

SUPPLEMENTAL MATERIAL

Table S1: Sequences of siRNAs.

	siRNA sequence
siTRPC1	CAGCCAGAUUCCUAGCUUU
siTRPC3	GGAAGAACUGCUCUCCUCAUUGCAA
siTRPC4	TCGAGGGACCAGCATACATG
siTRPC5	CCAAUGGACUGAACCGACGUUUACUU
siTRPC5	GGAGCUACCAUGUUUGGAATT
siTRPC6	TCGAGGGACCAGCATACATG
siHIF-1 α	AGTTCACCTGAGCCTAATA

Table S2: Primers used in PCR.

Gene		Sequence (5'-3')
TRPC1	Forward	AGCCTCTTGACAAACGAGGA
	Reverse	TCTTACAGGTGGGCTTACGG
TRPC3	Forward	GCCTTCATGTTCGGTGCTC
	Reverse	GCGTTCTGGCCCATGTAGT
TRPC4	Forward	GGCGGACTCCAGGATTACATC
	Reverse	CCATGATTCCCGTGGGTTCA
TRPC5	Forward	GGGCTGAGACTGAGCTGTC
	Reverse	TTGCGGATGGCGTAGAGTAAT
TRPC6	Forward	AGCCAGGACTATTGCTGATGG
	Reverse	AACCTTCTCCCTTCTCACGA
HIF-1 α	Forward	GCAGCAGGAATTGGAACATT
	Reverse	GCAGCAGGAATTGGAACATT
NF-kBp65	Forward	GGTCCACGGCGGACCGGT
	Reverse	GACCCCGAGAACGTGGTGC
HNF4 α	Forward	AGCCAACGATCACCAAGCAA
	Reverse	ACTGGTCCCTCGTGTACAT
STAT3	Forward	CCCATATCGTCTGAAACTC
	Reverse	TTGCTCCCTCTGCTCT
COX2	Forward	CCGGGCCAGGGACTATTCAA
	Reverse	TGCAGACTTCGGTCAGTCCA

FOXO1	Forward	TCGTCATAATCTGTCCCTACACA
	Reverse	CGGCTTCGGCTCTTAGCAAA
Smad3	Forward	TGTGCGGCTCTACTACATCG
	Reverse	GCAGCAAATTCTGGTTGTT
Mef2c	Forward	TCCTGGTGTAAACACATAGACCTC
	Reverse	TGGTAAAGTAGGAGTTGCTACGG
GAPDH	Forward	AACTTGCGATTGTGGAAGG
	Reverse	ACACATTGGGGTAGGAACA

Table S3: Potential HIF-1 α binding site on TRPC1 promoter.

Score	Relative score	Start	End	Strand	Predicted site sequence
7.282	0.883145232692922	82	89	1	CTGCGTGA
5.502	0.830043481576361	415	422	1	AGATGTGC
5.290	0.82371900335349	649	656	1	ACATGTGC
5.827	0.839739026021801	977	984	1	GGAGGTGC
5.011	0.815395751352636	1880	1887	1	AGACGCGC

Table S4: Primers for TRPC1 promoter construct.

Name	Sequences
TRPC1-1MluF	5'CG <u>ACGCGT</u> AGGTTGAACAACCAGCACTCTTATCCACT-3'
TRPC1-2MluF	5'CG <u>ACGCGT</u> TGTTATAAA <u>ACTAGTTGTGATGAAATCA</u> -3'
TRPC1-3MluF	5'CG <u>ACGCGT</u> TCCTCGGGAGTAAAGGGAGGAGAACATGAA-3'
TRPC1-4MluF	5'CG <u>ACGCGT</u> TAAGTATGTAGGTATTGTTAGAATGTCCA-3'
TRPC1-5MluF	5'CG <u>ACGCGT</u> ATATA <u>CAAATGTATGGCGCTCCTCTCC</u> -3'
TRPC1-6MluF	5'CG <u>ACGCGT</u> CCAAGATGCTAGGCGACCGACCAGAGAGGC-3'
TRPC1-XhoIR	5'CCG <u>CTCGAG</u> ACACTAA <u>ATTTATAGATGGCATATAAGTT</u> -3'

Figure S1: Effect of siRNA treatment on the mRNA expression of corresponding TRP channels. Scrambled siRNA and targeted siRNAs were added to mouse primary coronary artery endothelial cells. After 48 hours, RNA was extracted, and quantitative real time-polymerase chain reaction was performed to determine the mRNA level of corresponding TRP channels. One-way ANOVA and Dunnett's multiple comparison test, ** $P < 0.01$ versus ctl, n = 3. The data are presented as the mean \pm standard deviation.

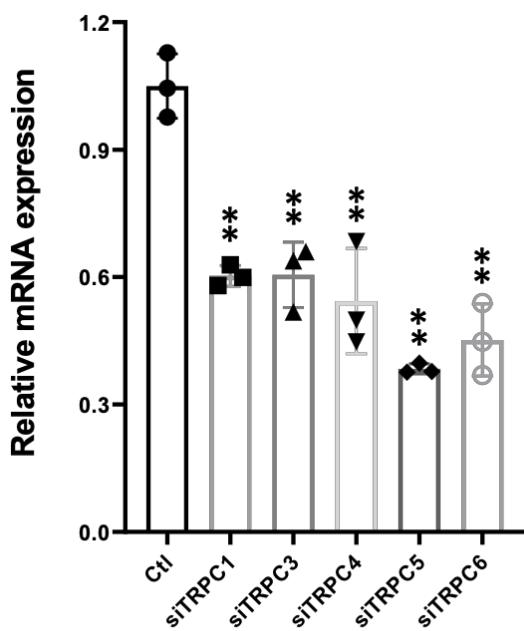


Figure S2: Gene identification of WT and TRPC1_{EC}^{-/-} mice. PCR analysis of the genomic DNA from WT and TRPC1_{EC}^{-/-} mice were performed by using two primer sequences (F1-R1, F2-R2) indicated by confirm insertion of the lox P sites. In the WT allele, 313-bp (F2-R2) fragment were amplified and in the TRPC1_{EC}^{-/-} allele, 390-bp and 415-bp (F2-R2) fragments were amplified.

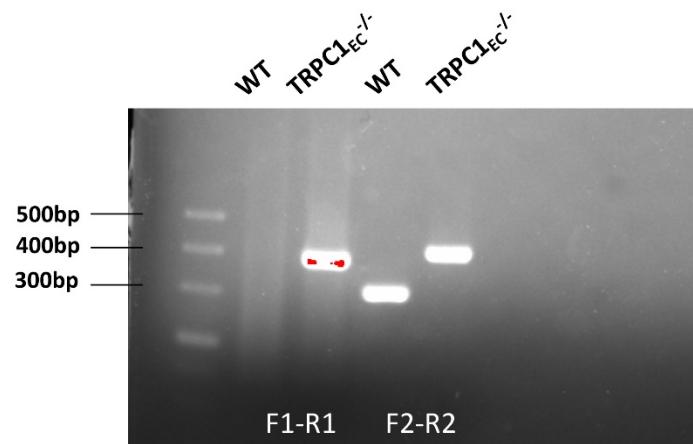


Figure S3: Clarifications of TRPC1_{EC}^{-/-} mice. A, Relative mRNA levels of TRPC1 in heart, mesentery and aorta tissues from TRPC1^{fl/fl} and TRPC1_{EC}^{-/-} mice (n = 3; *P < 0.05 and **P < 0.01 vs TRPC1^{fl/fl}, paired t test). B, Relative mRNA levels of TRPC1 in primary endothelial cells from TRPC1^{fl/fl} and TRPC1_{EC}^{-/-} mice (n = 3; *P < 0.05 and **P < 0.01 vs TRPC1^{fl/fl}, paired t test). C, Representative immunofluorescence images for TRPC1 in coronary arteries, mesenteric arteries and aortas from TRPC1_{EC}^{-/-} mice (yellow, CD31; red, TRPC1; blue, DAPI, scale bar, 10 μm).

