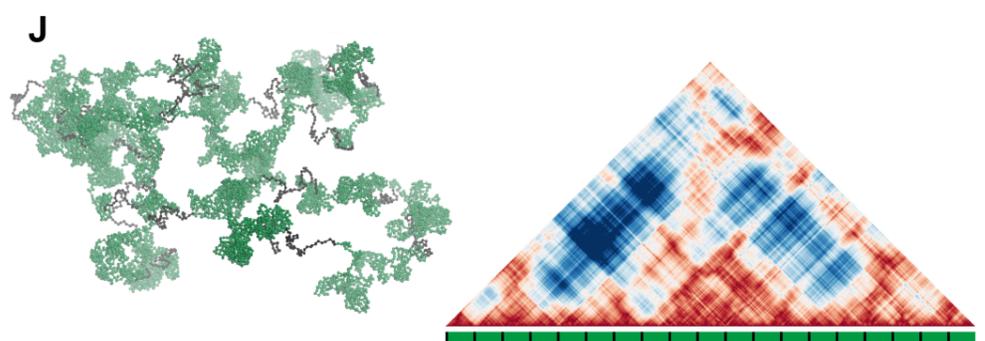
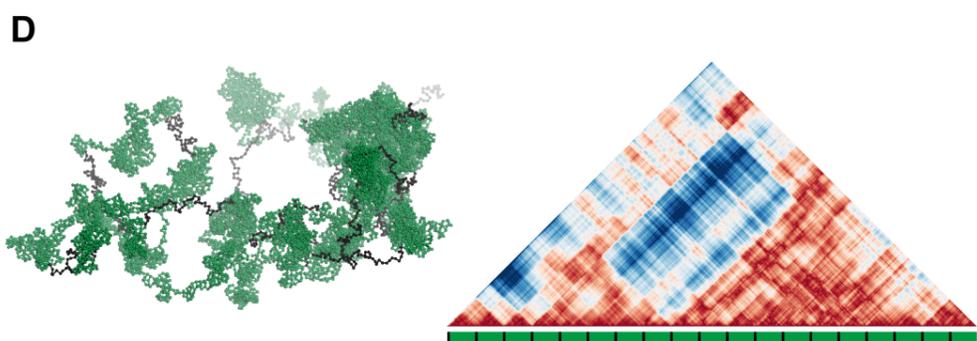
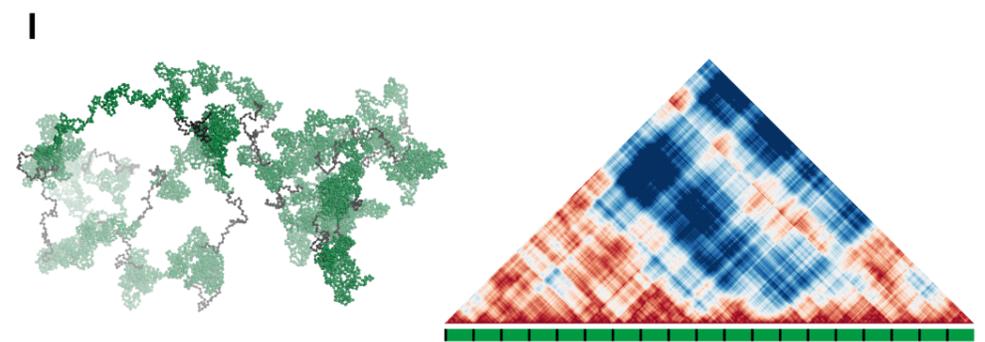
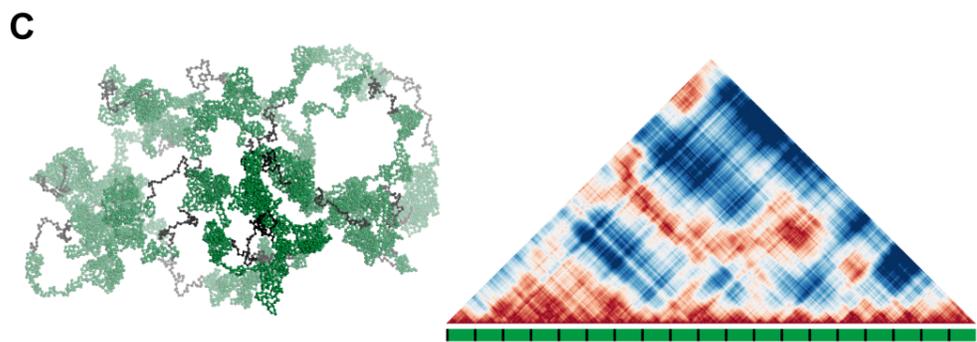
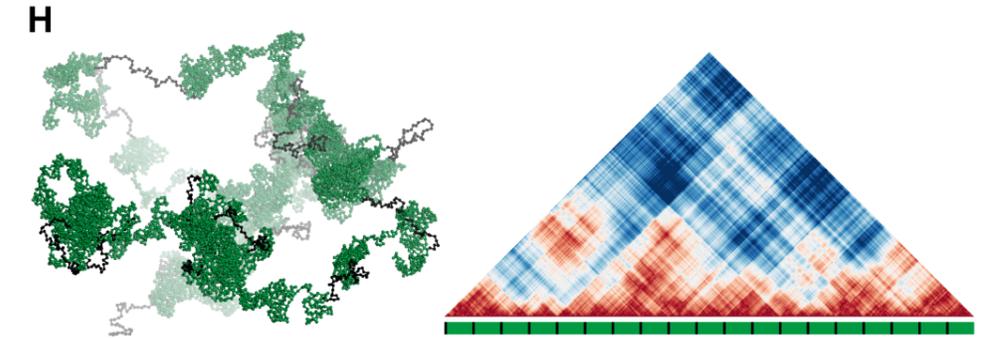
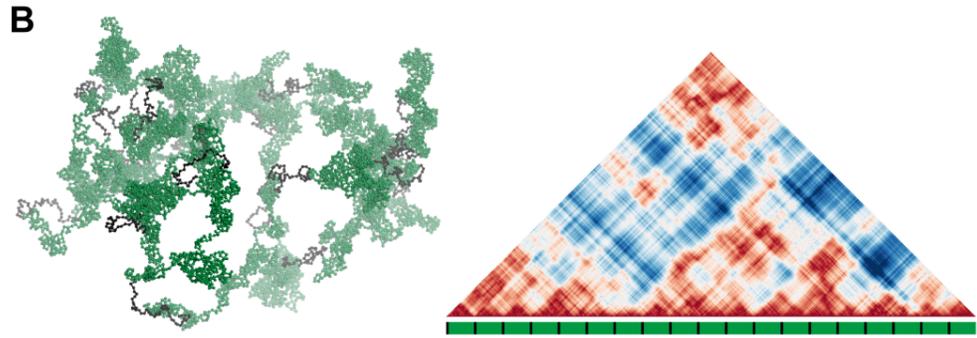
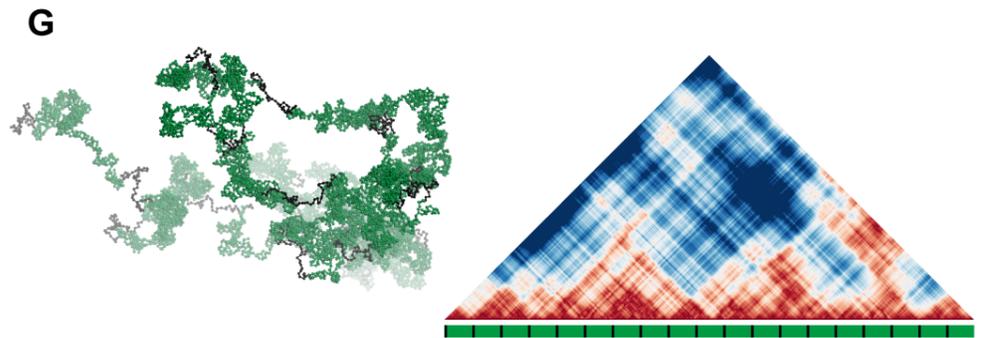
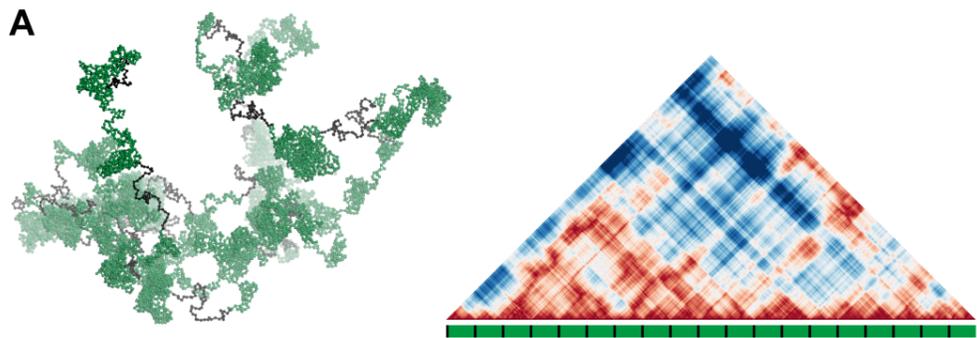


Schematic representation of the groups of bins:



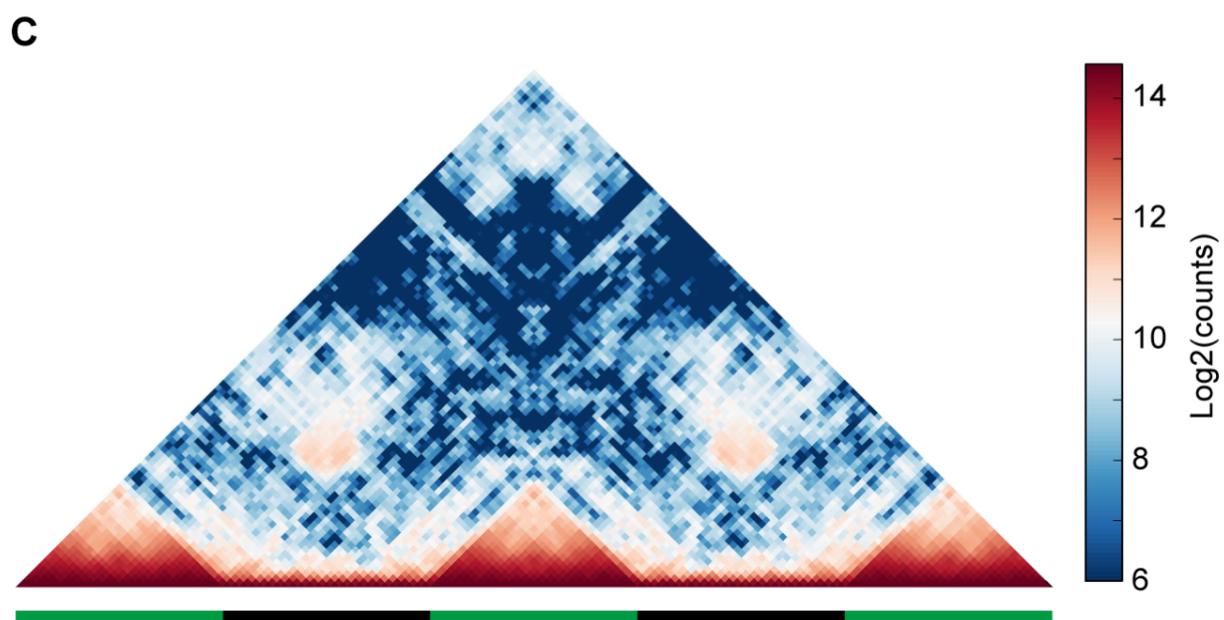
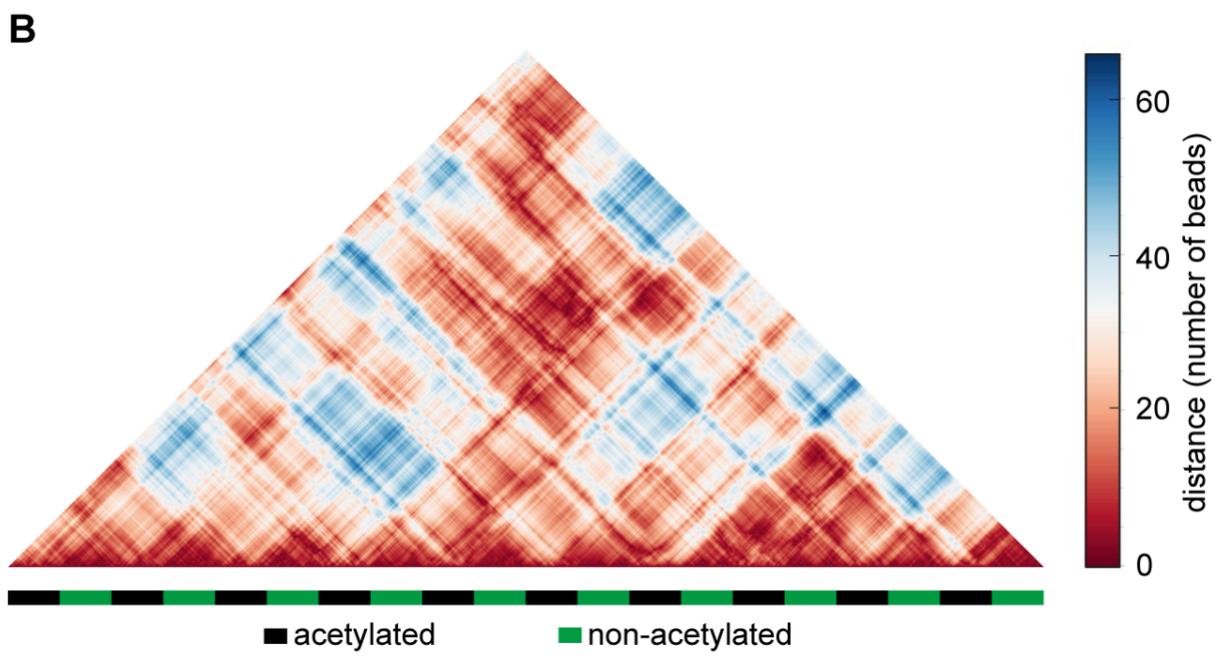
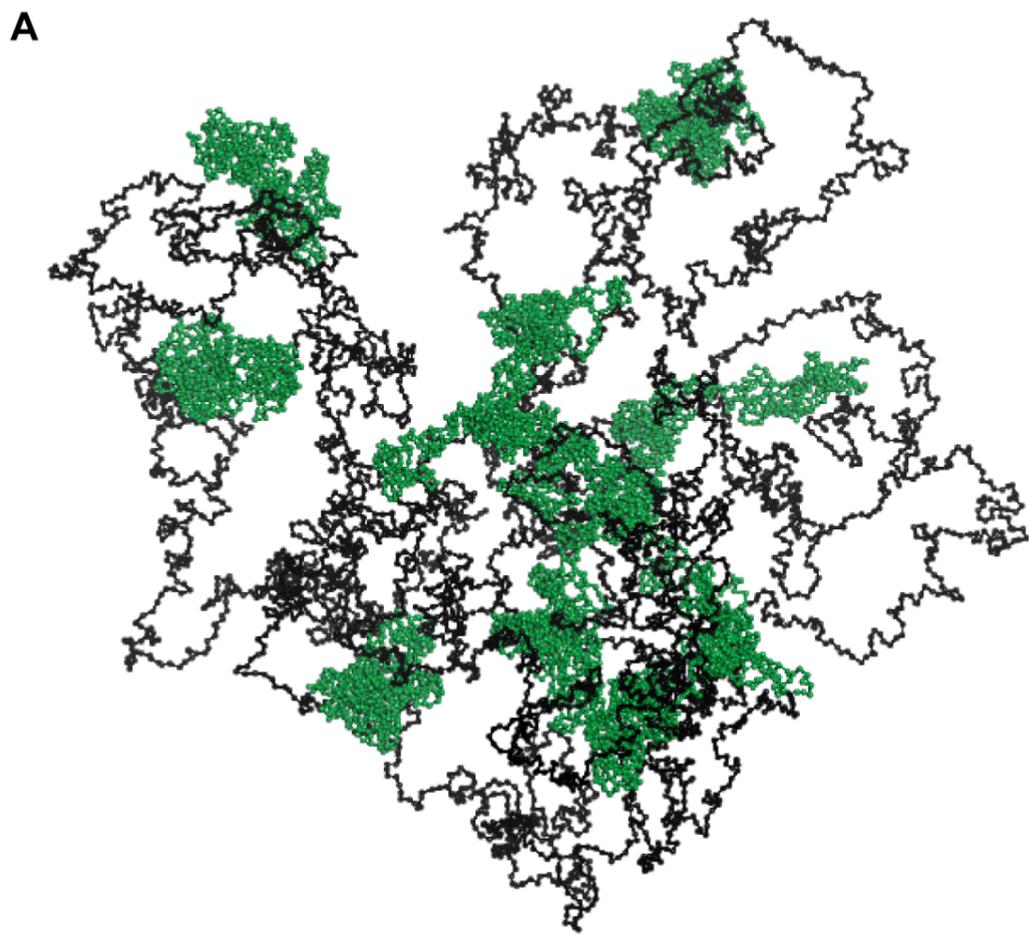


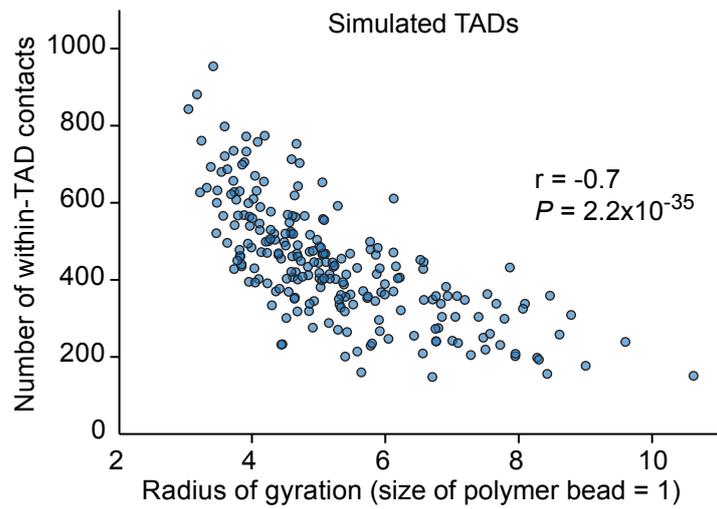
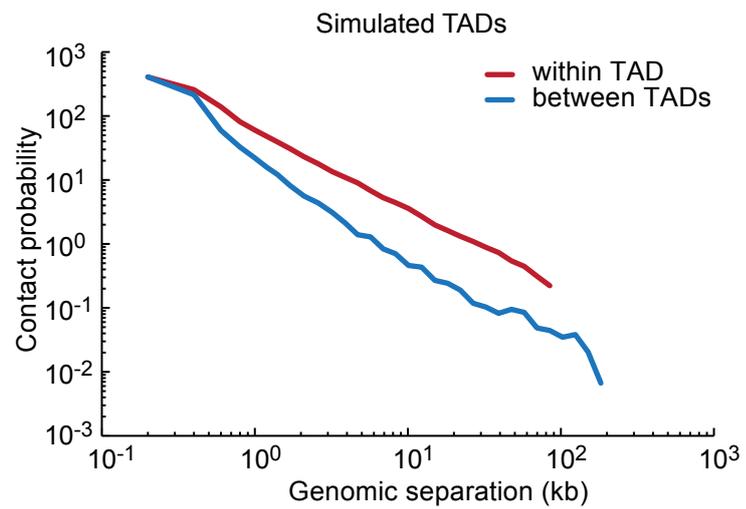


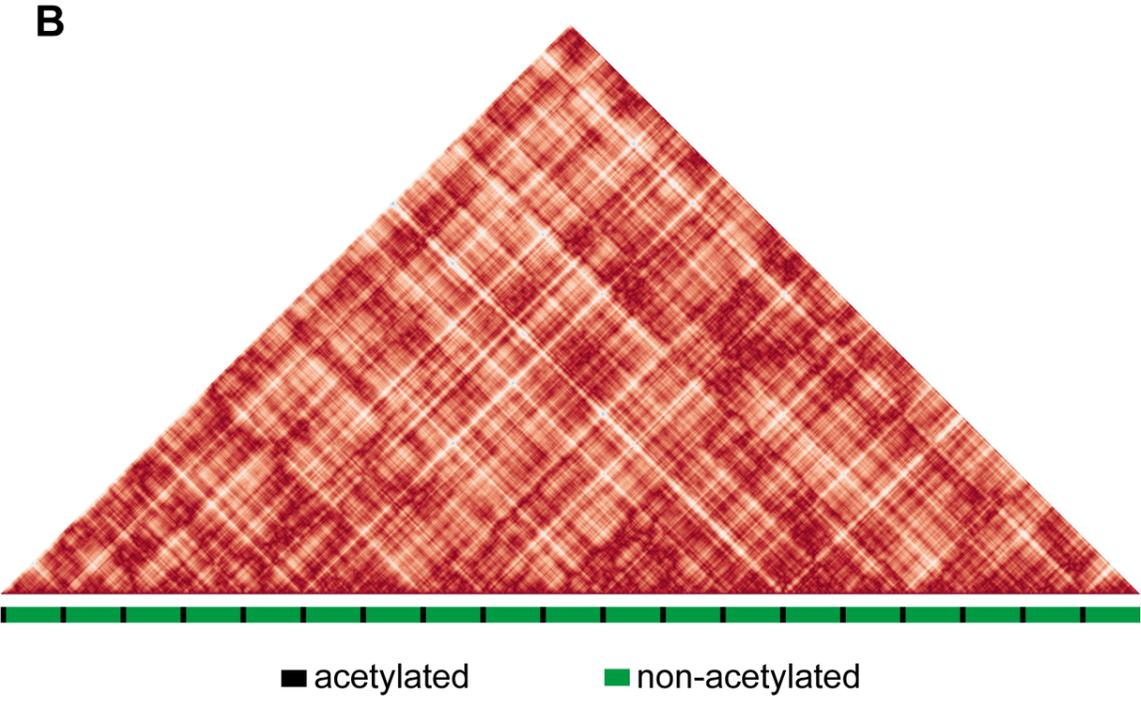
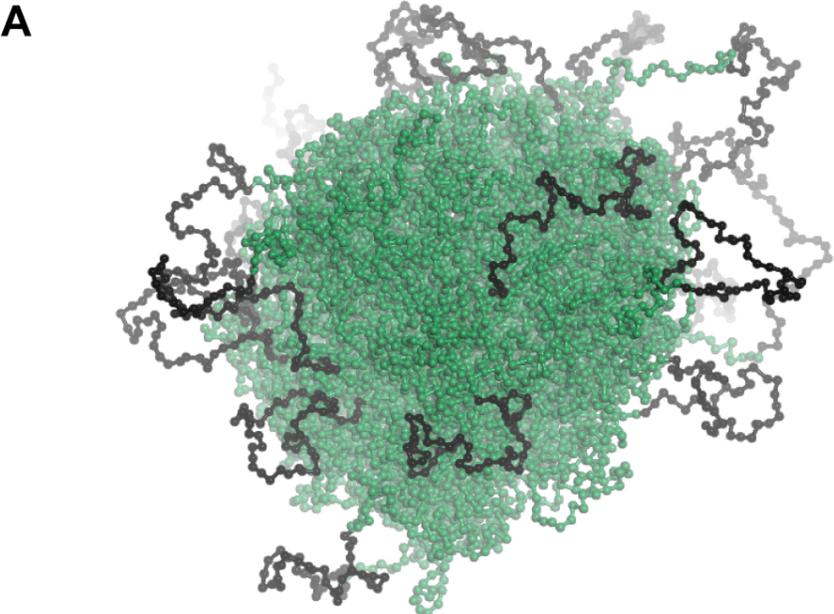


■ acetylated

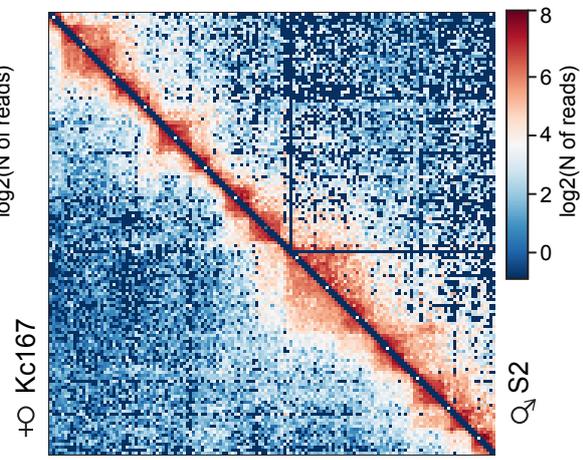
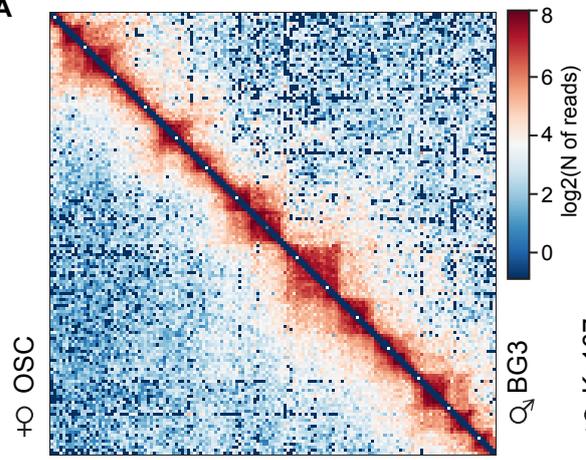
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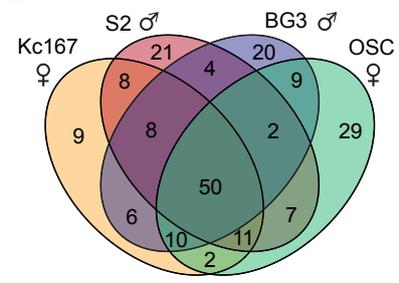
A**B**



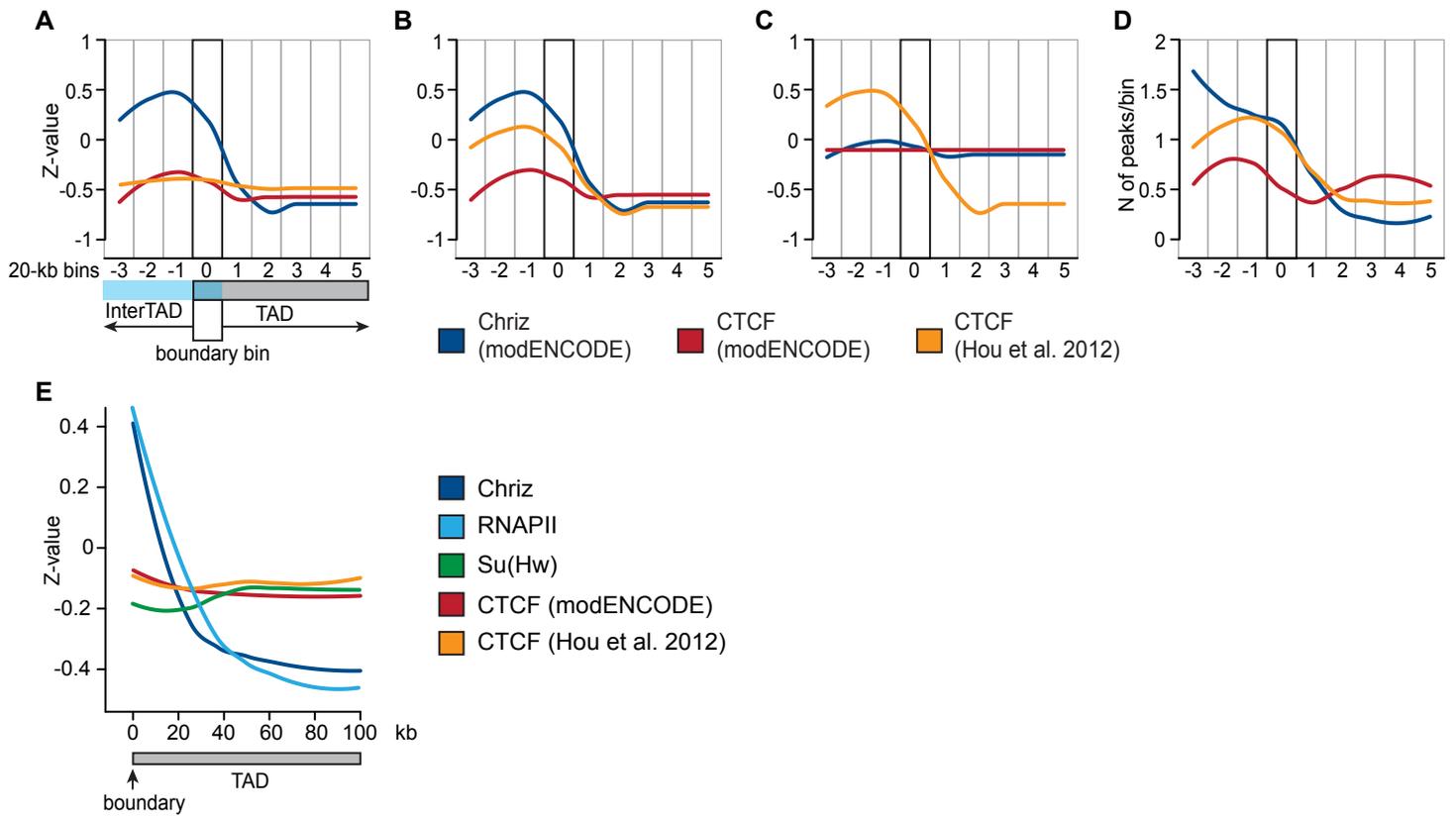
A



B



Ulianov_FigS18



SUPPLEMENTAL TABLE LEGENDS

Supplemental Table S1. Pearson correlation coefficients between biological replicates of the Hi-C experiments.

Supplemental Table S2. Numbers of Hi-C read pairs aligned to the *Drosophila* reference genome dm3 and filtered before the iterative correction step of the Hi-C data processing. Total reads – number of reads aligned to the reference genome; DS reads – read pairs in which both reads were aligned to the genome; self-circles – read pairs in which both reads were aligned to the same self-circularized restriction fragment; dangling ends – read pairs aligned at the same restriction site; duplicates – any two pairs of reads aligned to identical genomic positions.

Supplemental Table S3. Manually annotated long-range contacts on Hi-C heatmaps for all cell lines. Genomic positions of the midpoints of interacting regions are shown. Genomic rearrangements were annotated using the Delly and BreakDancer algorithms.

Supplemental Table S4. Number, genome coverage and sizes of TADs and inter-TADs in all cell lines, predicted by the Armatus algorithm.

Supplemental Table S5. Genomic positions of TADs in the four studied cell lines after the second step of TAD prediction procedure (splitting of TADs larger than 600 kb by the Armatus algorithm with the increased value of the scaling parameter gamma). The gamma values used at both steps of the procedure are shown.

Supplemental Table S6. P-values for the two-sided Mann-Whitney test used (i) to compare proportions of chromatin colors, as well as Z-values of epigenetic marks within the genomic bins belonging to TADs and inter-TADs (relative to Figures 2B, 2C, 5B and 5C); (ii) to compare proportions of chromatin colors within the genomic bins identified as belonging to inter-TADs at gamma values less than 1.5 and more than 1.5 (relative to Figure 3C and Supplemental Figure S5B).

Supplemental Table S7. Numbers of reads used for poly(A)+ RNA-seq.

Supplemental Table S8. Numbers of differentially and equally transcribed genomic bins in the four groups of bins identified in a pairwise comparison of TAD positions in the four

studied cell lines. Chi-square test and two-sided Fisher`s exact test were performed for the framed values.

Supplemental Table S9. Numbers of genomic bins containing different or equal proportion of active chromatin in the four groups of bins identified in a pairwise comparison of TAD positions in the four studied cell lines. Chi-square test and two-sided Fisher`s exact test were performed for the framed values.

Supplemental Table S10. Groups of tissue samples from Brown et al. (2013) used to compose a list of tissue-specific genes.

Supplemental Table S11. Genomic rearrangements annotated in the four studied cell lines by the Delly and BreakDancer algorithms. DEL – deletion, DUP – duplication, INV – inversion, TRA – translocation, INS – insertion, CTX – interchromosomal translocation, UNK – unknown, SUM - the number of all annotated genomic rearrangements.

Supplemental Table S1.

chr2L	BG3_rep1	BG3_rep2	OSC_rep1	OSC_rep2	S2_rep1	S2_rep2	Kc167_rep1	Kc167_rep2
BG3_rep1	1,000	0,950	0,924	0,932	0,893	0,883	0,899	0,887
BG3_rep2	0,950	1,000	0,926	0,933	0,893	0,883	0,899	0,887
OSC_rep1	0,924	0,926	1,000	0,945	0,881	0,871	0,886	0,876
OSC_rep2	0,932	0,933	0,945	1,000	0,883	0,874	0,894	0,882
S2_rep1	0,893	0,893	0,881	0,883	1,000	0,910	0,884	0,875
S2_rep2	0,883	0,883	0,871	0,874	0,910	1,000	0,874	0,864
Kc167_rep1	0,899	0,899	0,886	0,894	0,884	0,874	1,000	0,903
Kc167_rep2	0,887	0,887	0,876	0,882	0,875	0,864	0,903	1,000

chr2R	BG3_rep1	BG3_rep2	OSC_rep1	OSC_rep2	S2_rep1	S2_rep2	Kc167_rep1	Kc167_rep2
BG3_rep1	1,000	0,948	0,913	0,920	0,892	0,878	0,894	0,882
BG3_rep2	0,948	1,000	0,917	0,925	0,895	0,881	0,896	0,883
OSC_rep1	0,913	0,917	1,000	0,935	0,888	0,875	0,882	0,870
OSC_rep2	0,920	0,925	0,935	1,000	0,892	0,879	0,889	0,876
S2_rep1	0,892	0,895	0,888	0,892	1,000	0,904	0,879	0,866
S2_rep2	0,878	0,881	0,875	0,879	0,904	1,000	0,865	0,853
Kc167_rep1	0,894	0,896	0,882	0,889	0,879	0,865	1,000	0,888
Kc167_rep2	0,882	0,883	0,870	0,876	0,866	0,853	0,888	1,000

chr3L	BG3_rep1	BG3_rep2	OSC_rep1	OSC_rep2	S2_rep1	S2_rep2	Kc167_rep1	Kc167_rep2
BG3_rep1	1,000	0,947	0,902	0,908	0,889	0,874	0,886	0,875
BG3_rep2	0,947	1,000	0,902	0,909	0,890	0,874	0,887	0,876
OSC_rep1	0,902	0,902	1,000	0,938	0,875	0,864	0,874	0,860
OSC_rep2	0,908	0,909	0,938	1,000	0,880	0,868	0,882	0,870
S2_rep1	0,889	0,890	0,875	0,880	1,000	0,902	0,875	0,865
S2_rep2	0,874	0,874	0,864	0,868	0,902	1,000	0,864	0,853
Kc167_rep1	0,886	0,887	0,874	0,882	0,875	0,864	1,000	0,890
Kc167_rep2	0,875	0,876	0,860	0,870	0,865	0,853	0,890	1,000

chr3R	BG3_rep1	BG3_rep2	OSC_rep1	OSC_rep2	S2_rep1	S2_rep2	Kc167_rep1	Kc167_rep2
BG3_rep1	1,000	0,945	0,915	0,923	0,904	0,889	0,897	0,886
BG3_rep2	0,945	1,000	0,917	0,925	0,905	0,889	0,899	0,887
OSC_rep1	0,915	0,917	1,000	0,933	0,887	0,872	0,883	0,873
OSC_rep2	0,923	0,925	0,933	1,000	0,891	0,877	0,891	0,881
S2_rep1	0,904	0,905	0,887	0,891	1,000	0,903	0,881	0,872
S2_rep2	0,889	0,889	0,872	0,877	0,903	1,000	0,867	0,858
Kc167_rep1	0,897	0,899	0,883	0,891	0,881	0,867	1,000	0,890
Kc167_rep2	0,886	0,887	0,873	0,881	0,872	0,858	0,890	1,000

chr4	BG3_rep1	BG3_rep2	OSC_rep1	OSC_rep2	S2_rep1	S2_rep2	Kc167_rep1	Kc167_rep2
BG3_rep1	1,000	0,983	0,942	0,945	0,936	0,930	0,909	0,902
BG3_rep2	0,983	1,000	0,941	0,944	0,935	0,929	0,905	0,901
OSC_rep1	0,942	0,941	1,000	0,975	0,916	0,912	0,897	0,892
OSC_rep2	0,945	0,944	0,975	1,000	0,918	0,914	0,907	0,903
S2_rep1	0,936	0,935	0,916	0,918	1,000	0,942	0,885	0,883
S2_rep2	0,930	0,929	0,912	0,914	0,942	1,000	0,875	0,870
Kc167_rep1	0,909	0,905	0,897	0,907	0,885	0,875	1,000	0,919
Kc167_rep2	0,902	0,901	0,892	0,903	0,883	0,870	0,919	1,000

chrX	BG3_rep1	BG3_rep2	OSC_rep1	OSC_rep2	S2_rep1	S2_rep2	Kc167_rep1	Kc167_rep2
BG3_rep1	1,000	0,845	0,836	0,845	0,751	0,713	0,808	0,797
BG3_rep2	0,845	1,000	0,842	0,851	0,754	0,717	0,815	0,804
OSC_rep1	0,836	0,842	1,000	0,920	0,789	0,750	0,845	0,837
OSC_rep2	0,845	0,851	0,920	1,000	0,792	0,754	0,852	0,842
S2_rep1	0,751	0,754	0,789	0,792	1,000	0,732	0,777	0,764
S2_rep2	0,713	0,717	0,750	0,754	0,732	1,000	0,739	0,729
Kc167_rep1	0,808	0,815	0,845	0,852	0,777	0,739	1,000	0,854
Kc167_rep2	0,797	0,804	0,837	0,842	0,764	0,729	0,854	1,000

Supplemental Table S2.

Sample	Total reads	Total DS reads	Reads from the same fragment			Dangling ends of fragments separated by 500 bp or less	Valid pairs	Mapped near the restriction site	Duplicates	Reads from large (>100 kb) and small (<100 bp) fragments	0.5% of the most overrepresented fragments	N of reads after all filters
			Total	Self-circles	Dangling ends							
DmBG3-c2_combined	57 686 069	49 453 011	20 471 016	260 927	19 924 229	5 773 417	23 208 578	303 795	595 925	93 858	786 547	21 428 453
DmBG3-c2_rep1	30 300 932	26 214 543	11 969 368	127 680	11 684 813	2 926 955	11 318 220	146 564	287 558	47 703	379 271	10 457 124
DmBG3-c2_rep2	27 385 137	23 238 468	8 501 648	133 247	8 239 416	2 846 462	11 890 358	157 231	307 655	46 157	401 651	10 977 664
Kc167_combined	38 750 262	33 887 737	19 803 814	150 227	19 423 728	3 907 113	10 176 810	115 740	562 544	58 948	350 019	9 089 559
Kc167_rep1	16 696 812	14 650 515	8 811 499	57 819	8 657 210	1 772 010	4 067 006	49 405	230 004	24 305	140 959	3 622 333
Kc167_rep2	16 646 771	14 492 433	8 139 501	73 577	7 963 586	1 561 592	4 791 340	50 097	165 847	27 420	170 952	4 377 024
OSC_combined	57 526 086	48 264 746	21 192 418	239 524	20 693 278	5 867 448	21 204 880	366 284	481 319	87 304	596 641	19 673 332
OSC_rep1	24 771 372	20 764 840	8 852 306	99 949	8 643 955	2 536 283	9 376 251	142 571	167 572	35 299	266 212	8 764 597
OSC_rep2	32 754 714	27 499 906	12 340 112	139 575	12 049 323	3 331 165	11 828 629	223 713	313 183	52 008	326 891	10 912 834
S2_combined	42 656 628	34 317 102	14 014 451	172 352	13 591 531	5 427 129	14 875 522	160 109	1 140 083	74 735	608 789	12 891 806
S2_rep1	17 504 801	14 143 982	6 359 657	62 420	6 176 842	2 234 142	5 550 183	61 206	652 026	30 133	213 540	4 593 278
S2_rep2	17 503 379	14 017 647	5 118 208	79 494	4 954 171	2 219 157	6 680 282	70 454	110 004	32 313	296 818	6 170 693

Supplemental Table S3.

S2

No	Chr	Position of the left partner, kb	Position of the right partner, kb	Distance between the partners, kb	In the other datasets	Genomic rearrangements between the partners
1	2L	600	1640	1040	No	Deletion, inversion, deletion
2	2L	2240	2900	660	No	Duplication head-to-tail
3	2L	5620	9640	4020	No	Numerous deletions and inversions
4	2L	7480	21220	13740	No	Deletion
5	2L	13000	13600	600	No	Duplication head-to-tail
6	2R	3660	4920	1260	No	Deletion
7	2R	3680	4820	1140	No	Deletion
8	2R	7340	8780	1440	No	Inversion
9	2R	13260	14340	1080	No	Duplication head-to-tail
10	2R	15000	15340	340	No	Deletion
11	3L	1140	2880	1740	No	Deletion
12	3L	8680	10700	2020	No	Deletion
13	3L	9400	9880	480	No	Duplication head-to-tail
14	3L	9740	11240	1500	No	Deletion
15	3L	12600	16280	3680	No	Inversion
16	3L	18940	20880	1940	No	Inversion
17	3L	21040	21720	680	No	Duplication head-to-tail
18	3L	21100	22480	1380	No	Duplication head-to-tail
19	3R	15720	16880	1160	No	Duplication head-to-tail
20	3R	16820	17460	640	No	Duplication head-to-tail
21	3R	18400	19100	700	No	Duplication head-to-tail
22	X	900	15920	15020	BG3, OCS, Sexton et al., 2012	No
23	X	14080	16440	2360	No	Deletion
24	X	17900	19180	1280	No	Duplication head-to-tail
Kc167						
1	2L	5940	8780	2840	Hou et al., 2012	Deletion
2	2L	9180	13540	4360	No	Duplication head-to-tail
3	2R	6100	8840	2740	No	Duplication head-to-tail
4	2R	7020	11980	4960	No	Inversion
5	3L	8740	12980	4240	No	Deletion
6	3L	15920	19100	3180	No	Inversion
7	3R	2800	4660	1860	No	Inversion
8	3R	14480	14940	460	No	Deletion
9	3R	18120	18960	840	No	Duplication head-to-tail
10	3R	23600	24680	1080	No	Deletion
11	3R	26280	27100	820	No	Duplication head-to-tail
12	X	1160	2240	1080	No	Duplication head-to-tail
BG3						
1	2L	920	2600	1680	OSC, Sexton et al., 2012	No
2	2L	1980	4780	2800	OSC, Sexton et al., 2012	No

3	2L	6400	9080	2680	OSC, Sexton et al., 2012	No
4	2L	6400	9460	3060	OSC, Sexton et al., 2012	No
5	2L	9080	9460	380	OSC, Sexton et al., 2012	No
6	2L	20120	21680	1560	OSC, Sexton et al., 2012	No
7	2R	4660	10880	6220	OSC, Sexton et al., 2012	No
8	2R	20640	21080	440	No	Inversion
9	3L	200	1260	1060	No	No
10	3L	2720	5140	2420	No	Duplication, Inversion
11	3L	6640	9320	2680	OSC, Sexton et al., 2012, Hou et al., 2012	No
12	3R	7040	ChrX: 12060		No	Translocation
14	X	900	15920	15020	OSC, S2, Sexton et al., 2012	No
13	X	1500	4280	2780	No	Duplication head-to-tail
15	X	13240	19240	6000	OSC, Sexton et al., 2012	No

OSC

1	2L	920	2600	1680	BG3, Sexton et al., 2012	No
2	2L	1980	4780	2800	BG3, Sexton et al., 2012	No
3	2L	6400	9080	2680	BG3, Sexton et al., 2012	No
4	2L	6400	9460	3060	BG3, Sexton et al., 2012	No
5	2L	9080	9460	380	2BG3, Sexton et al., 2016	No
6	2L	20120	21680	1560	BG3, Sexton et al., 2012	No
7	2R	4660	10880	6220	BG3, Sexton et al., 2012	No
8	3L	1760	2200	440	No	Inversion
9	3L	1980	3560	1580	Hou et al., 2012	Duplication head-to-tail
10	3L	6640	9320	2680	BG3, Sexton et al., 2012, Hou et al., 2012	No
11	3L	7640	17240	9600	Sexton et al., 2012	No
12	3L	12840	20000	7160	Sexton et al., 2012	No
13	3R	980	7860	6880	Sexton et al., 2012	No
14	3R	22260	23060	800	No	Deletion
15	X	900	15920	15020	BG3, S2, Sexton et al., 2012	No
16	X	13240	19240	6000	BG3, Sexton et al., 2012	No

Supplemental Table S4.

Cell lines (gamma values used for the two-step TADs annotation)

S2 (1.26, 2.52)

Kc167 (1.21, 2.42)

BG3 (1.07, 2.14)

OSC (1.03, 2.06)

Total number	TADs	583	582	580	577
Genome coverage, %	TAD interior	67	66	63	63
	TAD interior + boundary bins	87	85	82	83
	inter-TADs + boundary bins	33	34	37	37
Average size, kb	TAD interior	137	134	129	131
	TAD interior + boundary bins	177	174	169	171
	inter-TADs + boundary bins	67	70	76	75
Median size, kb	TAD interior	100	100	100	100
	TAD interior + boundary bins	140	140	140	140
	inter-TADs + boundary bins	40	40	40	40
Minimal size, kb	TAD interior	40	40	40	40
	TAD interior + boundary bins	80	80	80	80
	inter-TADs + boundary bins	40	40	40	40
Maximal size, kb	TAD interior	540	640	540	540
	TAD interior + boundary bins	580	680	580	580
	inter-TADs + boundary bins	480	460	360	360

Supplemental Table S5.

S2, $\gamma1 = 1.26,$ $\gamma2 = 2.52$				Kc167, $\gamma1 = 1.21,$ $\gamma2 = 2.42$				BG3, $\gamma1 = 1.07,$ $\gamma2 = 2.14$				OSC, $\gamma1 = 1.03,$ $\gamma2 = 2.06$			
№	Chr	5'-end, kb	3'-end, kb	№	Chr	5'-end, kb	3'-end, kb	№	Chr	5'-end, kb	3'-end, kb	№	Chr	5'-end, kb	3'-end, kb
1	2L	0	120	1	2L	0	80	1	2L	0	80	1	2L	180	260
2	2L	120	280	2	2L	80	240	2	2L	160	260	2	2L	300	420
3	2L	280	440	3	2L	300	440	3	2L	300	420	3	2L	480	560
4	2L	580	820	4	2L	480	560	4	2L	480	560	4	2L	560	820
5	2L	860	980	5	2L	560	820	5	2L	560	820	5	2L	880	960
6	2L	1020	1180	6	2L	880	980	6	2L	860	960	6	2L	1000	1080
7	2L	1180	1640	7	2L	1020	1120	7	2L	1000	1080	7	2L	1140	1640
8	2L	1640	1720	8	2L	1240	1620	8	2L	1280	1620	8	2L	1640	1720
9	2L	1720	1940	9	2L	1620	1740	9	2L	1620	1720	9	2L	1720	1920
10	2L	1980	2160	10	2L	1740	1920	10	2L	1720	1980	10	2L	1980	2160
11	2L	2160	2240	11	2L	1980	2140	11	2L	2020	2160	11	2L	2220	2360
12	2L	2240	2360	12	2L	2220	2360	12	2L	2220	2360	12	2L	2360	2740
13	2L	2360	2620	13	2L	2360	2580	13	2L	2360	2600	13	2L	2880	2960
14	2L	2660	2740	14	2L	2640	2740	14	2L	2640	2720	14	2L	2960	3040
15	2L	2740	2860	15	2L	2740	2860	15	2L	2760	2860	15	2L	3140	3340
16	2L	2860	2980	16	2L	2860	2980	16	2L	2860	2980	16	2L	3340	3480
17	2L	3040	3160	17	2L	2980	3060	17	2L	3040	3140	17	2L	3480	3640
18	2L	3160	3360	18	2L	3160	3300	18	2L	3140	3360	18	2L	3640	3800
19	2L	3360	3460	19	2L	3360	3460	19	2L	3360	3460	19	2L	3800	4220
20	2L	3460	3660	20	2L	3520	3640	20	2L	3460	3640	20	2L	4220	4340
21	2L	3660	3800	21	2L	3640	3800	21	2L	3640	3780	21	2L	4460	4820
22	2L	3800	4340	22	2L	3860	4180	22	2L	3780	3860	22	2L	4820	4900
23	2L	4460	4820	23	2L	4180	4340	23	2L	3860	4380	23	2L	5100	5240
24	2L	4820	4960	24	2L	4340	4440	24	2L	4440	4820	24	2L	5240	5320
25	2L	5000	5080	25	2L	4440	4820	25	2L	4820	4960	25	2L	5320	5520
26	2L	5080	5280	26	2L	4820	4980	26	2L	5060	5280	26	2L	5520	5960
27	2L	5320	5520	27	2L	5100	5280	27	2L	5320	5540	27	2L	6100	6200
28	2L	5520	5980	28	2L	5320	5520	28	2L	5540	5800	28	2L	6240	6340
29	2L	5980	6060	29	2L	5520	5960	29	2L	5800	5940	29	2L	6340	6420
30	2L	6100	6340	30	2L	6080	6360	30	2L	5980	6060	30	2L	6720	6800
31	2L	6340	6460	31	2L	6360	6460	31	2L	6100	6340	31	2L	6800	6920
32	2L	6500	6640	32	2L	6500	6620	32	2L	6340	6460	32	2L	6960	7040
33	2L	6700	6960	33	2L	6660	6960	33	2L	6500	6620	33	2L	7040	7220
34	2L	6960	7040	34	2L	6960	7040	34	2L	6660	6960	34	2L	7220	7420
35	2L	7040	7220	35	2L	7040	7220	35	2L	7060	7220	35	2L	7460	7820
36	2L	7220	7420	36	2L	7220	7420	36	2L	7220	7420	36	2L	7880	7980
37	2L	7500	7820	37	2L	7420	7820	37	2L	7420	7540	37	2L	8220	8320
38	2L	7820	7980	38	2L	7820	7980	38	2L	7540	7800	38	2L	8420	8540
39	2L	8020	8100	39	2L	8040	8120	39	2L	7840	8000	39	2L	8540	8700
40	2L	8100	8240	40	2L	8120	8220	40	2L	8040	8220	40	2L	8700	8940
41	2L	8240	8340	41	2L	8220	8300	41	2L	8220	8320	41	2L	8940	9520
42	2L	8340	8420	42	2L	8300	8420	42	2L	8420	8540	42	2L	9580	9700
43	2L	8420	8540	43	2L	8420	8540	43	2L	8540	8700	43	2L	9700	9900
44	2L	8540	8680	44	2L	8540	8700	44	2L	8700	8940	44	2L	10020	10200

45	2L	8740	8940	45	2L	8700	8940	45	2L	9000	9080	45	2L	10420	10500
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4	2R	740	980	4	2R	1580	1900	4	2R	460	720	4	2R	720	1000
5	2R	1320	1480	5	2R	1900	2040	5	2R	720	1000	5	2R	1000	1260
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46	2R	9020	9120	46	2R	9380	9460	46	2R	9740	9820	46	2R	9120	9320
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4	3L	340	500	4	3L	340	520	4	3L	740	840	4	3L	340	520
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6	3L	660	840	6	3L	660	840	6	3L	1100	1180	6	3L	660	840
7	3L	900	980	7	3L	840	1080	7	3L	1180	1300	7	3L	840	1040
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9	3L	1200	1300	9	3L	1200	1300	9	3L	1600	1680	9	3L	1180	1300
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1	3R	0	140	1	3R	20	240	1	3R	0	240	1	3R	0	240
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3	3R	300	480	3	3R	480	640	3	3R	540	640	3	3R	540	640
4	3R	480	640	4	3R	640	740	4	3R	640	740	4	3R	640	740
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8	3R	1200	1300	8	3R	1300	1380	8	3R	1200	1320	8	3R	1320	1400
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17	3R	3720	3840	17	3R	3360	3500	17	3R	3080	3180	17	3R	3480	3580
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63	3R	13540	14000	63	3R	12180	12260	63	3R	14580	14720	63	3R	14480	14580
64	3R	14040	14120	64	3R	12300	12480	64	3R	14980	15480	64	3R	14580	14740
65	3R	14120	14240	65	3R	12480	12800	65	3R	15480	15680	65	3R	14740	14820
66	3R	14240	14400	66	3R	12800	12880	66	3R	15740	15840	66	3R	14980	15480
67	3R	14400	14480	67	3R	12940	13440	67	3R	15880	16080	67	3R	15480	15680
68	3R	14480	14580	68	3R	13540	14000	68	3R	16140	16280	68	3R	15740	15840
69	3R	14580	14740	69	3R	14140	14220	69	3R	16280	16360	69	3R	15880	15980
70	3R	14740	14820	70	3R	14260	14400	70	3R	16460	16600	70	3R	15980	16080
71	3R	14980	15480	71	3R	14400	14480	71	3R	16660	16780	71	3R	16080	16160
72	3R	15480	15680	72	3R	14480	14580	72	3R	16780	16900	72	3R	16160	16280
73	3R	15860	16080	73	3R	14580	14740	73	3R	17020	17120	73	3R	16280	16360
74	3R	16160	16360	74	3R	14820	14920	74	3R	17120	17200	74	3R	16460	16600
75	3R	16460	16580	75	3R	14980	15480	75	3R	17200	17320	75	3R	16660	16780
76	3R	16580	16660	76	3R	15480	15680	76	3R	17320	17440	76	3R	16780	16900
77	3R	16660	16780	77	3R	15740	16080	77	3R	17480	17700	77	3R	17020	17200
78	3R	16780	16880	78	3R	16140	16380	78	3R	17700	17860	78	3R	17200	17340
79	3R	16960	17040	79	3R	16380	16460	79	3R	17860	18200	79	3R	17340	17440
80	3R	17040	17120	80	3R	16460	16580	80	3R	18200	18280	80	3R	17480	17640
81	3R	17120	17200	81	3R	16580	16660	81	3R	18280	18420	81	3R	17700	17860
82	3R	17200	17320	82	3R	16660	16780	82	3R	18460	18540	82	3R	17860	18200
83	3R	17320	17420	83	3R	16780	16900	83	3R	18580	18960	83	3R	18200	18280
84	3R	17480	17700	84	3R	17040	17120	84	3R	19040	19120	84	3R	18280	18360
85	3R	17700	17860	85	3R	17120	17200	85	3R	19180	19540	85	3R	18560	18960
86	3R	17860	18200	86	3R	17200	17340	86	3R	19640	19720	86	3R	19160	19500

87	3R	18200	18280	87	3R	17340	17480	87	3R	19980	20060	87	3R	19640	19720
88	3R	18280	18400	88	3R	17480	17660	88	3R	20160	20360	88	3R	19760	19860
89	3R	18560	18960	89	3R	17700	17860	89	3R	20480	20580	89	3R	20120	20380
90	3R	19180	19540	90	3R	17860	18180	90	3R	20680	20760	90	3R	20480	20640
91	3R	19640	19760	91	3R	18280	18400	91	3R	20760	20860	91	3R	20760	20860
92	3R	19760	19880	92	3R	18580	18840	92	3R	20900	21080	92	3R	20920	21040
93	3R	19940	20080	93	3R	18880	18960	93	3R	21080	21320	93	3R	21080	21320
94	3R	20120	20380	94	3R	19160	19540	94	3R	21320	21460	94	3R	21320	21440
95	3R	20480	20580	95	3R	19640	19740	95	3R	21500	21800	95	3R	21500	21700
96	3R	20700	20880	96	3R	19780	19880	96	3R	21800	21880	96	3R	21700	21840
97	3R	20880	20960	97	3R	19940	20060	97	3R	21880	22280	97	3R	21940	22020
98	3R	20960	21040	98	3R	20100	20380	98	3R	22280	22380	98	3R	22020	22100
99	3R	21080	21320	99	3R	20460	20700	99	3R	22380	22700	99	3R	22100	22280
100	3R	21320	21460	100	3R	20700	20860	100	3R	22700	22780	100	3R	22280	22380
101	3R	21500	21700	101	3R	20900	21080	101	3R	22780	22940	101	3R	22380	22460
102	3R	21700	21800	102	3R	21080	21320	102	3R	22980	23060	102	3R	22460	22560
103	3R	21800	21880	103	3R	21320	21460	103	3R	23120	23380	103	3R	22560	22680
104	3R	21940	22080	104	3R	21500	21720	104	3R	23420	23500	104	3R	22820	22940
105	3R	22080	22280	105	3R	21720	21800	105	3R	23540	23740	105	3R	22980	23060
106	3R	22280	22360	106	3R	21800	21880	106	3R	23780	24120	106	3R	23160	23380
107	3R	22360	22700	107	3R	21940	22360	107	3R	24160	24360	107	3R	23420	23500
108	3R	22700	22780	108	3R	22360	22640	108	3R	24460	24660	108	3R	23580	23740
109	3R	22780	22980	109	3R	22700	22780	109	3R	24720	24860	109	3R	23780	24160
110	3R	22980	23060	110	3R	22780	22980	110	3R	24860	24960	110	3R	24160	24360
111	3R	23120	23380	111	3R	22980	23080	111	3R	25060	25140	111	3R	24360	24440
112	3R	23420	23500	112	3R	23120	23400	112	3R	25140	25300	112	3R	24500	24660
113	3R	23540	23740	113	3R	23400	23500	113	3R	25300	25560	113	3R	24700	24860
114	3R	23780	24140	114	3R	23500	23740	114	3R	25560	25640	114	3R	24860	24940
115	3R	24180	24360	115	3R	23780	24160	115	3R	25680	25840	115	3R	25040	25320
116	3R	24360	24440	116	3R	24160	24460	116	3R	25880	26020	116	3R	25320	25580
117	3R	24440	24660	117	3R	24500	24580	117	3R	26060	26280	117	3R	25680	25840
118	3R	24720	24860	118	3R	24720	24860	118	3R	26280	26620	118	3R	25880	26020
119	3R	25140	25320	119	3R	24860	24980	119	3R	26620	27060	119	3R	26060	26280
120	3R	25320	25560	120	3R	25140	25320	120	3R	27060	27220	120	3R	26280	26420
121	3R	25560	25640	121	3R	25320	25560	121	3R	27220	27440	121	3R	26420	26620
122	3R	25680	25880	122	3R	25680	25860	122	3R	27620	27880	122	3R	26620	26720
123	3R	25880	26020	123	3R	25900	26060					123	3R	26720	27060
124	3R	26060	26300	124	3R	26060	26280					124	3R	27060	27220
125	3R	26300	26600	125	3R	26280	26600					125	3R	27220	27440
126	3R	26640	27040	126	3R	26640	26720					126	3R	27560	27640
127	3R	27040	27400	127	3R	26720	27060					127	3R	27640	27800
128	3R	27400	27560	128	3R	27060	27220					128	3R	27800	27880
129	3R	27620	27880	129	3R	27280	27400								
				130	3R	27400	27500								
				131	3R	27620	27880								
1	X	20	100	1	X	20	100	1	X	40	120	1	X	20	100
2	X	140	380	2	X	160	380	2	X	160	380	2	X	140	540
3	X	380	540	3	X	380	540	3	X	380	540	3	X	540	660
4	X	540	680	4	X	540	660	4	X	540	660	4	X	660	1180

5	X	800	940	5	X	660	740	5	X	660	940	5	X	1180	1260
6	X	980	1080	6	X	800	940	6	X	980	1120	6	X	1260	1380
7	X	1180	1380	7	X	980	1100	7	X	1180	1260	7	X	1380	1760
8	X	1380	1760	8	X	1140	1260	8	X	1260	1360	8	X	1820	1900
9	X	1820	1980	9	X	1260	1380	9	X	1420	1520	9	X	1900	1980
10	X	1980	2060	10	X	1380	1780	10	X	1580	1700	10	X	2060	2140
11	X	2060	2160	11	X	1780	1980	11	X	1700	1780	11	X	2140	2240
12	X	2160	2240	12	X	2040	2160	12	X	1780	1980	12	X	2240	2340
13	X	2240	2480	13	X	2160	2240	13	X	2040	2160	13	X	2340	2520
14	X	2480	2620	14	X	2340	2420	14	X	2160	2240	14	X	2520	2620
15	X	2620	2700	15	X	2520	2620	15	X	2240	2520	15	X	2620	2700
16	X	2780	3240	16	X	2620	3060	16	X	2520	2620	16	X	2800	3060
17	X	3240	3360	17	X	3060	3240	17	X	2620	2840	17	X	3060	3240
18	X	3360	3580	18	X	3240	3360	18	X	2840	3060	18	X	3240	3360
19	X	3580	3680	19	X	3360	3580	19	X	3060	3240	19	X	3360	3580
20	X	3720	3840	20	X	3720	4000	20	X	3240	3360	20	X	3620	3700
21	X	3900	4100	21	X	4000	4240	21	X	3360	3580	21	X	3740	3860
22	X	4100	4280	22	X	4240	4440	22	X	3680	4020	22	X	3860	3980
23	X	4280	4440	23	X	4440	4540	23	X	4020	4260	23	X	4020	4100
24	X	4440	4560	24	X	4580	4800	24	X	4260	4540	24	X	4100	4200
25	X	4600	4800	25	X	4840	4960	25	X	4580	4820	25	X	4200	4280
26	X	4840	4960	26	X	5000	5220	26	X	4820	4960	26	X	4280	4440
27	X	5000	5200	27	X	5220	5320	27	X	5000	5220	27	X	4440	4540
28	X	5200	5420	28	X	5320	5420	28	X	5220	5320	28	X	4580	4820
29	X	5420	5560	29	X	5420	5520	29	X	5320	5580	29	X	4820	4960
30	X	5560	5660	30	X	5660	5780	30	X	5640	5780	30	X	5000	5200
31	X	5660	5780	31	X	5820	6120	31	X	5840	5980	31	X	5200	5320
32	X	5840	5920	32	X	6180	6260	32	X	5980	6120	32	X	5320	5400
33	X	5920	6120	33	X	6260	6420	33	X	6180	6260	33	X	5400	5580
34	X	6120	6200	34	X	6480	6560	34	X	6260	6420	34	X	5640	5800
35	X	6200	6480	35	X	6660	6740	35	X	6480	6560	35	X	5800	6180
36	X	6480	6560	36	X	6740	6900	36	X	6640	6740	36	X	6240	6440
37	X	6560	6640	37	X	7000	7180	37	X	6740	6900	37	X	6480	6560
38	X	6640	6740	38	X	7180	7580	38	X	7000	7180	38	X	6640	6740
39	X	6740	6900	39	X	7620	7800	39	X	7220	7580	39	X	6740	6900
40	X	7000	7180	40	X	7860	7940	40	X	7620	7800	40	X	6900	6980
41	X	7180	7580	41	X	8040	8140	41	X	7860	7940	41	X	6980	7180
42	X	7620	7800	42	X	8140	8300	42	X	8000	8080	42	X	7180	7600
43	X	7860	7940	43	X	8340	8460	43	X	8140	8300	43	X	7600	7820
44	X	7940	8060	44	X	8500	8600	44	X	8300	8480	44	X	7860	7940
45	X	8060	8140	45	X	8600	8760	45	X	8480	8600	45	X	8020	8320
46	X	8140	8300	46	X	8800	8980	46	X	8600	8780	46	X	8320	8420
47	X	8300	8480	47	X	8980	9060	47	X	8780	8980	47	X	8480	8600
48	X	8480	8600	48	X	9060	9180	48	X	8980	9060	48	X	8600	8780
49	X	8600	8760	49	X	9180	9460	49	X	9060	9140	49	X	8780	8980
50	X	8800	8980	50	X	9500	9580	50	X	9140	9240	50	X	8980	9060
51	X	9040	9180	51	X	9580	10120	51	X	9240	9460	51	X	9060	9140
52	X	9180	9440	52	X	10120	10220	52	X	9460	9580	52	X	9200	9480
53	X	9480	9580	53	X	10220	10360	53	X	9580	9660	53	X	9480	9560
54	X	9580	10140	54	X	10360	10500	54	X	9700	9980	54	X	9560	9660

55	X	10140	10240	55	X	10500	10640	55	X	9980	10260	55	X	9700	10220
56	X	10280	10360	56	X	10640	10720	56	X	10260	10360	56	X	10280	10360
57	X	10420	10500	57	X	10720	11020	57	X	10360	10500	57	X	10360	10680
58	X	10500	10640	58	X	11020	11240	58	X	10500	10640	58	X	10740	11000
59	X	10780	11000	59	X	11280	11360	59	X	10780	11000	59	X	11040	11240
60	X	11040	11220	60	X	11360	11460	60	X	11100	11220	60	X	11280	11360
61	X	11260	11340	61	X	11500	11580	61	X	11260	11360	61	X	11360	11460
62	X	11340	11460	62	X	11620	11700	62	X	11360	11460	62	X	11500	11580
63	X	11500	11580	63	X	11700	11780	63	X	11500	11580	63	X	11580	11720
64	X	11580	11700	64	X	11780	11920	64	X	11700	11780	64	X	11780	11900
65	X	11700	11780	65	X	11920	12480	65	X	11780	11900	65	X	11900	12480
66	X	11780	11920	66	X	12660	13020	66	X	11900	12480	66	X	12480	12560
67	X	11920	12480	67	X	13020	13160	67	X	12540	12660	67	X	12560	12660
68	X	12540	12660	68	X	13280	13520	68	X	12660	13060	68	X	12660	13020
69	X	12660	13020	69	X	13520	13620	69	X	13300	13520	69	X	13080	13180
70	X	13020	13140	70	X	13660	13740	70	X	13520	13640	70	X	13280	13520
71	X	13140	13240	71	X	13740	13900	71	X	13640	13720	71	X	13520	13640
72	X	13300	13520	72	X	13900	14100	72	X	13720	14120	72	X	13640	13720
73	X	13520	13640	73	X	14140	14720	73	X	14120	14500	73	X	13720	13900
74	X	13740	13900	74	X	14720	14980	74	X	14500	14720	74	X	13900	14100
75	X	13900	14100	75	X	14980	15220	75	X	14720	14980	75	X	14200	14400
76	X	14180	14640	76	X	15220	15360	76	X	14980	15220	76	X	14400	14480
77	X	14640	15220	77	X	15360	15600	77	X	15220	15360	77	X	14480	14560
78	X	15220	15360	78	X	15600	15700	78	X	15360	15480	78	X	14560	14660
79	X	15360	15600	79	X	15800	15900	79	X	15480	15600	79	X	14720	14980
80	X	15600	15780	80	X	15900	16000	80	X	15600	15800	80	X	14980	15220
81	X	15780	15900	81	X	16000	16180	81	X	15800	15900	81	X	15220	15380
82	X	15900	16000	82	X	16180	16280	82	X	15900	16000	82	X	15380	15480
83	X	16000	16180	83	X	16340	16440	83	X	16000	16160	83	X	15480	15600
84	X	16180	16260	84	X	16540	16680	84	X	16160	16340	84	X	15600	15680
85	X	16260	16340	85	X	16680	17040	85	X	16340	16460	85	X	15680	15760
86	X	16340	16440	86	X	17040	17520	86	X	16460	16540	86	X	15800	15900
87	X	16440	16560	87	X	17520	17600	87	X	16580	16680	87	X	15900	16000
88	X	16560	16680	88	X	17600	17740	88	X	16720	17040	88	X	16000	16180
89	X	16680	16840	89	X	17800	18260	89	X	17040	17180	89	X	16180	16320
90	X	16840	16980	90	X	18260	18400	90	X	17180	17340	90	X	16320	16480
91	X	16980	17060	91	X	18400	18540	91	X	17340	17460	91	X	16540	16640
92	X	17060	17420	92	X	18540	18660	92	X	17520	17600	92	X	16680	16840
93	X	17420	17540	93	X	18780	19060	93	X	17600	17740	93	X	16840	17000
94	X	17600	17740	94	X	19060	19220	94	X	17800	18000	94	X	17060	17180
95	X	17800	18260	95	X	19220	19360	95	X	18000	18260	95	X	17180	17340
96	X	18260	18400	96	X	19360	19540	96	X	18340	18720	96	X	17340	17420
97	X	18400	18540	97	X	19640	19760	97	X	18780	19080	97	X	17420	17540
98	X	18540	18720	98	X	19840	20060	98	X	19080	19160	98	X	17600	17760
99	X	18760	19060	99	X	20180	20380	99	X	19200	19360	99	X	17800	18020
100	X	19060	19160	100	X	20380	20920	100	X	19360	19560	100	X	18020	18280
101	X	19200	19360	101	X	20920	21200	101	X	19560	19640	101	X	18280	18400
102	X	19360	19520	102	X	21240	21400	102	X	19640	19740	102	X	18400	18540
103	X	19640	19740	103	X	21440	21520	103	X	19840	20060	103	X	18540	18680
104	X	19840	20060	104	X	21860	22400	104	X	20180	20260	104	X	18680	18760

105 X	20180	20260		105 X	20380	20820	105 X	18760	19080
106 X	20260	20380		106 X	20940	21200	106 X	19080	19160
107 X	20380	20920		107 X	21260	21400	107 X	19220	19380
108 X	20920	21180		108 X	21440	21540	108 X	19380	19500
109 X	21240	21400		109 X	21860	22400	109 X	19640	19800
110 X	21840	21960					110 X	19840	20060
111 X	21960	22400					111 X	20100	20280
							112 X	20400	20500
							113 X	20500	20640
							114 X	20640	20720
							115 X	20720	20900
							116 X	20940	21120
							117 X	21120	21200
							118 X	21240	21400
							119 X	21460	21540
							120 X	21860	22400

Supplemental Table S6.

Figure 2B

Kc167		BG3		S2	
Color	-log10(p-value)	Color	-log10(p-value)	Color	-log10(p-value)
RED	99,9	1	286,9	1	261,8
YELLOW	258,2	2	170,6	2	136,0
GREEN	22,9	3	94,8	3	80,8
BLUE	1,4	4	0,6	4	0,2
BLACK	169,8	5	20,0	5	15,4
		6	6,8	6	11,3
		7	0,2	7	0,1
		8	45,7	8	7,9
		9	20,2	9	113,6

Figure 2C

Chromatin mark	-log10(p-value)			-log10(bonf. corrected p-value)		
	S2	BG3	Kc167	S2	BG3	Kc167
H3K27ac	216,4	190,0	122,3	215,1	188,6	120,9
H3K27me3	185,3	35,5	NA	183,9	34,1	NA
H3K36me3	227,7	257,3	246,2	226,3	255,9	244,8
H3K4me1	178,5	133,0	228,8	177,1	131,6	227,4
H3K4me3	271,4	<300	280,8	270,0	<300	279,4
H4K16ac	195,9	149,4	247,8	194,5	148,0	246,4
polyA	164,6	NA	172,9	163,2	NA	171,6
totalRNA	222,3	215,1	216,7	221,0	213,7	215,3
CTCF	15,0	41,1	5,1	13,6	39,7	3,7
H1	93,2	190,5	217,4	91,8	189,1	216,0
ISWI	286,2	259,3	286,9	284,8	258,0	285,6
RNA-pol-II	282,3	267,1	290,3	280,9	265,8	288,9
Smc3	36,2	NA	37,6	34,8	NA	36,2
Su_Hw	41,3	44,1	22,2	39,9	42,7	20,9
Beaf-32	235,1	76,8	274,7	233,7	75,4	273,3
mod_mdg4	4,5	15,8	3,6	3,2	14,4	2,2
Chriz	304,8	<300	294,8	303,5	<300	293,4
Jil1	243,7	248,1	257,9	242,3	246,8	256,5
H3	103,2	152,6	191,4	101,8	151,2	190,0
WDS	281,9	262,1	264,4	280,5	260,7	263,0
MOF	223,8	184,1	224,5	222,5	182,7	223,1
NURF301	303,9	130,1	78,8	302,5	128,7	77,4
dmTopo-II	72,1	11,2	NA	70,7	9,8	NA
MRG15	287,8	263,6	297,4	286,4	262,2	296,0

Figure 3C

Kc167		BG3		S2	
Color	$-\log_{10}(\text{p-value})$	Color	$-\log_{10}(\text{p-value})$	Color	$-\log_{10}(\text{p-value})$
RED	123,9	1	274,3	1	238,5
YELLOW	290,9	2	181,4	2	142,8
GREEN	20,6	3	103,2	3	98,3
BLUE	0,2	4	3,5	4	3,7
BLACK	201,1	5	26,0	5	11,7
		6	4,5	6	13,0
		7	1,5	7	1,0
		8	69,2	8	11,3
		9	15,3	9	119,2

Figure 5B

Color	$-\log_{10}(\text{p-value})$
CYAN	<300
BLUE	247,0
GREEN	4,8
MAGENTA	258,9

Figure 5C

Color	$-\log_{10}(\text{p-value})$
CYAN	<300
BLUE	253,7
GREEN	24,3
MAGENTA	283,5

Supplemental Table S7.

Replicate	Total reads	Filtered out*	Uniq. mapped
BG3_rep1	16 429 383	9 702 016	5 252 957
BG3_rep2	16 623 342	9 806 995	5 296 560
KC_rep1	26 843 301	1 218 207	1 741 683
KC_rep2	17 205 118	803 813	1 159 250
OSC_rep1	17 316 612	3 820 182	4 505 649
OSC_rep2	18 719 578	4 231 838	5 028 219
S2_rep1	33 990 827	6 437 675	20 297 591
S2_rep2	20 677 615	3 301 936	13 045 928

* by TopHat2

Supplemental Table S8.

Cell line 1 vs Cell line 2

BG3 vs Kc167

Group	Cell line 1 Cell line 2	Transcription within the bin			
		No data	Cell line 1 < Cell line 2	Cell line 1 = Cell line 2	Cell line 1 > Cell line 2
T-T	TAD TAD	2841	25	349	65
I-I	inter-TAD inter-TAD	531	13	973	47
I-T	inter-TAD TAD	339	8	258	26
T-I	TAD inter-TAD	275	13	182	5
Sum		3986	59	1762	143

Chi-square test, p-value = 0,0006

Two-tailed Fisher's exact test, p-value = 0,001

Kc167 vs S2

Group	Cell line 1 Cell line 2	Transcription within the bin			
		No data	Cell line 1 < Cell line 2	Cell line 1 = Cell line 2	Cell line 1 > Cell line 2
T-T	TAD TAD	2942	34	446	42
I-I	inter-TAD inter-TAD	515	20	950	26
I-T	inter-TAD TAD	291	9	218	10
T-I	TAD inter-TAD	238	17	189	3
Sum		3986	80	1803	81

Chi-square test, p-value = 0,012

Two-tailed Fisher's exact test, p-value = 0,0187

BG3 vs S2

Group	Cell line 1 Cell line 2	Transcription within the bin			
		No data	Cell line 1 < Cell line 2	Cell line 1 = Cell line 2	Cell line 1 > Cell line 2
T-T	TAD TAD	2880	33	350	75
I-I	inter-TAD inter-TAD	517	30	943	51
I-T	inter-TAD TAD	353	7	259	35
T-I	TAD inter-TAD	236	11	157	13
Sum		3986	81	1709	174

Chi-square test, p-value = 0,01

Two-tailed Fisher's exact test, p-value = 0,0199

Kc167 vs OSC

Group	Cell line 1 Cell line 2	Transcription within the bin			
		No data	Cell line 1 < Cell line 2	Cell line 1 = Cell line 2	Cell line 1 > Cell line 2
T-T	TAD TAD	2850	49	366	32
I-I	inter-TAD inter-TAD	506	27	976	32
I-T	inter-TAD TAD	300	3	180	15
T-I	TAD inter-TAD	330	38	237	9
Sum		3986	117	1759	88

Chi-square test, p-value = 1,6e-06

Two-tailed Fisher's exact test, p-value = 0,0001

OSC vs S2

Group	Cell line 1 Cell line 2	Transcription within the bin			
		No data	Cell line 1 < Cell line 2	Cell line 1 = Cell line 2	Cell line 1 > Cell line 2
T-T	TAD TAD	2893	39	345	64
I-I	inter-TAD inter-TAD	496	32	940	36
I-T	inter-TAD TAD	340	12	265	34
T-I	TAD inter-TAD	257	17	176	4
Sum		3986	100	1726	138

Chi-square test, p-value = 2,6e-05

Two-tailed Fisher's exact test, p-value = 0,0001

BG3 vs OSC

Group	Cell line 1 Cell line 2	Transcription within the bin			
		No data	Cell line 1 < Cell line 2	Cell line 1 = Cell line 2	Cell line 1 > Cell line 2
T-T	TAD TAD	2849	35	328	45
I-I	inter-TAD inter-TAD	569	14	1027	47
I-T	inter-TAD TAD	301	4	211	22
T-I	TAD inter-TAD	267	16	202	13
Sum		3986	69	1768	127

Chi-square test, p-value = 0,00219

Two-tailed Fisher's exact test, p-value = 0,0044

Supplemental Table S9.

Cell line 1 vs Cell line 2

BG3 vs Kc167

Group Cell line 1 Cell line 2		Proportion of active chromatin within the bin			
		No data	Cell line 1 < Cell line 2	Cell line 1 = Cell line 2	Cell line 1 > Cell line 2
T-T	TAD TAD	1824	230	699	527
I-I	inter-TAD inter-TAD	155	90	1199	120
I-T	inter-TAD TAD	94	51	272	58
T-I	TAD inter-TAD	125	35	384	87
Sum		2198	406	2554	792

Chi-square test, p-value = 0,004

Two-tailed Fisher's exact test, p-value = 0,006

Kc167 vs S2

Group Cell line 1 Cell line 2		Proportion of active chromatin within the bin			
		No data	Cell line 1 < Cell line 2	Cell line 1 = Cell line 2	Cell line 1 > Cell line 2
T-T	TAD TAD	2005	404	845	210
I-I	inter-TAD inter-TAD	159	102	1173	77
I-T	inter-TAD TAD	88	54	316	48
T-I	TAD inter-TAD	110	57	269	33
Sum		2362	617	2603	368

Chi-square test, p-value = 0,145

Two-tailed Fisher's exact test, p-value = 0,187

BG3 vs S2

Group Cell line 1 Cell line 2		Proportion of active chromatin within the bin			
		No data	Cell line 1 < Cell line 2	Cell line 1 = Cell line 2	Cell line 1 > Cell line 2
T-T	TAD TAD	1801	236	948	353
I-I	inter-TAD inter-TAD	143	44	1297	57
I-T	inter-TAD TAD	133	24	441	56
T-I	TAD inter-TAD	83	37	265	32
Sum		2160	341	2951	498

Chi-square test, p-value = 0,003

Two-tailed Fisher's exact test, p-value = 0,004

Supplemental Table S10.

Group	1
	L3_Carcass L3_CNS L3_DigestiveSystem L3_FatBody L3_ImaginalDiscs L3_SalivaryGlands
Group	2
	WPP_2days_CNS WPP_2days_Fat WPP_FatBody WPP_SalivaryGlands
Group	3
	AdMatedM_Ecl_4days_Testes AdMatedM_Ecl_4days_AccessoryGlands AdMatedM_Ecl_4days_Heads
Group	4
	AdVirginF_Ecl_4days_Ovaries AdVirginF_Ecl_4days_Heads
Group	5
	AdMatedF_Ecl_4days_Ovaries AdMatedF_Ecl_4days_Heads
Group	6
	AdMixedMF_Ecl_1day_Carcass AdMixedMF_Ecl_1day_DigestiveSystem
Group	7
	AdMixedMF_Ecl_4days_Carcass AdMixedMF_Ecl_4day_DigestiveSystem
Group	8
	AdMixedMF_Ecl_20days_Carcass AdMixedMF_Ecl_20days_DigestiveSystem

Supplemental Table S11.

Cell line	Reads	Reads w. trimmed adaptor	Delly					BreakDancer					
			DEL	DUP	INV	TRA	SUM	DEL	INS	INV	CTX	UNK	SUM
S2	65 681 282	40 228 560	645	405	170	251	1471	1465	1183	938	17	1205	4808
Kc167	31 127 671	17 598 455	648	261	160	178	1247	340	504	205	53	459	1561
BG3	50 183 677	31 255 562	614	38	69	176	897	712	627	178	43	251	1811
OSC	32 372 349	19 698 432	623	28	104	168	923	573	528	196	66	179	1542