

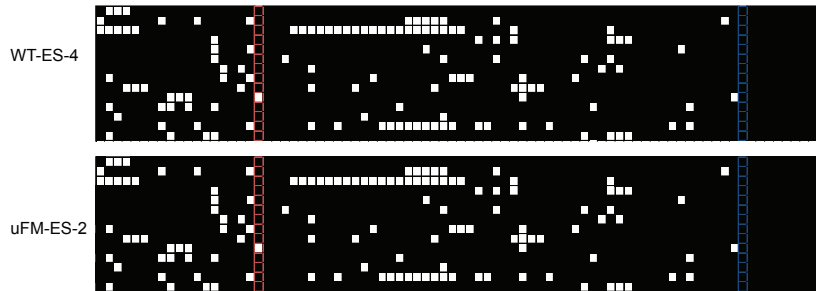
Figure S5

A

GAPDH

TSS

EX1-INTRON JUNCTION



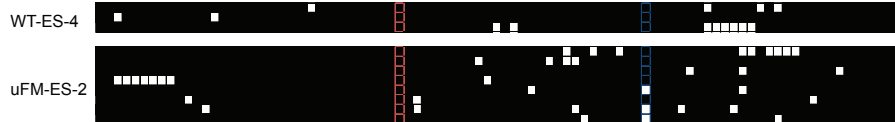
The template strand (C-rich)



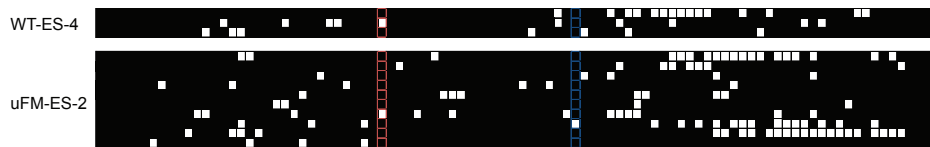
RPL13A

TSS

EX1-INTRON JUNCTION

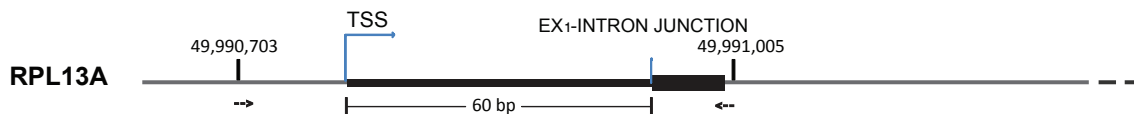


The template strand (C-rich)



● Unconverted C (dsDNA)  
○ Converted C (ssDNA)

B



WT-ES-4

The non-template strand (G-rich)

The template strand (C-rich)

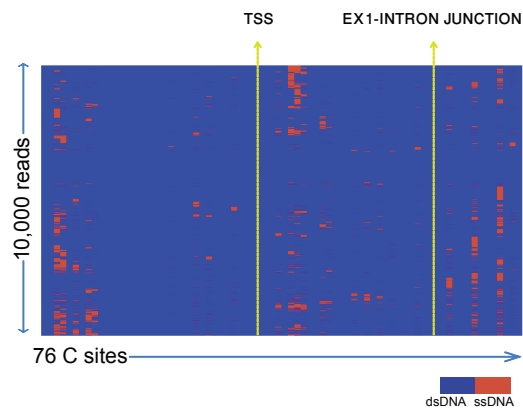
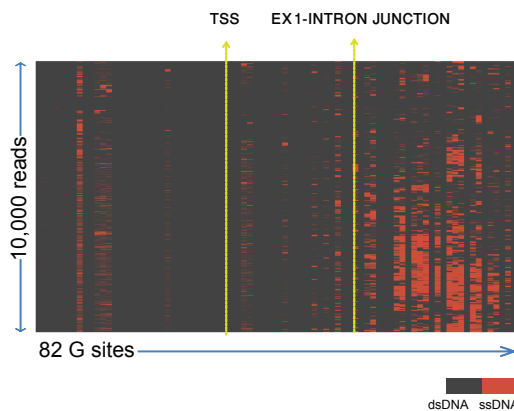


Figure S5. Bisulfite footprinting around the TSS in other loci. (A) DNA bisulfite footprinting by single colony sequencing using a pair of unconverted primers resulted in the amplification of the template and non-template DNA strands in two hESC lines (WT-ES-4 and uFM-ES-2) in the region that surrounds the TSS in other, non-repetitive, R-loop forming genes (*GAPDH* and *RPL13A*). TSS and first exon/intron boundaries are marked in red and blue, respectively. (B) Schematic diagram representing the 5'-UTR of *RPL13A* harboring the TSS and the exon/intron junction. DNA bisulfite footprinting by deep-sequencing across the TSS was carried out using unconverted primers in a single hESC line (WT-ES-4). This was followed by a bioinformatic analysis which separated the reads into non-template (G-rich; left) and template strands (C-rich; right), according to conversion patterns.