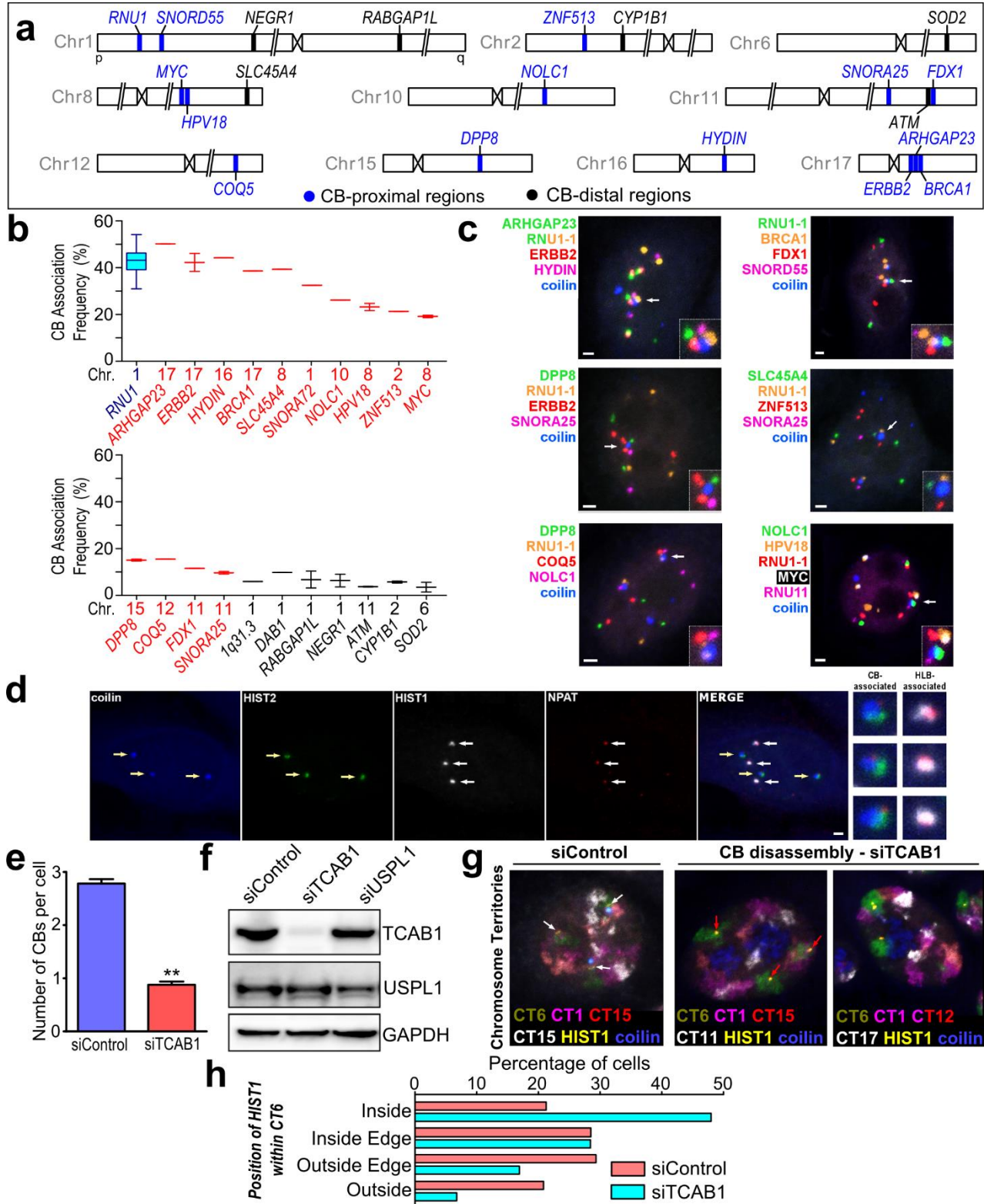


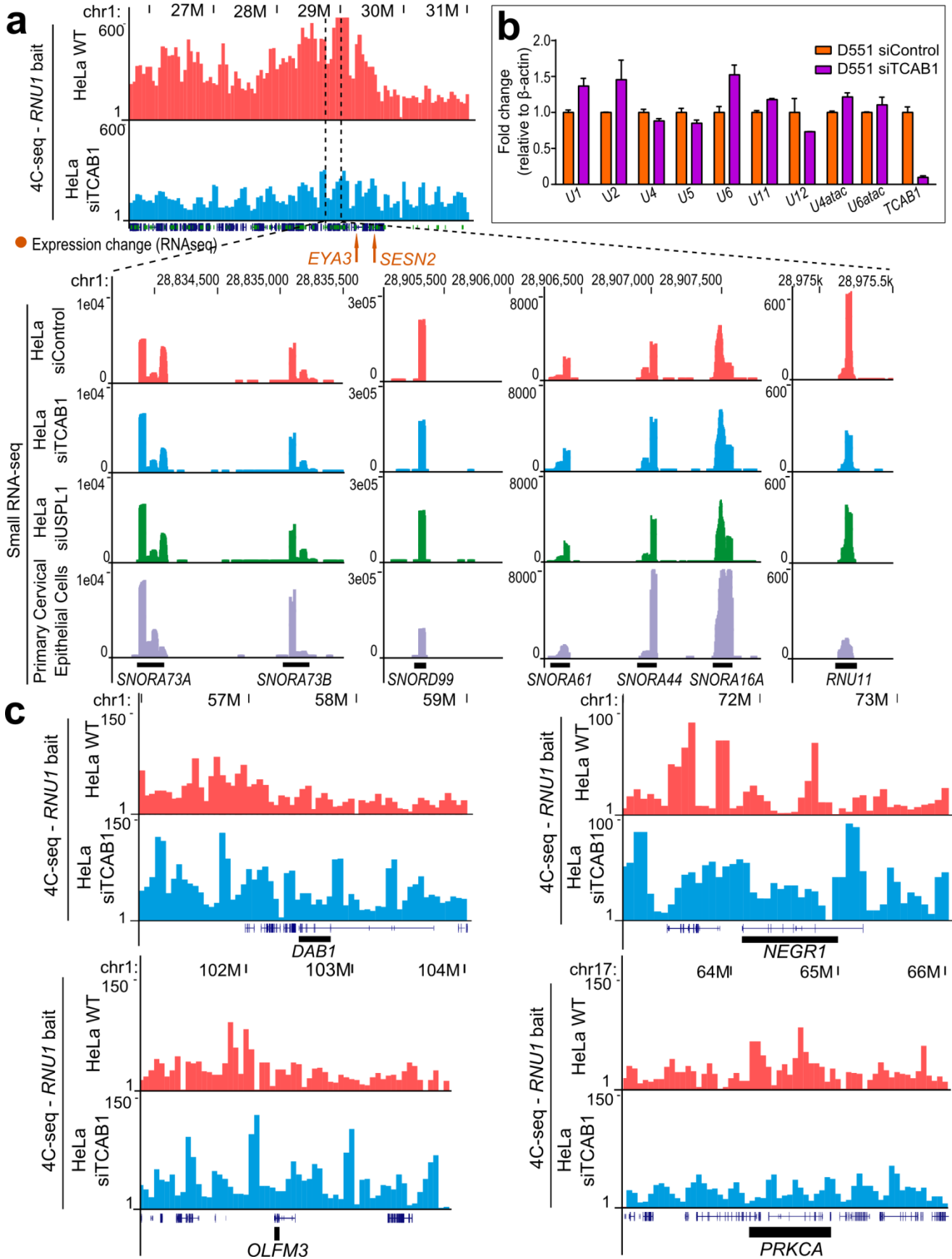
Supplementary Figure 1



(a) Approximate chromosomal location of BAC DNA-FISH probe target sequences used in this study. Blue text denotes CB interacting (4C contact positive) gene loci and black denotes non-CB interacting (4C contact negative gene) loci, 6-color fluorescent microscopy analysis of four chromosome territories

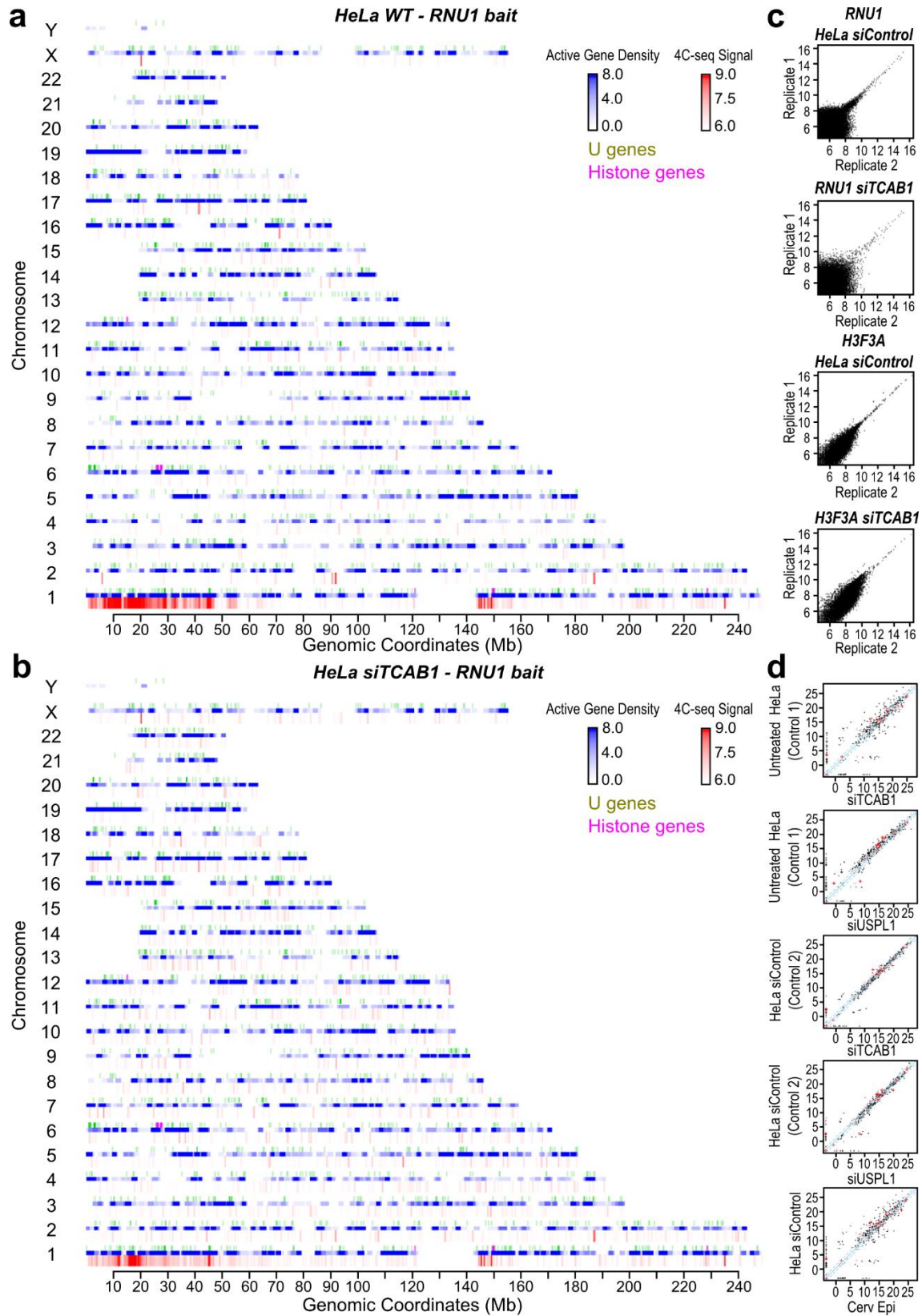
(CTs), CB interacting gene locus (*HIST1*) detected by DNA-FISH and CB localization visualized by CB marker protein coilin in aneuploid HeLa (CB-positive) and CB-deficient (siTCAB1) cells, (b) Box and whisker plot of DNA-FISH loci-CB association frequencies determined by manual scoring DNA FISH foci proximity to CBs using 6-color DNA FISH fluorescent microscopy images. Chromosome location is indicated below the X-axis. Black = non-CB interacting gene loci. Data represents assessment of at least 150 HeLa cells, (c) Validation of highly CB- interacting trans-chromosomal genomic regions identified by 4C analysis with the *RNU1* bait by using 6-color DNA FISH fluorescent microscopy analysis of CB-interacting loci (various) by DNA-FISH and coilin (blue) immunostaining in HeLa cells targeting the HLB marker protein NPAT (red) and CB marker coilin (blue), respectively, (d) Microscopic fluorescent analysis of the role of major histone locus (*HIST1*) on chromosome 6 and minor histone locus (*HIST2*) in the formation of CBs and HLBs in HeLa cells. *HIST1* (green) and *HIST2* (white) DNA FISH foci denote the major and minor histone gene cluster loci on chromosomes 6 and 1, respectively. Immunofluorescent detection of HLBs and CBs was achieved using specific antibodies, (e) Cajal body numbers in siControl and siTCAB1-treated HeLa cells. N=2 (400 cells), **= $p < 0.01$, significance was assessed by Student's T-test. Error bars represent SEM, (f) Representative TCAB1 and USPL1 immunoblots using extracts from HeLa cells exposed to indicated siRNA for up to 72 hours. GAPDH was used as a loading control, (g) (h) Quantitative assessment of HIST1 localization in control HeLa cells (red) and CB-deficient cells (siTCAB1, blue). Scoring reflected the relative position within the CT6 territory, from completely internal (inside), to positions reflecting more active states either within or on the outside edge of the CT (inside edge & outside edge, respectively) as well as gene completely looped out of the CT (Outside).

Supplementary Figure 2



(a) Representative 4C-seq contact profile of intra-chromosomal, CB-dependent interactions using a *RNU1* bait sequence for HeLa WT (pink) and siTCAB1 (blue) treatments. A number of small U RNA genes were enriched within this region (dotted lines) which were analyzed following siTCAB1 and siUSPL1 (green) treatment by small RNA-seq. The expression of these genes within human primary cervical epithelial cells is also shown (purple). A number of small U genes were determined to be insensitive to TCAB1 or USPL1kd, possibly distinguished from sensitive genes by their higher expression in primary epithelial cervical cells. The Y-axis displays the read counts/genome fragment, (b) Expression of histone genes in D551 primary diploid fibroblasts following depletion of TCAB1 by siRNA. N=2, error bars represent SEM. D551 siControl = orange, D551 siTCAB1 = purple, (c) Representative 4C-seq contact profile validations of non-CB-interacting (4C negative contact) loci used in DNA-FISH experiments as negative control. Profiles display *RNU1* bait to negative 4C region contacts on chromosome 1 and 17 in control (pink) and siTCAB1-treated (blue) HeLa cells. The Y-axis displays the read counts/genome fragment.

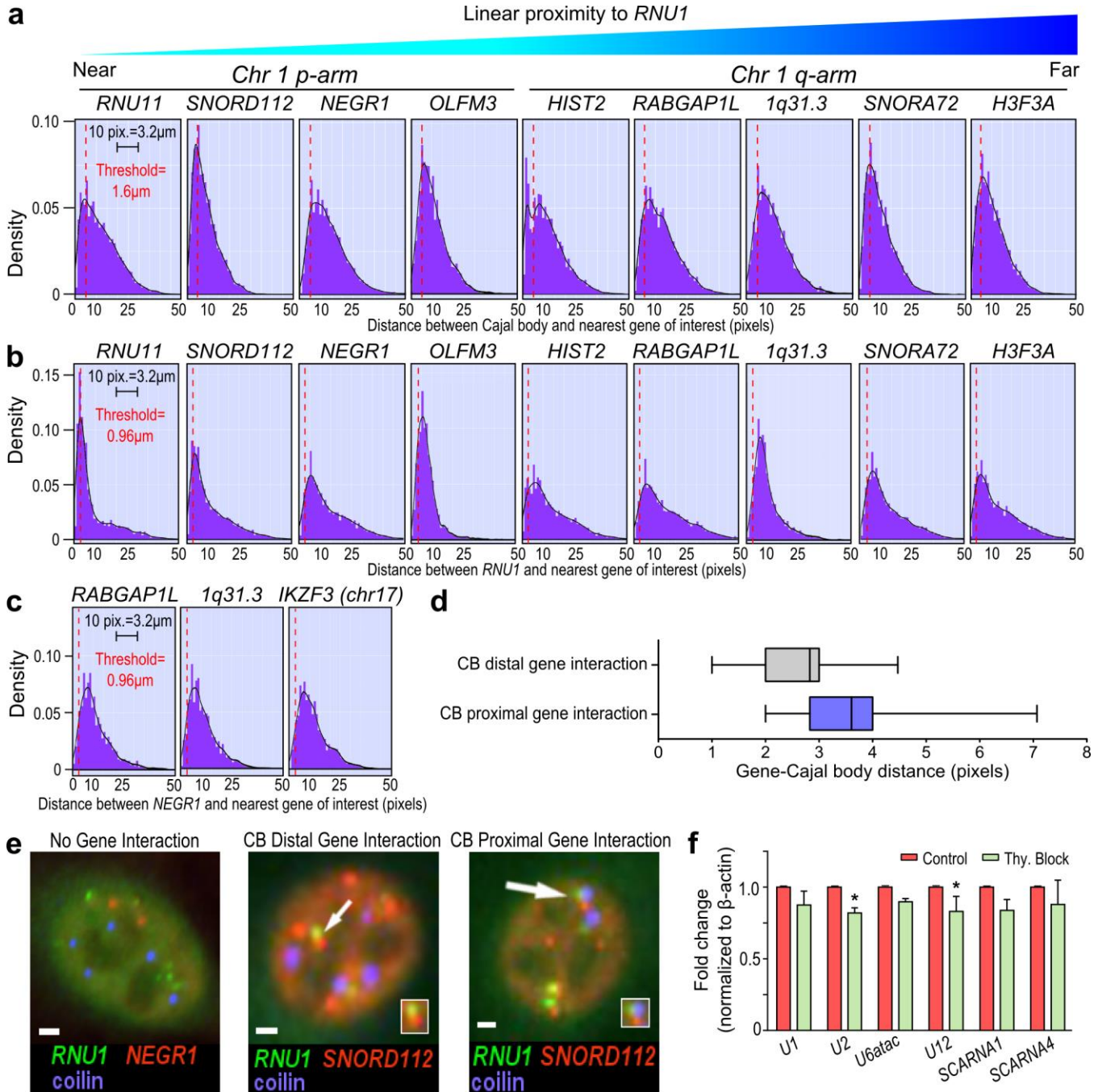
Supplementary Figure 3



(a)(b) Genome-wide *RNU1* 4C-seq physical contact profile (red, log10 scale) in WT HeLa cells following CB disassembly using TCAB1 siRNA (B). For comparison, active gene density is shown

adjacently (blue, log10 scale), as well as small U RNA gene and histone gene expression (green and red, respectively), (c) 4C-seq replicate reproducibility for *RNU1* and *H3F3A* bait analyses in HeLa cells treated with either control or TCAB1 siRNA treatments (siControl or siTCAB1, respectively). Spearman's correlation coefficients: *RNU1* bait – HeLa siControl = 0.182; *RNU1* bait – HeLa siTCAB1 = 0.080; *H3F3A* bait – HeLa siControl = 0.870; *H3F3A* bait – HeLa siTCAB1 = 0.903, (d) Small RNA-seq analysis comparing HeLa cells treated with either control or TCAB1 siRNA treatments (siControl or siTCAB1, respectively) and primary cervical epithelial cells (Cerv Epi). Red and pink crosses indicate loci within 10kb and 50kb of *RNU1* 4C contact regions in WT HeLa, respectively.

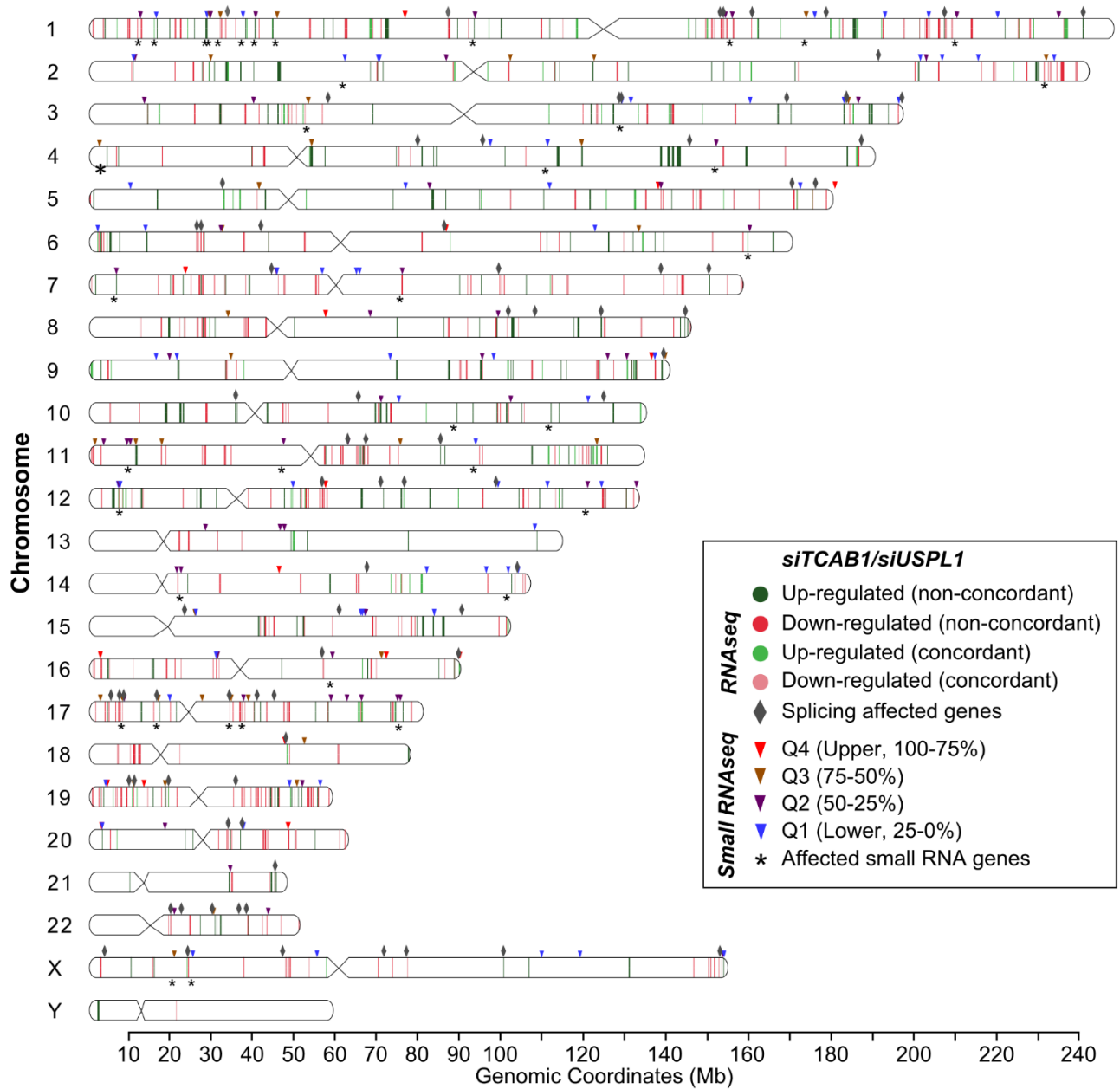
Supplementary Figure 4



(a) Histograms of minimum CB-gene of interest distances (in pixels), (b) minimum *RNU1*-gene of interest distances, and (c) minimum *NEGR1*-gene of interest distances, following high-throughput image analysis detection of chromosome 1-localized DNA FISH foci and CB immunofluorescence. Blue bars = binned data, black line = estimated density function. Histogram generated by analysis of >1000 HeLa cells/DNA FISH probe, (d) Box-and-whisker plot of CB-independent (*RNU1*-gene of interest, grey) and CB-dependent (*RNU1* or gene of interest – CB detected by coilin immunostaining, blue) distances (pixels) quantified through manual scoring of >150 foci-foci interactions in HeLa cells. Error bars

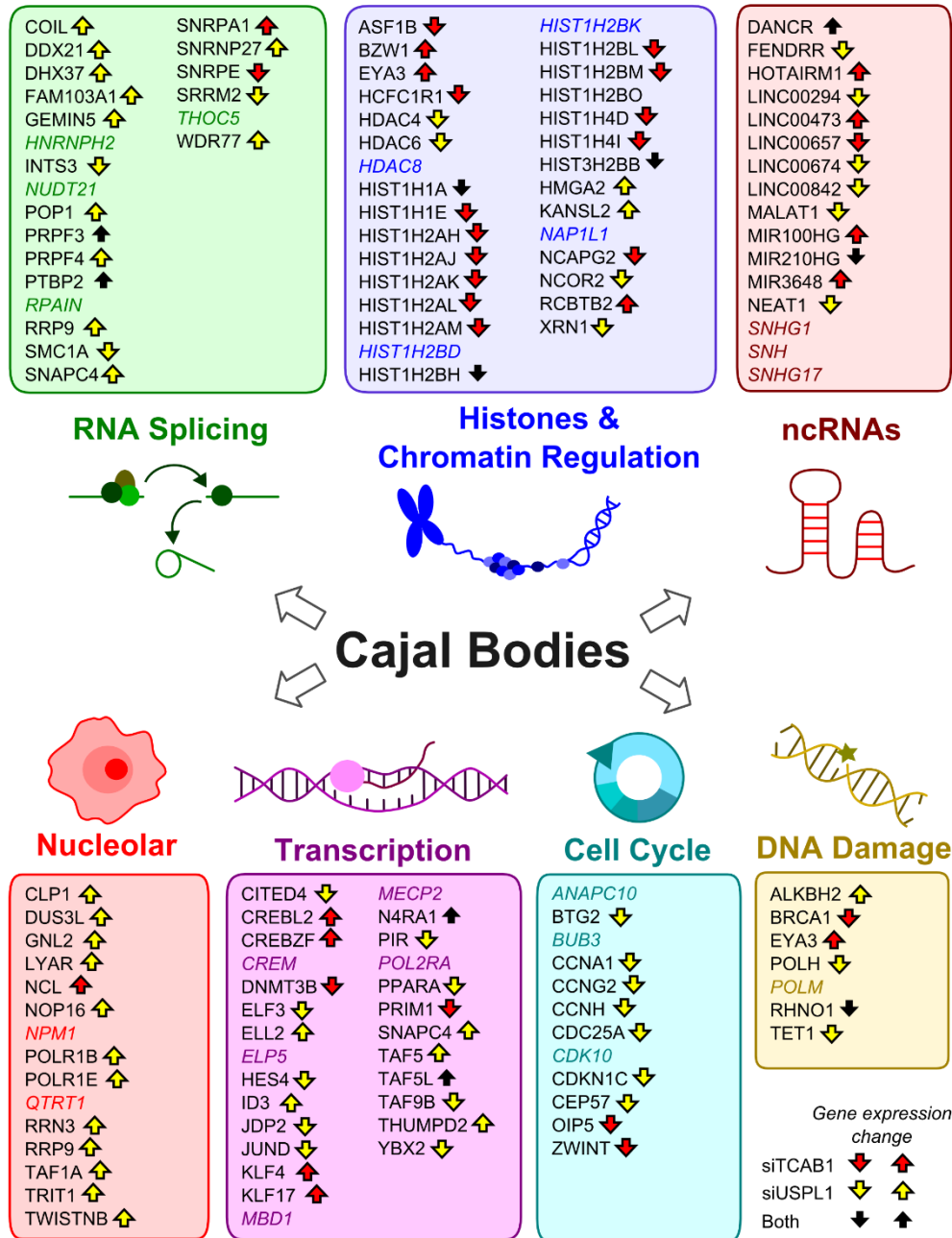
represent SEM, (e) Representative microscopy images acquired using the Opera hi-throughput imaging system depicting HeLa cells displaying either a lack of gene interactions (left), a CB distal gene interaction between *RNU1* and *SNORD112* (center) or a CB proximal gene interaction between *RNU1* and *SNORD112* (right). White scale bar = 2 μ m, (f) Expression of U sn/snoRNA genes and scaRNAs in cells treated with 5mM thymidine to induce a cell cycle block by qRT-PCR. N=2, *=p<0.05, **=p<0.01.

Supplementary Figure 5



Karyogram depicting the locations of RNA expression changes, small U gene expression levels and alternative splicing events in HeLa cells following CB disassembly (siTCAB1/siUSPL1). Up-regulated genes (total RNA-seq) are denoted by green chromosome bands (non-concordant = dark, concordant = light green) and down-regulated by red (non-concordant = dark, concordant = light red). Baseline (siControl) expression of small RNA genes are denoted by triangles (red = Q4 (upper, 100-75%), brown = Q3 (75-50%), purple = Q2 (50-25%) and blue = Q1 (lower, 0-25%)) Grey diamonds = alternative splicing events, * = small RNA expression change. Figure was produced using Idiographica¹

Supplementary Figure 6



Represented here are seven nuclear-related functional groups of potential CB targets in HeLa cells which show a significant ($p < 0.01$) reduction in RNA expression (identified by total RNA-seq) following TCAB1 or USPL1 siRNA treatment by CuffDiff or DEXseq analysis. Red or yellow arrows respectively identify TCAB1- or USPL1-only expression changes. Black arrows depict genes whose expression changed under both knockdown conditions. Genes that displayed alternative pre-mRNA splicing (assessed by Spanki and DEXseq) are italicized.

Supplementary Table 1

List of primers used in this study

Target	Forward	Reverse
<i>H3F3A</i> 4C bait	TAGCTCTTTATTGGAAAGAGTGATCT	TGCTTAAAACCAATACAACCACA
<i>RNU1</i> 4C bait	ATTGGAGTCCTTGTCAACTCTACA	ATGACAATCAGAGAAAGCTGA
<i>MCAM</i>	CTGCTGAGTGAACCACAGGA	TCAGGTCATGCAACTGAAGC
<i>MUTYH</i>	CTGTGATGGGATGATTGCTG	TCATCTTCTGCCCCGTTCT
<i>NSD1</i>	GCCAATTGTTTGCACCTCCT	TCTGTGACTGGCTGTTCCG
<i>PCNA</i>	CTGAGGGCTTCGACACCTAC	TTCCGTAACCGCTCATCTC
<i>POLR2A</i> Exon 22	AGGAAGGTGACTGCCAACAC	GTGAGCTTCCGGTCAGTCAT
<i>POLR2A</i> Exon 29	CTTCTTCTCCCCAAAAAC	TAGGTGCTCAGACCTCGTCA
<i>POLR2A</i> Exon 5	CGGAATGGAAGCACGTTAAT	GAGCCCTGCATCACAACAG
<i>RBBP4</i>	ATTCTTGCCACAGGATCAGC	CTGCATCTTCTGGGGATTGT
<i>SF3B3</i>	GAAATGGGTGGGACTGAGAA	GGCTTGACATGGCCAATACT
<i>TPM2</i>	GATGAAGCCAGYGCCAGAGYT	CGCACGTCAATGACATCACCT
<i>BECN1</i>	AGGAGAGACCCAGGAGGAAG	GTCAAAAAGGTCCCCAGTGA
<i>BRCA1</i>	GGAGTTGATCAAGGAACCTGTC	TGTCAAGCTGAAAAGCACAAAT
<i>DHX8</i>	ACCCTTGTAACCGCTATGA	CCAGCAGGGAAGAAATGAGA
<i>DMAP1</i>	ACCTGAGCCTGGTCTTCTT	GGGCCTCTTGAAAGTCAGTG
<i>DUSP3</i>	TAAAAACCCACCATTGGA	CTTCCCTGCTTGTCTTCTGG
<i>EIF2B3</i>	AGGCTGCTCTTCATGGCTAA	TGTTGTGAGGAAGCTGAGGA
<i>EIF6</i>	GGACATGGACTCGGTGGT	AACTGTGAGATCGGCTGCTT
<i>EXOC7</i>	TCACCTCAAGCCACCTTGG	AGCAGAGGACAGGCAAATG
<i>EZH1</i>	CTCAGTGCACACCCAACATC	TTCCAGCAAAGGAAGCAGT
<i>GNB1L</i>	CTCCAAGCCTGCCTAGGTG	CCTTCCCCATCTCCCTTGTAG
<i>GPNMB</i>	GCTTCAGCCATGTTGTGAAA	GCTGAAAGTATCCCGCCTCT
<i>H3F3A</i>	CATGGCTCGTACAAAGCAGA	GGGAAGTTTGCGAATCAGAA
<i>HECTD3</i>	GAGAACCTGGGTAGCGTGAA	CCGCTTTGGCATAAAGTTGT
<i>HIST1H2AK</i>	GGCTAAGACCCGTTCTTCAA	GATGATGCGGGTCTTCTTGT
<i>HIST1H2BD</i>	ATGGGGCTCATGAATTCCTTC	TTGGAAGTGGTGTACTTGGTGA
<i>HIST1H2BK</i>	GCTCGTCTCAGGCTCGTAGT	TGATTCCCATGGCCTTAGAG
<i>HIST1H3A</i>	AAACAGACCTGCGTTCCAG	GCTTGGACCGTCAGAGAGAC
<i>HIST1H3E</i>	GGTGCAGAGAAATAGCTCAGG	CAAAACAATTCACGCCCTCT
<i>HIST2H2AA3</i>	CGGTCCTCGAGTATCTGACC	TCCGTCTTCTTAGGGAGCAG
<i>HIST3H2A</i>	AAATGTCCGGTCGTGGTAAG	CAGGATCTCGGCAGTCAAGT
<i>HIST3H3</i>	GAGCTGCTAATCCGCAAGTT	TGTCCTTAGGCATGATGGTG
<i>Histone H3.2</i>	GCTACCAGAAGTCCACGGAG	GATGTCCTTGGGCATAATGG
<i>Histone H31.B</i>	GCTGCTGATTCGCAAACCTGC	GCATAGGTTGGTGCCTCAAACAG
<i>Histone H3C</i>	CGCTGGTAAGCCTGTTTTT	CTCCGTGGACTTCTGTAGC
<i>HMGB1</i>	CTTCGGCCTTCTTCTCTTC	CAGGCTTTCCTTGTAGCTCGAT

<i>HMGB1</i>	CTTCGGCCTTCTTCCTCTTC	CAGGCTTTCCTTTAGCTCGAT
<i>HNRNP3</i>	CGACCGGGACCATATGATAG	CTCCACCATAGCCATGTCCT
<i>MAD2L2</i>	TCACACGACAAGACCTCAACTT	CAGCGTGTCTGGATACTGA
<i>MCAM</i>	CTGCTGAGTGAACCACAGGA	TCAGGTCATGCAACTGAAGC
<i>MTOR</i>	TTTGACAGAGTTGGAAGACAGT	AATTCTCCTATTGTTGCCAGGA
<i>MUTYH</i>	CTGTGATGGGATGATTGCTG	TCATCTTCTGCCCGTCTTCT
<i>MYNN</i>	TCACTGTCAGCTGTGAATAGCC	TTCCAAGTCAAGGAAACAAG
<i>NBR1</i>	GGCATTGAAATAACACCCAGA	CATTGCTCAATAGGTGCTGGT
<i>NCAPH2</i>	CTGTGTTCTCCCTCTG	TCTTGGCTCTAGGCTGGGTA
<i>NOTCH1</i>	GGCTACAACCTGCGTGTGTGT	AGTTGGAGCCCTCGTTACAG
<i>NPM1</i>	TGCAGGAAGATGCAGAGTCA	TTTCTTCACTGGCGCTTTTT
<i>PCNA</i>	CTGAGGGCTTCGACACCTAC	TTCCGTAACCGCTCATCTC
<i>PSMC3IP</i>	CCTGCCAGATATCAAACCTGA	AGTGTTCCTGCATTTCTCC
<i>RAB2A</i>	GCAAGAATCCTTTCGTTCCA	GAGTCCATGTTCTCGTGCAA
<i>RAN</i>	TGTGTGGCAACAAAGTGGAT	AAAGCTGGGTCCATGACAAC
<i>RBBP4</i>	ATTCTTGCCACAGGATCAGC	CTGCATCTTCTGGGGATTGT
<i>RNU1</i>	GATACCATGATCACGAAGGTGGTT	CACAAATTATGCAGTCGAGTTTCC
<i>RNU11</i>	GTGCGGAATCGACATCAAGAG	CGCCGGGACCAACGAT
<i>RNU12</i>	AACTTATGAGTAAGGAAAATAACGATTCCG	CGACCTTTACCCGCTCAAAA
<i>RNU2</i>	TTTGGCTAAGATCAAGTGTAGTATCTGTTC	AATCCATTTAATATATTGTCCTCGGATAGA
<i>RNU3</i>	GAACGTGTAGAGCACCGAAAA	TCAGACCGGTTTCTCTCC
<i>RNU4</i>	GGCAGTATCGTAGCCAATGAG	CCGTAGAGACTGTCAAAAATTGC
<i>RNU5</i>	GGTTTCTCTTCAGATCGCATAAATC	CTCAAAAAATTGGGTTAAGACTCAGA
<i>RNU6</i>	GCTTCGGCAGCACATATACTAAAAT	ACGAATTTGCGTGTATCCTT
<i>RNU7</i>	GCATAAGCTTAGTGTACAGCTCTTTTAGAA TTTGTC	CGTAGAATTCAGGGGCTTTCCGGTAAAAAG CCAG
<i>RPS24</i>	AAGGCGACAGTGCCTAAGAC	TCTCCAGCTCACTGTCTTGC
<i>S100A4</i>	CATCTGTCCTTTTCCCAAG	TCTCTCCTCAGCGCTTCTTC
<i>SCARNA1</i>	GCACTTGATACTAACCGAGCTG	CTGGGCTTAAAAGACTCATGG
<i>SCARNA2</i>	GTGCAGGGTGAGTGTGAGTG	TCGCAGGAGGAGAGCTTTT
<i>SCARNA4</i>	CTGGAGGACTAAGAAGGCTGAG	GGTAGTACAGTCAGCCCTGTGATA
<i>SCARNA7</i>	TGTGTGCTTGTGGTGGCTA	TGGATCAGATCACAGGACCA
<i>SF3B3</i>	GAAATGGGTGGGACTGAGAA	GGCTTGACATGGCCAATACT
<i>SNORA18</i>	CTATCCCGATGGGGCTTT	TGTCTTGAATTCCTTCCCACA
<i>SNORA36B</i>	CCAAAGTGTAAAGTTCAGTTCAGG	TGTAGGCTGGGGATAAATGAA
<i>SNORA48</i>	TCCCTGACCTGGGTAGAGTG	TGTCTTGAATTCCTTCCCACA
<i>SNORA55</i>	AGCACCTGAATCTTTCCATT	CTGTAGAGACAGGCCCAAG
<i>SNORA74B</i>	AGAACCGTTCCTGTGATGGT	AGCAGCCAAAGTGAATGCTTA
<i>SNORA78</i>	ACTCTGGTTTCTTTCAGATCGT	GTCGGAACAAGGCCTCAAA
<i>SNORA7B</i>	GGGATCGCATCTGGAGACT	CGCAGAGTGTCTTCCAATCA
<i>STAT3</i>	ACAGCTCCTTGGTGCTTTA	TCTGCCAGCCTTACTCACT
<i>STAT5A</i>	CTGAACAACCTGCTGCGTGAT	GTGGACGATGACAACCACAG
<i>TBP</i>	TATAATCCCAAGCGGTTTGC	CACAGCTCCCCACCATATTC
<i>TCAB1</i>	AAGGCTGTAAGTCGGTCTT	GGGCTGAGGACATCAGAGAA
<i>THOC5</i>	TCAAACCTTTGCTGGCTCT	ATGGAGTGTGTTGGGGAAGT

<i>TP53BP1</i>	CCAGAGCCTGAGGTTTTGTC	GCATCAGGAGAAGGAGCAAC
<i>TPM2</i>	GATGAAGCCAGTGCCAGAGT	CGCATCAATGACATCACCT
<i>TUBA1A</i>	ATTGGCAGCATCTTCTTTG	CCAGTGCCAAGTGACAAGA
<i>TUBA3D</i>	GCTTCCTCATCTTCCACAGC	CCATGAAGGCACAGTCAGAA
<i>U4atac</i>	GCGCATAGTGAGGGCAGTACT	GCACCAAATAAAGCAAAGCTCTA
<i>U6atac</i>	AGGTTAGCACTCCCCTTGACAA	TGGCAATGCCTTAACCGTATG
<i>USPL1</i>	TTCTGGAATGGCAGACCT	TTCGAGCACAGTCCAGTCAC
<i>β-actin</i>	TCCGGCATGTGCAAGGCCGGCTTCG	GTACTTCAGGGTGAGGATGCCTCTCTTG

Supplementary References

- 1 Kin, T. & Ono, Y. Idiographica: a general-purpose web application to build idiograms on-demand for human, mouse and rat. *Bioinformatics* **23**, 2945-2946, doi:10.1093/bioinformatics/btm455 (2007).