Supplemental Figures

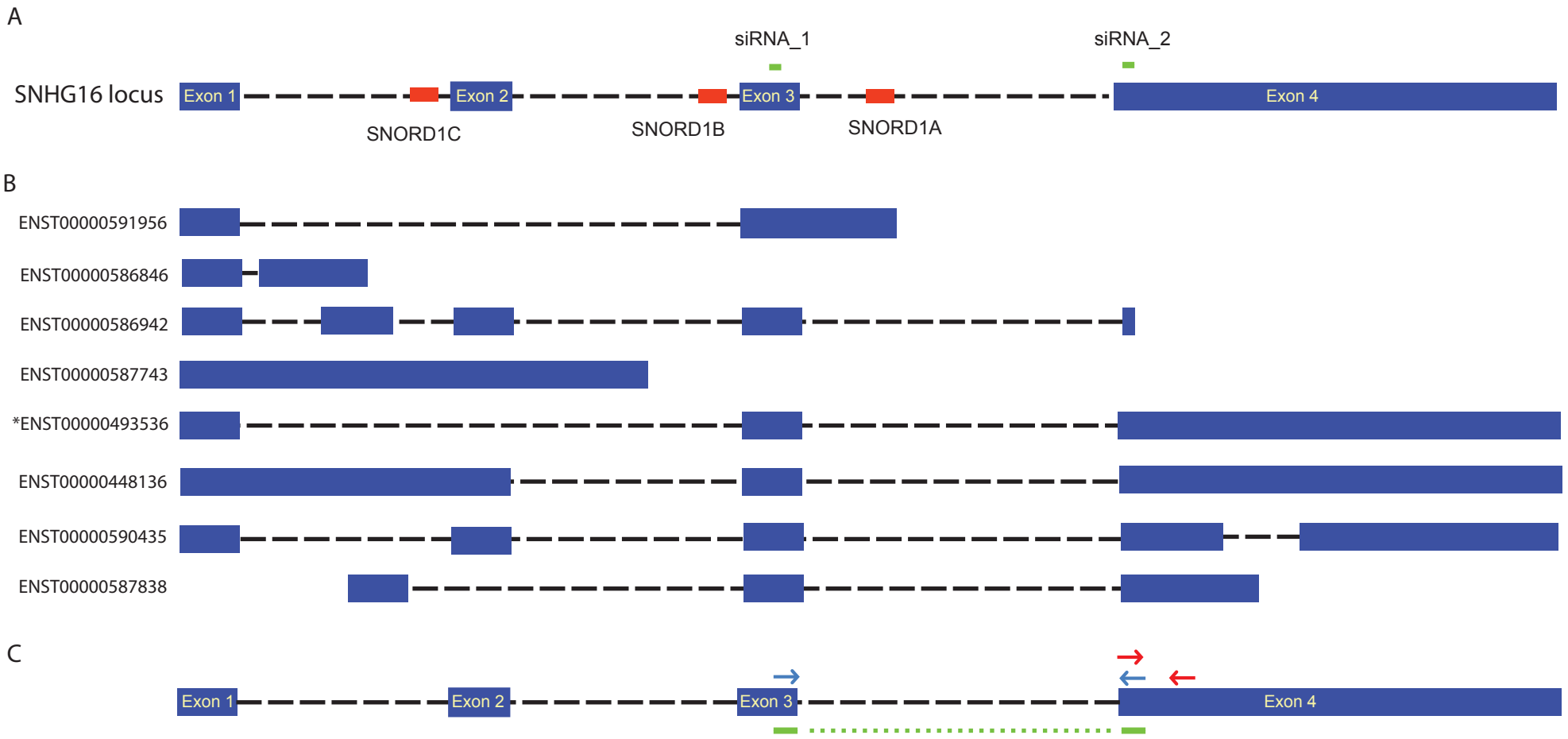


Figure 1. Schematic presentation of the SNGH16 locus. The genomic organization of SNHG16 including the snoRNAs hosted by SNHG16 (red bars) and the position of the siRNAs used for SNHG16 knock-down (green bars) (A). The eight TopHat defined SNHG16 isoforms. *Highly expressed isoform in colorectal cancer (B). Localization of primers used for RT-qPCR detection of Nbla10727(blue arrows) and Nbla12061(red arrows) in the study by Qi et al., 2015. The TaqMan probe used in the present study spans the exon 3 and 4 junction (green bars)(C).

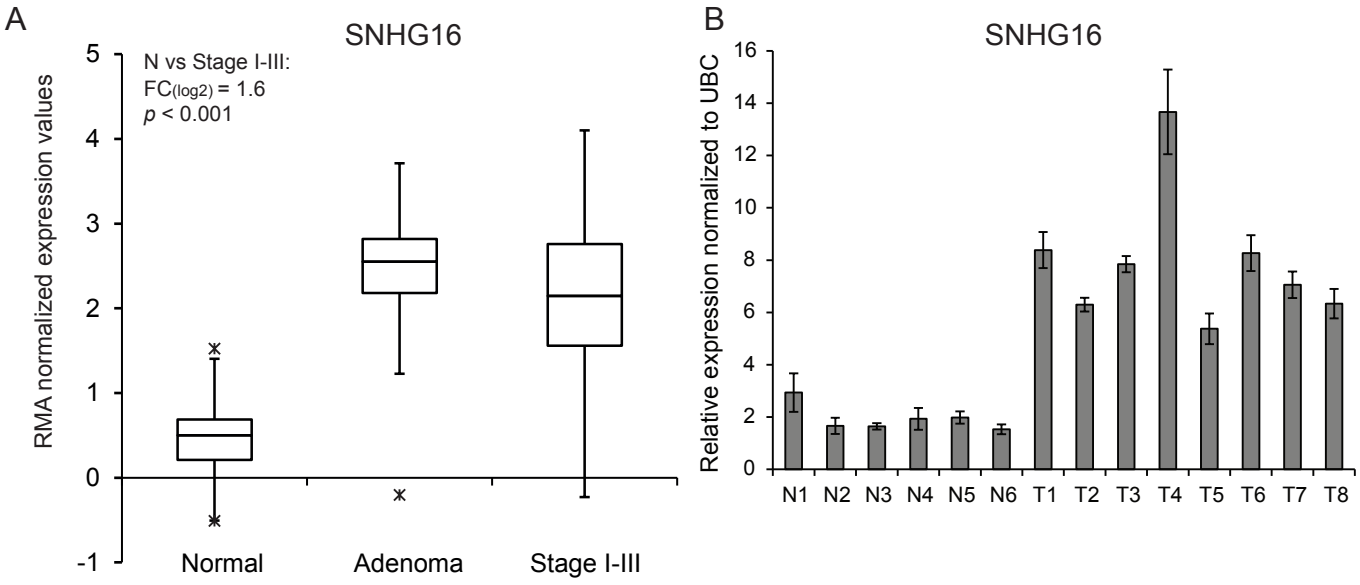


Figure 2. Validation of the up-regulation of SNHG16 in CRC tissue samples. Expression of SNHG16 in the validation cohort using custom-made microarrays. Normal colon mucosa (n=20), Adenoma (n=39) and adenocarcinoma (stage I-III)(n=44))(A). Expresson of SNHG16 in a subset of the samples from the validation cohort using RT-qPCR. Normal colon mucosa (n=6) and adenocacinoma (stage II and III) (n=8). The columns represents the mean of 3 replicates ± sd (B).

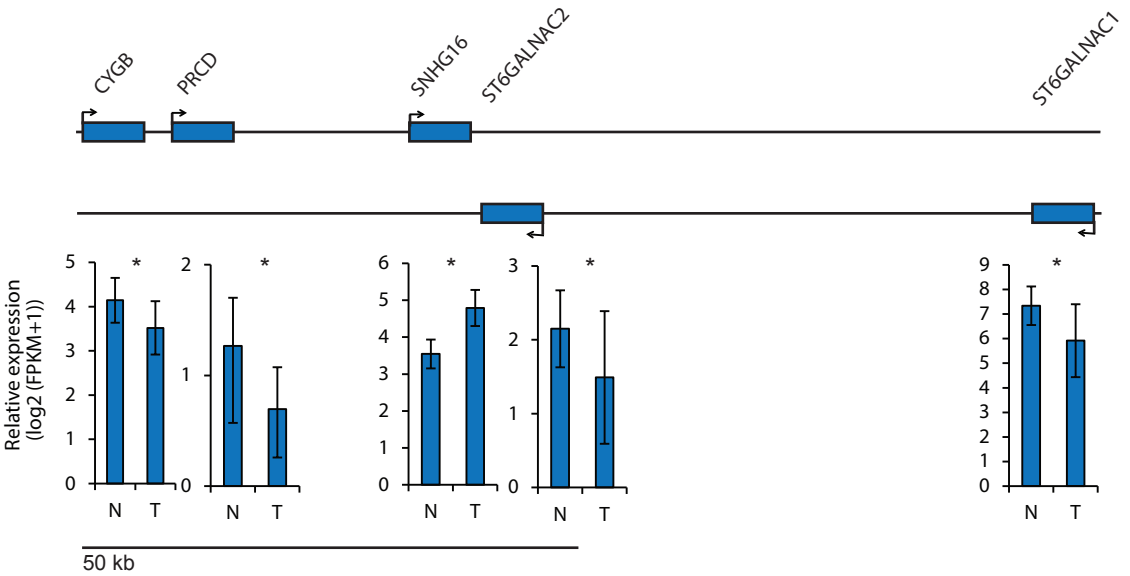


Figure 3. The expression pattern of SNHG16 is distinct from the surrounding genes. Shown is the SNHG16 locus and the surrounding genes within 50 kbs up- or downstream. SNHG16 is significantly upregulated and CYGB, PRCD, ST6GALNAC2 and ST6GALNAC1 are all significantly down-regulated (*$p < 0.001$).The analysis is based upon analysis of the normal mucosa and adenocarcinoma samples of the largeRNAseq cohort.

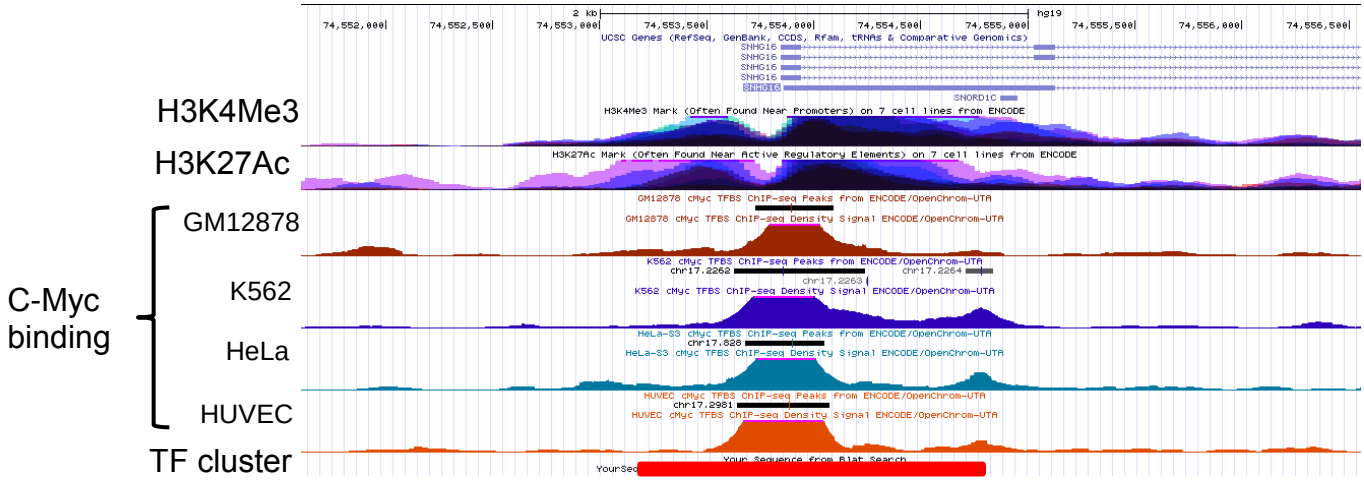


Figure 4. c-Myc binds the SNHG16 promoter in various cell lines. ENCODE/open Chrom (UT Austin) open chromatin TFBS by ChIP-seq data showing strong binding of c-Myc to the promoter region of SNHG16 in various cells lines such as GM12878 (blood), K562 (blood), HeLa (cervix), HUVEC (blood vessel). The TF binding cluster identified in Lovo cells at the SNHG16 TSS is shown as red box (Yan et al. Cell 2013;154:801-813.)

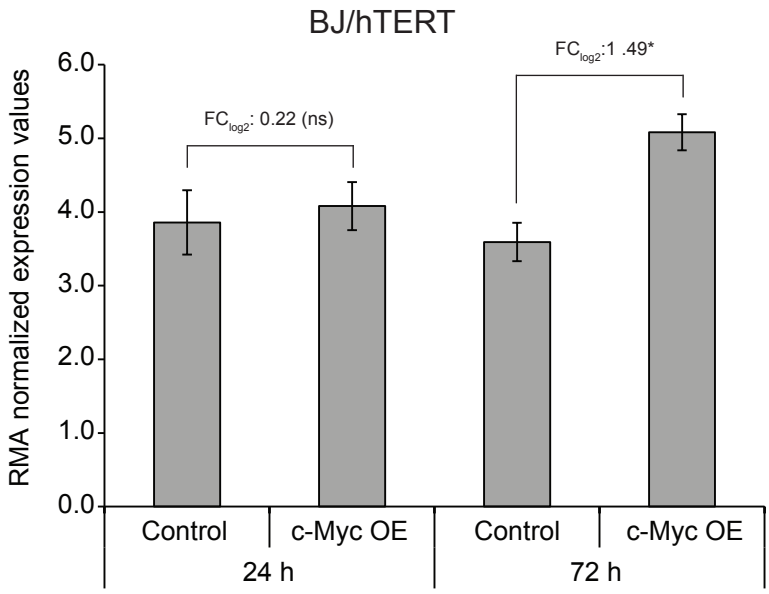


Figure 5. Expression of SNHG16 in human immortalized fibroblast cells (BJ/hTERT) over-expressing c-Myc or control vectors for 24 h or 72 h. The expression of SNHG16 was measeaured using custom-made microarrays. The columns represents the mean of 3 independent biological replicates ± sd. OE: over-expression and ns: non signifcant. *p<0.005.

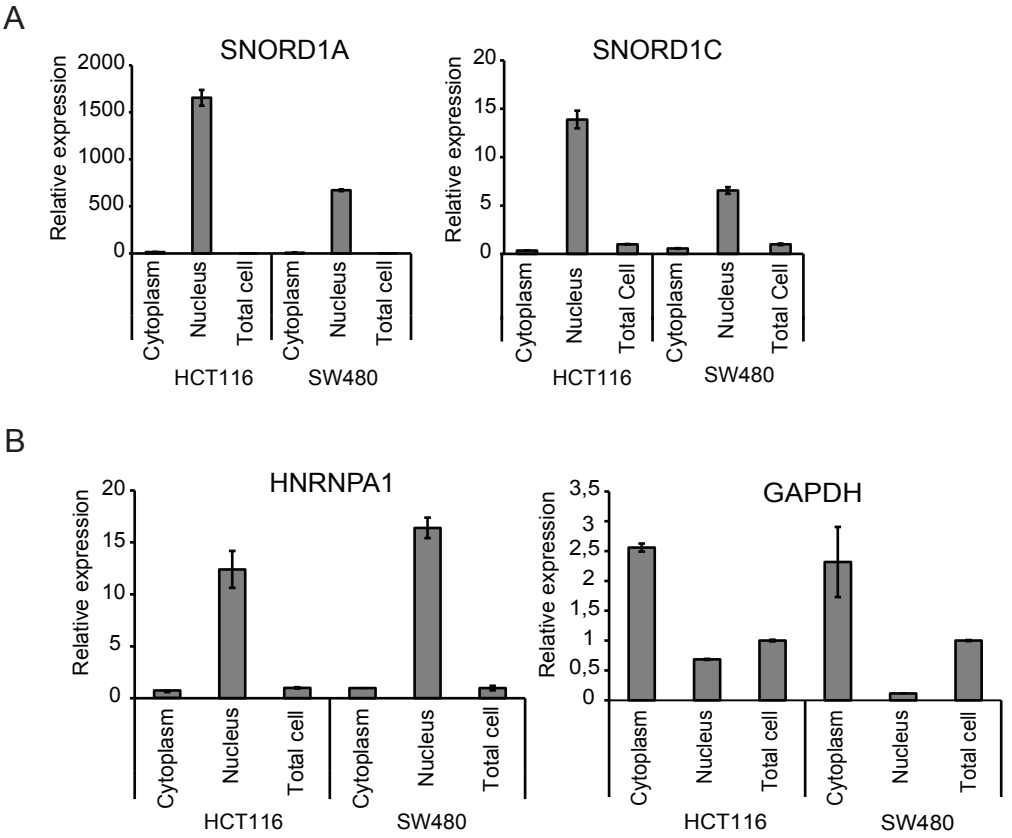


Figure 6. Expression of snoRNAs and fractionation controls in CRC cell lines. Expression of snoRD1A and snoRD1C (A) and HNRNPA1 (non-coding isoform)(nuclear marker) and GAPDH (cytoplasmic marker) (controls) (B) in fractionated HCT116 and SW480 cells. The cells were fractionated into cytoplasmic and nuclear fractions.The expression was measured in triplicates using RT-qPCR. The data are presented as the relative expression in the nuclear/cytoplasmic fractions normalized to the expression in unfractionated cells (total cells). Expression in total cell =1.The result of one representative experiment ±sd is shown.

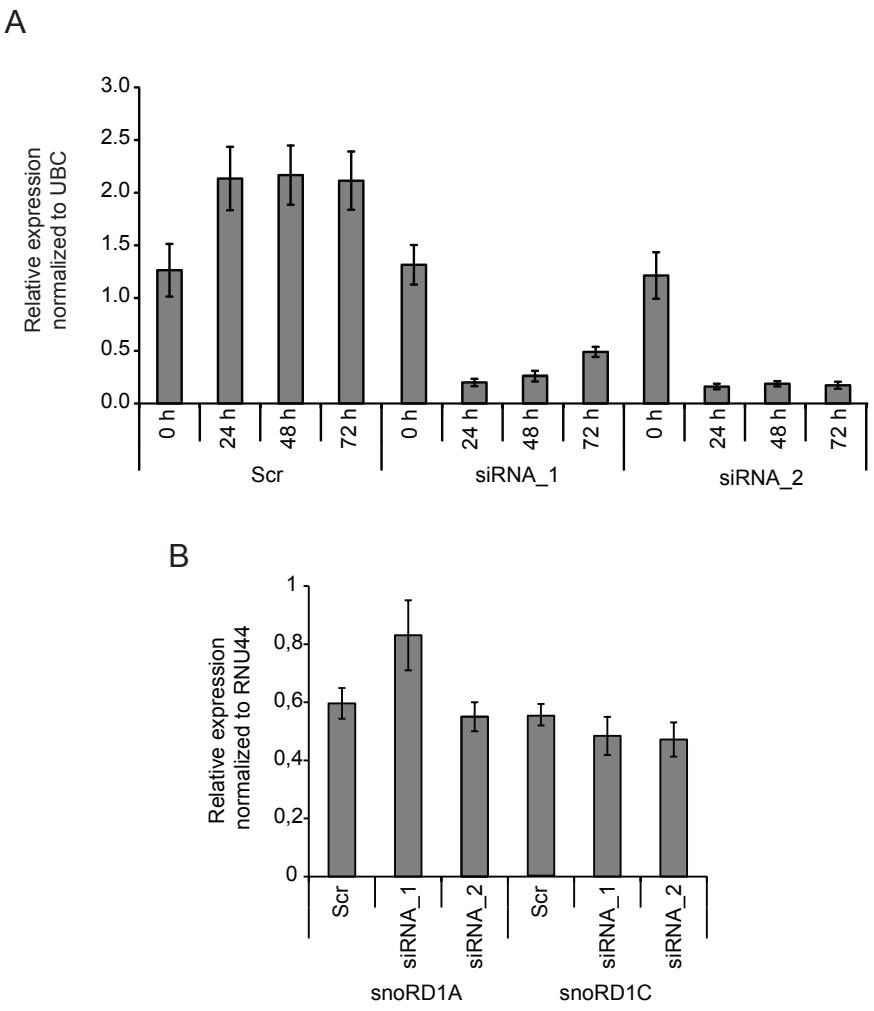
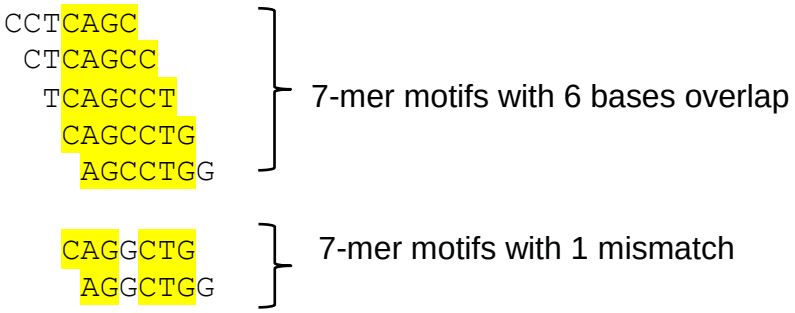


Figure 7. The relative expression of SNHG16 RNA in HCT116 cells transfected with siRNA_1 and siRNA_2 (20 nM). The cells were harvested at four different time points.The columns represent the mean of 3 replicates ± sd (A).The relative expression of snoRD1A and snoRD1C in HCT116 cells transfected with siRNA_ 1 and siRNA_2 (20 nM) for 48 hours. The columns represents the mean of 3 replicates ± sd (B).

A



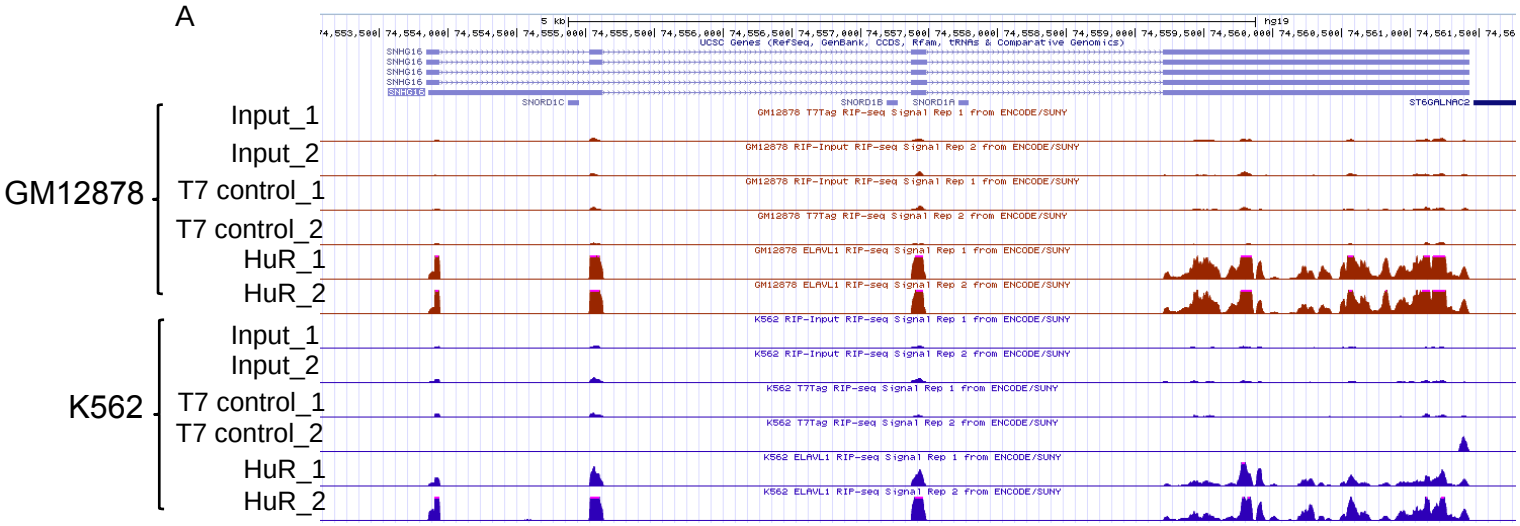
CAG(G/C)CTG 7-mer consensus sequence

B SNHG16 (reverse complement sequence)

1171 CAGCCTG (position_1)
1516 CAGCCTG (position_2)

Figure 8. Identification of common motifs in the genes affected by SNHG16 knock-down. Five 7-mer motifs overlapped with 6 bases and two motifs had only a single mismatch to the 7-mers, thus defining a 7-mer consensus sequence (A). Matching of the SNHG16 reverse complement to the overlapping 7-mers identified matches at 2 positions in SNHG16. The numbers are defined by the first base in the reverse complement of the SNHG16 RNA (NR_038108.1) (B). Bases matching the 7-mer consensus sequence are marked in yellow.

A



B

	HuR binding probability (RPIseq)	
RNA	RF classifier	SVM classifier
SNHG16	0.75	0.80
c-Myc	0.80	0.76

Figure 9. HuR binding analyses. The SNHG16 transcript binds HuR in GM12878 and K562 cells. ENCODE/SUNY Albani RNA immunoprecipitation (RIP)-seq data showing enrichment of SNHG16 in immunopreciptates from GM12878 (blood) and K562 (blood) cells in a HuR dependent manner (A). Interaction probability of HuR/SNHG16 and HuR/c-Myc (positive control) using the RNA-protein interaction prediction program (RPIseq). RIPseq range 0-1 (positive range >0.5). The following protein and mRNA sequences where used: HuR; Q15717 (UniProt), SNHG16; NR_038108.1 (RefSeq) and c-Myc; NM_002467 (RefSeq) (B).

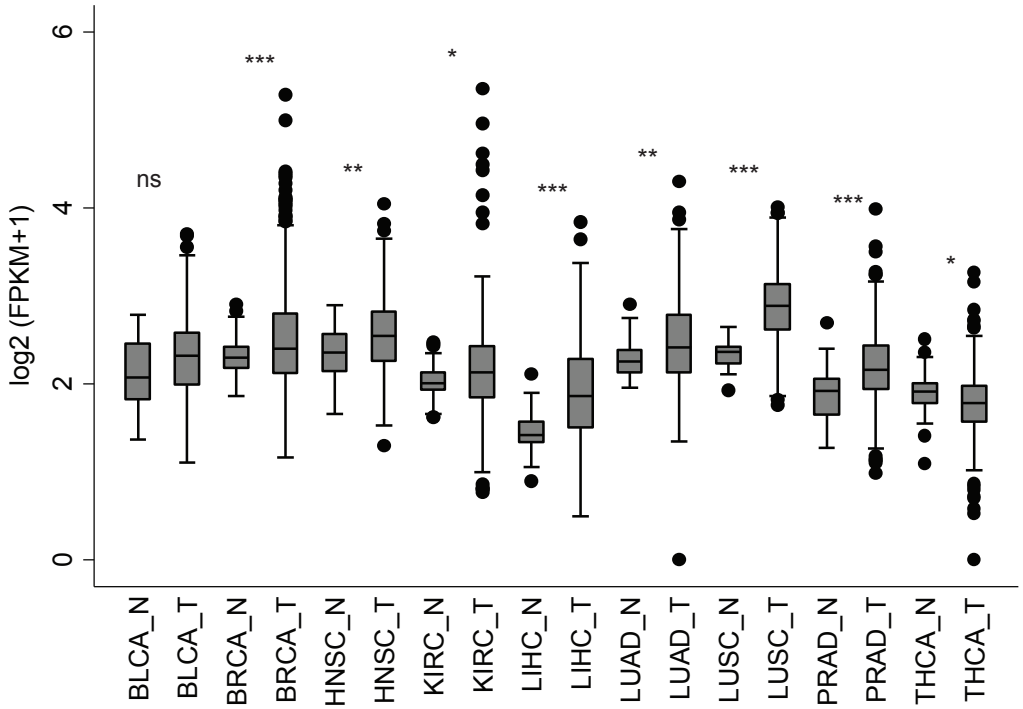


Figure 10. Expression of SNHG16 based on data from TCGA and downloaded from Tanric. BLCA: bladder urothelial carcinoma (n=19 (N) and n=252 (T)), BRCA: breast invasive carcinoma (n=105 (N) and n=837 (T)), HNSC: head and neck squamous carcinoma (n=42 (N) and n=426 (T)), KIRC: kidney renal clear cell carcinoma (n=67 (N) and n=448 (T)), LIHC: liver hepatocellular carcinoma (n=50 (N) and n=200 (T)), LUAD: lung adenocarcinma (n=58 (N) and n=488 (T)), LUSC: lung squamous cell carcinoma (n=17 (N) and n=220 (T)), PRAD: prostate adenocarcinoma ¨ (n=52 (N) and n=374 (T)) and THVA: thyroid carcinoma (n=59 (N) and n=497 (T)). N: normal, T: tumor and ns: not significant. *p < 0.05, **p < 0.01 and ***p <0.001 (Student's unpaired *t*-test).