



Figure S3. H3K27me3 gain upon DNAm depletion is focused on CpG-rich regions. A) Comparison of CpG density in regions gain/loss H3K27me3 in *Nsd1*-KO and *Dnmt*-TKO, indicating the H3K27me3 gain in TKO occurs especially in CpG-rich regions. P-values are based on t-test. Here we use H3K27me3 replicates (Replicate 2), showing similar trend as in Figure 4B. B) H3K27me3 redistribution in the absence of DNAm, using CHIP-seq replicates of H3K27me3 (Replicate 2), showing similar trend as in Figure 4C; note the “dip” in the centre of unmethylated CGIs appears in all four samples, is due to technical artifact, most likely a GC bias in the library preparation for this batch of samples. Data used in (A) and (B) are from this study. C) H3K27me3 redistribution in 2i medium versus serum centred on highly methylated CGIs (n = 642) and un-methylated CGIs (n = 3152) in serum respectively. As known, 2i leads to DNA hypomethylation compared to serum culture. Similar to *Dnmt*-TKO, H3K27me3 upon DNAm loss in 2i is enriched in CGIs with high DNAm in serum. However, the artifact “dip” in the plots around unmethylated CGIs makes it hard to estimate the depletion of H3K27me3 from those CGIs in 2i medium. The CHIP-seq data of H3K27me3 and WGBS data used in (C) are from Marks et al. 2012 and Habibi et al. 2013. D) Aggregate plots (normalized by read-depth and MS ratios) of H3K27me3 (Replicate 1 and 2 from this study) centred on CGIs with different DNAm levels in WT. Note that the plots on CGIs in the bottom and top ranges (i.e. [0, 0.01] and [0.8, 1]) are already shown in Figure 4C and Figure S3B. Similar trends are shown between the plots in [0.01, 0.2] and [0, 0.01], although the change in the former is far less dramatic. Same is true for the plot in [0.4, 0.8] compared to the plot in [0.8, 1]. The decreased peaks in *Nsd1*-KO for all groups of CGIs are due to the uniform spread of H3K27me3 across the genome.