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Supplemental Information

A Cohesin-Mediated Intrachromosomal Loop Drives Oncogenic *ROR* lncRNA to Accelerate Tumorigenesis

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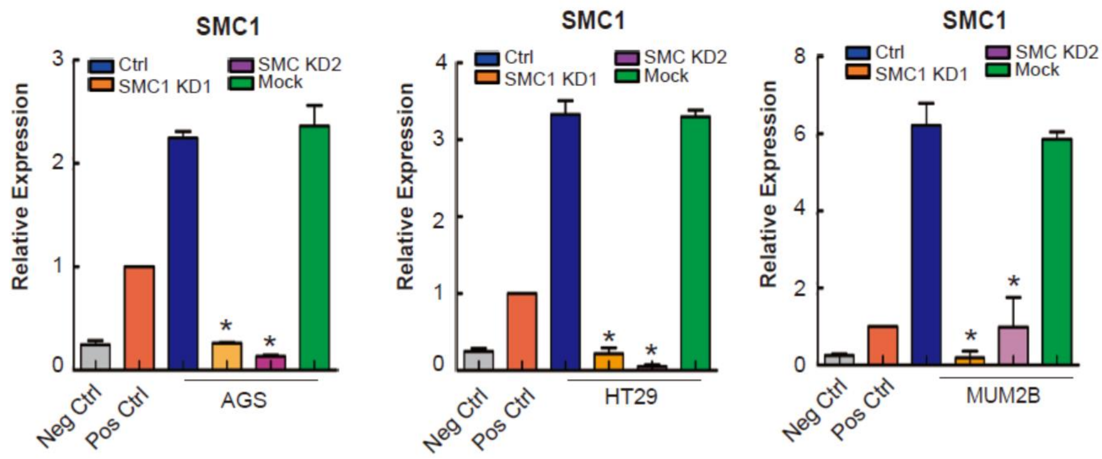

Figure S1. Validation of two shRNA mediated knockdowns of SMC1

Real-time PCR demonstrating that shRNA efficiently silenced *SMC1* at the RNA expression level in AGS, HT29 and MUM2Bcells. *P<0.05: compared with the control (ctrl) and mock. Mock: empty pGIPZ vector. 293T cell and fibroblast used as positive and negative control respectively.

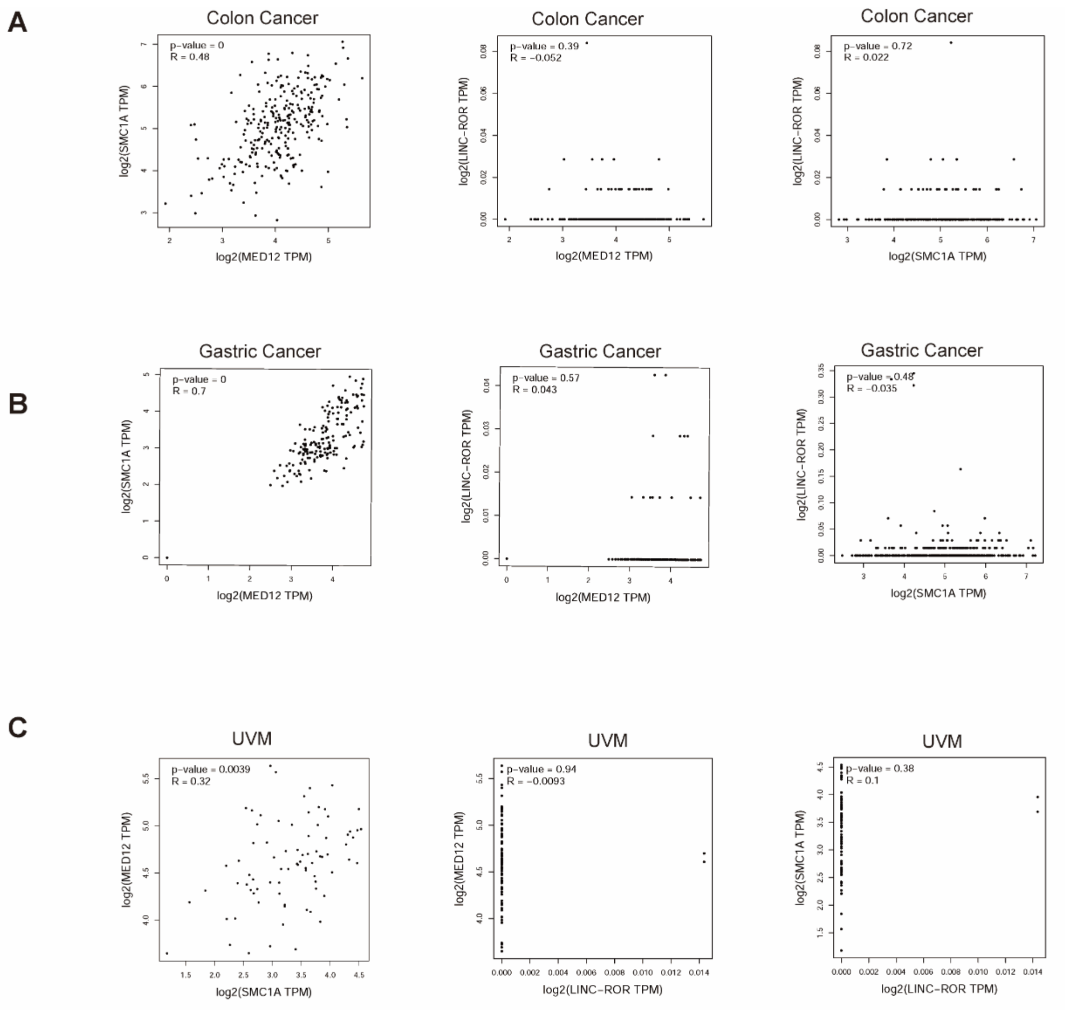

Figure S2. Pearson correlation between *ROR* and the epigenetic regulators (*SMC1* and *MED12*) mRNA expression in the GEPIA dataset.

Gene expression analysis using the GEPIA dataset validated the positive correlation between *SMC1* and *MED12* in colon cancer, gastric cancer and uveal melanoma (UVM). But there was no correlation between the expression of *SMC1* and *MED12* with *ROR* lncRNA.

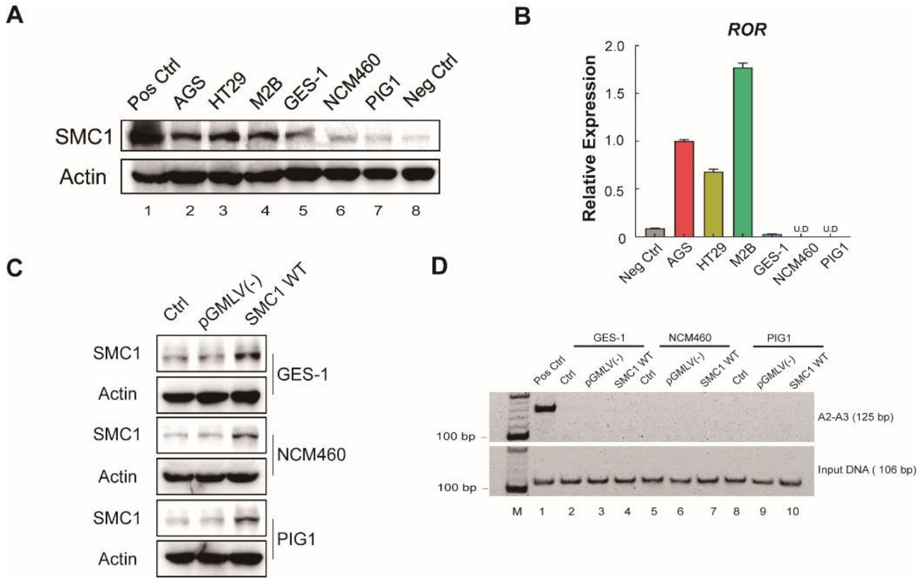

Figure S3. Rescue study of *SMC1* in low *ROR* expression cells

A. *SMC1* expression in different tumors were measured by Western blot. *SMC1* presented higher expression in a series of tumor cells than in three normal cells (NCM460, GES-1 and PIG1). 293T cell: positive control. Fibroblast: negative controls.

B. Real-time PCR showed *ROR* expression in tumors and normal cells. There was almost no expression of *ROR* in three normal cells (NCM460, GES-1 and PIG1). Fibroblast was used as negative controls.

C. The protein levels of SMC1 in the normal cells treated with wild-type SMC1 and empty vector were measured with Western blotting. *SMC1* was overexpression in the presence of the overexpressed wild-type SMC1. SMC1 WT: SMC1 ORF was cloned into pGMLV plasmids for stable expression. pGMLV(-): empty pGIPZ vector.

D. Chromosome conformation capture (3C) detected the intrachromosomal loop in the normal cells treated with wild-type SMC1. The cohesin-orchestrated intrachromosomal looping was not restored in the *SMC1* overexpression cells. MUM2B cell was used as a positive control.

Table S1. Primers for 3C

Primer	Sequence (5’-3’)
P1A	TAACAGAACCATCAGCCCTGAATGTC
P1B	ACCATCAGCCCTGAATGTCCCCCTACT
P2A	TGCAAAGAGCTGGCCAGAGGCATC
P2B	TGGCCAGAGGCATCCTGGCAGT
P3A	TGATCCACTGGTCAGATCCCAGGTC
P3B	ACTGGTCAGATCCCAGGTCTCATGACTC
P4A	TTCTGACACACCTCCAGTGGATCTG
P4B	TCCAGTGGATCTGGGCGCACTCCAG
P5A	GATGACCAGTGGCCCTGGAGGTC
P5B	GCCCTGGAGGTCCCAAAGGTCAAAGATG
P6A	TGTGCATTAATTCCAACCACCTGCTC
P6B	TAATTCCAACCACCTGCTCTGTGGAGC
R1	TCAGCTGTCACTCAGCCACAGTG
R2	CAGTGAGAGGATGAACACCTCGCA

Table S2. Primers for ChIP and qRT-PCR

Gene	Primer Sequence (5’-3’)	Purpose
ROR- pro1F (L1)	ACCACTCATTGTTGGCGCATTCACTG	ChIP
ROR- pro1R (L2)	GTACTCTTCCCACCCCTACTGCCA	ChIP
ROR-pro2F (L3)	AGCGCCCTTAAGCAGGGTCATTC	ChIP
ROR-pro2R (L4)	CTGTGCTCGCTCCCTTGGGAG	ChIP
ROR-pro3F (L5)	CAGATCCCAGGTCTCATGACTCCCAG	ChIP
ROR-pro3R (L6)	GCACAATGGCACTGCAGCACTGT	ChIP
SMC1-F	AGAGGTTCACCGCCATCATTGGAC	qRT-PCR
SMC1-R	CACAGGAGCTCCATGGATCAGGTC	qRT-PCR
ROR-F	CTTGGCTTAGCGGCTGAAGACTGACG	qRT-PCR
ROR-R	TGGCCATGCACCAGGTAGAAATCTGTAG	qRT-PCR

Table S3. Primers for enhancer cloning and RNAi

Gene	shRNA CN / Primer Sequence （5'-3'）
Smc1a	H4 (V3LHS_637850)
Smc1a	H10 (V3LHS_637855)
ROR-Enhancer-F	GGGTACCACAATCATTGAAAGCTTTCATG
ROR-Enhancer-R	GCTCGAGCACTGGCTCAGGATAACTAG

The underline indicated the restriction enzyme site. The CN indicated the catalog number of Open Biosystems.

Table S4. Primers for MED12 RNAi

Primer	Sequence (5'-3')
si*MED12*-1	CGCUGGUCUUUCGAUAAAUTT
si*MED12*-2	CCAGCACCUUUCACAUUAUTT
si*MED12*-3	CCAGGGCUAUACUCCUUAUTT
Negative Control	UUCUCCGAACGUGUCACGUTT