Cell Reports, Volume *32*

Supplemental Information

The Human Integrator Complex Facilitates Transcriptional Elongation by Endonucleolytic Cleavage of Nascent Transcripts

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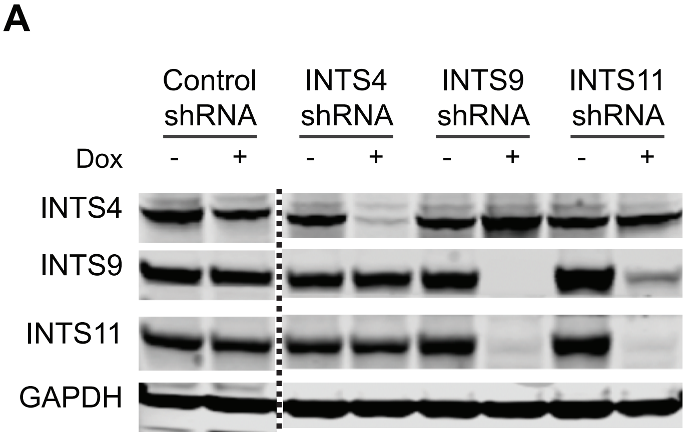

Figure S1, Related to figure 1. (A) Immunoblot of INTS11, INTS9, INTS4, and GAPDH without and with shRNA induction.

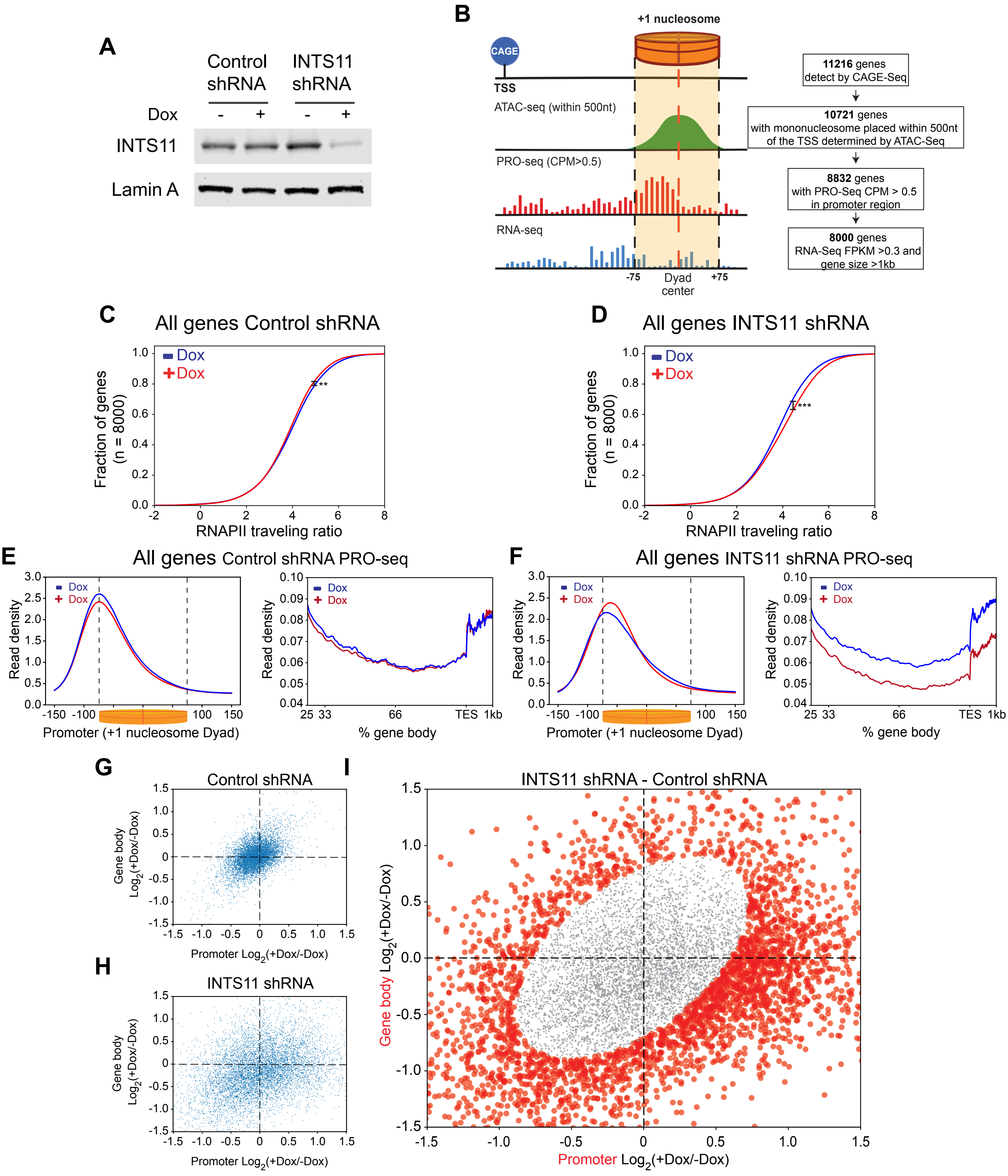

Figure S2, Related to figure 2.(A) Immunoblot of INTS11 and Lamin A without and with shRNA induction in Control shRNA cells and INTS11 shRNA cells. (B) Schematic of the criteria used to curate genes. (C and D) PRO-seq TR at all expressed genes (n = 8000) in (C) Control shRNA cells (D) and INTS11 shRNA cells without and with shRNA induction. KS test ** = p-value < 0.01 and *** = p-value < 0.001. (E and F) PRO-seq promoter proximal read density and gene body profile at all expressed genes (n = 8000) in (E) Control shRNA cells (F) and INTS11 shRNA cells without and with shRNA induction. PRO-seq promoter profiles are centered on the +1 nucleosome dyad. The gene body length is normalized using the region corresponding to 25% of the total annotated gene length up to the transcription end site (TES) plus an additional 1kb. (G and H) Dotted TM of (G) Control shRNA cells (H) and INTS11 shRNA cells where each dot represents one gene. (I) Dotted differential TM of INTS11 shRNA cells.

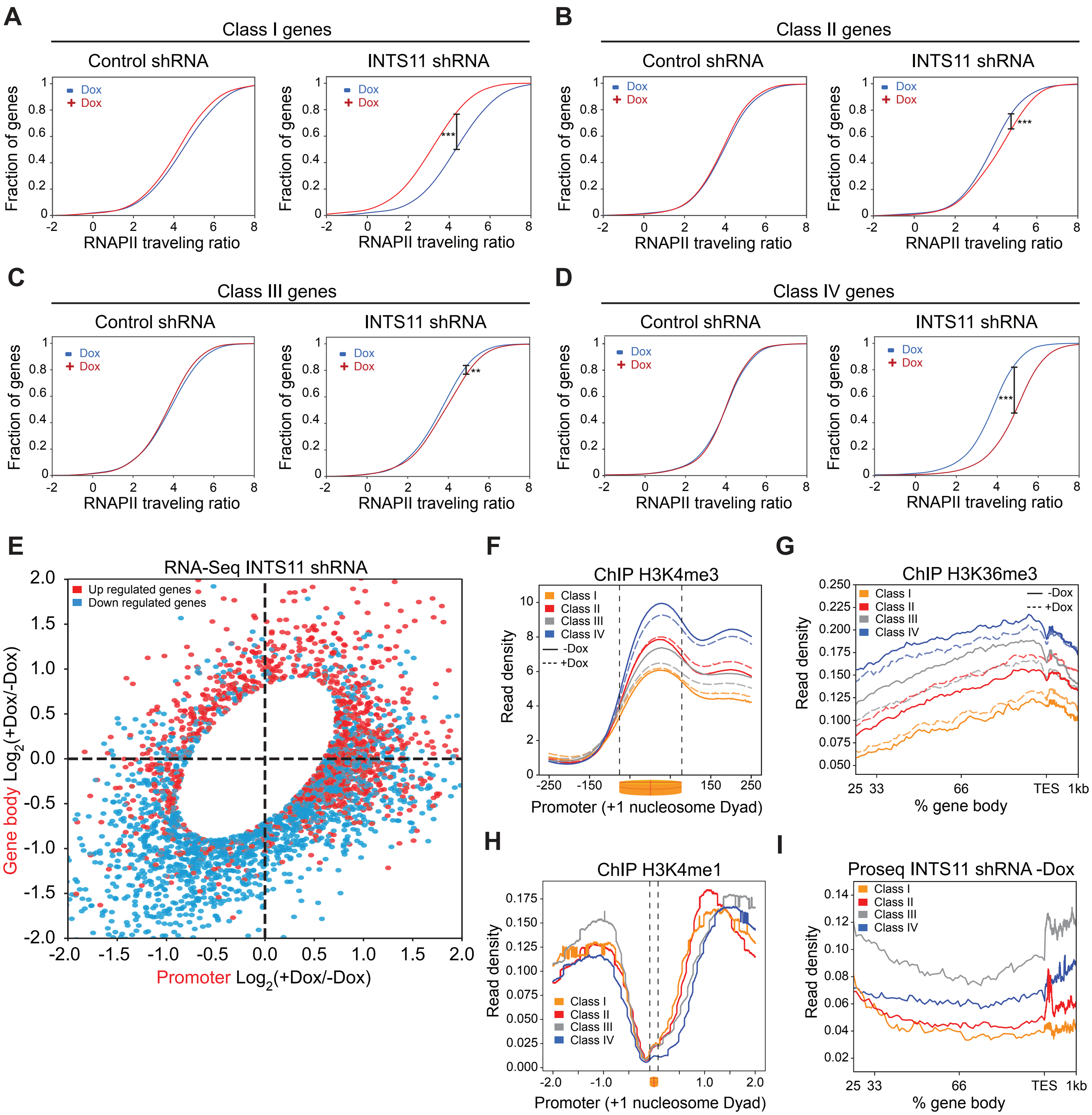

Figure S3, Related to figure 3. (A-D) PRO-Seq TR of (A) Class I genes, (B) Class II genes, (C) Class III genes, (D) and Class IV genes in Control shRNA cells and INTS11 shRNA cells without and with shRNA induction. (E) Projection of differentially expressed genes calculated by RNA-seq on the differential TM of INTS11 shRNA cells. Blue dots are downregulated genes and red dots are upregulated genes determined by RNA-seq. (F) ChIP-seq promoter profile centered on the +1 nucleosome dyad at classes defined by the TM of H3K4me3 without and with shRNA induction in shINTS11 cells. KS test p-value < 0.001 for classes paired against Class IV. (G) ChIP-seq gene body profile of H3K36me3 without and with shRNA induction in shINTS11 cells. KS test p-value < 0.001 for classes paired against Class IV. (H) ChIP-seq promoter profile centered on the +1 nucleosome dyad at classes defined by the TM of H3K4me1 in shINTS11 cells. (I) Gene body PRO-seq profile of INTS11 shRNA cells at the classes defined by the TM, KS test p-value < 0.0001 for classes paired against Class IV.

Figure S4

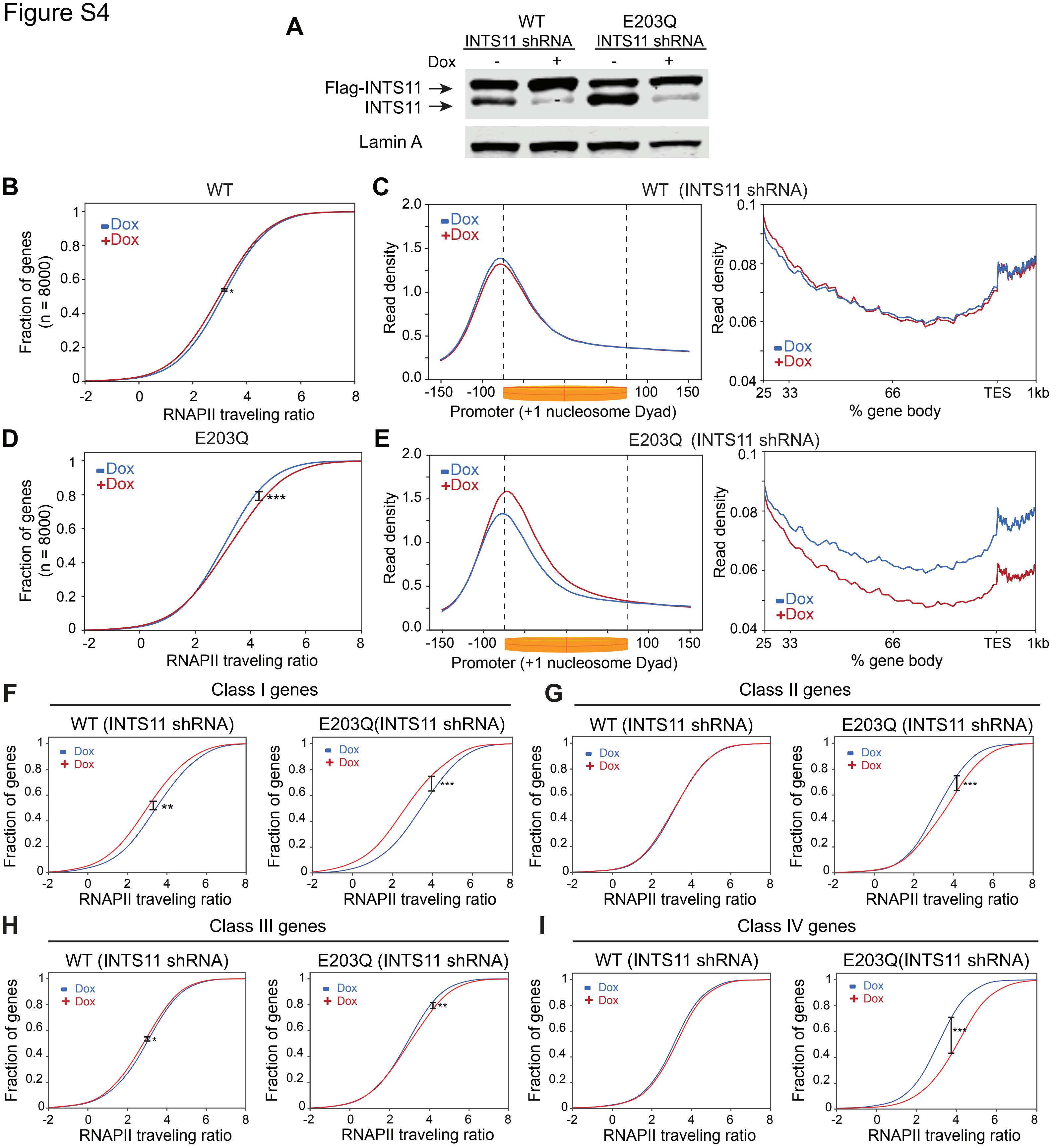

Figure S4, Related to figure 4. (A) Immunoblot of INTS11 and Lamin A without and with shRNA induction in cells expressing WT-INTS11 and E203Q-INTS11. The upper band denotes stable expression of Flag-tagged WT-INTS11 or E203Q-INTS11 from a sequence refractory to INTS11 shRNA. (B) PRO-seq TR at all expressed genes (n = 8000) in INTS11 shRNA cells expressing WT-INTS11 without and with shRNA induction. (C) PRO-seq promoter proximal read density and gene body profile at all expressed genes (n = 8000) in INTS11 shRNA cells expressing WT-INTS11 without and with shRNA induction. PRO-seq promoter profiles are centered on the +1 nucleosome dyad. (D) PRO-seq TR at all expressed genes (n = 8000) in INTS11 shRNA cells expressing E203Q-INTS11 without and with shRNA induction. (E) PRO-seq promoter proximal read density and gene body profile at all expressed genes (n = 8000) in INTS11 shRNA cells expressing E203Q-INTS11 without and with shRNA induction. PRO-seq promoter profiles are centered on the +1 nucleosome dyad. (F-I) PRO-Seq TR (F) of Class I genes, (G) Class II genes, (H) Class III genes, (I) and Class IV genes in INTS11 shRNA cells expressing WT-INTS11 and INTS11 shRNA cells expressing E203Q-INTS11 without and with shRNA induction. KS test; * = p-value < 0.05, **= p-value < 0.01 and *** = p-value < 0.001.

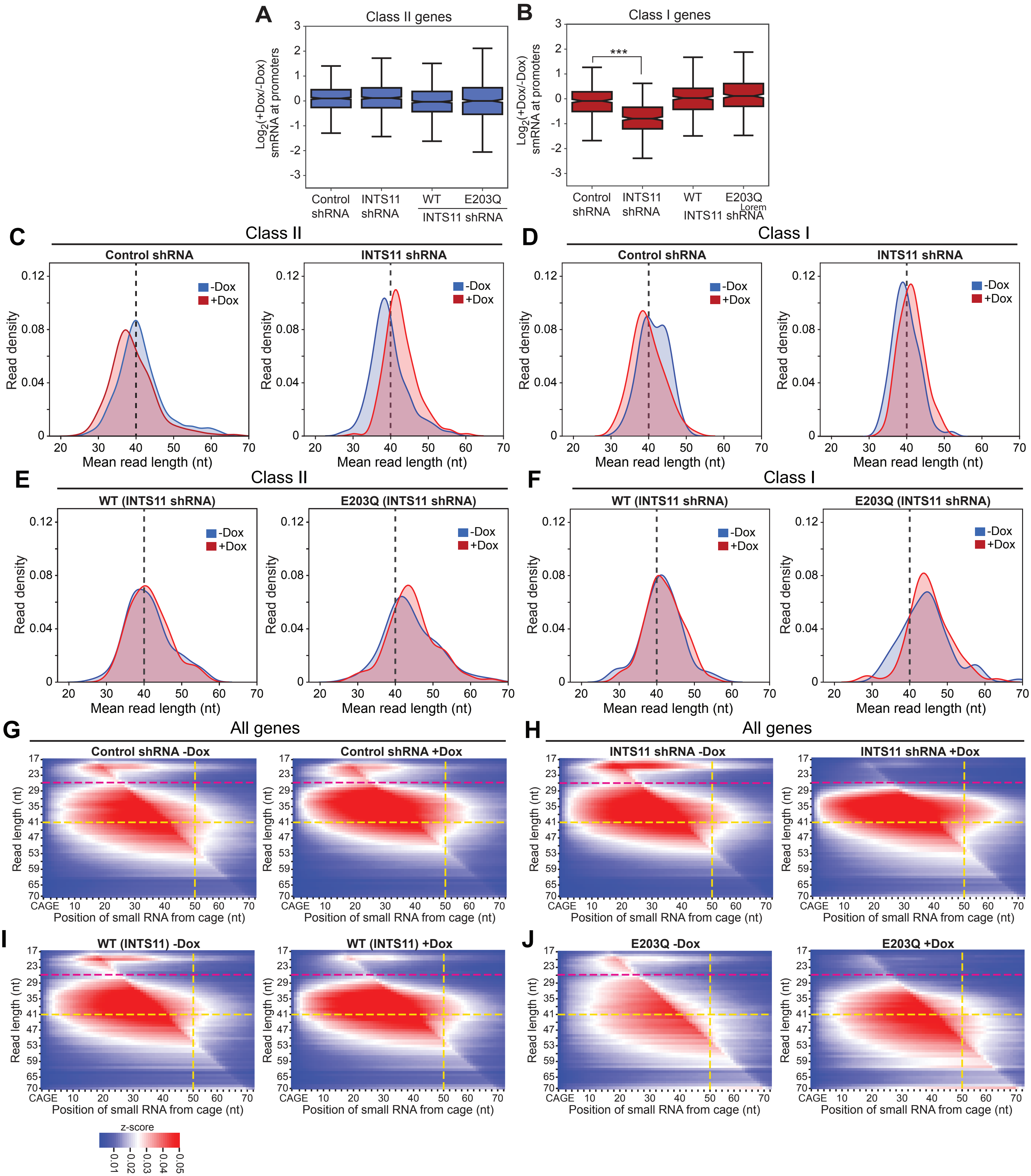

Figure S5, Related to figure 5. (A and B) Box plot of log2(+Dox/-Dox) smRNA CPM at (A) Class II (B) and Class I genes. (C and D) smRNA mean size distribution centered on the TSSs of (C) Class II (D) and Class I genes in Control shRNA cells and INTS11 shRNA cells without and with shRNA induction. (E and F) smRNA mean size distribution centered on the TSSs of (E) Class II (F) and Class I genes in INTS11 shRNA cells expressing WT-INTS11 and INTS11 shRNA cells expressing E203Q-INTS11 without and with shRNA induction. (G-J) Two-dimensional heatmap of smRNA distance from TSS vs. read length in (G) Control shRNA cells, (H) INTS11 shRNA cells, (I) INTS11 shRNA cells expressing WT-INTS11 (J) and INTS11 shRNA cells expressing E203Q-INTS11 without and with shRNA induction. The magenta dotted line partitions smRNAs into two populations of shorter and longer than 25 nucleotides. The yellow dotted lines serve as visual placeholders.

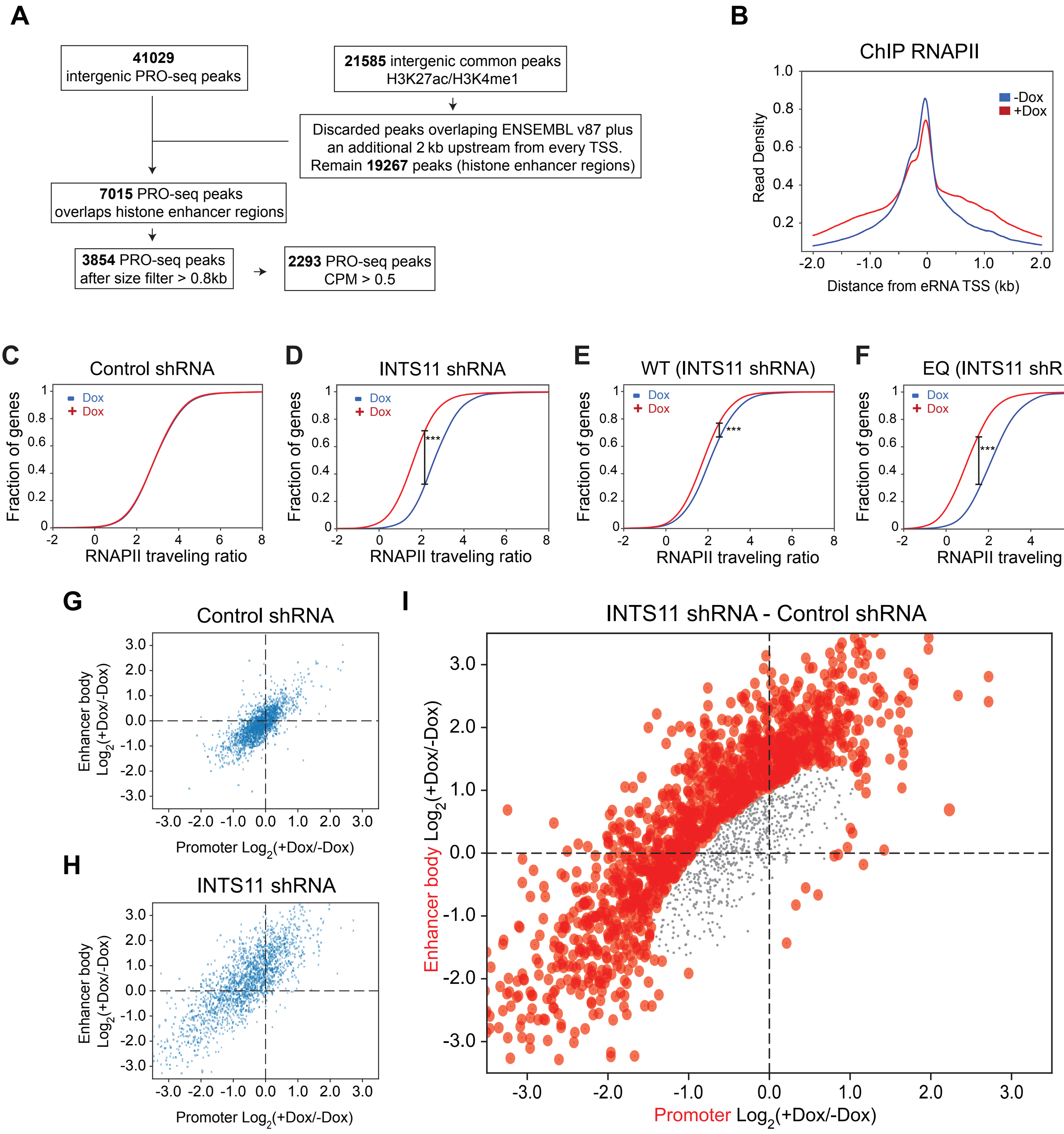

Figure S6, Related to figure 6. (A) Enhancer analysis schematic. (B) RNAPII ChIP-seq profile at enhancers (n = 2293) without and with INTS11 shRNA induction. (C-F) Enhancer TR (C) in Control shRNA cells (D) INTS11 shRNA cells (E) INTS11 shRNA cells expressing WT-INTS11 (F) and INTS11 shRNA cells expressing E203Q-INTS11 without and with shRNA induction. KS test; *** = p-value < 0.001. (G and H) Dotted enhancer TM (G) of Control shRNA cells (H) and INTS11 shRNA cells where each dot represents one enhancer locus. (I) Dotted enhancer differential TM of INTS11 shRNA cells.

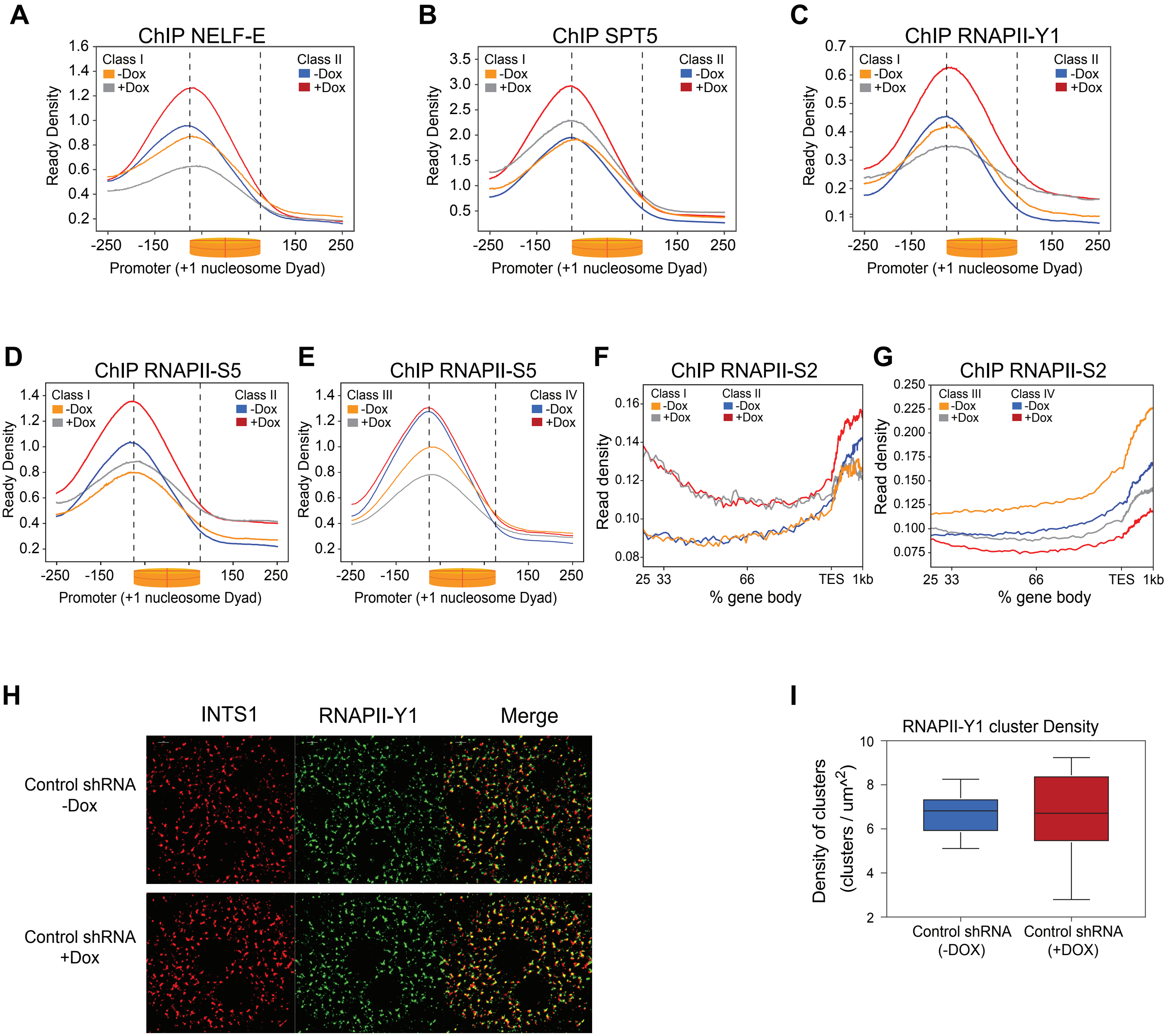

Figure S7. Related to figure 7. (A) NELF-E ChIP-seq promoter profile at Class I and Class II genes without and with shRNA induction in INTS11 shRNA cells. KS test p-value < 0.0001 for Class I and II (–Dox vs. +Dox), respectively. (B) SPT5 ChIP-seq promoter profile at Class I and Class II genes without and with shRNA induction in INTS11 shRNA cells. KS test p-value < 0.0001 for Class I and II (–Dox vs. +Dox). (C) RNAPII-Y1 ChIP-seq promoter profile at Class I and Class II genes without and with shRNA induction in INTS11 shRNA cells. KS test p-value < 0.0001 for Class I and II (–Dox vs. +Dox) (D and E) RNAPII-S5 ChIP-seq promoter profile (D) at Class I and Class II genes without and with shRNA induction in INTS11 shRNA cells. RNAPII-S5 ChIP-seq promoter profile (E) at Class III and Class IV genes without and with shRNA induction in INTS11 shRNA cells. KS test p-value < 0.0001 for Class III (–Dox vs. +Dox). (F and G) RNAPII-S2 ChIP-seq gene body profile (F) at Class I and Class II genes without and with shRNA induction in INTS11 shRNA cells. KS test p-value < 0.0001 for Class I and II (–Dox vs. +Dox). RNAPII-S2 ChIP-seq gene body profile (G) at Class IV and Class III genes without and with shRNA induction in INTS11 shRNA cells. KS test p-value < 0.0001 for Class III and IV (–Dox vs. +Dox). (H) STORM images of INTS1, RNAPII-Y1 and a merge without and with shRNA induction in Control shRNA cells. (I) Quantification of RNAPII-Y1 cluster density without and with shRNA induction in Control shRNA cells (n = 8).