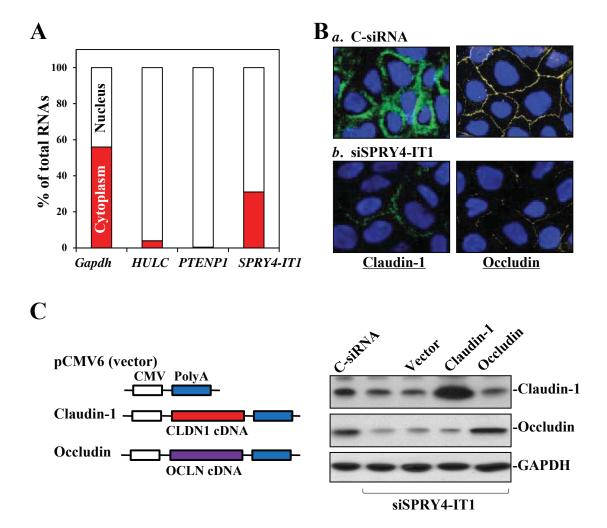
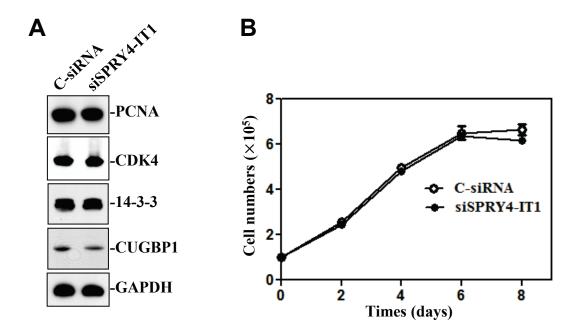
Supplemental Materials Molecular Biology of the Cell

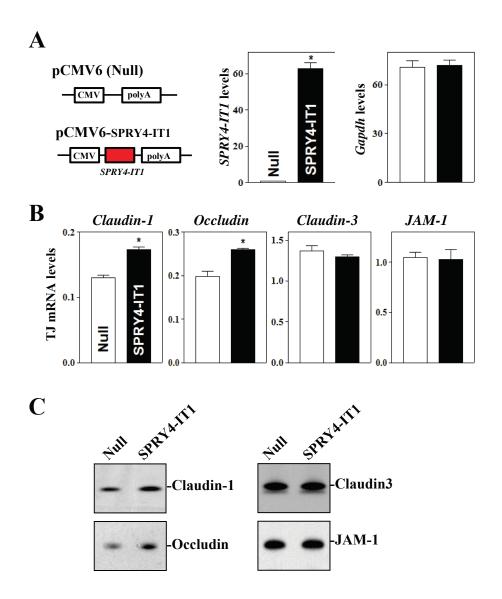
Xiao et al.



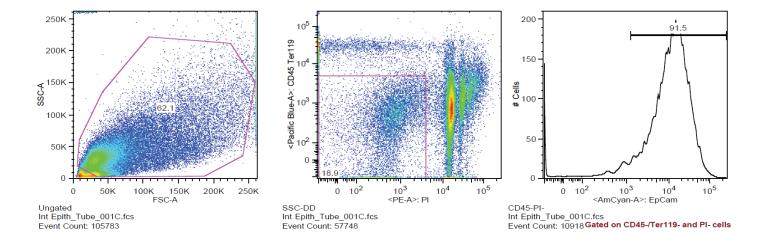
Supplementary Figure 1. Distribution of *SPRY4-IT1* and changes in the levels of claudin-1 and occludin after different treatments. A) Levels of nuclear and cytoplasmic *Gapdh* mRNA and lncRNAs *SPRY4-IT1*, *HULC*, and *PTENP1* in Caco-2 cells as measured by Q-PCR analysis. Values are the means \pm SEM (n=3). B) Cellular distribution of claudin-1 and occludin in SPRY4-IT1-silenced cells. After Caco-2 cells were transfected with siSPRY4-IT1 and C-siRNA for 48 h, cells were fixed, permeabilized, incubated with the antibody against occludin or claudin-1, and then with anti-IgG conjugated with fluorescein isothiocyanate. C) Representative immunoblots of claudin-1 and occludin in SPRY4-IT1-silenced cells transfected with the claudin-1 or occludin expression vector.



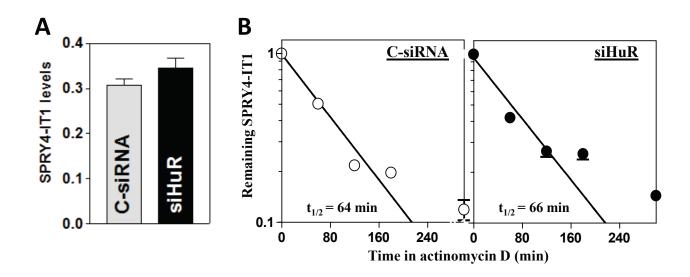
Supplementary Figure 2. Effect of SPRY4-IT1 silencing on Caco-2 cell proliferation. *A*) Representative immunoblots of PCNA (proliferating cell nuclear antigen), CDK4 (cyclindependent kinase 4), 14-3-3, and CUGBP1 (CUG-binding protein-1) proteins in Caco-2 cells 48 h after transfection with siSPRY4-IT1 or C-siRNA. B) Cell numbers at different times after transfected with siSPRY4-IT1 or C-siRNA. Values are the means \pm SEM (n = 6).



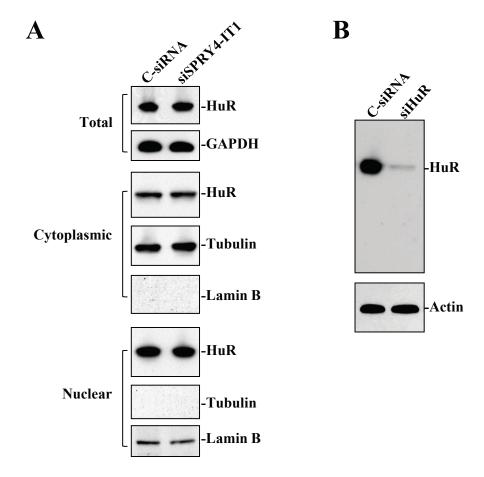
Supplementary Figure 3. Ectopic overexpression of SPRY4-IT1 on TJ expression in Caco-2 cells. A) SPRY4-IT1 levels 48 h after transfecting cells with the SPRY4-IT1 expression or control (null) vector. Values are means \pm SEM from three separate experiments. $^*P < 0.05$ compared with Null. B) Changes in the levels of TJ mRNAs in cells described in A. C) Representative immunoblots of TJ proteins in cells described in A.



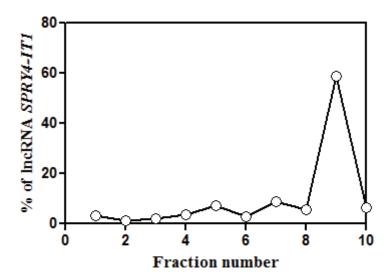
Supplementary Figure 4. **Population of IECs in the extracted tissue as measured by fluorescence-activated cell sorting analysis**. The small intestinal mucosa was scraped from the underlying smooth muscles using a glass microscope slide and incubated in stain buffer (BD biosciences) containing FBS and 0.5 mM EDTA for 15 min at 37°C. Cells were filtered with a 70-μm cell strainer (BD biosciences) and blocked with Fc Block (2.4G2) for 5 min at room temperature, followed by incubated with antibodies and other reagents for 30 min 4°C. Dead cells were identified and excluded after staining with propidium iodide (PI). The epithelial cell fraction was defined by PI-CD45-TER119-EpCAM+ gating. Samples were analyzed by using a BD LSR II Flow Cytometer System. Experiments were repeated six times and showed similar results.



Supplementary Figure 5. Effect of HuR silencing on SPRY4-IT1 content and its stability. A) Levels of SPRY4-IT1 48 h after transfection with siHuR or C-siRNA. Values are means \pm SEM from three separate experiments. B) Stability of SPRY4-IT1 in cells described in A. The levels of SPRY4-IT1 were examined at different times after administration with actinomycin D.



Supplementary Figure 6. Effect of SPRY4-IT1 silencing on cellular HuR abundance and HuR levels after transfection with siHuR. A) The levels of total, cytoplasmic, and nuclear HuR were examined 48 h after cells were transfected with siSPRY4-IT or C-siRNA. Three separate experiments showed similar results. B) HuR levels in cells transfected with siHuR or C-siRNA for 48 h.



Supplementary Figure 7. Distribution of *SPRY4-IT1* in each gradient fraction prepared from polysomes isolated from Caco-2 cells. Total RNA was isolated from different fractions, and the levels of *SPRY4-IT1* were measured and plotted as a percentage of total *SPRY4-IT1* levels in each sample.

Supplementary Table 1: Sequence alignments with highest score between *SPRY4-IT1* and TJ mRNAs

Score	Expect	Identities	Gaps	Strand	
21.1 bits	1.0	11/11(100%)	0/11(0%)	Plus/Plus	
SPRY4-IT1	24 TTGGTCAGGCT 34				

SPRY4-IT1 24 TTGGTCAGGCT 34
CLDN1 722 TTGGTCAGGCT 732

Score		Expect	Identities	Gaps	Strand
107 bits		2e-26	97/120(81%)	3/120(2%)	Plus/Plus
SPRY4-IT1 OCLN	2 3452			GTCTTGAACTCCTGACC	
SPRY4-IT1 OCLN	62			ACAGGCATGAGCCACTGC	

Score	Expect	Identities	Gaps	Strand	
19.3 bits	1.4	13/15(87%)	0/15(0%)	Plus/Plus	
CDDAYA ITTI 624 CTCACCATTCCCCCAT 638					

SPRY4-IT1 624 CTGAGCATTGCCCAT 638 | III III III III III CLDN3 290 CTGCGCGTTGCCCAT 304

Score		Expect	Identities	Gaps	Strand
107 bits		1e-26	97/120(81%)	3/120(2%)	Plus/Plus
SPRY4-IT1 JAM-1	1 3439	GTAGAGATGGGGGTTTCATCCTGTTGGTCAGGCTGGTCTTGAACTCCTGACCTCAAGTGA 60			
SPRY4-IT1 JAM-1	61 3496	11 1111 11111111	111111 1 11 11111	CAGGCATGAGCCACTGCGC	Ī