

Nucleation and spreading rejuvenate polycomb domains every cell cycle

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Supplemental Materials

Figure S1

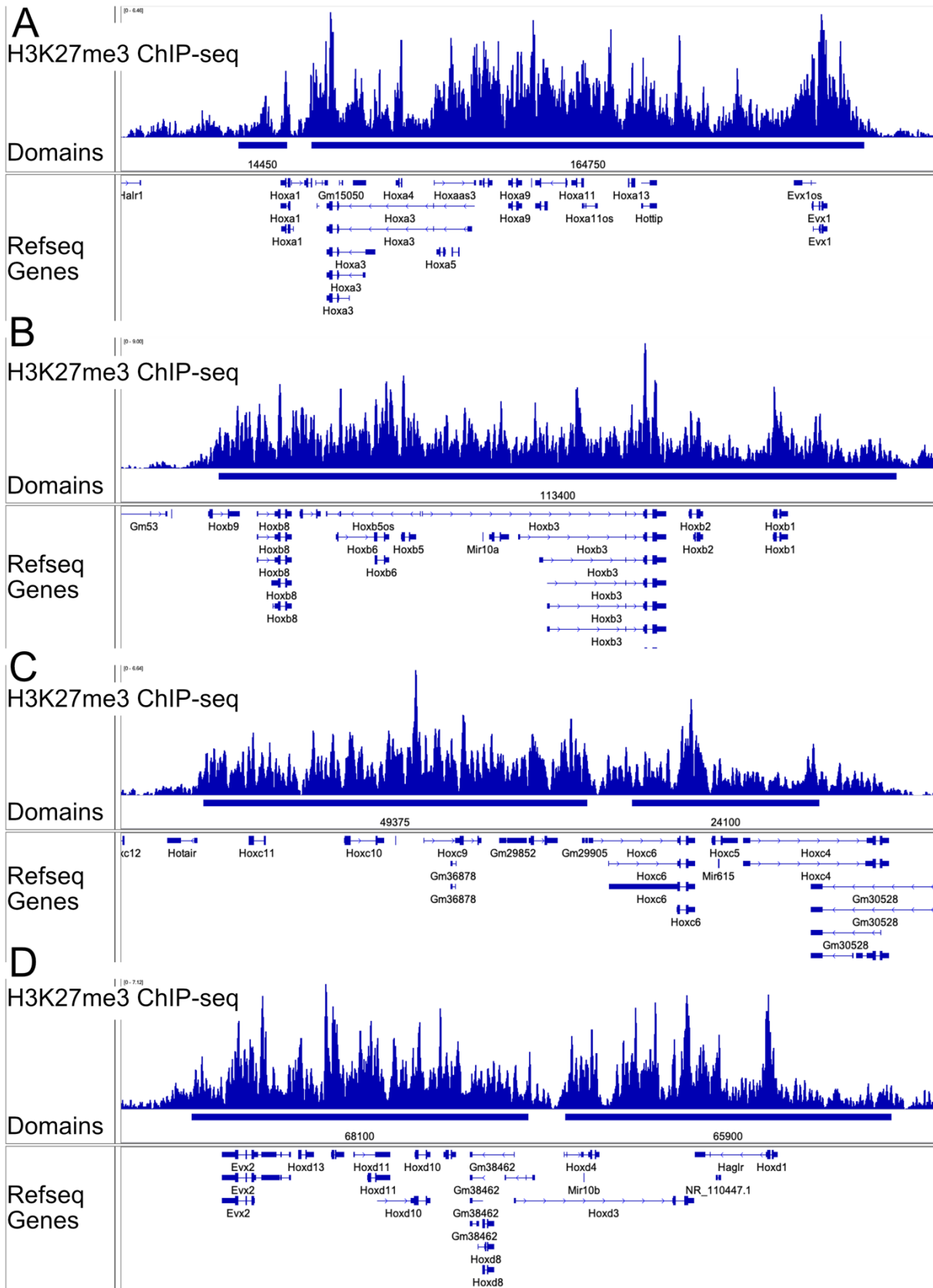


Figure S1. Large domains defined at Hox loci. **A)** Our algorithm identifies 164 kb domain at the Hoxa locus (chr6:52,120,00-52,340,000). **B)** A 113 kb domain is defined at the Hoxb locus (chr11:96,260,000-96,390). **C)** Two domains of 49 kb and 24 kb respectively are defined at the HoxC locus (chr15:102,940,000-103,040,000). **D)** Two domains of 68 kb and 66 kb respectively are defined at the HoxD locus (chr2:74,640,000-74,790,000).

Table S1. Critical resources used in this study

Resource	Reference
Cell lines	
E14 mouse embryonic stem cells	Gift from Dr. Peter J. Koch
Antibodies	
Anti-trimethyl-Histone H3 (Lys27) C36B11 rabbit monoclonal (1 mg/ml)	Cell Signaling Technology; Cat # 9733; RRID: AB_2616029
Normal IgG rabbit polyclonal (1 mg/ml)	R&D Biosciences; Cat # AB-105-C; RRID: AB_354266
Anti-rabbit goat mixed monoclonal (1 mg/ml)	Epiccypher; Cat # 13-0047
Chemicals	
2% 4elatina solution	Millipore-Sigma; Cat # G1393
KnockOut DMEM	Gibco; Cat # 10829018
FBS, ES-qualified	Gibco; Cat # 16141079
Glutamax (100X)	Gibco; Cat # 35050061
Penicillin-streptomycin (10,000 U/ml)	Gibco; Cat # 15140122
MEM non-essential amino acids (100X)	Gibco; Cat # 11140050
2-mercaptoethanol (1000X)	Gibco; Cat # 21985023
ESGRO-LIF	Millipore-Sigma; Cat # ESG1107
Trypsin-EDTA (0.25%), phenol red	Gibco; Cat# 25200056
Digitonin	Millipore-Sigma; Cat # 300410
Spermidine	Millipore-Sigma; Cat # S2626
Roche Complete Protease Inhibitor mini EDTA-Free tablets	Millipore-Sigma; Cat # 04693159001
TAPS buffer 0.2 M	Boston Scientific; Cat # BB-2372
CUTANA pAG-Tn5	Epiccypher; Cat # 15-1017
AMPure XP SPRI beads	Beckman Coulter; Cat # A63881
Propidium Iodide	Millipore-Sigma; Cat # P4170
RNase A, DNase and protease-free (10 mg/mL)	ThermoFisher; Cat # EN0531
Commercial Assays	
NEBNext High-Fidelity 2× PCR Master Mix	NEB; Cat # M0541
Qubit dsDNA HS Assay Kit	ThermoFisher; Cat # Q32854
Tapestation High Sensitivity D1000 ScreenTapes	Agilent; Cat # 5067-5584
Tapestation High Sensitivity D1000 Sample Buffer & Ladder	Agilent; Cat # 5067-5585
Oligonucleotides	
Universal i5 primer for CUT&tag: 5' AATGATACGGCGACCACCGAGATCTACACTAGATCGCTCGTCGG CAGCGTCAGATGTGTAT 3'	Epiccypher
Barcoded i7 primers for CUT&Tag: 5' CAAGCAGAAGACGGCATAACGAGATNNNNNNNGTCTCGTGGGCT CGGAGATGTG 3'	Epiccypher
Equipments	
Countess 3 Automated Cell Counter	ThermoFisher; Cat # AMQAX2000
Magnetic Separation Rack, 0.2 mL Tubes	Epiccypher; Cat # 10-0008
4200 TapeStation System	Agilent; Cat # G2991BA
Qubit 4 Fluorometer	ThermoFisher; Cat # Q33238
MoFlo Astrios EQ Cell Sorter	Beckman Coulter; Cat # B52102

Table S2. External datasets used in this study

S. No.	Dataset	Figures	GEO Accession ID	Reference
1	H3K27me3 ChIPseq	Figure 1	GSM1033638	(Banaszynski et al., 2013)
2	H3K27me3 ChIPseq	Figure 1	GSM487549	(Banaszynski et al., 2013)
3	H3K27me3 ChIPseq	Figure 2, 3	GSM2779214	(Hojfeldt et al., 2018)
4	H3K27me3 ChIPseq, 7-day treatment with EPZ6438	Figure 2, 3	GSM2779215	(Hojfeldt et al., 2018)
5	H3K27me3 ChIPseq 4 hours after washout of EPZ6438	Figure 2, 3	GSM2779216	(Hojfeldt et al., 2018)
6	H3K27me3 ChIPseq 8 hours after washout of EPZ6438	Figure 2, 3	GSM2779217	(Hojfeldt et al., 2018)
7	H3K27me3 ChIPseq 16 hours after washout of EPZ6438	Figure 2, 3	GSM2779218	(Hojfeldt et al., 2018)
8	H3K27me3 ChIPseq 24 hours after washout of EPZ6438	Figure 2, 3	GSM2779219	(Hojfeldt et al., 2018)
9	H3K27me3 ChIPseq 48 hours after washout of EPZ6438	Figure 2, 3	GSM2779220	(Hojfeldt et al., 2018)
10	H3K27me3 ChIPseq 96 hours after washout of EPZ6438	Figure 2, 3	GSM2779221	(Hojfeldt et al., 2018)
11	Jarid2 ChIP-seq	Figure 3	GSM3021208	(Perino et al., 2018)
12	Jarid2 ChIP-seq	Figure 3	GSM3021209	(Perino et al., 2018)
13	PCL2 ChIP-seq	Figure 3	GSM2472747	(Perino et al., 2018)
14	PCL2 ChIP-seq	Figure 3	GSM2472748	(Perino et al., 2018)
15	Ezh2 ChIP-seq	Figure 3	GSM2472741	(Perino et al., 2018)
16	Ezh2 ChIP-seq	Figure 3	GSM2472742	(Perino et al., 2018)
17	Suz12 ChIP-seq	Figure 3	GSM2779242	(Hojfeldt et al., 2018)
18	H2AK119ubiquitin ChIP-seq	Figure 3	GSE119618	(Fursova et al., 2019)
19	ATAC-seq	Figure 3	GSM1941479	(de Dieuleveult et al., 2016)
20	ATAC-seq	Figure 3	GSM1941480	(de Dieuleveult et al., 2016)
21	ATAC-seq	Figure 3	GSM1941487	(de Dieuleveult et al., 2016)
22	ATAC-seq	Figure 3	GSM1941488	(de Dieuleveult et al., 2016)
23	H3.3 ChIP-seq	Figure 3	GSM423355	(Goldberg et al., 2010)
24	H3.3 ChIP-seq	Figure 3	GSM1207786	(Banaszynski et al., 2013)
25	H3.3 ChIP-seq	Figure 3	GSM487544	(Goldberg et al., 2010)
26	H3.3 ChIP-seq	Figure 3	GSM487551	(Goldberg et al., 2010)

27	H3K27me3 ChIP-seq (Suz12 Δ N-term)	Figure 4	GSM2779202	(Hojfeldt et al., 2018)
28	H3K27me3 ChIP-seq (RING1A/B KO)	Figure 4	GSE119618	(Fursova et al., 2019)
29	H3K27me3 ChIP-seq (PCGF1/3/5/6 KO)	Figure 4	GSE119618	(Fursova et al., 2019)
30	H3K27me3 ChIP-seq (PCGF2/4 KO)	Figure 4	GSE119618	(Fursova et al., 2019)
31	H3K27me3 ChIP-seq (EED Y365A)	Figure 4	GSM2475241	(Oksuz et al., 2018)
32	H3K27me3 ChIP-seq (EED Y365A)	Figure 4	GSM2475248	(Oksuz et al., 2018)
33	H3K27me3 ChIP-seq (EED F97A)	Figure 4	GSM2475240	(Oksuz et al., 2018)
34	H3K27me3 ChIP-seq (EED F97A)	Figure 4	GSM2475247	(Oksuz et al., 2018)
35	H3K27me3 ChIP-seq (Aebp2 KO)	Figure 4	GSM3639119	(Hojfeldt et al., 2019)
36	H3K27me3 ChIP-seq (Jarid2 KO)	Figure 4	GSM3639121	(Hojfeldt et al., 2019)
37	H3K27me3 ChIP-seq (Epop2 KO)	Figure 4	GSM3639120	(Hojfeldt et al., 2019)
38	H3K27me3 ChIP-seq (PCL2 KO)	Figure 4	GSM3639122	(Hojfeldt et al., 2019)
39	H3K27me3 ChIP-seq (P123 KO)	Figure 4	GSM3639149	(Hojfeldt et al., 2019)
40	H3K27me3 ChIP-seq (P123 KO)	Figure 4	GSM3639148	(Hojfeldt et al., 2019)
41	H3K27me3 ChIP-seq (JA KO)	Figure 4	GSM3639151	(Hojfeldt et al., 2019)
42	H3K27me3 ChIP-seq (JA KO)	Figure 4	GSM3639152	(Hojfeldt et al., 2019)
43	H3K27me3 ChIP-seq (PE KO)	Figure 4	GSM3639149	(Hojfeldt et al., 2019)
44	H3K27me3 ChIP-seq (PE KO)	Figure 4	GSM3639150	(Hojfeldt et al., 2019)
45	H3K27me3 ChIP-seq (JAE KO)	Figure 4	GSM3639169	(Hojfeldt et al., 2019)
46	H3K27me3 ChIP-seq (PJ KO)	Figure 4	GSM3639153	(Hojfeldt et al., 2019)
47	H3K27me3 ChIP-seq (PJ KO)	Figure 4	GSM3639154	(Hojfeldt et al., 2019)
48	H3K27me3 ChIP-seq (PJA KO)	Figure 4	GSM3639155	(Hojfeldt et al., 2019)
49	H3K27me3 ChIP-seq (PJA KO)	Figure 4	GSM3639155	(Hojfeldt et al., 2019)
50	H3K27me3 ChIP-seq (PEA KO)	Figure 4	GSM3639171	(Hojfeldt et al., 2019)
51	H3K27me3 ChIP-seq (PEJA KO)	Figure 4	GSM3639157	(Hojfeldt et al., 2019)
52	H3K27me3 ChIP-seq (PEJA KO)	Figure 4	GSM3639158	(Hojfeldt et al., 2019)
53	mESC Replication Timing	Figure 5, 6, 7	GSE137764	(Zhao et al., 2020)
54	H3K27me3 CUT&Flow	Figure 5, 6, 7	This study	