

**Supplemental Figure 1.** Scatter plots showing the pair wise correlation of the two replicate ChIP samples for each of the PcG proteins and Trx-C. According to these plots cut-offs for TSS window enrichments were defined (see methods section). Cut-off values, number of Refseq TSS above these cut-offs and percentage of all non-overlapping TSSs (n=8977) are indicated.

**Supplemental Figure 2.** Genome browser view of ChIP-seq and RNA-seq tag densities for three different genomic regions. (A) Promoters of two inactive transcription units (*Scr*) are occupied by PRC1 proteins and Trx-C. (B) Promoters of active genes (*Psc* and *Su(z)2*) are occupied by PRC1 proteins and Trx-C. (C) Promoter of active gene (CG6643) is bound by Trx-C, while PRC1 proteins are absent.

**Supplemental Figure 3.** Overview of 5'-MACE (Massive amplification of cDNA ends) data. (A) Enrichment of 5'-MACE and RNA-seq read alignments (number given in parenthesis) in genomic region types relative to their sizes. (B) Pairwise scatter plots between the log2 number of 5'-MACE read alignments at RefSeq TSS windows and transcript expression levels (below the diagonal), Pearson correlation coefficients (above the diagonal) and histogram of read numbers (in the diagonal). (C) Schematic representation of the technical workflow.

**Supplemental Figure 4.** Comparison of PcG target genes found in this study (red circles) with other *Drosophila* genome-wide datasets (blue circles) (Oktaba et al. 2008; Schuettengruber et al. 2009; Schwartz et al. 2010). The gene lists used for these Venn diagrams can be found in Supplemental Table 4. The gene list of this study comprises 1067 genes where at least three of the four proteins Pc, Ph, Psc and Trx-C co-localize at the promoter. Our target gene set covers 48%, 47% and 33% of the genes identified in Oktaba et al. (“core-PhoRC bound genes”), Schuettengruber et al. (“PcG gene targets”) and Schwartz et al. (“Class I high-confidence PcG target genes”), respectively. Now, a sum of 1371 target genes has been identified in *Drosophila*, with 55 genes common in all 4 datasets.

**Supplemental Table 1.** Summary statistics of DNA library sequence reads.

See methods for details on filtering and alignment of sequence reads. “Single hit”, single hits in genome and annotation database; “Single hit genome”, single hits in genomic sequence.

**Supplemental Table 2.** Enrichments of PcG proteins at Refseq TSS.

**Supplemental Table 3.** PcG binding sites in *Drosophila* S2 cells. Table containing chromosomal coordinates of all PcG binding sites used in this study and the distance to the nearest Refseq TSS and TSS cluster (as determined by 5'-MACE).

**Supplemental Table 4.** Combined list of PcG target genes identified in this and other genome-wide studies (Oktaba et al., 2008; Schuettengruber et al., 2009; Schwartz et al., 2010).

**Supplemental Table 5.** *Drosophila* miRNAs targeted by PcG proteins. Listing of all 41 miRNA loci considered as PcG targets in this study with corresponding genomic coordinates. We include information on the mature miRNA locus and if the corresponding PcG site was found in S2 cells or embryos (see Methods). In addition we note, if the PcG site contains a signal from 5'-MACE oriented towards the mature miRNA and if the attempt to clone a primary transcript for this promoter was successful.

**Supplemental Text 1.** Non-coding RNAs at PREs in the Bithorax Complex. Primer sequences and cDNA clones from 5'- and 3'-RACE experiments detecting transcripts over the Fab-7 PRE in *Drosophila* SF4 cells.

**Supplemental Text 2.** Intergenic pri-miRNA transcripts at PcG binding sites. Sequences of pri-miRNA cDNA clones and PCR primers used for analysis.

**Supplemental File 1.** UCSC genome browser track of PcG binding sites in S2 cells. Chromosomal coordinates of all PcG binding sites used in this study in the BED format compatible with the UCSC Genome Browser.

**Supplemental File 2.** UCSC genome browser track of pooled TSS clusters. Chromosomal coordinates of all TSS clusters recognized by 5'-MACE in a pooled dataset from S2 cells and 0-16h embryos. This file uses the BED format compatible with the UCSC Genome Browser.

**Supplemental File 3.** UCSC genome browser track of ncRNAs. BLAT alignment file of all cDNAs described in this study in the PSL format compatible with the UCSC Genome Browser.