Supplementary Information for
A molecular complex of Ca$_v$1.2/CaMKK2/CaMK1α in caveolae is responsible for vascular remodeling via excitation-transcription coupling

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Materials and Methods

Animals
Wild type (C57BL/6) and cav1-KO mice (8-12 weeks, male) were obtained from Japan SLC and Jackson Labs, respectively (1). All experiments were approved by the Ethics Committee of Nagoya City University (H30-P-1) and conducted in accordance with the Guide for the Care and Use of Laboratory Animals of the Japanese Pharmacological Society.

Mesenteric arterial bed preparation.
The mesenteric arterial bed was removed from the intestine after banding distal arterioles. It was placed in ice cold Ca\(^{2+}/Mg^{2+}\)-free Hanks solution: (mM) 137 NaCl, 5.4 KCl, 0.39 Na\(_2\)HPO\(_4\), 0.44 KH\(_2\)PO\(_4\), 4.2 NaHCO\(_3\), and 5.6 glucose and a smoothened needle (0.6 mm in diameter) was inserted into the first branch of the mesenteric arteries. The perfusion rate was maintained at a 0.15 ml/min to create physiological shear force (~5 dyn/cm\(^2\)) and to avoid endothelial cell activation using a peristaltic pump (MP-3B, TOKYO RIKAKIKAI). The mesenteric arterial bed preparation was perfused with Ca\(^{2+}/Mg^{2+}\)-free Hanks solution to remove blood for 15 min and then switched to standard Hanks solution additionally containing 2.2 mM CaCl\(_2\) and 1.2 mM MgCl\(_2\) with or without drugs. The tissue was then transferred to a dish containing the same solution and was incubated without perfusion for an additional 45 min at 37\(^\circ\)C. The first order of mesenteric arteries was ligated at center and the distal side of the arteries were harvested for a 5 mM K\(^+\) control. Remaining tissue connected to the needle was attached to the peristaltic pump and perfused with 60 mM K\(^+\) Hanks solution containing 82 mM NaCl and 60 mM KCl for 30 to 60 min. In experiments in which the endothelium was removed, arteries were perfused with 0.2% sodium deoxycholate dissolved in ice cold PBS for 120 sec (34). Endothelial removal was confirmed by (i) confocal imaging, (ii) qPCR analysis of the endothelium marker, von Willebrand factor (vWF), and (iii) pressure recording. Harvested tissues were immersed in or perfused with ice cold PBS with 4% paraformaldehyde (PFA) for immunostaining. In the case of RNA or protein extraction, tissues were flash-frozen with liquid N\(_2\) after removal of connective tissues. High (80 mmHg) or low (40 mmHg) pressure was loaded to mesenteric arteries by adjusting the flow speed using a peristaltic pump (2). Perfusion pressure was monitored using a pressure-displacement transducer (Baxter) connected to a needle. All solutions contained 100 nM tetrodotoxin (LATOXAN) to avoid the activation of autonomic nerves by 60 mM K\(^+\) solution. In the present study, second and third-order mesenteric arteries were used unless otherwise stated.

Single cell isolation and unpassaged culture
To isolate single mouse mesenteric artery myocytes (1), second and third-order mesenteric arteries were incubated in Ca\(^{2+}/Mg^{2+}\)-free Hanks solution containing 0.4% collagenase and 0.1% papain (MilliporeSigma) at 37\(^\circ\)C for 40 min. After the incubation, tissues were washed in ice cold Ca\(^{2+}/Mg^{2+}\)-free Hanks solution and treated by gentle agitation with a glass pipette. For unpassaged cultured mouse aorta myocytes (3), mouse aortas were dissected out and placed in ice-cold DMEM containing 1×penicillin/streptomycin/amphotericin B. Connective tissue was removed in sterile PBS and incubated in a DMEM solution containing 0.2% collagenase at 37\(^\circ\)C for 45 minutes. Then the adventitia was removed in PBS and transferred to a DMEM solution containing 0.2% collagenase and 0.2% papain at 37 for 60 min to disperse myocytes. Before starting the incubation, the tissue was cut into 2-5 mm segments. The digested tissue was then centrifuged for 5 minutes at 3,000 rpm and the solution was replaced with fresh DMEM solution containing 10% FBS (Gibco). The pellet was gently resuspended, which resulted in dispersion of individual arterial myocytes. Cells were then seeded on glass bottom dishes (Matsunami) coated with 20 µg/mL laminin (MilliporeSigma) for 2 hours and kept in an incubator at 37\(^\circ\)C with 5% CO\(_2\) for 5-7 days to expand semi-confluent. Then, medium was switched to DMEM with 0.5% FBS, 1×ITS (insulin, transferrin and selenium) supplement and 200 µM L(+)-ascorbic acid sodium for additive 2-3 days. siRNA or viral vectors were applied during this period. For experiments, cells were incubated with standard (5 mM K\(^+\)) HEPES-buffered solution: (mM) 137 NaCl, 5.9 KCl, 2.2 CaCl\(_2\), 1.2 MgCl\(_2\), 14 glucose, and 10 HEPES or 60 mM K\(^+\) HEPES-buffered solution: (mM) 83 NaCl, 60 KCl, 2.2 CaCl\(_2\), 1.2 MgCl\(_2\), 14 glucose, and 10 HEPES. When the smooth muscle myocytes were stimulated with BayK8644, CaCl\(_2\) concentration was 20 mM and NaCl concentration was reduced to 105 mM (4). The pH was adjusted to 7.4 with NaOH.

Mesenteric artery ligation model
WT and cav1-KO mice (male or female, 12 weeks) were anesthetized by chloral hydrate. From three adjacent second-order mesenteric arteries, the first and third arteries were ligated at the distal end of the vessels using 8-0
nylon suture (Natsume Seisakusho) (5). This creates high pressure in the middle vessel. Control vessels were second-order mesenteric arteries obtained along the intestine, remote from the ligated vessels. The mesenteric artery was isolated 2d, 7d or 14 d after surgery. To evaluate the effects of STO609 in vivo, mice were treated with STO609 (1 mg/kg) intraperitoneally on the day of surgery (at least 2 hours before surgery) and then three time per week. STO609 was dissolved in DMSO at 10 mg/mL and stored at -20°C until usage. This stock solution was diluted to 0.2 mg/mL with sterile PBS. A PBS solution containing 2% DMSO was used as a vehicle control.

**Immunostaining**

Mesenteric arteries, freshly isolated and primary vascular myocytes were fixed with 4% PFA in PBS. These were treated with 0.2% Triton X-100, 3% bovine serum albumin (BSA, MilliporeSigma), and anti-Phospho-CREB (Ser133) rabbit monoclonal antibody (1:300, 87G3, CST), anti-CaMKI (phospho T177) rabbit antibody (1:300, ab62215, Abcam), anti-CaMKK2 rabbit antibody (1:200, GTX115950, GeneTex), anti-CaMKK2 mouse antibody (1:200, 6/CaM Kinase, 610544, BD Biosciences), anti-cav1 rabbit antibody (1:200, GTX109650, GeneTex), anti-Ca1.2 mouse monoclonal antibody (1:200, N263/31, NeuroMab) or anti-smooth muscle actin (SMA) mouse monoclonal antibody (1:1000, 1A4, CST) at 4°C for 12 h. After washing repeatedly, the preparations were treated with Alexa 488-, Alexa 594- or Alexa 647-conjugated secondary antibodies (1:500, Molecular Probes) at room temperature for 1 h. In some experiments, cav1 antibody conjugated with HiLyte Fluor 555 (Dojindo) was used for dual staining with another rabbit antibodies. For detection of macrophages, PE conjugated anti F4/80 antibody (1:100, REA126, Miltenyi Biotec) was utilized. Nuclei were stained with TO-PRO3 (1:500, Thermo Fisher Scientific) or Hoechst (1:500, Thermo Fisher Scientific) at room temperature for 30 min. The mesenteric artery was mounted on coverslips (Matsunami) using Dako fluorescence mounting medium (Agilent Technologies). Fluorescently labeled samples were observed using a confocal or TIRF imaging system.

**ELISA**

The P-CREB levels in arteries were quantified using ELISA. Two types of ELISA detecting (i) phosphorylated CREB at S133 (KHO0241, Thermo Fisher Scientific) and (ii) total CREB (KHO0231, Thermo Fisher Scientific) were performed using arterial lysates according to protocols provided by the company. Second and third-order mesenteric arteries were flash-frozen in liquid N₂. The frozen samples were pulverized and proteins were extracted using a cell lysis buffer (100 µL) containing 10 mM Tris, pH 7.4, 100 mM NaCl, 1 mM EDTA, 1 mM EGTA, 1 mM NaF, 20 mM Na₃P₂O₇, 2 mM Na₃VO₄, 1% Triton X-100, 10% glycerol, 0.1% SDS, 0.5% deoxycholate (Thermo Fisher Scientific). Protease inhibitor cocktail (MilliporeSigma) and 1 mM PMSF (TCI) were added just prior to experiments. The lysates were sonicated briefly and centrifuged to remove debris. The supernatant was diluted to 1:20 and subjected to ELISA. Both phosphorylated and total CREB amounts of each sample were calculated from standard curves, and the relative P-CREB level to total CREB of each sample was obtained.

**Histological analysis**

Isolated arteries were fixed in 4% PFA, embedded in paraffin, and cut into 5-µm thick sections for routine hematoxylin-eosin (HE) staining (Biopathology Institute). Medial thickness was measured at 4 different points and averaged per sample. Image analysis was performed using NIS Elements (Nikon).

**Transfection using virus vectors**

Cav1 (NM_001753) and CaMKK2 (XM_006530233) was labeled with mCherry or GGECO1.1 (Addgene plasmids #32445) at the N termini. pCMV-PV-NLS-GFP was obtained from Addgene (#17300). These constructs were cloned into pENTR1A and obtained pAd/CMV/V5-DEST or BacMam pCMV-DEST vectors using Gateway system (Thermo Fisher Scientific). Adenovirus and baculovirus vector were harvested from 293A and Sf9 cells after introducing linearized pAd/CMV/V5-DEST and bacmid, respectively. Adenovirus was used for knock-in or overexpression experiments (Fig. 2C, 4D and 5J-L). On the other hand, baculovirus was utilized for TIRF imaging (Fig. 3) because of relatively low expression efficiency compared with adenovirus and high signal to noise ratio. Primary vascular myocytes at semiconfluent condition were infected for 12 hours and then washed. Experiments were performed 48-72 hours after infection.

**siRNA knockdown.**

Genes were knocked down using specific siRNA (NIPPON GENE). Cells were transfected with siRNA using Lipofectamine RNAiMAX Reagent (Thermo Fisher Scientific). Two siRNA (#1 and #2, 100 nM each) targeting different regions of the same gene were simultaneously transfected. Negative control siRNA was also purchased.
from NIPPON GENE. All experiments were performed between 48 h and 72 h after siRNA treatment. The sequence of gene specific siRNA was as follows. siCaMKK2 #1: GGAUUGUGGUGCCUGAAAU, siCaMKK2 #2: GCGACGCCUUGCUGCUUA, siCaMK1a #1: GAACGAGAUUGCCGUCUUA and siCaMK1a #2: CCCACCCUUUUAUGAUGA.

Real-time qPCR.
Total RNA extraction, reverse transcription and real-time quantitative PCR were carried out as previously reported (6). The sequence of primers was listed in Table S2. For normalization of gene expression among each samples, *Ppia* and *Hprt* were utilized as endogenous housekeeping genes because these expression was similar between 5 mM and 60 mM K⁺ conditions. Averaged Cq value of these genes was used for calculation.

RNA sequence analysis
RNA was extracted from samples using TRlzol and PureLink RNA spin columns (Thermo Fisher Scientific). Genomic DNA was digested using TURBO™ DNase (Thermo Fisher Scientific). Conventional RNA-Seq was performed by Novogene. The RNA-seq reads were aligned with the mouse reference genome mm9 using TopHat2 software. Prior to differential gene expression analysis, the read counts were adjusted for each sequenced library by edgeR program package through one scaling normalized factor. Differential expression analysis of two conditions was performed using the DEGSeq R package. The P values were adjusted using the Benjamini & Hochberg methods. Corrected qvalue of 0.005 and |log₂ (Fold Change)| of 1 were set as the threshold for significantly differential expression.

TIRF imaging
Two-dimensional single-molecular imaging were done using a TIRF imaging system (Nikon), which consists of a fluorescent microscope (Ti2; Nikon), objective lens (CFI Plan Apo TIRF 60× or 100×; Nikon), EM-CCD camera (C9100-12; Hamamatsu Photonics), and NIS Elements software (Nikon) (1, 6). For co-localization analysis, two molecules were labeled with green or orange indicators, and corresponding fluorescence puncta having higher intensity than background levels were converted to binary images. Then, co-localized puncta were extracted by image arithmetic operations using the NIS Elements software. The ratio of the number of co-localized puncta to the total number of puncta was calculated. For GGECO imaging, fluorescence signals from GGECO1.1 are described as F/F₀, where F is the sum fluorescence intensity within the regions of interest (ROI, 2 µm square) in the TIRF area during measurements, and F₀ is the baseline F value obtained as the average intensity of the ROI for 5 sec before stimulation. GGECO1.1 puncta whose maximal increase in fluorescence intensity due to depolarization stimulus was higher than 5×SD of baseline values were accepted as positive GGECO1.1 puncta. Images were collected at an interval of 800 msec for two color imaging, i.e. GG-CaMKK2 and mCh-cav1 or 100 msec for single color imaging, i.e. GG-CaMKK2 only. Immunolabeling images were collected by exposure for 100 msec. The resolution of images was 270 or 160 nm per pixel (x−y) and less than 200 nm (z). All experiments were performed at room temperature.

Confocal imaging
Confocal images were obtained using a laser scanning confocal fluorescent microscope (A1R, Nikon) equipped with a fluorescent unit (ECLIPSE Ti), objective lens (Plan Apo 60× or 20×), and NIS Elements software (Nikon). For P-CREB analysis, fluorescence signals from secondary antibodies binding to P-CREB primary antibodies and nuclear indicators (Hoechst or TO-PRO3) were converted into binary images. Next, fluorescence signals of nuclei co-localized with P-CREB signals were extracted by image arithmetic operations using the NIS Elements software. The ratio of the number of nuclei co-localized with P-CREB signals to the total number of nuclei was calculated. For each artery, three image sections (215 µm×215 µm) were acquired and the calculated ratio values were averaged.

Ca²⁺ imaging
Intracellular Ca²⁺ imaging was performed using a confocal microscope. For monitoring [Ca²⁺], mesenteric artery myocytes were incubated with 10 µM Fluo-4/AM for 20 min at room temperature. The excitation and emission wavelength were 488 nm and 500-530 nm, respectively. When the amplitude of [Ca²⁺] elevation in primary vascular myocytes of WT and cav1-KO cells were recorded, cells were loaded with 10 µM Fluo-4/AM and 10 µM Fura Red/AM for 30 min to perform ratio metric measurement. These indicators were excited at 488 nm and emission was collected at 500-530 nm (Fluo-4) and 662-737 nm (Fura-Red). Fratio was calculated by dividing fluorescence intensity of Fluo-4 by that of Fura-Red. When Ca²⁺ responses of myocytes transfected with PV-NLS-GFP were recorded, cells were loaded with 6.7 µM CaSiR/AM (Goryo Chemical) and Hoechst. These indicators were excited at 640 and 405 nm and emission was collected at 662-737 nm (CaSiR) and 425-475 nm (Hoechst). ROIs
corresponding to whole cell regions and nuclei were created according to the fluorescence of CaSiR and Hoechst, respectively. The sum of CaSiR intensity and area corresponding to cytosol were calculated by subtracting these values of nuclei from those of whole cell regions.

**In situ PLA**
To clarify whether cav1/Ca,1.2/CaM KK2/CaMK1a co-localizes within 40 nm in arterial myocytes, a PLA was performed using a PLA kit (Duolink, MilliporeSigma) (6). Myocytes were fixed with 4% paraformaldehyde in PBS and treated with 0.2% Triton X-100. Cells were then labeled with primary antibodies mentioned above at 4°C for 12 h. After washing repeatedly, cells were incubated in a humidified chamber at 37°C for 1 h with secondary anti-mouse PLUS and anti-rabbit MINUS PLA probes and then washed in Duolink Wash Buffer A. The preparations were incubated in ligase solution at 37°C for 30 min in a humidifier chamber and then washed repeatedly in Wash Buffer A. Samples were incubated in Amplification Polymerase solution at 37°C for 100 min in a humidifier chamber and then washed repeatedly in Duolink Wash Buffer B. Fluorescence images were observed using a confocal imaging system. When two PLAs probes were within 40 nm, positive signals (green puncta) were generated. The excitation of fluorescent puncta was illuminated at 488 nm. Negative control experiments were performed using myocytes treated with either of antibodies. PLA puncta were extracted by binary image processing and the relative area of these puncta to the whole cell area was calculated.

**Data notation and statistical analysis**
Pooled data are shown as the mean ± SEM. The significance of differences between two groups was evaluated using the two-tailed t test after the application of the F test. Data from more than two groups were compared using one-way or two-way analysis of variance (ANOVA) followed by Tukey or Dunnett tests. In all cases, p values <0.05 were considered to be significant. All data were obtained from at least three independent experiments.

**Drugs**
Drugs were purchased from FUJIFILM Wako Pure Chemical except for BayK8644 and nicardipine (MilliporeSigma), EGTA-AM (Setareh), BAPTA-AM and HEPES (Dojindo), KN93 and MβCD (TCI), and STO609 (Cayman). All hydrophobic compounds were dissolved in dimethyl sulfoxide (DMSO) at a concentration of 10-30 mM as a stock solution.

**Data Availability.**
The datasets generated in this study are available at https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE183588
Supplementary Figures

**Fig. S1. Expression profiles of CaMK family genes in mesenteric artery and primary vascular myocytes.**

(A and B) Splice variants of CaMK2γ (Camk2g, A) and CaMK2δ (Camk2d, B) in primary vascular myocytes, mesenteric artery (MA), and brain were identified using reverse-transcription PCR analysis. Negative control data was obtained by adding water instead of cDNA (non-template control, NTC). In both Panel (A) and (B), predicted band size for each splice variant is indicated within the box below. Representative photos from three independent experiments are shown.
Fig. S2. Effects of CaMK inhibitors on 60 mM K+ superfusate-induced [Ca2+]i elevation by Ca2+ influx through voltage-dependent Ca2+ channels in mesenteric artery myocytes.

(A) Traces of fluorescence signals from Fluo-4/AM in mesenteric artery myocytes. Myocytes were stimulated by 60 mM K+ solution twice, and drugs were applied at the second 60 mM K+ stimulation. (B) Relative Fluo-4 intensity normalized to untreated control (the first 60 mM K+ stimulation). Data were obtained from 19-43 cells from three independent experiments. Statistical analysis was performed using one-way ANOVA followed by Tukey test (**p<0.01).
Fig. S3. Illustration and validation of essential features of unpassaged primary vascular myocytes.
(A) Unpassaged primary vascular myocytes were stained with anti-smooth muscle actin antibody. Nuclei were stained with Hoechst. Representative pictures from three independent experiments are shown. (B) Traces of mean and standard error of fluorescence ratio (F_{ratio}, Fluo-4/Fura-Red) in primary vascular myocytes from WT and cav1-KO mice are shown (297 and 423 cells from WT and cav1-KO mice). (C) Peak amplitude of F_{ratio} (left) and resting F_{ratio} (right) are compared between vascular myocytes from WT and cav1-KO mice. Data were obtained from 297 (WT) and 423 (cav1-KO) cells from four sections (635 µm×635 µm). (D) Knockdown efficiency by siRNA was confirmed by qPCR. Data were obtained from five independent primary (unpassaged) cultures per group. (E) mRNA levels of CaMK family genes in mesenteric artery (left) and primary vascular myocytes (right) are compared between WT and cav1-KO mice. Data were obtained from five mice (mesenteric artery) and four independent unpassaged cultures (primary vascular myocytes). (F) The cav1 expression (green) in cav1-KO myocytes expressing mCherry (cav1-KO, upper) or mCh-cav1 (cav1-KI, middle) or in WT myocytes (WT, bottom). Nuclei and mCherry are shown in blue and red, respectively. Representative pictures from three independent experiments are shown. Statistical analysis was performed using two-tailed t test (*p<0.05, **p<0.01).
Fig. S4. Cav1.2 is coupled to cav1 in mesenteric artery myocytes.
PLA was performed to obtain evidence for direct coupling of Cav1.2 with cav1 in mesenteric artery myocytes (15 cells). Cav1 was labeled with two different antibodies from mouse and rabbit and then subjected to PLA to obtain positive control data (8 cells).
Fig. S5. Ca\textsubscript{v}1.2 and CaMKK2 are coupled with cav3 in primary vascular myocytes from WT and cav1-KO mice.
PLA was performed to seek evidence for direct coupling of cav3 with Ca\textsubscript{v}1.2 and CaMKK2 in primary myocytes. WT cells labeled with only the cav3 antibody were used for a negative control. Each data set was obtained from 9-18 cells. Statistical analysis was performed using one-way ANOVA followed by Tukey test (**p<0.01 vs. WT cav3).
Fig. S6. CaMK1a is coupled with cav1, CaMKK2 and Ca\textsubscript{v}1.2 in mesenteric artery myocytes.
PLA was performed to seek evidence for direct coupling of CaMK1a with cav1, CaMKK2 and Ca\textsubscript{v}1.2 in mesenteric artery myocytes from WT mice. To obtain negative control data, myocytes were treated only with the anti-CaMK1a antibody and then subjected to PLA. Each data set was obtained from 14-19 cells. Statistical analysis was performed using one-way ANOVA followed by Dunnett test (*p<0.05 vs. CaMK1a).
Fig. S7. Endothelium-denuded mesenteric arteries can exhibit depolarization-induced gene transcription very similar to the pattern obtained in intact arteries. 
Endothelium of mesenteric artery beds were removed by perfusing 0.2% sodium deoxycholate (SD) dissolved in ice cold PBS for 120 sec. (A) Confocal images of endothelial layers were obtained from control (treated with PBS) and SD groups (9 arteries from 3 mice per group). Vessels were stained with Hoechst (left). The density of endothelial cells (ECs) is compared (right). Note that the number of ECs was reduced, and ECs exhibited morphologic abnormality i.e. round, not elliptic, shape (indicated by arrowheads), and these ECs often aggregated in the SD group. For each artery, three image sections (215 µm×215 µm) were acquired and the calculated EC density values were averaged. (B) The gene expression of an endothelium marker, Von Willebrand Factor (VWF) was compared by qPCR. Data were obtained from 5 mice per group. (C) Acetylcholine (ACh)-induced vasodilation was recorded in before (Control) and after SD treatment. Vasoconstriction was induced by 30 mM K+ Hanks solution and then arterial beds were dilated by 1 µM ACh. ACh-induced vasodilation (%) was estimated from ACh-induced reversal of KCl-mediated vasoconstriction. Data were obtained from 5 mice. (D) Gene transcription after depolarizing stimulus for 1 hour was confirmed by qPCR. Data were obtained 5 mice per group. Statistical analysis was performed using two-tailed t test (**p<0.01).
Fig. S8. Sustained increase in transmural pressure in mesenteric artery causes CREB phosphorylation in vascular myocytes by activating the Ca$_{1.2}$/CaMKK2/CaMK1α axis.

The possibility that increased perfusion pressure can cause CREB phosphorylation was addressed using ex vivo model. (A) Mesenteric artery preparations were perfused with 5 mM K$^+$ solution at 0, 40 and 80 mmHg for 30 min, and then fixed and stained with P-CREB antibody. The effect of the L-type Ca$^{2+}$ channel blocker nicardipine (Nic) on 80 mmHg-induced CREB phosphorylation also examined. The P-CREB positive myocyte ratio in vivo was estimated using acute fixation by perfusing ice cold 4% PFA from heart under deep anesthesia. Data were obtained from 9-16 arteries. (B) The effect of CaMK inhibitors KN93 and STO609 on 80 mmHg-induced CREB phosphorylation was examined. Data were obtained from 16-20 arteries. (C) High pressure (80 mmHg) was applied to mesenteric artery of WT and cav1-KO mice. Mesenteric artery treated with methyl-β-cyclodextrin (MβCD) was also utilized. Data were obtained from 15 arteries per group. Statistical analysis was performed using one-way (A) or two-way ANOVA (B and C) followed by Tukey test (**p<0.01).
Fig. S9. Changes in cross-section area (CSA) of mesenteric arteries preloaded with high pressure.

(A) Changes in CSA of second-order mesenteric arteries from WT (left) and cav1-KO (right) mice 2, 7 and 14 days after ligation. Control vessels were second-order mesenteric arteries located along the intestine, remote from the ligated vessels. Data were collected from 4-7 arteries.

(B) Changes in CSA of second-order mesenteric arteries from mice treated with DMSO (left) or STO609 (right) 2, 7 and 14 days after ligation was plotted. Data were collected from 5-6 arteries. Statistical analysis was performed using two-way ANOVA followed by Tukey test (*p<0.05, **p<0.01; #p<0.05, ##p<0.01 vs. HP group at the same day).
Table S1. The list of differentially expressed genes.

| Gene symbol | Gene name | Gene ID          | log2 Fold change. | qvalue       |
|-------------|-----------|------------------|-------------------|--------------|
| Ackr3       | atypical chemokine receptor 3 [Source:MGI Symbol;Acc:MGI:109562] | ENSMUSG00000044337 | 1.0597          | 1.36E-05     |
| Adams4      | a disintegrin-like and metallopeptidase (reprolysin type) with thrombospondin type 1 motif, 4 [Source:MGI Symbol;Acc:MGI:1339949] | ENSMUSG00000006403 | 2.708           | 4.25E-84     |
| Adams9      | a disintegrin-like and metallopeptidase (reprolysin type) with thrombospondin type 1 motif, 9 [Source:MGI Symbol;Acc:MGI:1916320] | ENSMUSG00000030022 | 2.0148          | 4.25E-12     |
| Adgrg1      | adhesion G protein-coupled receptor G1 [Source:MGI Symbol;Acc:MGI:1340051] | ENSMUSG00000031785 | 1.3945          | 0.0021643    |
| Agap3       | ArfGAP with GTPase domain, ankyrin repeat and PH domain 3 [Source:MGI Symbol;Acc:MGI:2183446] | ENSMUSG00000023353 | 1.1596          | 2.72E-05     |
| Akap12      | a disintegrin (PRKA) anchor protein (gravin) 12 [Source:MGI Symbol;Acc:MGI:1932576] | ENSMUSG00000038587 | 1.2534          | 9.16E-27     |
| Alpl        | alkaline phosphatase, liver/bone/kidney [Source:MGI Symbol;Acc:MGI:88067] | ENSMUSG00000028766 | 1.1803          | 0.00079298   |
| Arc         | activity regulated cytoskeletal-associated protein [Source:MGI Symbol;Acc:MGI:88067] | ENSMUSG00000022602 | 1.68            | 2.20E-06     |
| Arih1       | ariadne RBR E3 ubiquitin protein ligase 1 [Source:MGI Symbol;Acc:MGI:1344363] | ENSMUSG00000025234 | 1.0438          | 1.85E-05     |
| Atf4        | activating transcription factor 4 [Source:MGI Symbol;Acc:MGI:1859216] | ENSMUSG00000042406 | 1.2373          | 3.56E-20     |
| Atxn7       | ataxin 7 [Source:MGI Symbol;Acc:MGI:2179277] | ENSMUSG00000021738 | 1.23            | 0.0037518    |
| Avpr1a      | arginine vasopressin receptor 1A [Source:MGI Symbol;Acc:MGI:1859216] | ENSMUSG00000020123 | 1.1303          | 4.55E-07     |
| B4galt5     | UDP-Gal:betaGlcNAc beta 1,4-galactosyltransferase, polypeptide 5 [Source:MGI Symbol;Acc:MGI:1927169] | ENSMUSG00000017929 | 1.9999          | 1.04E-08     |
| Bag3        | BCL2-associated athanogene 3 [Source:MGI Symbol;Acc:MGI:1352493] | ENSMUSG00000030847 | 1.1922          | 3.72E-13     |
| Bcl3        | B cell leukemia/lymphoma 3 [Source:MGI Symbol;Acc:MGI:88140] | ENSMUSG00000053175 | 2.6521          | 2.73E-10     |
| Bcr         | breakpoint cluster region [Source:MGI Symbol;Acc:MGI:1344363] | ENSMUSG00000096811 | 1.3652          | 3.96E-40     |
| Bpnt1       | bisphosphate 3'-nucleotidase 1 [Source:MGI Symbol;Acc:MGI:1338800] | ENSMUSG00000026617 | 1.1158          | 0.00065801   |
| Btg1        | B cell translocation gene 1, anti-proliferative [Source:MGI Symbol;Acc:MGI:88215] | ENSMUSG00000036478 | 1.0345          | 3.08E-09     |
| Cbarp       | calcium channel, voltage-dependent, beta subunit associated regulatory protein [Source:MGI Symbol;Acc:MGI:1354170] | ENSMUSG00000035640 | 1.2007          | 7.13E-05     |
| Ccl2        | chemokine (C-C motif) ligand 2 [Source:MGI Symbol;Acc:MGI:98259] | ENSMUSG00000035385 | 2.9814          | 0.00021177   |
| Ccl7        | chemokine (C-C motif) ligand 7 [Source:MGI Symbol;Acc:MGI:99512] | ENSMUSG00000035373 | 2.6699          | 7.30E-06     |
| Cdo42ep4    | CDC42 effector protein (Rho GTPase binding) 4 [Source:MGI Symbol;Acc:MGI:104556] | ENSMUSG000000031825 | 1.4015          | 3.05E-223    |
| Cdkn1a      | cyclin-dependent kinase inhibitor 1A (P21) [Source:MGI Symbol;Acc:MGI:104556] | ENSMUSG00000023067 | 1.3586          | 2.72E-31     |
| Crispd2     | cysteine-rich secretory protein LCCL domain containing 2 (Source:MGI Symbol;Acc:MGI:1926142) | ENSMUSG000000314599 | 1.0836          | 2.43E-05     |
| Csf1        | colony stimulating factor 1 (macrophage) [Source:MGI Symbol;Acc:MGI:1339753] | ENSMUSG00000018920 | 1.873           | 0.0032762    |
| Cxcl1       | chemokine (C-X-C motif) ligand 1 [Source:MGI Symbol;Acc:MGI:108068] | ENSMUSG000000058427 | 2.9805          | 4.20E-15     |
| Gene   | Description                                                                 | Symbol/Access Number                                      | Log2 Fold Change | P-value         |
|--------|------------------------------------------------------------------------------|----------------------------------------------------------|-----------------|-----------------|
| Dbp    | D site albumin promoter binding protein                                      | ENSMUSG00000059824                                       | -1.0969         | 9.50E-10        |
| Dnaj1  | DnaJ heat shock protein family (Hsp40) member B1                             | ENSMUSG0000005483                                       | 2.0748          | 5.06E-22        |
| Dusp14 | dual specificity phosphatase 14                                              | ENSMUSG00000018648                                      | 1.5717          | 0.0016669       |
| Dusp5  | dual specificity phosphatase 5                                               | ENSMUSG00000034765                                      | 2.5077          | 0.00015392      |
| Edn1   | endothelin 1                                                                  | ENSMUSG00000021367                                      | 1.1433          | 0.0024699       |
| Efhd2  | elongation factor RNA polymerase II                                          | ENSMUSG00000040659                                      | 1.4353          | 1.15E-37        |
| Ell2   | elongation factor RNA polymerase II 2                                        | ENSMUSG0000001542                                       | 1.1949          | 0.0014532       |
| Emp1   | epithelial membrane protein 1                                                | ENSMUSG00000030208                                      | 1.2917          | 1.85E-31        |
| Eprs   | glutamyl-prolyl-tRNA synthetase                                               | ENSMUSG00000026615                                      | 1.9194          | 2.17E-64        |
| Ets1   | E26 avian leukemia oncogene 1, 5' domain                                    | ENSMUSG00000032035                                      | 1.7823          | 4.21E-07        |
| Foxs1  | forkhead box S1                                                              | ENSMUSG00000074676                                      | 1.6844          | 0.0032904       |
| Gbp5   | guanylate binding protein 5                                                  | ENSMUSG000000105504                                      | 1.6845          | 0.0002694       |
| Gem    | GTP binding protein (gene overexpressed in skeletal muscle)                 | ENSMUSG00000028214                                      | 1.7555          | 1.59E-34        |
| Gm13889| predicted gene 13889                                                        | ENSMUSG000000087006                                      | 1.9546          | 6.22E-115       |
| Gm48878| predicted gene, 48878                                                       | ENSMUSG000000112794                                      | 2.3573          | 0.0019815       |
| Gm9973 | predicted gene 9973                                                         | ENSMUSG000000113175                                      | 1.6745          | 1.65E-07        |
| Gpr4   | G protein-coupled receptor 4                                                 | ENSMUSG000000044317                                      | 2.2243          | 0.00059977      |
| H3f3b  | H3 histone, family 3B                                                        | ENSMUSG00000016559                                      | 1.3075          | 3.00E-56        |
| Has1   | hyaluronan synthase 1                                                        | ENSMUSG00000003665                                       | 1.5313          | 5.29E-05        |
| Hbegf  | heparin-binding EGF-like growth factor                                        | ENSMUSG00000024486                                      | 1.0841          | 1.57E-10        |
| Hist1h1c| histone cluster 1, H1c                                                       | ENSMUSG00000036181                                      | 1.0529          | 7.56E-07        |
| Hk2    | hexokinase 2                                                                 | ENSMUSG0000000628                                       | 3.2305          | 1.25E-25        |
| Hmgb2  | high mobility group box 2                                                    | ENSMUSG00000054717                                      | 1.1939          | 0.00097572      |
| Hr     | lysine demethylase and nuclear receptor corepressor                         | ENSMUSG00000022096                                      | 1.0624          | 4.84E-08        |
| Hspa1a | heat shock protein 1A                                                        | ENSMUSG00000091971                                      | 2.9825          | 6.48E-126       |
| Hspa1b | heat shock protein 1B                                                        | ENSMUSG00000090877                                      | 3.573           | 2.26E-234       |
| Hspa5  | heat shock protein 5                                                         | ENSMUSG00000026864                                      | 1.8551          | 6.22E-115       |
| Hspb1  | heat shock protein 1                                                         | ENSMUSG0000004951                                       | 1.3568          | 7.87E-104       |
| Hsps1  | heat shock protein 1 (chaperonin 10)                                         | ENSMUSG00000073676                                      | 1.4587          | 4.27E-05        |
| Hsph1  | heat shock 105kDa/110kDa protein 1                                           | ENSMUSG00000029657                                      | 1.2356          | 2.33E-07        |
| Icam1  | intercellular adhesion molecule 1                                            | ENSMUSG00000037405                                      | 2.9417          | 1.76E-91        |
| Icosl  | icos ligand                                                                  | ENSMUSG00000000732                                       | 1.4928          | 0.00025112      |
| Id1    | inhibitor of DNA binding 1, HLH protein                                     | ENSMUSG00000042745                                      | 1.0985          | 5.94E-13        |
| Id3  | inhibitor of DNA binding 3 [Source:MGI Symbol;Acc:MGI:96398] | ENSMUSG00000007872 | 1.4255 | 1.69E-46 |
|------|---------------------------------------------------------------|---------------------|--------|----------|
| lEr3 | immediate early response 3 [Source:MGI Symbol;Acc:MGI:104814] | ENSMUSG00000003541 | 1.2464 | 2.66E-18 |
| lEr5 | immediate early response 5 [Source:MGI Symbol;Acc:MGI:1337072] | ENSMUSG000000056708 | 1.3744 | 2.39E-13 |
| lEr5l| immediate early response 5-like [Source:MGI Symbol;Acc:MGI:1919750] | ENSMUSG000000089762 | 1.4474 | 0.0016154 |
| lI4ra| interleukin 4 receptor, alpha [Source:MGI Symbol;Acc:MGI:105367] | ENSMUSG000000030748 | 1.8812 | 2.36E-08 |
| lI6  | interleukin 6 [Source:MGI Symbol;Acc:MGI:96559] | ENSMUSG000000025746 | 2.422  | 1.96E-32 |
| IrF1 | interferon regulatory factor 1 [Source:MGI Symbol;Acc:MGI:96590] | ENSMUSG00000018899 | 1.0562 | 0.0001643 |
| lTpk1| inositol 1,3,4-triphosphate 5/6 kinase [Source:MGI Symbol;Acc:MGI:1929512] | ENSMUSG000000057963 | 1.2224 | 3.53E-12 |
| JmY  | junction-mediating and regulatory protein [Source:MGI Symbol;Acc:MGI:1913096] | ENSMUSG000000021690 | 1.0568 | 0.00058379 |
| Jund | jun D proto-oncogene | ENSMUSG000000071076 | 1.0731 | 4.42E-06 |
| lI4ra| interleukin 4 receptor, alpha | ENSMUSG000000034394 | 2.064  | 0.0007255 |
| LitaF| LPS-induced TN factor [Source:MGI Symbol;Acc:MGI:1929512] | ENSMUSG000000022500 | 1.6033 | 1.45E-13 |
| lPep1| lipin 1 [Source:MGI Symbol;Acc:MGI:1891340] | ENSMUSG000000020593 | 1.1171 | 1.45E-06 |
| Map2K3| mitogen-activated protein kinase kinase 3 [Source:MGI Symbol;Acc:MGI:1346868] | ENSMUSG00000018932 | 1.1502 | 1.96E-05 |
| MapkapK2| MAP kinase-activated protein kinase 2 [Source:MGI Symbol;Acc:MGI:109298] | ENSMUSG00000016528 | 1.0647 | 1.78E-07 |
| Mcl1 | myeloid cell leukemia sequence 1 [Source:MGI Symbol;Acc:MGI:101769] | ENSMUSG000000038612 | 1.1695 | 3.40E-39 |
| MidN| midnolin [Source:MGI Symbol;Acc:MGI:1890222] | ENSMUSG000000035621 | 1.0411 | 3.51E-11 |
| MT1 | metallothionein 1 [Source:MGI Symbol;Acc:MGI:977171] | ENSMUSG000000031765 | 1.234  | 1.98E-50 |
| mt-RNr1| mitochondrially encoded 12S rRNA [Source:MGI Symbol;Acc:MGI:102493] | ENSMUSG000000064337 | 1.0849 | 5.83E-08 |
| mt-RNr2| mitochondrially encoded 16S rRNA [Source:MGI Symbol;Acc:MGI:102492] | ENSMUSG000000064339 | 1.3532 | 1.41E-33 |
| Myc  | myelocytomatosis oncogene [Source:MGI Symbol;Acc:MGI:97250] | ENSMUSG00000022346 | 1.681  | 4.42E-06 |
| NcOr2| nuclear receptor co-repressor 2 [Source:MGI Symbol;Acc:MGI:1337080] | ENSMUSG00000029478 | 1.1405 | 2.75E-09 |
| Nfil3| nuclear factor, interleukin 3, regulated [Source:MGI Symbol;Acc:MGI:109495] | ENSMUSG000000056749 | 1.5059 | 7.62E-08 |
| NFkB | nuclear factor of kappa light polypeptide gene enhancer in B cells inhibitor, alpha [Source:MGI Symbol;Acc:MGI:104741] | ENSMUSG00000021025 | 1.6169 | 1.86E-36 |
| Ngf  | nerve growth factor [Source:MGI Symbol;Acc:MGI:97321] | ENSMUSG00000027859 | 1.3443 | 0.0012134 |
| Nos3 | nitric oxide synthase 3, endothelial cell [Source:MGI Symbol;Acc:MGI:97362] | ENSMUSG00000028978 | 1.1007 | 3.86E-05 |
| Nr1D1| nuclear receptor subfamily 1, group D, member 1 [Source:MGI Symbol;Acc:MGI:2444210] | ENSMUSG00000020889 | -1.8606 | 6.56E-24 |
| Nup98| nucleoporin 98 [Source:MGI Symbol;Acc:MGI:109404] | ENSMUSG000000063550 | 1.1863 | 0.001609 |
| Odc1 | ornithine decarboxylase, structural 1 [Source:MGI Symbol;Acc:MGI:97402] | ENSMUSG00000011179 | 1.1525 | 2.58E-09 |
| PDE4b| phosphodiesterase 4B, cAMP specific [Source:MGI Symbol;Acc:MGI:99557] | ENSMUSG00000028525 | 1.7624 | 9.28E-47 |
| Peg3 | paternally expressed 3 [Source:MGI Symbol;Acc:MGI:104748] | ENSMUSG00000002265 | -1.0855 | 0.0034048 |
| Pim1 | proviral integration site 1 [Source:MGI Symbol;Acc:MGI:97584] | ENSMUSG00000024014 | 1.9542 | 2.81E-23 |
| Plat | plasminogen activator, tissue [Source:MGI Symbol;Acc:MGI:97610] | ENSMUSG00000031538 | 1.3295 | 8.55E-15 |
| Gene   | Description                                                                 | Symbol          | EntrezGene ID | Ratio   | p-value     |
|--------|-----------------------------------------------------------------------------|-----------------|---------------|---------|-------------|
| Plau   | plasminogen activator, urokinase                                             | ENSMUSG00000021822 | 1.3454        | 1.71E-07 |
| Plaur  | plasminogen activator, urokinase receptor                                   | ENSMUSG00000046223 | 1.8866        | 4.39E-06 |
| Por    | P450 (cytochrome) oxidoreductase                                             | ENSMUSG00000005514 | 1.1298        | 8.96E-06 |
| Pqlc1  | PQ loop repeat containing 1                                                 | ENSMUSG00000034006 | 1.7352        | 8.73E-10 |
| Procr  | protein C receptor, endothelial                                             | ENSMUSG00000027611 | 1.9788        | 2.33E-22 |
| Ptgs2  | prostaglandin-endoperoxide synthase 2                                       | ENSMUSG00000032487 | 1.4719        | 1.92E-17 |
| Ptn1   | protein tyrosine phosphatase, non-receptor type 1                           | ENSMUSG00000027540 | 1.2378        | 5.29E-05 |
| Ptn2   | protein tyrosine phosphatase, non-receptor type 2                           | ENSMUSG00000024539 | 1.5295        | 0.0004601 |
| Pvr    | poliovirus receptor                                                          | ENSMUSG00000040511 | 1.7311        | 2.97E-07 |
| Rcan1  | regulator of calcineurin 1                                                  | ENSMUSG00000022951 | 1.3662        | 2.45E-13 |
| Rcc2   | regulator of chromosome condensation 2                                      | ENSMUSG00000040945 | 1.3071        | 1.26E-06 |
| Rnd1   | Rho family GTPase 1                                                         | ENSMUSG00000054855 | 2.4898        | 2.28E-41 |
| Rnf152 | ring finger protein 15Z                                                      | ENSMUSG00000047496 | -1.4452       | 2.38E-05 |
| Rnf19b | ring finger protein 19B                                                      | ENSMUSG00000028793 | 1.4218        | 0.0006166 |
| Sh2b17 | ring finger protein 217                                                     | ENSMUSG00000063760 | 1.5687        | 0.0010808 |
| Rrad   | Ras-related associated with diabetes                                         | ENSMUSG00000031880 | 1.1934        | 4.44E-20 |
| Sbn2   | strawberry notch 2                                                           | ENSMUSG00000035673 | 1.6347        | 4.14E-40 |
| Sdc4   | syndecan 4                                                                  | ENSMUSG00000017009 | 1.0984        | 7.97E-32 |
| Sele   | selectin, endothelial cell                                                  | ENSMUSG00000026582 | 4.2624        | 2.36E-23 |
| Selp   | selectin, platelet                                                          | ENSMUSG00000026580 | 2.7379        | 6.56E-24 |
| Sgms2  | sphingomyelin synthase 2                                                     | ENSMUSG00000050931 | 2.2309        | 2.55E-07 |
| Sh2b3  | SH2B adaptor protein 3                                                       | ENSMUSG00000042594 | 1.3165        | 3.40E-06 |
| Slc16a3| solute carrier family 16 (monocarboxylic acid transporters), member 3       | ENSMUSG00000025161 | 1.4582        | 3.20E-07 |
| Slc25a25| solute carrier family 25 (mitochondrial carrier, phosphate carrier), member 25 | ENSMUSG00000026819 | 1.0335        | 8.73E-10 |
| Slc2a1 | solute carrier family 2 (facilitated glucose transporter), member 1         | ENSMUSG00000028645 | 1.9585        | 1.27E-07 |
| Slc35e4| solute carrier family 35, member E4                                         | ENSMUSG00000048807 | 1.4866        | 0.0006791 |
| Slc41a3| solute carrier family 41, member 3                                          | ENSMUSG00000030089 | 1.3387        | 0.0023525 |
| Slc7a2 | solute carrier family 7 (cationsic amino acid transporter, y+ system), member 2 | ENSMUSG00000031596 | -1.1298       | 2.81E-10 |
| Socs3  | suppressor of cytokine signaling 3                                           | ENSMUSG00000053113 | 1.61          | 3.22E-05 |
| Sphk1  | sphingosine kinase 1                                                         | ENSMUSG00000061878 | 1.3674        | 8.69E-07 |
| Sqstm1 | sequestosome 1                                                               | ENSMUSG00000015837 | 1.13          | 2.10E-26 |
| Gene   | Description                                                                 | Symbol/Accession          | ENSMUSG00000020077 | 1.0165 | 0.0039384 |
|--------|------------------------------------------------------------------------------|---------------------------|--------------------|--------|------------|
| Srgn   | serglycin [Source:MGI Symbol;Acc:MGI:97756]                                  | ENSMUSG00000032802        | 1.3111             | 4.89E-05 |            |
| Srxn1  | sulfiredoxin 1 homolog (S. cerevisiae) [Source:MGI Symbol;Acc:MGI:104971]    | ENSMUSG000000013846       | 1.2355             | 0.00037098 |            |
| St3gal1| ST3 beta-galactoside alpha-2,3-sialyltransferase 1 [Source:MGI Symbol;Acc:MGI:98304] | ENSMUSG00000039232        | 3.4356             | 4.92E-07 |            |
| Stx11  | syntaxin 11 [Source:MGI Symbol;Acc:MGI:1921982]                               | ENSMUSG00000006301        | 1.029              | 2.23E-09 |            |
| Tmbim1 | transmembrane BAX inhibitor motif containing 1 [Source:MGI Symbol;Acc:MGI:1916910] | ENSMUSG00000021281        | 2.1137             | 5.12E-44 |            |
| Tnfaip2| tumor necrosis factor, alpha-induced protein 2 [Source:MGI Symbol;Acc:MGI:104960] | ENSMUSG00000019850        | 2.5895             | 6.96E-31 |            |
| Tnfaip3| tumor necrosis factor, alpha-induced protein 3 [Source:MGI Symbol;Acc:MGI:1196377] | ENSMUSG00000032501        | 1.3871             | 2.39E-07 |            |
| Trib1  | tribbles pseudokinase 1 [Source:MGI Symbol;Acc:MGI:2443397]                 | ENSMUSG00000014158        | 1.1583             | 5.86E-05 |            |
| Trpv4  | transient receptor potential cation channel, subfamily V, member 4 [Source:MGI Symbol;Acc:MGI:1926945] | ENSMUSG00000001473        | 1.7196             | 5.76E-14 |            |
| Tubb6  | tubulin, beta 6 class V [Source:MGI Symbol;Acc:MGI:1915201]                 | ENSMUSG00000000759        | 1.2478             | 4.42E-07 |            |
| Tubgcp3| tubulin, gamma complex associated protein 3 [Source:MGI Symbol;Acc:MGI:2183752] | ENSMUSG00000020250        | 1.194              | 4.36E-06 |            |
| Txrd1  | thioredoxin reductase 1 [Source:MGI Symbol;Acc:MGI:1354175]                 | ENSMUSG000000026670       | 2.1732             | 4.27E-70 |            |
| Uap1   | UDP-N-acetylglucosamine pyrophosphorylase 1 [Source:MGI Symbol;Acc:MGI:1334459] | ENSMUSG00000008348        | 1.5107             | 2.47E-54 |            |
| Ubc    | ubiquitin C [Source:MGI Symbol;Acc:MGI:98889]                                | ENSMUSG00000033364        | 1.4754             | 0.00027371 |            |
| Usp37  | ubiquitin specific peptidase 37 [Source:MGI Symbol;Acc:MGI:2442483]         | ENSMUSG00000027962        | 2.3003             | 3.93E-122 |            |
| Vcam1  | vascular cell adhesion molecule 1 [Source:MGI Symbol;Acc:MGI:98926]         | ENSMUSG00000042677        | 1.6734             | 1.76E-07 |            |
| Zfp703 | zinc finger protein 703 [Source:MGI Symbol;Acc:MGI:2662729]                 | ENSMUSG00000085795        | 1.4066             | 0.0025267 |            |
| Gene   | sense                  | antisense                  | size (bp) |
|--------|------------------------|----------------------------|-----------|
| Camk1  | AAGCAGGCGGAAGACATTAGG | AGTTTCTGAGTCTCTTGTCTT     | 104       |
| Camk1d | TTCGCAGTGAAGTGACATCCC | TTTCTAAACGACCGCAATTCG     | 80        |
| Camk1g | ATGGGGCGTAAGGAGAAGGAGG | ACACGCATCTACCTCTTGTAGGC   | 212       |
| Camk2a | TGCAAGGACATCAGTCTCT    | TGGAAGTCTCCGAGGCAACCA     | 112       |
| Camk2b | GCACGTACCTTGCGGAGGAGG  | ACACGGCTTTCCGAGGACTGG     | 103       |
| camk2d | TGATCTTTCGCTGGGATACCC  | GTAAGCTCCGCTTGTACCT      | 81        |
| camk2g | ACACAGGACTACGCATTCTTC  | GCACATATTCCCTGTGAGAAT     | 95        |
| camk4  | GAGAACCTCTCGCCCGGAGTAC | AAGGAGTTCCCTTCGAGCATC     | 106       |
| camkk1 | TGCCACCTTGGGGAAGAGGC   | GGGAAGCTTTCTAGCCGAGAGG    | 149       |
| camkk2 | TGACGTGCTCTAGCCAGCC    | TGAAAGTCCATCCCTCCAC       | 157       |
| camk2d for splice variant | CGATTTTCCGTACCAGAAT       | TGAAGTCACCCTCCCTCC        |           |
| camk2g for splice variant | GCAAGAGACGGGTAGAGTGC     | ACGTGGACGAGGCTTTTAG       |           |
| Adams4 | ATGGCCTCAATCCATCCAG    | AAGCAGGTTGGAACTTTTCG      | 107       |
| Adams9 | GAATTTGCTGGGAAGGTAAGG  | TCAGTCTCAGGGGATGTAATCTG   | 105       |
| Ccl2   | TTAACGACCTGGGACCGACCA  | GCACCATCTGCAGATTTCGAGGT   | 121       |
| Ccl7   | GCTGCTTTCAGCATCCAGT    | CCACGGACACGGACTCTG        | 135       |
| Cxcl1  | CTGGGATTTCACTTACGAGAT  | CAGGGTCAAGAGCGCTCCCTA     | 117       |
| Cxcl2  | CCAACGACCGAGGCTACAGG  | CAGTCACACTCAAGCTCTG       | 108       |
| Hk2    | TGATCGCTTGCTTATCCAGG   | AACCAGCTGAAATCTCCAGAG     | 112       |
| Icam1  | GTGATGCTCAGGATCCATCCA  | CACAGTTCTCAAAGACAGCG      | 213       |
| Il6    | AGACAAAGCAAGATCCTCCAGAG | GGCACCTCTCAGTGACTCCAGA   | 146       |
| Sele   | ATGCCTCAGCTTGGTCCTCT   | GATGCCGCCTGAGATGC         | 128       |
| Selp   | CATCTGGGATGCTTTTGATGT  | ACGGCTGAGTTATCCAGATGG     | 105       |
| Vcam1  | AGTGGGGATTTGCTGTTTCT  | CCCCTTACCTTACCCACCC       | 112       |
| Hprt   | TCAAGACCTGCCAGGACATAAA | GGGGCTGTACTGCTTAAACAG     | 142       |
| Ppia   | GACGTTTGCTGCAGAACAGT   | CCGTCAGGATACCTTG         | 125       |
| Gapdh  | CATGGCCTCCGTGTTCTCT    | CCTGCTCCACCACCTTCTGA      | 104       |
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