

Supplementary Material

Methods

Animals and ethics statement

Human IL-32 α -expressing transgenic (hIL-32 α -Tg) mice on a C57BL/6 background[1] (Disease Animal Model Research Center, Korea Research Institute of Bioscience and Biotechnology, Daejeon, Korea) were crossed with ApoE^{-/-} mice (B6.129P2-Apoe^{tm1Unc}/J, Jackson Laboratory, Bar Harbor, ME, USA) to generate ApoE^{-/-}/hIL-32 α -Tg mice. All genotyping was performed by polymerase chain reaction (PCR) amplification of tail-tip DNA to identify hIL-32 α -Tg and ApoE^{-/-}/hIL-32 α -Tg mice with primers generating a 450-bp amplicon in Tg mice (primer 1: 5'-GAAGGT CCTCTCTGATGACA-3', primer 2: 5'-GGGGTTCAGAGCACTTCT-3'). The ApoE null and wild-type alleles were detected by PCR amplification using a set of 3 primers (primer 1: 5'-GCCTAGCCGAGGGAGAGCCG-3', primer 2: 5'-TGTGACTTGGGAGCTCTGCAGC-3', primer 3: 5'-GCCGCCCGACTGCATCT-3') that amplify a 155 bp wild-type band and a 245 bp targeted (null) band. All experiments were performed with littermate controls. This study was carried out in strict accordance with the Guide for the Care and Use of Laboratory Animals of Chungbuk National University (Cheongju, Chungbuk, Korea). All animal protocols were reviewed and approved by the Chungbuk National University Institutional Animal Care and Use Committee and compiled with the Korean National Institute of Health Guide for the Care and Use of Laboratory Animals (CBNUA-792-15-01).

Mouse partial carotid ligation surgery

Male mice were fed *ad libitum* with standard chow diet until surgery at 8–9 weeks of age. Animals were anaesthetized with 3.5% isoflurane initially; this dose was then reduced to 1.5–2% during the entire procedure while the mice were subjected to partial ligation of the left carotid artery (LCA). The LCA bifurcation was exposed by blunt dissection and 3 of 4 caudal LCA branches (the left external carotid, internal carotid, and occipital arteries) were ligated with 6-0 silk sutures, leaving the superior thyroid artery intact. The contralateral right carotid artery (RCA) was left intact as an internal control. Following surgery, analgesic buprenorphine (0.1 mg kg⁻¹) was administered. The RCA and LCA were isolated at the indicated experimental end-points.

Microarray procedures

Endothelial-enriched total carotid intimal RNAs were separately obtained from the LCA and RCA of hIL-32 α -Tg and non-Tg mice after partial ligation as described previously[2]. At 48 h after partial ligation, intimal RNA samples from the carotid arteries of 4 mice were pooled to obtain ~50 ng of total RNA as 1 array sample, yielding 3 array samples from 12 mice per group. All RNA samples used for the DNA microarray study passed an initial quality control test by a BioAnalyzer 2100 instrument (Agilent Technologies, Santa Clara, CA, USA) and each sample was linearly amplified, as we described previously[3]. Each sample was then used for microarray studies using GeneChip[®] Mouse Gene 2.0 ST arrays (Affymetrix, Santa Clara, CA, USA) at BioCore Co. (Seoul, Korea). After hybridization in a GeneChip[®] Hybridization Oven 645 (Affymetrix), washing and staining steps proceeded in a GeneChip[®] Fluidics Station 450 (Affymetrix), and then the GeneChip[®] arrays were scanned on a GeneChip[®] 3000 7G

scanner (Affymetrix) to determine the probe fluorescence intensity. The raw probe intensities were then normalized by the quantile normalization algorithm[4].

Microarray data analysis and bioinformatics

The normalized microarray data were statistically analyzed by GeneSpring GX 13.0 software (Agilent Technologies). The differentially expressed genes between LCA and RCA that showed more than 2.0-fold changes at *p*-values of less than 0.05 by 1-way analysis of variance were identified. The lists of differentially expressed genes were interrogated for statistically significant overrepresented cellular functions and disorders using the Cluster 3.0, TreeView, DAVID, KEGG pathway, and GeneSpring GX 13.0 software programs.

Comparative network analysis and functional annotation

We constructed co-expression networks using microarray data obtained by analysis of the LCAs of hIL-32 α -Tg and non-Tg mice as well as publicly available microarray data from TNF- α treated HUVEC samples with and without exposure to apple OPC (dp3.9 apple procyanidin extract)[5]. The microarray data (GSE9647) from this study were downloaded from the GEO database (<http://www.ncbi.nlm.nih.gov/geo/>). The normalized microarray data were used to generate gene co-expression networks using WGCNA[6]. DAVID (<http://david.abcc.ncifcrf.gov/home.jsp>) was used to identify the biological processes that were significantly enriched in the genes included in the co-expression modules[7]. *p*-values < 0.05 were considered significant.

Quantitative real-time PCR

Total RNA was collected from mouse carotid endothelium or EC cultures in 700 μ L of QIAzol[®] and purified using the miRNeasy Mini Kit (Qiagen GmbH, Hilden, Germany) according to the manufacturer's protocol. Total RNA was reverse transcribed into complementary DNA (cDNA) using a High Capacity RNA-to-cDNA kit (Applied Biosystems, Foster City, CA), and then subjected to quantitative real-time PCR (qPCR) using QuantiFast[®] SYBR[®] Green PCR master mix (Qiagen) with custom-designed specific primers using 18S as house-keeping control on a StepOnePlus[™] Real-Time PCR System (Applied Biosystems). Primer sequences can be found in Supplementary Table 9. For the detection of miRNA, cDNA was prepared in a reverse transcription reaction using a miScript II RT Kit (Qiagen). Mature miRNA expression was determined using a miRNA-specific miScript Primer Assay and a miScript SYBR[®] Green PCR Kit (Qiagen) on a StepOnePlus[™] Real-Time PCR System (Applied Biosystems). Specific mature and pre-mature-miRNA primers were purchased from Qiagen. Relative fold-changes in target gene expression were calculated using *Rnu6B* as an internal control. The qPCR experiments were performed as described previously[8, 9]. The fold change between groups was determined for all targets using the $2^{\Delta\Delta Ct}$ method.

Dual-luciferase activity assay

Measurement of firefly luciferase activity was obtained at RT using the Luc-Pair miR Luciferase Assay Kit (GeneCopoeia, Rockville, MD, USA) and a luminometer (1420 Luminescence Counter, PerkinElmer, Waltham, MA, USA) as recently described[8]. Standard 3'-untranslated region (UTR) luciferase assays were performed to validate *in silico* target predictions. Dual-luciferase reporter constructs containing the Timp3 3'UTR containing a miR-205-binding site (5'- UGG AAG G-3') and Reck 3'UTR with miR-205 binding site (5'- AUG AAG G-3') (kindly provided by Dr. Hanjoong Jo of Georgia Institute of Technology and Emory University, Atlanta, GA, USA) or Timp3 3'UTR with mutated miR-205-binding site (5'- GAU

UCA AGG AAA GUC GGU U-3') and Reck 3'UTR with mutated miR-205-binding site (5'- GUC GGU U-3') (custom cloned at Cosmogenetech Co., Seoul, Korea) were transfected into iMAECs using a Nucleofection Kit (Lonza, Walkersville, MD, USA) and were allowed to recover for 24 h. The second transfection was performed using increasing concentrations of miR-205 mimic (*mirVana*TM, Ambion, Foster City, CA, USA) and control-miR (Ambion) with the Lipofectamine[®] RNAiMAX transfection reagent (ThermoFisher Scientific, Waltham, MA, USA). Firefly and *Renilla* luciferase activities were measured using a Luciferase assay system (Promega, Madison, WI, USA) as per manufacturer recommendation.

Arterial wall thickening and atherosclerotic plaque formation assessment

For studies on arterial wall thickening, animals were sacrificed by CO₂ gas inhalation, then aorta and carotid arteries from partial carotid-ligated hIL-32 α -Tg and littermate non-Tg mice fed a high-fat diet for 4 weeks were carefully excised and the surrounding fat tissue was removed by dissection. Aortas and carotid arteries were photographed using a digital microscope device (ToupCam TPH02000A, ToupTek, Hangzhou, China) with a scale bar for reference. LCA and RCA tissues were embedded in Tissue-Tek[®] optimal cutting temperature compound (Sakura Finetek, Torrance, CA, USA), and cryosections (7- μ m) were prepared as previously described[8]. Sections were stained with hematoxylin and eosin, then micrographs were taken using a light microscope (Axio Imager A2, Carl Zeiss, Oberkochen, Germany) at 10 \times magnification and images were analyzed using NIH ImageJ software. Lesion size (cross-sectional areas; intima-media area) and wall thickness (intima media thickness) were calculated using three sections from each LCA segment as described previously[10].

For studies of atherosclerotic plaque formation, aorta and carotid arteries from partial carotid-ligated ApoE^{-/-}/hIL-32 α -Tg, littermate ApoE^{-/-}, and recombinant human IL-32 α protein (rhIL-32 α ; R&D systems, 1 μ g/mouse, administered intraperitoneally every 2 days)-injected ApoE^{-/-} mice fed a high-fat diet [11] for 2 weeks were carefully excised, and the surrounding fat tissue was removed by dissection. The RCA and LCA were photographed using a digital microscope device (ToupCam TPH02000A) as described above, and the opaque area covered by plaque and total artery area of LCA were quantified using NIH ImageJ software as described previously[8]. To visualize atherosclerotic plaques, oil red O staining of tissue sections (7- μ m thick) was carried out, then micrographs were taken with a light microscope at 10 \times magnification and images were analyzed using NIH ImageJ software. The atherosclerotic lesion area and plaque size were calculated as described previously[8].

Immunohistochemical and Immunofluorescence staining

Cryosections were fixed in a 1:1 mixture of methanol/acetone for 5 min at -20 °C and then blocked (1 h, at room temperature) using 10% (v/v) donkey serum in phosphate-buffered saline (PBS). Immunohistochemical staining was carried out using the following antibodies: IL-32 (KU-32-52, 1:100), α -SMA (Abcam, Cambridge, UK, 1:200), CD31 (Abcam, 1:100), VCAM-1 (BD Biosciences, Franklin Lakes, NJ, USA, 1:50), ICAM-1 (Santa Cruz Biotechnology, Dallas, TX, USA, 1:100), MOMA2 (Abcam, 1: 100), TIMP3 (Abcam, 1:50), and (Cell Signaling Technology, Danvers, MA, USA, 1:100) overnight at 4 °C. The sections were incubated with biotinylated secondary antibodies for 2 h at room temperature. After washing in PBS, the immunocomplex was visualized using 3,3'-diaminobenzidine solution (2 mg/10 mL) containing 0.08% (v/v) hydrogen peroxide in PBS. Sections were dehydrated in a series of graded alcohols, cleared in xylene, and mounted with coverslips using PermountTM (Fisher Scientific, Suwanee, GA, USA).

Immunohistological images were acquired using a light microscope (Axio Imager A2, Carl Zeiss).

For immunofluorescence analysis, all sections were incubated with primary antibodies (VCAM-1 and ICAM-1 from Santa Cruz Biotechnology used at 1:200 dilution; CD31 from Abcam at 1:500) overnight at 4 °C, followed by staining with Alexa Flour 568-conjugated donkey anti-goat secondary antibody (Invitrogen, Carlsbad, CA, USA, 1:300) for VCAM-1 and ICAM-1. Alexa Flour 680-conjugated donkey anti-rat secondary antibody (Invitrogen, 1:300) was used for CD31. Nuclei were counterstained with DAPI and mounted with coverslips using an anti-fade fluorescent mounting media (DAKO, Santa Clara, CA, USA). Samples were imaged using Zeiss LSM 710 confocal microscope (Carl Zeiss).

ECs and VSMCs culture and treatment with siRNAs and miRNA mimic

Human umbilical vein endothelial cells (HUVECs; Lonza) were cultured and maintained as described previously[12]. Immortalized mouse aortic ECs (iMAECs) were generated from C57BL/6 mice as previously described[13]. Primary mouse arterial VSMCs were isolated from the thoracic aorta of hIL-32 α -Tg mice and their littermate control (non-Tg) mice, and maintained as described previously[14]. Timp3, Reck, Dgcr8, Drosha, Dicer1, and Rprd2 siRNAs (Stealth RNAi™, ThermoFisher Scientific) or miR-205 mimic (Ambion) were transfected into cells using Lipofectamine® RNAiMAX transfection reagent (ThermoFisher Scientific) in normal culture medium, following the manufacturer's protocol.

Transient expression of hIL-32 α in ECs

For the examination of gene expression and tests of cell functions *in vitro*, iMAECs or HUVECs were transiently transfected with the pcDNA3.1⁺-6 \times Myc-hIL-32 α vector or a control vector using Lipofectamine® 3000 (Life Technologies, Carlsbad, CA, USA), following the manufacturer's protocol as described previously[15].

Smooth muscle cell proliferation and migration assay

VSMCs isolated from hIL-32 α -Tg or non-Tg mice were used. Cells were seeded in a 96-well plate at a density of 5×10^3 cells/mL in Dulbecco's modified Eagle's medium for 24 h, and then transfected with siRNAs, miR-205 mimic, or rhIL-32 α for 24 h before starving in serum-free medium for 24 h. The transfected cells were stimulated with 25 ng/mL of platelet-derived growth factor (PDGF)-BB (PeproTech, Rocky Hill, NJ, USA) for 24 h, and cell proliferation was determined using a BrdU cell proliferation assay kit (BioVision, Milpitas, CA, USA) according to the manufacturer's instructions. Briefly, cells were pulsed with BrdU for 2 h after 20 h PDGF-BB stimulation. ELISAs were performed using antibody for BrdU and the absorbance was measured by a plate reader at 450 nm. BrdU incorporation was calculated as the percentage of BrdU incorporated with respect to the untreated serum-free control. To determine migration, VSMCs were seeded in a μ -Dish with culture inserts (ibidi, Martinsried, Germany) and then maintained in serum-free media for 24 h before performing the migration assays. VSMCs were stimulated with PDGF-BB (25 ng/mL), and then cell migration was monitored for 24 h by taking a picture every 3 h. The area of the migrated cells was quantified using NIH ImageJ software.

Preparation of whole-cell lysates and immunoblotting

Following treatment, cells were washed 3 times with ice-cold PBS and lysed with radioimmunoprecipitation (RIPA) assay buffer. The protein content of each sample was determined using a DC assay kit (Bio-Rad, Hercules, CA, USA). Aliquots of cell lysates were resolved by 10% SDS-PAGE

and subsequently transferred to a polyvinylidene difluoride membrane (EMD Millipore, Billerica, MA, USA). The membrane was incubated with the following antibodies: Myc (Abcam; 1:1,000), IL-32 (KU-32-52; BioLegend, San Diego, CA, USA, 1:1,000), VCAM-1 (Santa Cruz Biotechnology; 1:500), ICAM-1 (Santa Cruz Biotechnology, 1:500), PCNA (Cell Signaling Technology; 1:500), p21 (Santa Cruz Biotechnology; 1:500), phospho-ERK1/2 (Cell Signaling Technology; 1:500), total-ERK1/2 (Cell Signaling Technology; 1:500), Timp3 (Abcam; 1:1,000), and Reck (Cell Signaling Technology; 1:1,000) overnight at 4 °C, followed by secondary antibody (1:2,000) for 1 h at room temperature. Immunoreactive bands were then detected by chemiluminescence. Full-sized scans of all western blots are provided in Supplementary Figs. 14–16.

Monocyte-EC adhesion assay

Following the transfection of iMAECs or HUVECs with hIL-32 α -expressing vector, siRNAs, or miR-205 mimic, or treatment with rhIL-32 α for 24 h, the cells were treated with TNF α (10 ng/mL) for 24 h, and a monocyte-EC binding assay was performed using fluorescently labeled THP-1 cells as described previously[16]. Adherent cells were quantified by fluorescence microscopy[12].

Enzyme-linked immunosorbent assay (ELISA)

Human IL-32 α (hIL-32 α) was transiently expressed in iMAECs as described above. Cells were then transfected with siRNAs for Timp3 or Reck or combination (final concentration, 100 nM) for 24 h, then starved in serum-free medium for 24 h and stimulated with 25 ng/mL of platelet-derived growth factor (PDGF)-BB for 24 h, the stimulated with IL-1 β (10 ng/mL) for 24 h. Soluble-TNF α or IL-10 in conditioned culture medium was measured using mouse soluble-TNF α or IL-10 ELISA kits (R&D Systems, Minneapolis, MN, USA) following the manufacturer's protocol.

***In situ* zymography**

To measure the activity of MMPs in mouse carotid frozen sections, *in situ* zymography was performed in unfixed sections (30- μ m thick) using DQTM-gelatin substrate (Molecular Probes, Eugene, OR, USA) as described previously[8, 9]. DQTM-gelatin was dissolved in a concentration of 1 mg/mL in water and then diluted 1:10 in reaction buffer according to the manufacturer's protocol. The mixture (50 μ L) was put on top of the sections and incubated for 6 h at room temperature. Proteolytic activity was detected as green fluorescence (530 nm) by fluorescence microscopy.

MMP activity analysis

A cell-based ELISA using the DQTM-gelatin assay was employed to assess MMP activity as described previously[9]. VSMCs from non-Tg and hIL-32 α -Tg mice aorta were seeded in a 96-well plate and treated with PDGF-BB (25 ng/mL) in serum-free medium for 24 h, then DQTM-gelatin (100 μ g/mL) was added and incubated for 12 h. Product formation was determined by excitation 495 nm and emission at 515 nm (SpectraMax[®] Gemini EM Fluorescence Microplate Reader; Molecular Devices, Sunnyvale, CA, USA).

Gelatin zymography

For *in vitro* gelatinolytic activity of MMP-2 or MMP-9 assay, a gelatin zymography assay using the SDS-acrylamide gels containing 0.1% gelatin was employed. VSMCs from non-Tg and hIL-32 α -Tg mice aorta were cultured in 6-well plate to 90% confluence. Cells were then washed with phosphate-buffered saline

(PBS), and incubated with serum-free culture medium with or without PDGF-BB (25 ng/mL) for 24 h. Conditioned culture medium was then collected and their gelatinolytic activity was measured by gelatin zymography as described previously[17]. Gels were washed in renaturing buffer for 30 min, and at least 24 h in developing buffer at 37°C. Gels were staining with 0.5% (w/v) Coomassie brilliant blue R-250 solution for 30 min at room temperature, and destained with a destaining solution (methanol/acetic acid/water, 50/10/40, v/v) MMP activity in the gel was detected as white bands against a dark blue background. Band intensity was quantified using NIH ImageJ software. A full-sized scan of the gel is provided in Supplementary Fig. 17.

In situ ADAMs activity assay

ADAM activity was assessed by detection of a fragment peptide (DPEAAE) produced from the digestion of versican s by immunofluorescence staining. Sections were fixed in cold acetone for 5 min at -20 °C, and incubated with a primary antibody against the versican fragment (Abcam, 1:300) overnight at 4 °C, followed by DyLight® 549-conjugated donkey anti-rabbit secondary antibody (Jackson ImmunoResearch, West Grove, PA, USA, 1:250). Nuclei were counterstained with DAPI. Samples were imaged using fluorescence microscopy

Statistical analysis

Statistical analysis was carried out using GraphPad Prism 5.0 (GraphPad Software Inc., La Jolla CA, USA). Pairwise comparisons were performed using 1-way Student's *t*-tests. Data are presented as means ± standard error of the mean (SEM) of the indicated number of experiments. Differences between groups were considered significant at $p < 0.05$.

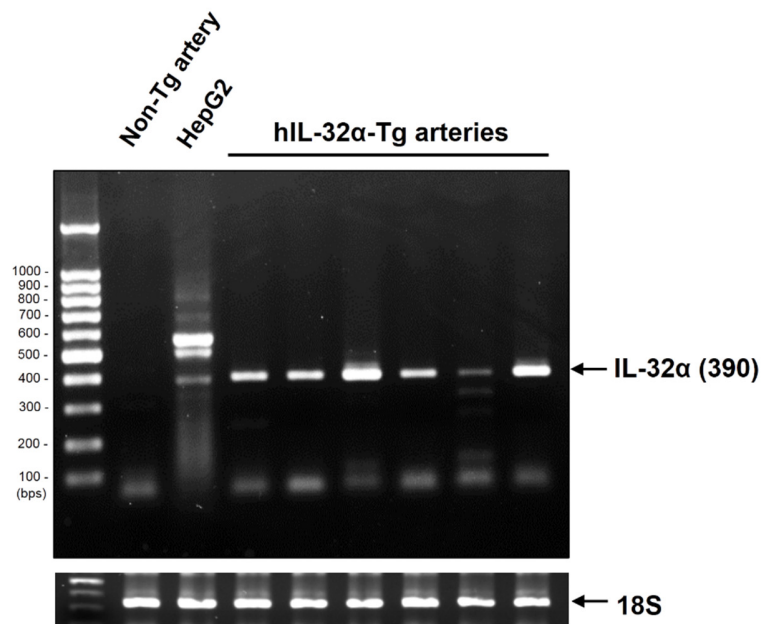
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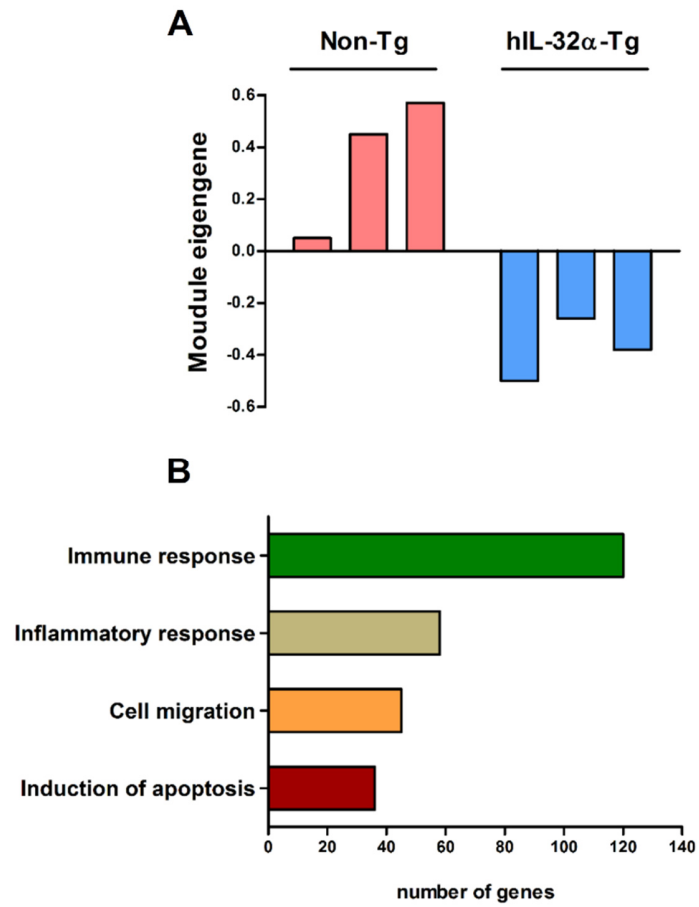
Supplementary Figures and Legends

Supplementary Figure 1.



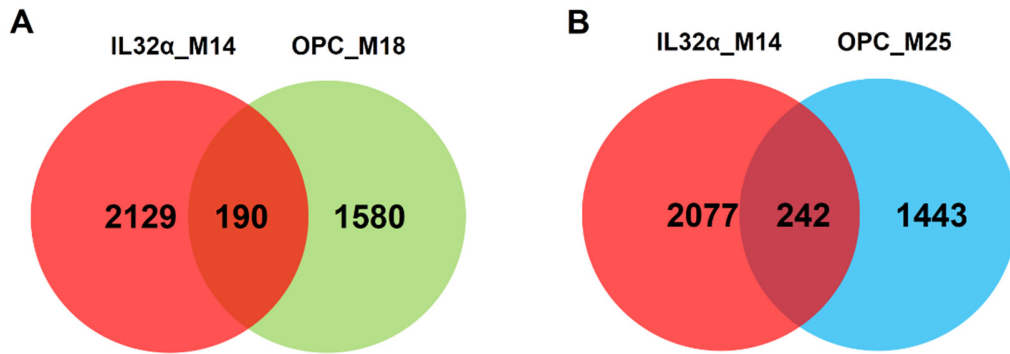
Supplementary Figure 1. Confirmation of hIL-32 α mRNA expression in aorta tissue from hIL-32 α -Tg mice. Total RNA samples were extracted from thoracic and abdominal aorta of hIL-32 α -Tg and non-Tg littermate control mice and analyzed by RT-PCR. Non-Tg littermate control mice aorta tissue and HepG2 cells served as negative and positive controls, respectively.

Supplementary Figure 2.



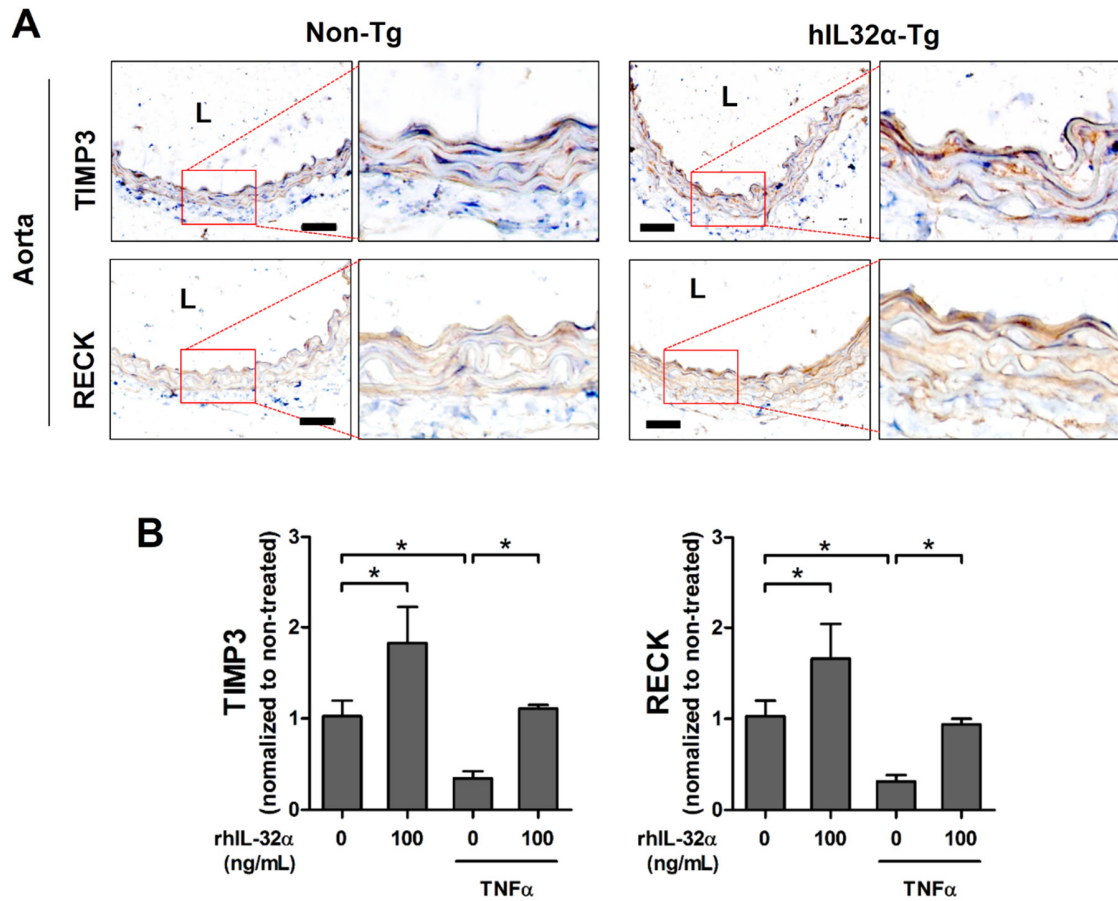
Supplementary Figure 2. Co-expression module associated with hIL-32 α . (A) The eigengene values across samples in the IL-32 α _M14. (B) Major biological processes (gene ontology) significantly enriched in the genes in the co-expression module.

Supplementary Figure 3.



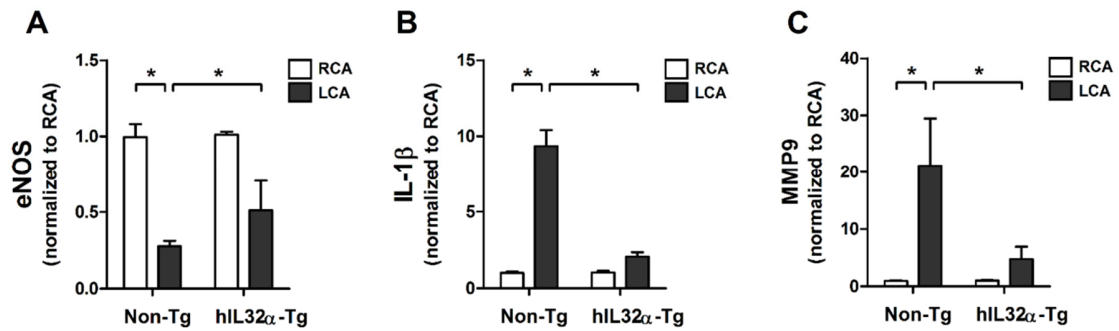
Supplementary Figure 3. Comparative network analysis of the hIL-32 α associated module. (a) Venn diagram showing the number of common and unique genes between the hIL-32 α -associated module IL-32 α _M14 and the two modules, OPC_M18 (**A**) and OPC_M25 (**B**), which associated with apple oligomeric procyanidin (OPC) treatment.

Supplementary Figure 4.



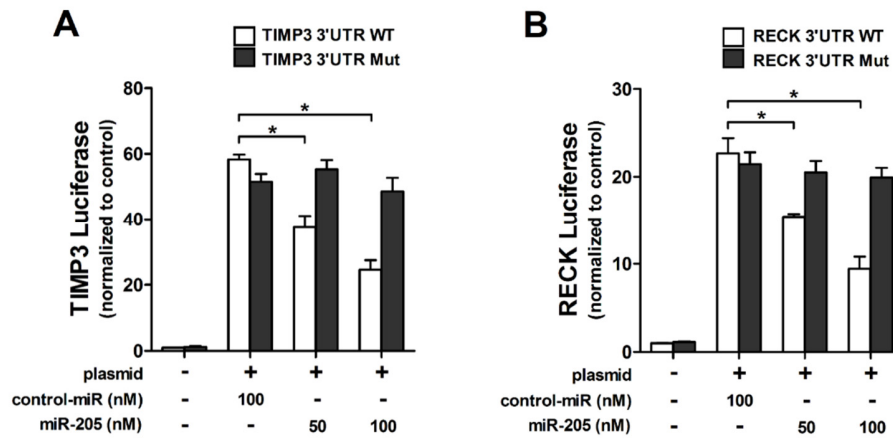
Supplementary Figure 4. *Timp3* and *Reck* expression in the hIL-32 α -Tg mice aorta and hIL-32 α -overexpressing iMAECs. (A) Frozen sections of thoracic aorta tissues from hIL-32 α -Tg and littermate non-Tg control mice were stained with *Timp3* and *Reck* antibodies. Representative microscopy images are shown. The red rectangle indicates the magnified area shown in the lower panel. Nuclei (blue); protein expression (brown). L, lumen. Scale bar, 100 μ m. (B) iMAECs were cultured in a 6-well culture plate for 24 h, then treated with recombinant human IL-32 α protein (rhIL-32 α). After 24 h, cells were treated with tumor necrosis factor α (TNF α) (10 ng/mL; 24 h), and *Timp3* and *Reck* mRNA expression was determined by qPCR (data shown as mean \pm SEM; $n=3$, * $P<0.05$ as determined by Student's t -test).

Supplementary Figure 5.



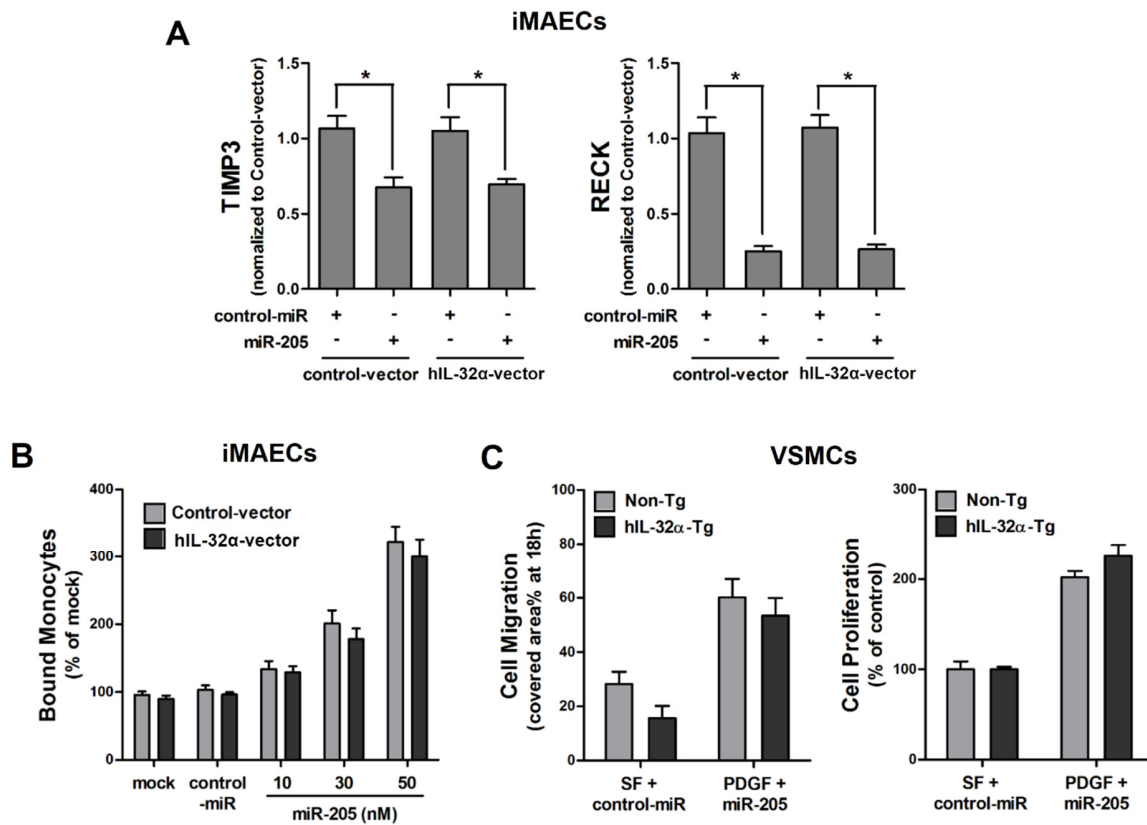
Supplementary Figure 5. eNOS, IL-1 β , and MMP9 expression in the hIL-32 α -Tg mice carotid endothelium. eNOS (*Nos3*) (A), IL-1 β (*Il1b*) (B), and MMP9 (*Mmp9*) (C) mRNA expression was determined by qPCR using endothelial-enriched RNA obtained from left carotid artery (LCA) and right carotid artery (RCA) following partial carotid ligation in hIL-32 α -Tg or non-Tg mice at 2 days post-ligation (data shown as mean \pm SEM; $n = 5$ each, $*P < 0.05$ as determined by Student's *t*-test).

Supplementary Figure 6.



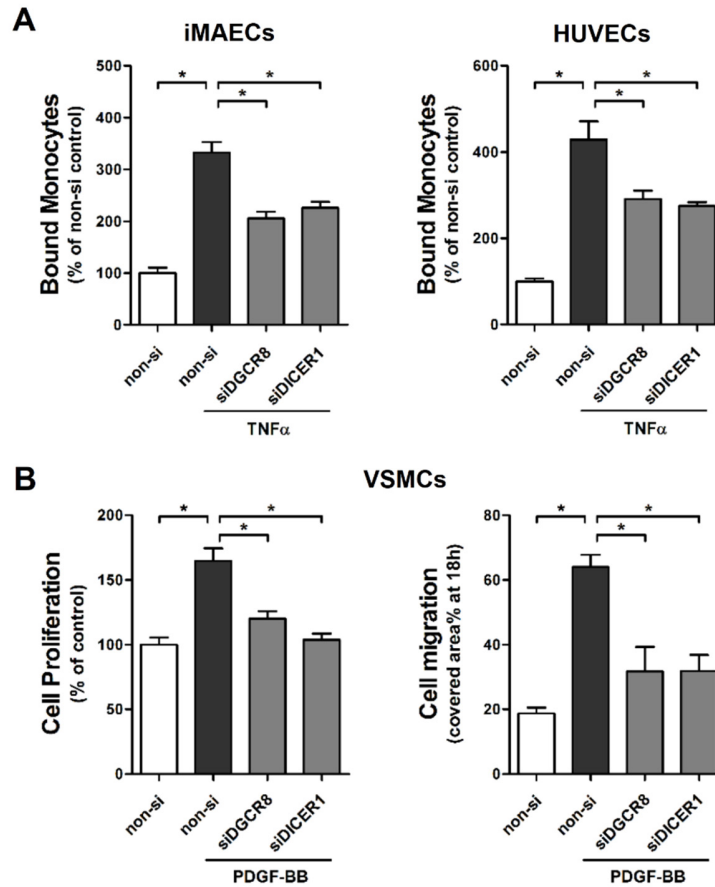
Supplementary Figure 6. miR-205 directly targets Timp3 and Reck. (A) iMAECs were cultured, then transfected with dual-luciferase reporter plasmids containing wild-type (WT) or mutant (Mut) of (A) Timp3-3'UTR and (B) Reck-3'UTR by electroporation. Cells were allowed to recover for 24 h, and treated with mature-miR-205 mimic or control-miR mimic. Firefly luciferase activity (normalized to control *Renilla* luciferase) indicating Timp3 and Reck expression was determined using a Luciferase assay system. ($n = 6$). All data shown as mean \pm SEM, $*P < 0.05$ as determined by Student's *t*-test.

Supplementary Figure 7.



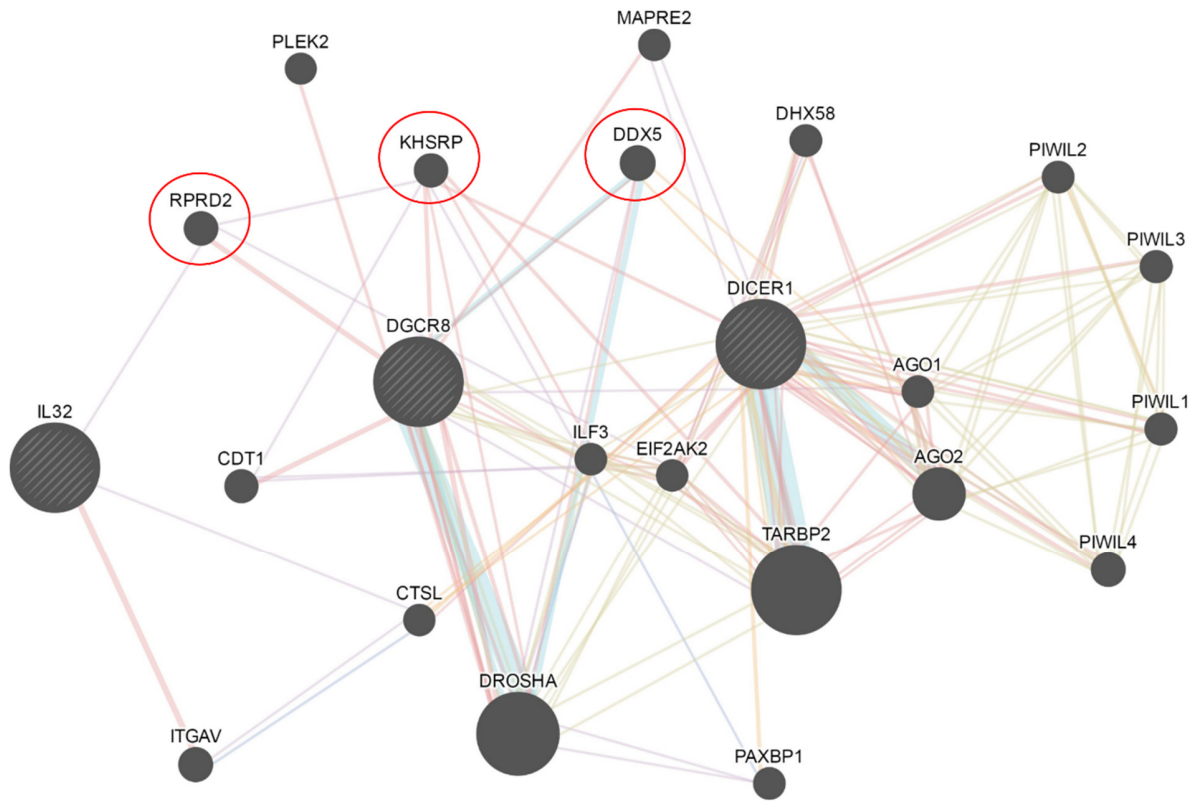
Supplementary Figure 7. hIL-32 α expression does not alter miR-205 biological function. (A) iMAECs were transfected with hIL-32 α -expressing pcDNA3.1⁺-6 \times Myc or control vector. After 24 h, cells were then transfected with mature-miR-205 mimic (50 nM; 24 h) or control-miR mimic, and *Timp3* and *Reck* mRNA expression was determined by qPCR ($n = 4$). (B) iMAECs were transfected with hIL-32 α or control vector, then treated with mature-miR-205 mimic (10–50 nM; 24 h), and THP-1 monocyte adhesion to iMAECs was determined ($n = 3$). (C) Primary cultured VSMCs from hIL-32 α -Tg or non-Tg mice aorta were transfected with mature-miR-205 mimic (50 nM) or control-miR mimic. Cells were then treated with PDGF-BB (25 ng/mL) in serum-free media, and cell migration ($n = 3$) and cell proliferation ($n = 5$) were determined. All data shown as mean \pm SEM, * $P < 0.05$ as determined by Student's *t*-test.

Supplementary Figure 8.



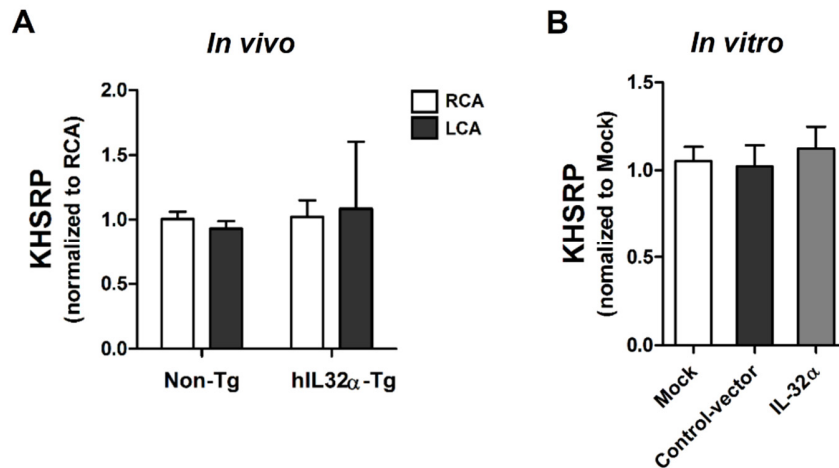
Supplementary Figure 8. Knockdown of Dgcr8 and Dicer1 inhibits EC inflammation and VSMC proliferation and migration. (A) iMAECs and HUVECs were transfected with Dgcr8 and Dicer1 siRNA (siDgcr8 or siDicer1, 150 nM) for 24 h. Cells were then treated with TNF α (10 ng/mL; 24 h), and monocyte-EC adherence was determined ($n = 6$ each). (B) VSMCs isolated from the thoracic aortas of non-Tg mice were transfected with siDgcr8 or siDicer1 (150 nM), then VSMC proliferation and migration were determined by wound healing ($n = 3$) and BrdU incorporation assays ($n = 6$), respectively, as described in the Methods. All data shown as mean \pm SEM, $*p < 0.05$ as determined by Student's t -test.

Supplementary Figure 9.



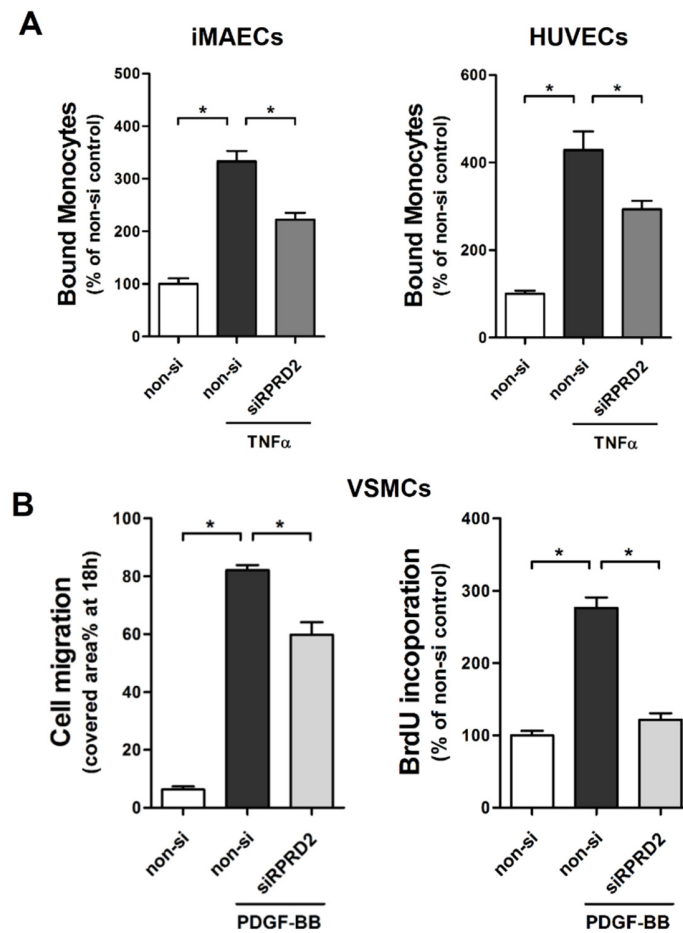
Supplementary Figure 9. Gene network analysis using GeneMANIA. The relationships between IL-32 and Dgcr8/Drosha/Dicer1 are shown based on known functional association networks.

Supplementary Figure 10.



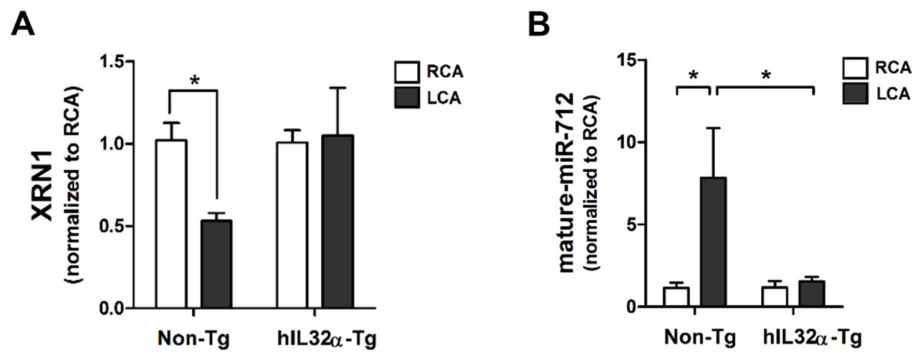
Supplementary Figure 10. *Khsrp* expression in hIL-32 α -Tg mice carotid endothelium and hIL-32 α -expressed iMAECs. (A) *Khsrp* mRNA expression was determined by qPCR using endothelial-enriched RNA obtained from left carotid artery (LCA) and right carotid artery (RCA) following partial carotid ligation in hIL-32 α -Tg or non-Tg mice at 2 days post-ligation ($n = 5$). (B) iMAECs were cultured in a 12-well culture plate for 24 h, then transfected with human IL-32 α expressing pcDNA3.1⁺-6 \times Myc vector (IL-32 α) or control-vector. After 24 h, cells were then treated with TNF α (10 ng/mL; 24 h), and *Khsrp* mRNA expression was determined by qPCR. All data shown as mean \pm SEM, * $P < 0.05$ as determined by Student's *t*-test.

Supplementary Figure 11.



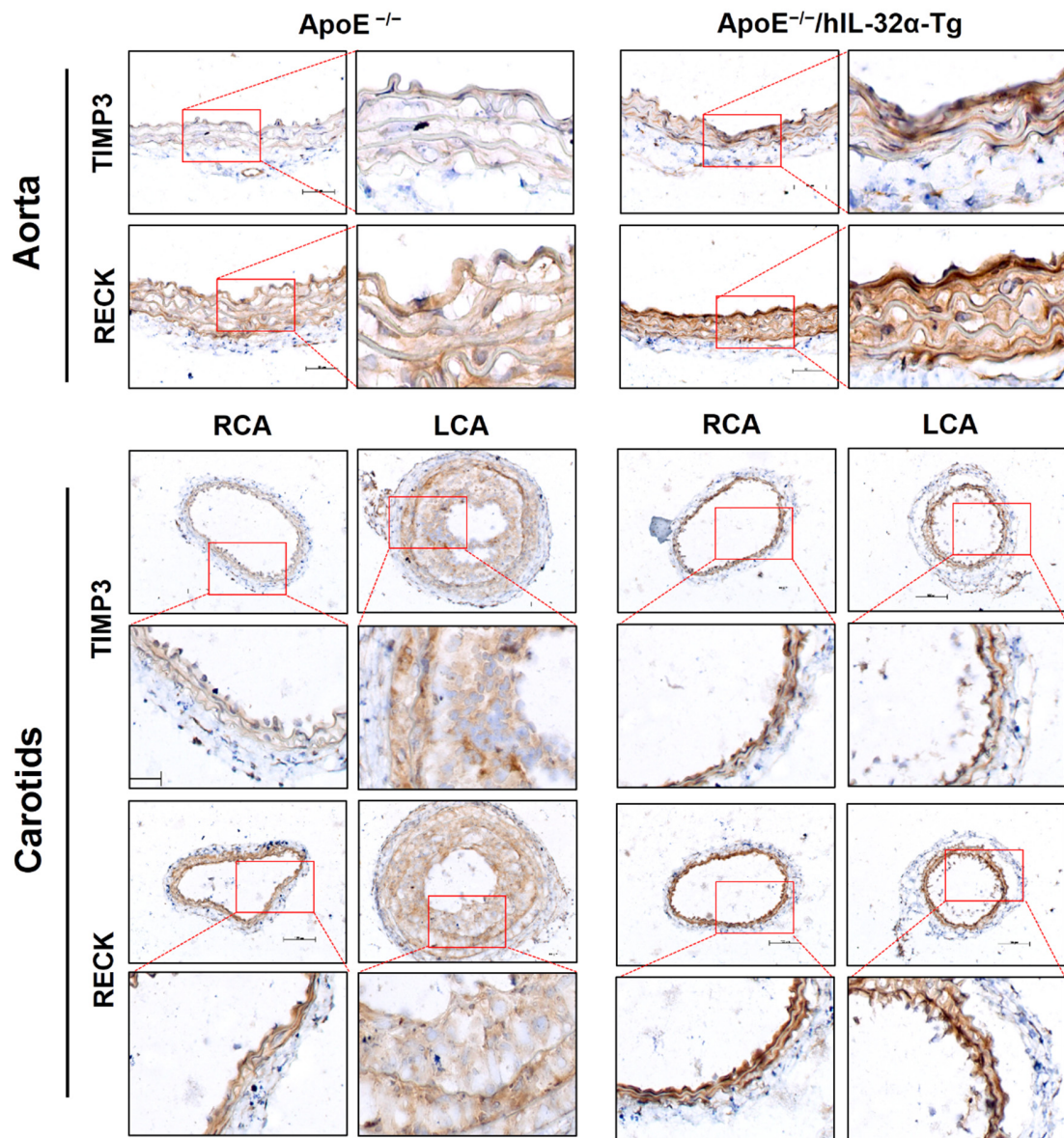
Supplementary Figure 11. Knockdown of Rpr2 inhibits EC inflammation and VSMC proliferation and migration. (A) iMAECs and HUVECs were transfected with Rpr2 siRNA (siRpr2, 200 nM) for 24 h. Cells were then treated with TNF α (10 ng/mL; 24 h), and monocyte-EC adherence was determined ($n = 6$ each). (B) VSMCs isolated from the thoracic aortas of non-Tg mice were transfected with siRpr2 (200 nM), then VSMC proliferation and migration were determined by wound healing ($n = 3$) and BrdU incorporation assays ($n = 6$), respectively, as described in the Methods. All data shown as mean \pm SEM, $*p < 0.05$ as determined by paired t -test.

Supplementary Figure 12.



Supplementary Figure 12. Effect of IL-32 α on expression of the noncanonical microRNA processor XRN1 and mature-microRNA-712 in the mouse endothelium. Expression of (A) *Xrn1* mRNA and (B) mature-microRNA-712 (miR-712) were determined by qPCR using endothelial-enriched RNA obtained from left carotid artery (LCA) and right carotid artery (RCA) following partial carotid ligation in hIL-32 α -Tg or non-Tg mice at 2 days post-ligation ($n = 5$ each, data shown as mean \pm SEM, $*P < 0.05$ as determined by Student's *t*-test).

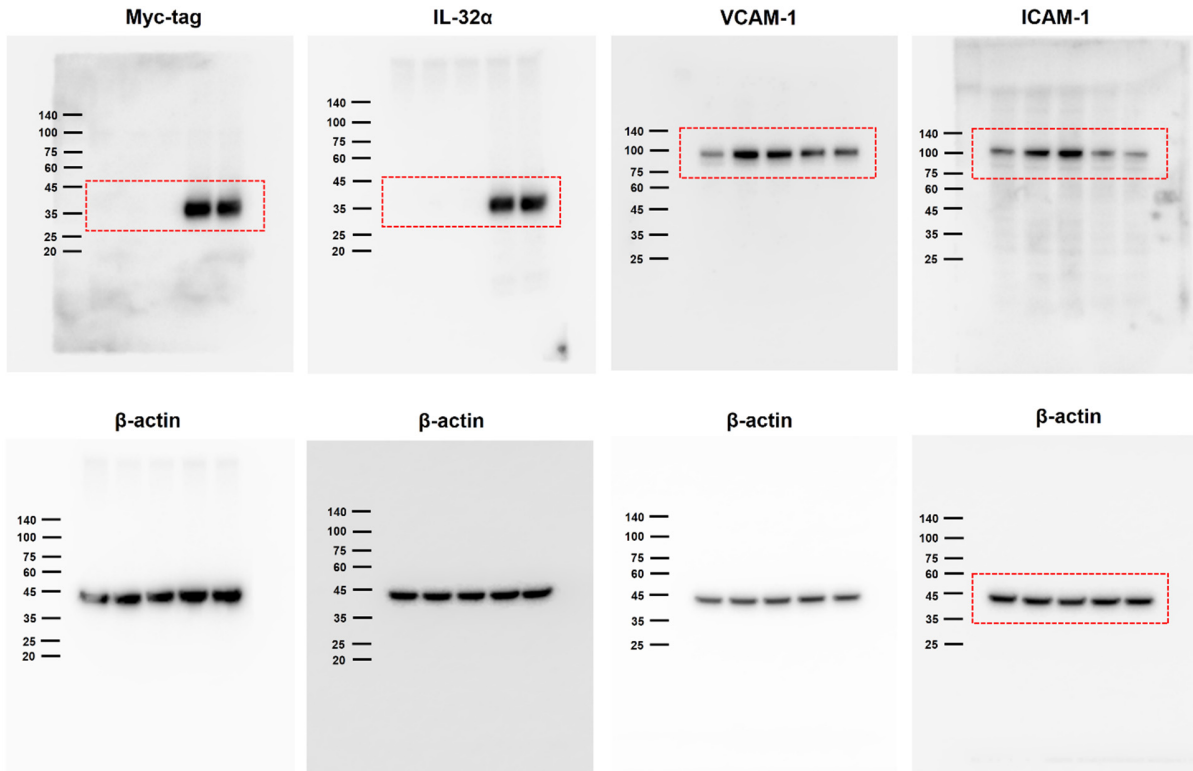
Supplementary Figure 13.



Supplementary Figure 13. Timp3 and Reck expression in ApoE^{-/-}/hIL-32α-Tg mice aorta and carotid arteries. ApoE^{-/-}/hIL-32α-Tg and littermate ApoE^{-/-}/non-Tg control mice were partially ligated and fed a high-fat diet for 2 weeks. Frozen sections of thoracic aorta and carotid tissues from hIL-32α-Tg and littermate non-Tg control mice were stained with Timp3 and Reck antibodies. Images shown are representative microscopy images. The red rectangle indicates the magnified area shown in the lower panel. Nuclei (blue); protein expression (brown).

Supplementary Figure 14.

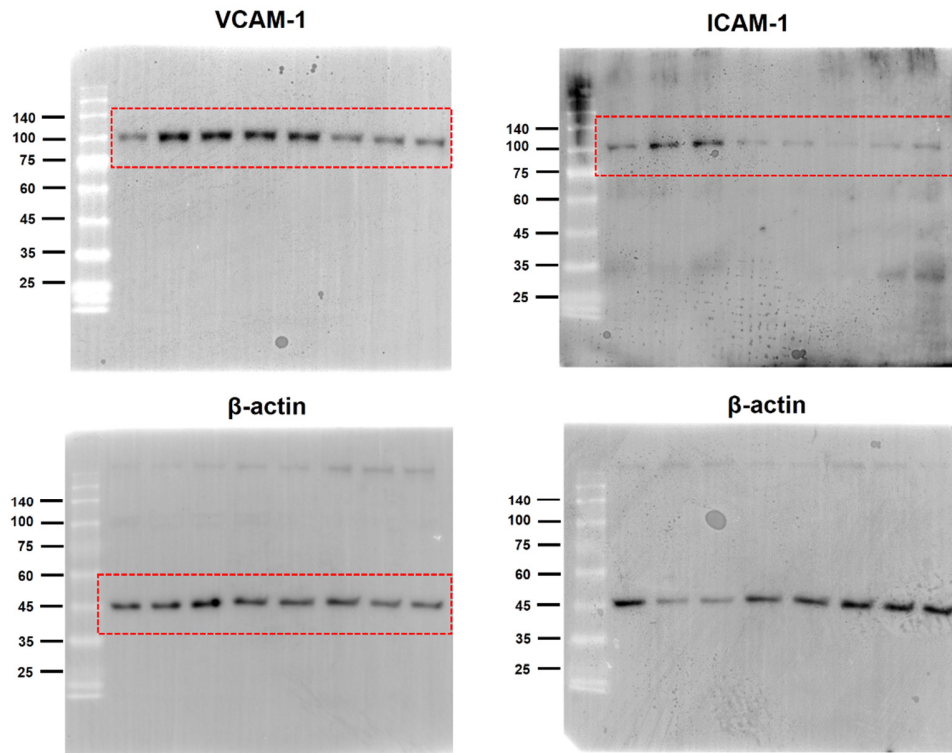
Fig. 1B



Supplementary Figure 14. Full western blot images for Myc-tag, IL-32, VCAM-1, ICAM-1, and respective β -actin control bands shown in Figure 1B. The red dotted rectangle indicates the area shown in Fig. 1B.

Supplementary Figure 15.

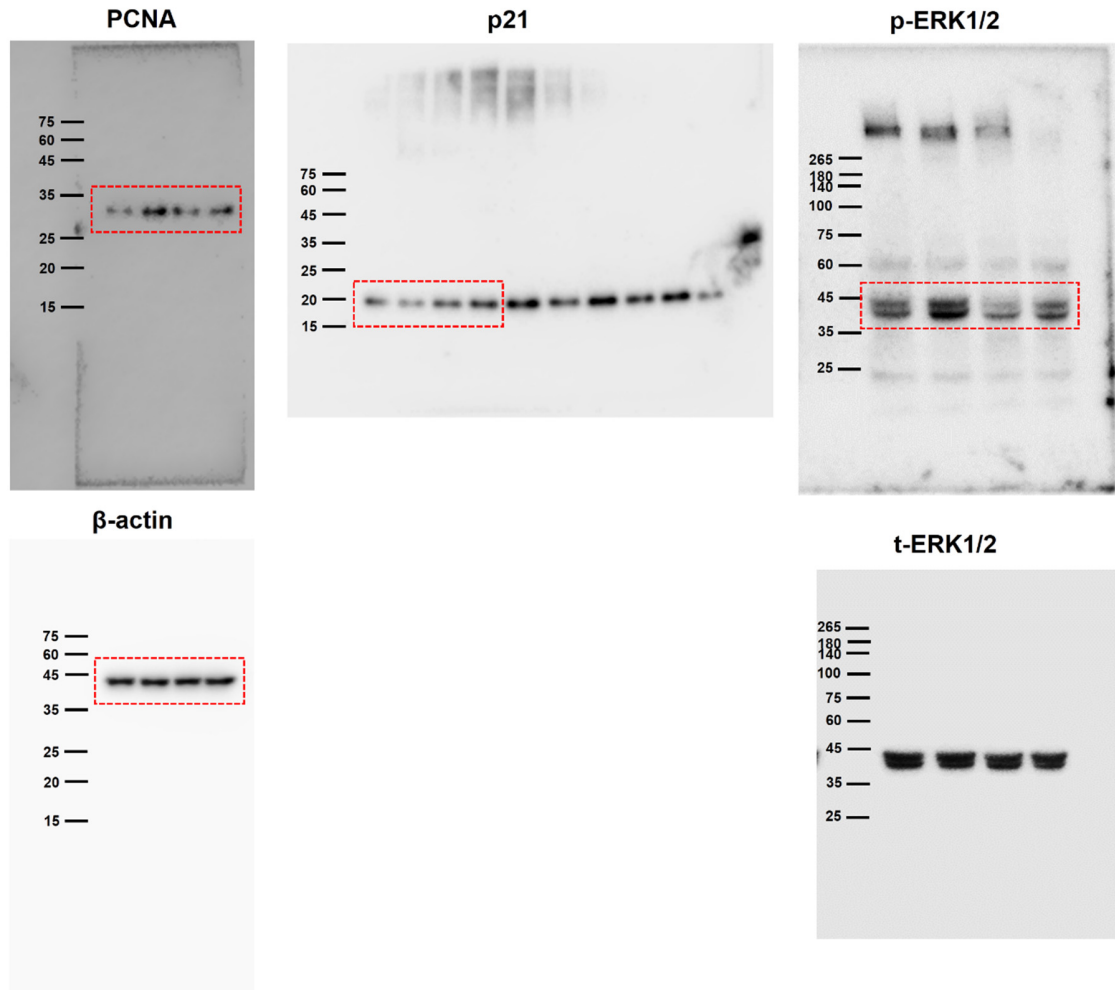
Fig. 1C



Supplementary Figure 15. Full western blot images for VCAM-1, ICAM-1, and the respective β -actin control bands shown in Figure 1C. The red dotted rectangle indicates the area shown in Fig. 1C.

Supplementary Figure 16.

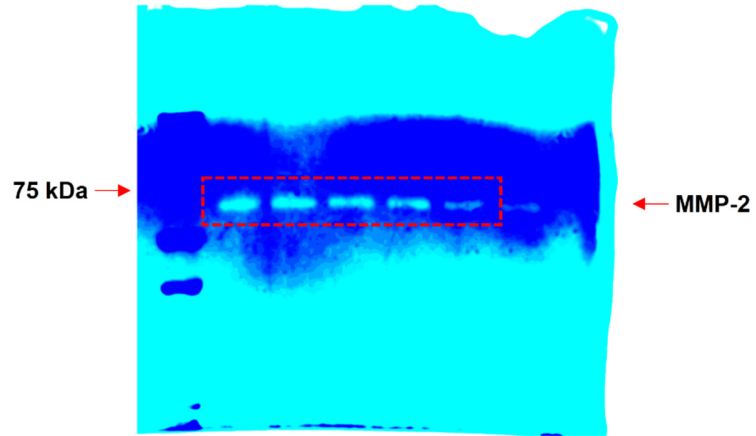
Fig. 2E



Supplementary Figure 16. Full western blot images for PCNA, p32, phospho-ERK1/2, total-ERK1/2, and the respective β -actin control bands in Figure 2E. The red dotted rectangle indicates the area shown in Fig. 2E.

Supplementary Figure 17.

Fig. 2G



Supplementary Figure 17. Full gelatin zymography gel image for MMP activity in Figure 2G. The red dotted rectangle indicates the area shown in Fig. 2G.

Supplementary Tables

Supplementary Table 1. Fold-change expression of endothelial genes in hIL-32 α -Tg mice LCA compared to non-Tg LCA *in vivo*.

Probe ID	Gene Symbol	IL-32 α LCA-1	IL-32 α LCA-2	IL-32 α LCA-3	Non-Tg LCA-1	Non-Tg LCA-2	Non-Tg LCA-3	IL-32 α LCA/Non-Tg LCA (Fold-Changes)	p-value
17459423	---	59.41696461	80.90236656	78.762773	6.642581391	5.161223121	8.978534601	10.542	1.19E-04
17248276	Hba-a2	342.9277874	461.0578596	445.9332609	37.32493907	38.67544537	49.39444851	9.968	9.14E-05
17491193	Saa3	36.63223957	37.15622425	31.47176096	6.270571777	7.073464484	6.001890507	5.441	4.21E-05
17290010	Esm1	180.5427904	220.3180675	228.9462302	43.32503471	42.43812466	46.88085577	4.748	3.42E-05
17302054	Snora31	150.796486	206.1879198	170.0398404	40.59508049	34.22356156	38.39738084	4.655	1.15E-04
17227892	Prg4	61.00155018	80.53148322	77.45690765	17.85330172	22.94630575	17.5222605	3.755	2.39E-04
17236182	Timp3	264.5308186	351.5239071	314.3382444	83.6673595	101.1526987	90.26301101	3.382	2.30E-04
17467458	---	16.45858041	8.925704224	12.6568106	3.413718271	3.934390073	4.091321755	3.325	1.03E-02
17284460	---	6.305164454	14.09138	16.1931528	3.806977386	4.70020278	2.921005349	3.202	4.35E-02
17232731	Rnu3a	69.21280168	85.61822373	71.39608197	18.8826717	27.71524777	29.11629739	2.988	1.31E-03
17508591	Snord13	638.6558895	707.5013056	593.4879797	175.3477368	259.1455841	243.2358382	2.862	1.05E-03
17547616	---	20.75508593	42.40492714	37.18469413	11.97940211	13.44114373	12.1087413	2.674	2.30E-02
17428477	Cyp4b1	93.78589118	76.49327437	75.83957635	23.30161429	36.900079	34.89007999	2.588	3.02E-03
17425523	Snora17	67.49089592	70.46075361	88.31049526	31.75326416	33.66606036	22.78963637	2.565	1.82E-03
17300929	---	83.81514206	66.92410337	90.45021331	29.9198907	31.47754235	32.78863641	2.561	9.29E-04
17225169	Snora75	70.01504209	84.42503692	84.09003847	28.69988399	29.44004558	36.37652939	2.524	4.83E-04
17469339	---	18.07946398	9.028854731	15.45940619	5.872044901	5.217797663	6.170498303	2.466	2.86E-02
17218927	Dpt	14.58304412	16.19661023	17.34164678	6.129481331	7.595258148	6.315259426	2.401	3.74E-04
17387517	Serpig1	31.41659671	38.9540115	30.10126057	12.78716908	15.02438466	15.09442609	2.342	1.18E-03
17280493	---	125.9595763	136.0514007	121.6465842	50.65848405	65.13576426	48.58876977	2.334	8.73E-04
17385838	---	24.22392939	35.48912255	25.1584825	9.490742765	11.54305344	15.78261108	2.305	1.13E-02
17419405	Snora16a	115.8920666	111.9112297	107.8259039	46.915346	49.71550649	54.96094862	2.214	7.14E-05
17399176	583041710Rik	27.75177223	16.42275202	15.66666286	11.73149525	6.600123408	8.801216828	2.205	4.36E-02
17547719	Ahnak2	54.43882413	84.89949104	76.34686435	31.8842712	33.94400756	32.38747294	2.196	5.90E-03
17383129	Snora17	112.4861057	112.4081633	106.2909533	43.35321259	57.72718618	50.88543973	2.179	5.08E-04
17284334	Ighg	10.82002554	9.971503154	8.084580972	5.184443004	4.399211613	3.802177806	2.157	4.37E-03
17281142	Ctl2	396.033044	414.3318743	458.539752	196.2629445	198.9153775	198.9088973	2.136	5.50E-05
17401066	Ngf	58.28274815	75.07340429	83.60903808	28.66864871	40.91383876	32.11599302	2.133	6.79E-03
17301340	---	69.67103064	75.59489993	105.5068604	29.60487793	38.23743913	50.41848979	2.121	1.70E-02
17513854	---	77.94469879	92.4222944	111.1197925	50.35094674	35.67792978	48.36474878	2.094	7.25E-03
17276906	Smoc1	172.8334917	200.5078625	171.3980636	81.70151922	88.6642738	91.6469523	2.079	2.10E-04
17257591	Snord104	234.4060507	235.3682645	225.9434619	94.53980606	119.0483653	121.5333122	2.076	5.96E-04
17512852	---	56.63547115	54.97093068	48.06687145	32.36088146	22.26911006	22.69439306	2.065	4.03E-03

Probe Set ID	Gene Symbol	IL-32 α LCA-1	IL-32 α LCA-2	IL-32 α LCA-3	Non-Tg LCA-1	Non-Tg LCA-2	Non-Tg LCA-3	IL-32 α LCA/Non-Tg LCA Fold-Changes	p-value
17358266	Pgm5	13.55672695	13.25979899	10.23648143	93.62494104	119.7442856	170.8802675	0.096429554	4.98E-04
17528586	Ccnb2	12.4519346	21.09593316	19.74847027	136.7301886	165.4919234	160.3906242	0.115207243	5.67E-05
17277876	---	15.40183136	21.07313425	23.05094513	123.622197	130.8441851	147.3420081	0.148145017	4.94E-05
17504138	Ccl17	6.359811475	8.058693703	8.504208687	50.59005842	55.24216667	47.52421857	0.149473431	1.54E-05
17483615	Itgax	10.7753513	18.03351848	17.15566328	65.35741417	94.01975396	93.82652962	0.18153184	8.21E-04
17514495	Mmp12	7.375930183	11.76409146	9.109949142	41.62669173	55.14235585	57.58287833	0.183023118	3.53E-04
17321768	---	29.74390165	49.38664295	37.04453964	231.0156854	194.9964434	202.8984453	0.18472433	2.17E-04
17454166	Pilra	14.46006367	16.32446719	21.13808827	83.9196642	83.30908919	106.4897675	0.189693481	0.00020295
17271724	Cd300ld	21.32531283	31.37250884	27.8649952	135.7728304	122.4550957	135.9541142	0.204379725	8.96E-05
17277794	Gpr65	8.773235528	9.841976011	11.12949325	40.54482836	60.14025167	43.58331413	0.206176169	0.000671995
17325608	Cd80	46.94865752	63.323057	43.70934943	251.7972995	220.0262739	271.8580993	0.207052385	0.000193798
17404217	LOC10003894	11.63237678	23.34223825	11.95284382	52.81090649	77.57565135	92.48810976	0.210555373	0.003885379
17490149	Cd33 // Cd33	46.1102953	80.28814076	92.56391631	362.6273677	342.1601655	321.1910762	0.213418048	0.001290414
17405082	Slc7a11	6.381355909	8.533573454	6.332025213	24.71186518	36.46958809	37.90932859	0.214419083	0.001559442
17260474	Igfbp3	17.03435009	36.11962476	27.14229863	99.41578276	112.8528467	155.7943388	0.218159066	2.69E-03
17248809	Havcr2	11.55534957	17.66665499	13.94153008	51.35276312	67.55759195	77.38312039	0.219892865	7.22E-04
17418485	Csf3r	79.17653352	101.8766066	104.2935408	378.6176877	424.9752462	467.3880377	0.224509011	0.000159703
17485510	Tarm1	40.81526729	48.34282912	46.23245281	190.0072951	190.7963815	220.2511894	0.225254892	2.38E-05
17212174	Il1r2	35.2749712	35.89569685	37.49374425	141.7595383	166.1634194	173.4154948	0.225754688	2.66E-05
17333731	Fpr2	4.257300719	3.369968252	3.232505893	21.37608878	12.44877605	14.15128346	0.226357789	6.98E-03
17494370	Trim30b	6.948063793	6.471189835	5.177038331	24.86076869	31.79449303	24.85646102	0.228142546	0.000393958
17517097	1600029D21Ri	7.92160629	8.496513734	8.224961923	28.54564983	43.43915096	35.95655755	0.228300647	6.38E-04
17325324	Stfa21l	9.086205911	12.41394712	11.28135926	40.25346305	46.83983212	52.22841438	0.235293641	0.000216839
17408856	I830077J02Rik	26.8826415	34.9792633	36.66376402	142.9644861	119.4591871	143.8987251	0.24248151	0.000194631
17536383	F630028O10R	19.05571716	33.93927872	30.5374142	114.4589923	123.0994922	106.6847385	0.242655206	6.48E-04

17262065	Cyfp2	11.33125695	7.029241394	10.70969879	20.15842474	21.60505502	18.65817533	0.481122159	0.004234064
17501311	---	50.69343431	52.02769906	52.37447601	126.0935789	88.75989157	106.5412277	0.482570529	0.00277724
17400599	Txnip	238.6912714	262.007264	268.2116327	528.2975833	525.7914967	537.0028743	0.483259416	4.56E-05
17338378	Trem14	16.80800976	19.62175256	24.02770581	30.72923614	43.91077219	50.38568483	0.483560352	0.017313072
17316956	Trps1	15.1312262	22.24035637	20.08467206	31.84303627	40.58748384	46.33710478	0.483770343	0.008660136
17532257	Csrp1	15.72595799	16.12032584	19.92541683	37.89351329	36.42439236	32.52508676	0.484558692	0.000903211
17505148	Cdh1	12.05703278	11.60949248	11.72554439	23.21069794	22.93264728	26.75480298	0.485500256	0.000249658
17374632	D2Erd750e	19.11905087	17.37145872	14.2268563	32.11770717	35.89223856	36.42095887	0.48565476	0.001241064
17276776	Arg2	14.79312557	16.64280843	16.50953979	31.25744605	37.10037849	30.23913571	0.486277403	0.000729789
17219248	Adams4	11.54453373	10.40549722	8.18116331	24.64981647	14.06025642	23.14314459	0.487140289	0.033949554
17454179	Pilrb1	16.8029193	15.52297313	14.56338683	27.82380844	35.09739379	33.29422377	0.487336399	0.001436232
17229782	Slamf7	11.95994624	9.470719284	8.683718747	19.49542904	24.07867332	18.19826491	0.487505751	0.005647957
17406279	Tlr2	12.08051455	15.14716612	15.57067951	22.90498956	28.28930836	36.56706676	0.487667441	0.015318361
17232112	H60a // H60a	7.582433815	6.193010963	7.638985219	11.36779864	14.41531043	18.11316811	0.487841598	0.016340595
17471528	Clec1a	12.0034482	20.1823463	22.96793106	28.90772159	39.40183733	44.60343765	0.488462154	0.028491344
17391056	Atp8b4	5.65162601	9.948437817	9.277441507	15.9875393	18.43742148	16.47817998	0.488722404	0.00570665
17261107	Rel	25.91398532	36.98963091	43.35062836	72.873784	73.52846693	70.8517214	0.489078489	0.005559218
17373550	Trp53i11	19.69219465	26.64847371	20.81693842	43.98699071	45.66493897	47.53456326	0.489535124	0.001266611
17400555	Hist2h3c2 // H	48.39077031	50.14100472	51.7945358	102.3038054	112.9538786	91.66785436	0.489781045	0.000516886
17430861	Sesn2	12.76696766	9.696364952	7.748102288	18.42270497	22.66221545	20.57145596	0.489996926	0.006583059
17285867	Hist1h2ab	4.787840195	4.453380864	5.629871525	6.754626513	11.39138295	12.19144308	0.490189233	0.037324276
17321597	Racgap1	14.21409163	12.53201069	12.22519863	23.47053788	21.79709548	34.2006064	0.490400958	0.014593916
17302141	Epst1	13.2500693	13.27377666	9.47747667	18.18992892	28.49604906	26.70733281	0.490525938	0.017099334
17436607	Sh3bp2	15.37301252	15.6097443	14.61714856	27.3896704	26.95027333	38.61498752	0.490559293	0.007922712
17532137	Myd88	39.6674843	35.87719016	34.3238148	69.55175434	68.1920665	86.12970591	0.490761417	0.001823127
17451297	Adrbk2	10.36028654	9.804037483	7.536715844	15.26115238	22.75211883	18.40070325	0.491031524	0.009080079
17432674	Tnfrsf1b	28.04072994	41.68446749	30.42674289	65.05797444	75.62047467	63.20649894	0.491217921	0.005259261
17454187	Pilrb2	8.92146107	7.956213552	7.57668096	17.53419145	20.97649892	11.22558714	0.491680456	0.038764507
17312341	Grina	98.4731839	112.1604319	95.64383014	194.3282723	220.3173039	208.1468622	0.491780932	0.000265231
17317486	Fam49b	64.35302229	59.08811212	74.97158511	116.3753745	151.6420442	135.2683024	0.49199044	0.002224009
17338388	Trem12	4.565921322	6.849311623	6.170960243	9.174630145	13.62286273	12.90635761	0.492557328	0.016853053
17454283	BC037034	25.75034756	19.52252832	26.1724849	39.70384988	43.63618285	61.69561292	0.492605528	0.015538089
17309935	Dab2	59.91569237	66.61194488	72.05696588	144.7170405	131.2141048	126.8147722	0.493076638	0.000555823
17324745	Bex6 // Bex6	4.582913322	4.810983995	3.437028942	5.753991142	9.98301781	10.22884964	0.494146042	0.04093412
17321078	Vdr	11.28541057	13.05136684	15.41839833	26.81460321	27.12766559	26.47535273	0.49435901	0.000690923
17449939	Anrx2	32.31578204	35.10152974	40.12156566	64.25442917	73.4991673	79.5890472	0.494789589	0.001082409
17277387	Fos	24.62379123	36.30750781	28.35411064	50.30280691	60.69038216	69.26084098	0.495331004	0.007773755
17236787	Btg1	43.31581629	50.0679333	56.8324892	94.80366973	110.7808728	97.61344709	0.495439429	0.001234206
17548348	Actg1 // Actg1	106.138702	122.6335573	136.4892956	245.0421296	251.8041063	239.8949537	0.495780011	0.000500888
17294738	Vcan	24.01471863	25.10834444	24.61839836	39.80202701	55.05554618	53.78988502	0.496082895	0.003350976
17283380	Fbln5	105.1648666	109.9353841	103.5836866	195.9568235	214.2857346	230.0242256	0.497736171	0.000101105
17489304	Ffar2	8.890151471	9.50103709	7.672990667	17.51395494	14.08279707	20.74891511	0.497924292	0.009809245
17284247	Gpr132	16.30066594	15.46635149	24.02092829	34.02386192	31.12554083	46.62190339	0.499125828	0.02234895
17246803	Osm	13.16298697	13.41728625	12.98597111	21.5469386	22.4819458	35.23814317	0.499151356	0.024790684
17391920	Rassf2	16.44210378	18.27266303	15.22186262	32.72993881	29.57132536	37.67929326	0.499463403	0.001603643
17494081	Rhog	53.24418254	63.24563464	58.72634723	106.6135501	117.4928374	126.6753496	0.499501958	0.000548526
17379938	Cebpb	28.99809054	28.83118041	26.32393026	60.67229589	48.89232137	58.84573701	0.499691373	0.001250896

Supplementary Table 2. Correlation coefficients and p-values between co-expression modules and human IL-32 α .

Module	R	P-value
IL32 α _M1	0.28	0.59
IL32 α _M2	0.70	0.12
IL32 α _M3	0.05	0.93
IL32 α _M4	0.03	0.96
IL32 α _M5	0.28	0.60
IL32 α _M6	0.72	0.11
IL32 α _M7	0.19	0.72
IL32 α _M8	-0.43	0.40
IL32 α _M9	-0.22	0.68
IL32 α _M10	-0.74	0.09
IL32 α _M11	-0.67	0.15
IL32 α _M12	-0.59	0.22
IL32 α _M13	-0.38	0.46
IL32α_M14	-0.90	0.01
IL32 α _M15	0.11	0.84
IL32 α _M16	0.63	0.18
IL32 α _M17	-0.04	0.95
IL32 α _M18	-0.11	0.83
IL32 α _M19	0.00	1.00
IL32 α _M20	0.11	0.84
IL32 α _M21	0.55	0.26
IL32 α _M22	0.46	0.36
IL32 α _M23	0.08	0.87

Supplementary Table 3. Biological processes enriched in the genes in the IL32 α _M14 module

Term	Count	p-Value
GO:0006955~immune response	121	1.21E-20
GO:0001775~cell activation	72	1.58E-15
GO:0045321~leukocyte activation	63	2.61E-13
GO:0007242~intracellular signaling cascade	168	7.63E-13
GO:0002684~positive regulation of immune system process	58	6.72E-12
GO:0006952~defense response	97	1.16E-11
GO:0046649~lymphocyte activation	53	1.09E-10
GO:0006954~inflammatory response	57	8.71E-10
GO:0006796~phosphate metabolic process	149	2.29E-09
GO:0006793~phosphorus metabolic process	149	2.29E-09
GO:0050865~regulation of cell activation	44	2.94E-09
GO:0016310~phosphorylation	128	4.17E-09
GO:0002694~regulation of leukocyte activation	43	6.33E-09
GO:0042110~T cell activation	36	6.73E-09
GO:0009611~response to wounding	74	8.27E-09
GO:0048534~hemopoietic or lymphoid organ development	63	1.74E-08
GO:0002252~immune effector process	37	2.03E-08
GO:0051249~regulation of lymphocyte activation	40	2.70E-08
GO:0002520~immune system development	64	4.88E-08
GO:0050778~positive regulation of immune response	38	5.36E-08
GO:0006935~chemotaxis	33	6.09E-08
GO:0042330~taxis	33	6.09E-08
GO:0006468~protein amino acid phosphorylation	113	6.49E-08
GO:0002250~adaptive immune response	28	8.24E-08
GO:0002460~adaptive immune response based on somatic recombination of immune receptors built from immunoglobulin superfamily domains	28	8.24E-08
GO:0001817~regulation of cytokine production	38	9.93E-08
GO:0043067~regulation of programmed cell death	101	1.26E-07
GO:0045087~innate immune response	32	1.36E-07
GO:0010941~regulation of cell death	101	1.64E-07
GO:0019884~antigen processing and presentation of exogenous antigen	15	2.23E-07
GO:0042981~regulation of apoptosis	99	2.44E-07
GO:0030097~hemopoiesis	55	3.43E-07
GO:0050867~positive regulation of cell activation	30	4.22E-07
GO:0002449~lymphocyte mediated immunity	25	6.18E-07
GO:0048584~positive regulation of response to stimulus	44	7.07E-07
GO:0002764~immune response-regulating signal transduction	20	7.43E-07
GO:0044092~negative regulation of molecular function	35	7.49E-07
GO:0019886~antigen processing and presentation of exogenous peptide antigen via MHC class II	11	8.18E-07
GO:0002495~antigen processing and presentation of peptide antigen via MHC class II	11	8.18E-07
GO:0002478~antigen processing and presentation of exogenous peptide antigen	13	9.23E-07
GO:0002757~immune response-activating signal transduction	19	9.30E-07
GO:0002696~positive regulation of leukocyte activation	29	9.33E-07
GO:0051251~positive regulation of lymphocyte activation	28	1.04E-06
GO:0002443~leukocyte mediated immunity	27	1.15E-06
GO:0002237~response to molecule of bacterial origin	19	1.32E-06
GO:0002253~activation of immune response	26	2.03E-06
GO:0009617~response to bacterium	38	2.55E-06
GO:0002697~regulation of immune effector process	26	3.23E-06
GO:0001816~cytokine production	17	4.21E-06
GO:0050900~leukocyte migration	17	4.21E-06
GO:0002521~leukocyte differentiation	35	4.43E-06

GO:0048002~antigen processing and presentation of peptide antigen	15	6.25E-06
GO:0002504~antigen processing and presentation of peptide or polysaccharide antigen via MHC class II	11	6.97E-06
GO:0007243~protein kinase cascade	49	7.84E-06
GO:0030595~leukocyte chemotaxis	13	7.89E-06
GO:0060326~cell chemotaxis	13	7.89E-06
GO:0070663~regulation of leukocyte proliferation	24	9.98E-06
GO:0019221~cytokine-mediated signaling pathway	19	1.17E-05
GO:0042098~T cell proliferation	13	1.25E-05
GO:0016477~cell migration	49	1.26E-05
GO:0031349~positive regulation of defense response	19	1.54E-05
GO:0042127~regulation of cell proliferation	90	1.54E-05
GO:0030029~actin filament-based process	39	1.67E-05
GO:0030098~lymphocyte differentiation	29	1.82E-05
GO:0070661~leukocyte proliferation	16	2.08E-05
GO:0032943~mononuclear cell proliferation	16	2.08E-05
GO:0032944~regulation of mononuclear cell proliferation	23	2.18E-05
GO:0050670~regulation of lymphocyte proliferation	23	2.18E-05
GO:0008283~cell proliferation	49	2.76E-05
GO:0002218~activation of innate immune response	9	2.76E-05
GO:0002703~regulation of leukocyte mediated immunity	21	2.78E-05
GO:0019724~B cell mediated immunity	20	3.08E-05
GO:0002274~myeloid leukocyte activation	14	3.43E-05
GO:0043068~positive regulation of programmed cell death	49	3.80E-05
GO:0043086~negative regulation of catalytic activity	26	4.38E-05
GO:0010942~positive regulation of cell death	49	4.69E-05
GO:0045088~regulation of innate immune response	16	5.15E-05
GO:0050863~regulation of T cell activation	27	5.18E-05
GO:0030036~actin cytoskeleton organization	36	5.23E-05
GO:0007049~cell cycle	97	5.82E-05
GO:0042325~regulation of phosphorylation	54	6.19E-05
GO:0043065~positive regulation of apoptosis	48	6.38E-05
GO:0051101~regulation of DNA binding	21	6.65E-05
GO:0016064~immunoglobulin mediated immune response	19	6.90E-05
GO:0046651~lymphocyte proliferation	15	6.94E-05
GO:0042108~positive regulation of cytokine biosynthetic process	15	6.94E-05
GO:0002758~innate immune response-activating signal transduction	8	7.61E-05
GO:0019220~regulation of phosphate metabolic process	55	8.84E-05
GO:0051174~regulation of phosphorus metabolic process	55	8.84E-05
GO:0009891~positive regulation of biosynthetic process	89	9.79E-05
GO:0007626~locomotory behavior	46	1.06E-04
GO:0002706~regulation of lymphocyte mediated immunity	19	1.08E-04
GO:0002768~immune response-regulating cell surface receptor signaling pathway	15	1.23E-04
GO:0051726~regulation of cell cycle	42	1.43E-04
GO:0030593~neutrophil chemotaxis	9	1.66E-04
GO:0009620~response to fungus	9	1.66E-04
GO:0001819~positive regulation of cytokine production	18	1.90E-04
GO:0031328~positive regulation of cellular biosynthetic process	87	1.91E-04
GO:0002221~pattern recognition receptor signaling pathway	7	2.11E-04
GO:0010604~positive regulation of macromolecule metabolic process	97	2.16E-04
GO:0002429~immune response-activating cell surface receptor signaling pathway	14	2.22E-04
GO:0045089~positive regulation of innate immune response	13	2.28E-04
GO:0048514~blood vessel morphogenesis	39	2.37E-04
GO:0048870~cell motility	51	2.54E-04
GO:0051674~localization of cell	51	2.54E-04
GO:0043549~regulation of kinase activity	38	2.62E-04

GO:0051338~regulation of transferase activity	39	2.64E-04
GO:0042035~regulation of cytokine biosynthetic process	18	2.88E-04
GO:0043388~positive regulation of DNA binding	14	2.91E-04
GO:0002683~negative regulation of immune system process	20	3.05E-04
GO:0051090~regulation of transcription factor activity	17	3.34E-04
GO:0048585~negative regulation of response to stimulus	18	3.52E-04
GO:0010324~membrane invagination	37	3.57E-04
GO:0006897~endocytosis	37	3.57E-04
GO:0007010~cytoskeleton organization	56	3.89E-04
GO:0032496~response to lipopolysaccharide	13	4.02E-04
GO:0051130~positive regulation of cellular component organization	27	4.25E-04
GO:0006909~phagocytosis	15	4.33E-04
GO:0042113~B cell activation	20	4.35E-04
GO:0045576~mast cell activation	8	4.63E-04
GO:0051240~positive regulation of multicellular organismal process	33	4.72E-04
GO:0006928~cell motion	61	5.04E-04
GO:0010557~positive regulation of macromolecule biosynthetic process	82	5.41E-04
GO:0051091~positive regulation of transcription factor activity	12	5.53E-04
GO:0051094~positive regulation of developmental process	40	5.84E-04
GO:0001944~vasculature development	45	5.84E-04
GO:0002819~regulation of adaptive immune response	16	5.86E-04
GO:0002822~regulation of adaptive immune response based on somatic recombination of immune receptors built from immunoglobulin superfamily domains	16	5.86E-04
GO:0010648~negative regulation of cell communication	36	5.99E-04
GO:0045859~regulation of protein kinase activity	36	5.99E-04
GO:0051099~positive regulation of binding	14	6.13E-04
GO:0000278~mitotic cell cycle	44	6.52E-04
GO:0001568~blood vessel development	44	6.52E-04
GO:0000087~M phase of mitotic cell cycle	37	6.61E-04
GO:0050764~regulation of phagocytosis	10	7.33E-04
GO:0051092~positive regulation of NF-kappaB transcription factor activity	10	7.33E-04
GO:0045577~regulation of B cell differentiation	8	7.47E-04
GO:0002700~regulation of production of molecular mediator of immune response	11	7.54E-04
GO:0010647~positive regulation of cell communication	36	8.10E-04
GO:0044093~positive regulation of molecular function	52	8.30E-04
GO:0070665~positive regulation of leukocyte proliferation	15	8.36E-04
GO:0030217~T cell differentiation	19	8.71E-04
GO:0045619~regulation of lymphocyte differentiation	16	8.74E-04
GO:0010627~regulation of protein kinase cascade	31	8.90E-04
GO:0050866~negative regulation of cell activation	15	0.001027
GO:0002695~negative regulation of leukocyte activation	15	0.001027
GO:0030099~myeloid cell differentiation	22	0.001029
GO:0051173~positive regulation of nitrogen compound metabolic process	80	0.001045
GO:0051098~regulation of binding	21	0.001092
GO:0009967~positive regulation of signal transduction	33	0.001225
GO:0051254~positive regulation of RNA metabolic process	66	0.001242
GO:0050870~positive regulation of T cell activation	17	0.001269
GO:0032675~regulation of interleukin-6 production	11	0.001328
GO:0008284~positive regulation of cell proliferation	48	0.001511
GO:0050864~regulation of B cell activation	15	0.001517
GO:0045123~cellular extravasation	5	0.001627
GO:0045597~positive regulation of cell differentiation	33	0.001643
GO:0045893~positive regulation of transcription, DNA-dependent	65	0.001661
GO:0019882~antigen processing and presentation	20	0.001785
GO:0010628~positive regulation of gene expression	74	0.001789
GO:0032946~positive regulation of mononuclear cell proliferation	14	0.001797

GO:0050671~positive regulation of lymphocyte proliferation	14	0.001797
GO:0050766~positive regulation of phagocytosis	9	0.001867
GO:0050830~defense response to Gram-positive bacterium	9	0.001867
GO:0007264~small GTPase mediated signal transduction	44	0.002041
GO:0045941~positive regulation of transcription	72	0.002076
GO:0042742~defense response to bacterium	23	0.002105
GO:0006469~negative regulation of protein kinase activity	14	0.00218
GO:0033673~negative regulation of kinase activity	14	0.00218
GO:0007610~behavior	63	0.002189
GO:0009968~negative regulation of signal transduction	32	0.002215
GO:0043122~regulation of I-kappaB kinase/NF-kappaB cascade	12	0.002453
GO:0045428~regulation of nitric oxide biosynthetic process	8	0.002468
GO:0002285~lymphocyte activation during immune response	8	0.002468
GO:0030183~B cell differentiation	13	0.002583
GO:0051250~negative regulation of lymphocyte activation	14	0.002628
GO:0008285~negative regulation of cell proliferation	39	0.002629
GO:0043405~regulation of MAP kinase activity	19	0.002901
GO:0006917~induction of apoptosis	31	0.002978
GO:0012502~induction of programmed cell death	31	0.002978
GO:0007265~Ras protein signal transduction	15	0.00309
GO:0042129~regulation of T cell proliferation	15	0.00309
GO:0051607~defense response to virus	7	0.003134
GO:0032655~regulation of interleukin-12 production	7	0.003134
GO:0002712~regulation of B cell mediated immunity	10	0.003141
GO:0002889~regulation of immunoglobulin mediated immune response	10	0.003141
GO:0051348~negative regulation of transferase activity	14	0.003148
GO:0008219~cell death	75	0.003163
GO:0016044~membrane organization	45	0.003269
GO:0007067~mitosis	34	0.003323
GO:0000280~nuclear division	34	0.003323
GO:0002335~mature B cell differentiation	5	0.003475
GO:0051301~cell division	46	0.003595
GO:0032844~regulation of homeostatic process	15	0.003643
GO:0032494~response to peptidoglycan	6	0.003666
GO:0051640~organelle localization	14	0.003749
GO:0045935~positive regulation of nucleobase, nucleoside, nucleotide and nucleic acid metabolic process	75	0.003757
GO:0016265~death	76	0.003857
GO:0043069~negative regulation of programmed cell death	41	0.00386
GO:0000279~M phase	46	0.004116
GO:0060548~negative regulation of cell death	41	0.004151
GO:0051347~positive regulation of transferase activity	26	0.004233
GO:0010876~lipid localization	25	0.004254
GO:0012501~programmed cell death	70	0.004331
GO:0050871~positive regulation of B cell activation	11	0.004373
GO:0050777~negative regulation of immune response	9	0.004448
GO:0043392~negative regulation of DNA binding	9	0.004448
GO:0030101~natural killer cell activation	7	0.004565
GO:0043487~regulation of RNA stability	7	0.004565
GO:0009142~nucleoside triphosphate biosynthetic process	20	0.005052
GO:0032649~regulation of interferon-gamma production	10	0.005065
GO:0033674~positive regulation of kinase activity	25	0.005209
GO:0006754~ATP biosynthetic process	18	0.005336
GO:0030003~cellular cation homeostasis	28	0.005468
GO:0044087~regulation of cellular component biogenesis	19	0.00554
GO:0032680~regulation of tumor necrosis factor production	9	0.00574

GO:0048285~organelle fission	34	0.005878
GO:0046034~ATP metabolic process	19	0.006252
GO:0043123~positive regulation of I-kappaB kinase/NF-kappaB cascade	10	0.006317
GO:0045059~positive thymic T cell selection	5	0.006364
GO:0050732~negative regulation of peptidyl-tyrosine phosphorylation	5	0.006364
GO:0007059~chromosome segregation	15	0.006713
GO:0007249~I-kappaB kinase/NF-kappaB cascade	9	0.007298
GO:0010638~positive regulation of organelle organization	13	0.007523
GO:0030005~cellular di-, tri-valent inorganic cation homeostasis	25	0.007654
GO:0043254~regulation of protein complex assembly	15	0.007736
GO:0051656~establishment of organelle localization	10	0.007794
GO:0045807~positive regulation of endocytosis	10	0.007794
GO:0045621~positive regulation of lymphocyte differentiation	10	0.007794
GO:0002699~positive regulation of immune effector process	12	0.007802
GO:0002708~positive regulation of lymphocyte mediated immunity	11	0.007922
GO:0002705~positive regulation of leukocyte mediated immunity	11	0.007922
GO:0043066~negative regulation of apoptosis	39	0.007924
GO:0051051~negative regulation of transport	16	0.008183
GO:0002823~negative regulation of adaptive immune response based on somatic recombination of immune receptors built from immunoglobulin superfamily domains	6	0.008512
GO:0043488~regulation of mRNA stability	6	0.008512
GO:0050854~regulation of antigen receptor-mediated signaling pathway	6	0.008512
GO:0042089~cytokine biosynthetic process	6	0.008512
GO:0002820~negative regulation of adaptive immune response	6	0.008512
GO:0019915~lipid storage	6	0.008512
GO:0032755~positive regulation of interleukin-6 production	7	0.008782
GO:0043408~regulation of MAPKKK cascade	19	0.008844
GO:0009206~purine ribonucleoside triphosphate biosynthetic process	19	0.008844
GO:0009201~ribonucleoside triphosphate biosynthetic process	19	0.008844
GO:0009152~purine ribonucleotide biosynthetic process	21	0.008879
GO:0045449~regulation of transcription	271	0.009062
GO:0006334~nucleosome assembly	16	0.00931
GO:0022403~cell cycle phase	50	0.009477
GO:0050672~negative regulation of lymphocyte proliferation	10	0.00952
GO:0032945~negative regulation of mononuclear cell proliferation	10	0.00952
GO:0070664~negative regulation of leukocyte proliferation	10	0.00952
GO:0006915~apoptosis	67	0.009554
GO:0009145~purine nucleoside triphosphate biosynthetic process	19	0.009878
GO:0009205~purine ribonucleoside triphosphate metabolic process	20	0.009888
GO:0006323~DNA packaging	20	0.009888
GO:0022402~cell cycle process	58	0.010037
GO:0045351~type I interferon biosynthetic process	4	0.010294
GO:0032606~type I interferon production	4	0.010294
GO:0002312~B cell activation during immune response	4	0.010294
GO:0002313~mature B cell differentiation during immune response	4	0.010294
GO:0045736~negative regulation of cyclin-dependent protein kinase activity	5	0.010495
GO:0045075~regulation of interleukin-12 biosynthetic process	5	0.010495
GO:0045580~regulation of T cell differentiation	12	0.010845
GO:0010740~positive regulation of protein kinase cascade	18	0.010961
GO:0009199~ribonucleoside triphosphate metabolic process	20	0.010971
GO:0034220~ion transmembrane transport	11	0.011303
GO:0002260~lymphocyte homeostasis	9	0.01134
GO:0031341~regulation of cell killing	9	0.01134
GO:0001910~regulation of leukocyte mediated cytotoxicity	9	0.01134
GO:0051100~negative regulation of binding	9	0.01134
GO:0050851~antigen receptor-mediated signaling pathway	10	0.01152

GO:0006672~ceramide metabolic process	10	0.01152
GO:0045860~positive regulation of protein kinase activity	23	0.011545
GO:0055066~di-, tri-valent inorganic cation homeostasis	26	0.01159
GO:0048535~lymph node development	7	0.011697
GO:0031497~chromatin assembly	16	0.01193
GO:0007159~leukocyte adhesion	6	0.012094
GO:0006825~copper ion transport	6	0.012094
GO:0042107~cytokine metabolic process	6	0.012094
GO:0051050~positive regulation of transport	24	0.01231
GO:0006665~sphingolipid metabolic process	14	0.012611
GO:0045944~positive regulation of transcription from RNA polymerase II promoter	53	0.013106
GO:0009260~ribonucleotide biosynthetic process	21	0.013206
GO:0007585~respiratory gaseous exchange	8	0.013312
GO:0009615~response to virus	16	0.013439
GO:0065004~protein-DNA complex assembly	16	0.013439
GO:0034728~nucleosome organization	16	0.013439
GO:0007015~actin filament organization	13	0.013677
GO:0016049~cell growth	10	0.013817
GO:0015986~ATP synthesis coupled proton transport	10	0.013817
GO:0015985~energy coupled proton transport, down electrochemical gradient	10	0.013817
GO:0002366~leukocyte activation during immune response	9	0.013888
GO:0007595~lactation	9	0.013888
GO:0002263~cell activation during immune response	9	0.013888
GO:0009150~purine ribonucleotide metabolic process	22	0.014184
GO:0006357~regulation of transcription from RNA polymerase II promoter	84	0.014555
GO:0003018~vascular process in circulatory system	12	0.014721
GO:0002698~negative regulation of immune effector process	7	0.015231
GO:0050727~regulation of inflammatory response	13	0.01569
GO:0048872~homeostasis of number of cells	21	0.015921
GO:0043368~positive T cell selection	5	0.01603
GO:0009144~purine nucleoside triphosphate metabolic process	20	0.016279
GO:0006643~membrane lipid metabolic process	14	0.016336
GO:0046519~sphingoid metabolic process	10	0.016436
GO:0032760~positive regulation of tumor necrosis factor production	6	0.016574
GO:0045840~positive regulation of mitosis	6	0.016574
GO:0006911~phagocytosis, engulfment	6	0.016574
GO:0051785~positive regulation of nuclear division	6	0.016574
GO:0070201~regulation of establishment of protein localization	15	0.016721
GO:0045582~positive regulation of T cell differentiation	9	0.016829
GO:0052547~regulation of peptidase activity	12	0.017012
GO:0051056~regulation of small GTPase mediated signal transduction	36	0.017492
GO:0048732~gland development	32	0.017608
GO:0010033~response to organic substance	70	0.018116
GO:0046328~regulation of JNK cascade	11	0.018283
GO:0032886~regulation of microtubule-based process	11	0.018283
GO:0051223~regulation of protein transport	14	0.018486
GO:0002467~germinal center formation	4	0.018961
GO:0009141~nucleoside triphosphate metabolic process	21	0.019056
GO:0051252~regulation of RNA metabolic process	184	0.019116
GO:0001776~leukocyte homeostasis	10	0.019401
GO:0060322~head development	7	0.019441
GO:0030100~regulation of endocytosis	12	0.019557
GO:0051235~maintenance of location	9	0.020193
GO:0043406~positive regulation of MAP kinase activity	13	0.020378
GO:0070302~regulation of stress-activated protein kinase signaling pathway	11	0.021194

GO:0033043~regulation of organelle organization	26	0.021708
GO:0008354~germ cell migration	5	0.023096
GO:0046823~negative regulation of nucleocytoplasmic transport	5	0.023096
GO:0009259~ribonucleotide metabolic process	22	0.02373
GO:0007033~vacuole organization	9	0.024008
GO:0050885~neuromuscular process controlling balance	9	0.024008
GO:0002824~positive regulation of adaptive immune response based on somatic recombination of immune receptors built from immunoglobulin superfamily domains	9	0.024008
GO:0045787~positive regulation of cell cycle	9	0.024008
GO:0002821~positive regulation of adaptive immune response	9	0.024008
GO:0002573~myeloid leukocyte differentiation	9	0.024008
GO:0032101~regulation of response to external stimulus	19	0.024124
GO:0043433~negative regulation of transcription factor activity	7	0.024379
GO:0001974~blood vessel remodeling	7	0.024379
GO:0048771~tissue remodeling	11	0.024429
GO:0019362~pyridine nucleotide metabolic process	8	0.024728
GO:0045185~maintenance of protein location	8	0.024728
GO:0080135~regulation of cellular response to stress	15	0.026124
GO:0006869~lipid transport	21	0.02674
GO:0051336~regulation of hydrolase activity	31	0.027234
GO:0009725~response to hormone stimulus	27	0.027474
GO:0032880~regulation of protein localization	17	0.027894
GO:0035150~regulation of tube size	11	0.028003
GO:0050880~regulation of blood vessel size	11	0.028003
GO:0042102~positive regulation of T cell proliferation	9	0.028301
GO:0046578~regulation of Ras protein signal transduction	29	0.028353
GO:0060627~regulation of vesicle-mediated transport	16	0.028515
GO:0016051~carbohydrate biosynthetic process	16	0.028515
GO:0060324~face development	6	0.028535
GO:0045766~positive regulation of angiogenesis	6	0.028535
GO:0032729~positive regulation of interferon-gamma production	6	0.028535
GO:0043933~macromolecular complex subunit organization	52	0.02951
GO:0043085~positive regulation of catalytic activity	39	0.029513
GO:0048536~spleen development	7	0.030088
GO:0031343~positive regulation of cell killing	7	0.030088
GO:0001912~positive regulation of leukocyte mediated cytotoxicity	7	0.030088
GO:0001933~negative regulation of protein amino acid phosphorylation	7	0.030088
GO:0002224~toll-like receptor signaling pathway	4	0.030576
GO:0045084~positive regulation of interleukin-12 biosynthetic process	4	0.030576
GO:0010812~negative regulation of cell-substrate adhesion	4	0.030576
GO:0002718~regulation of cytokine production during immune response	4	0.030576
GO:0002456~T cell mediated immunity	4	0.030576
GO:0050798~activated T cell proliferation	4	0.030576
GO:0051452~intracellular pH reduction	4	0.030576
GO:0050868~negative regulation of T cell activation	10	0.030588
GO:0032763~regulation of mast cell cytokine production	3	0.031757
GO:0046633~alpha-beta T cell proliferation	3	0.031757
GO:0002238~response to molecule of fungal origin	3	0.031757
GO:0006021~inositol biosynthetic process	3	0.031757
GO:0031113~regulation of microtubule polymerization	5	0.031775
GO:0002920~regulation of humoral immune response	5	0.031775
GO:0032387~negative regulation of intracellular transport	5	0.031775
GO:0010829~negative regulation of glucose transport	5	0.031775
GO:0046486~glycerolipid metabolic process	22	0.032419
GO:0034637~cellular carbohydrate biosynthetic process	12	0.032595
GO:0006119~oxidative phosphorylation	12	0.032595

GO:0070507~regulation of microtubule cytoskeleton organization	9	0.033095
GO:0051969~regulation of transmission of nerve impulse	19	0.034028
GO:0055080~cation homeostasis	29	0.03423
GO:0001570~vasculogenesis	10	0.03515
GO:0065003~macromolecular complex assembly	48	0.035248
GO:0033157~regulation of intracellular protein transport	8	0.035265
GO:0060341~regulation of cellular localization	26	0.035293
GO:0007167~enzyme linked receptor protein signaling pathway	40	0.036024
GO:0043281~regulation of caspase activity	11	0.036238
GO:0052548~regulation of endopeptidase activity	11	0.036238
GO:0001818~negative regulation of cytokine production	7	0.036605
GO:0050729~positive regulation of inflammatory response	7	0.036605
GO:0009820~alkaloid metabolic process	7	0.036605
GO:0031646~positive regulation of neurological system process	7	0.036605
GO:0046496~nicotinamide nucleotide metabolic process	7	0.036605
GO:0006769~nicotinamide metabolic process	7	0.036605
GO:0032271~regulation of protein polymerization	12	0.036637
GO:0018212~peptidyl-tyrosine modification	9	0.038413
GO:0046165~alcohol biosynthetic process	9	0.038413
GO:0018108~peptidyl-tyrosine phosphorylation	9	0.038413
GO:0006333~chromatin assembly or disassembly	19	0.039977
GO:0045637~regulation of myeloid cell differentiation	11	0.040928
GO:0007163~establishment or maintenance of cell polarity	8	0.041501
GO:0006910~phagocytosis, recognition	5	0.042114
GO:0045408~regulation of interleukin-6 biosynthetic process	5	0.042114
GO:0055074~calcium ion homeostasis	17	0.043456
GO:0010741~negative regulation of protein kinase cascade	7	0.043961
GO:0002637~regulation of immunoglobulin production	7	0.043961
GO:0051651~maintenance of location in cell	7	0.043961
GO:0051224~negative regulation of protein transport	6	0.04486
GO:0033032~regulation of myeloid cell apoptosis	4	0.045103
GO:0045579~positive regulation of B cell differentiation	4	0.045103
GO:0001810~regulation of type I hypersensitivity	4	0.045103
GO:0042308~negative regulation of protein import into nucleus	4	0.045103
GO:0015992~proton transport	11	0.046016
GO:0051276~chromosome organization	55	0.048184
GO:0009165~nucleotide biosynthetic process	27	0.048542

Supplementary Table 4. Correlation coefficients and p-values between co-expression modules and oligomeric procyanidins treatment.

Module	R	P-value
OPC_M1	0.15	0.77
OPC_M2	0.41	0.42
OPC_M3	0.36	0.48
OPC_M4	0.23	0.66
OPC_M5	-0.37	0.47
OPC_M6	-0.35	0.49
OPC_M7	0.79	0.06
OPC_M8	0.15	0.78
OPC_M9	0.15	0.78
OPC_M10	-0.45	0.38
OPC_M11	-0.02	0.98
OPC_M12	0.12	0.82
OPC_M13	0.06	0.91
OPC_M14	0.63	0.18
OPC_M15	-0.25	0.64
OPC_M16	-0.65	0.16
OPC_M17	-0.66	0.15
OPC_M18	-0.94	0.006
OPC_M19	-0.46	0.36
OPC_M20	0.49	0.32
OPC_M21	-0.43	0.39
OPC_M22	-0.08	0.87
OPC_M23	-0.21	0.70
OPC_M24	0.54	0.27
OPC_M25	0.97	0.001

Supplementary Table 5. Biological processes enriched in the genes in the OPC_M18 module

Term	Count	PValue
GO:0008219~cell death	107	5.05E-06
GO:0016265~death	107	6.92E-06
GO:0006468~protein amino acid phosphorylation	100	7.79E-06
GO:0009725~response to hormone stimulus	63	8.21E-06
GO:0045859~regulation of protein kinase activity	60	9.23E-06
GO:0051338~regulation of transferase activity	63	1.27E-05
GO:0043549~regulation of kinase activity	61	1.35E-05
GO:0019220~regulation of phosphate metabolic process	77	1.37E-05
GO:0051174~regulation of phosphorus metabolic process	77	1.37E-05
GO:0010033~response to organic substance	105	1.64E-05
GO:0006793~phosphorus metabolic process	134	1.67E-05
GO:0006796~phosphate metabolic process	134	1.67E-05
GO:0012501~programmed cell death	91	2.73E-05
GO:0007242~intracellular signaling cascade	164	3.35E-05
GO:0042325~regulation of phosphorylation	73	3.71E-05
GO:0043627~response to estrogen stimulus	25	4.83E-05
GO:0009719~response to endogenous stimulus	65	5.03E-05
GO:0006915~apoptosis	88	7.46E-05
GO:0006396~RNA processing	80	1.61E-04
GO:0048545~response to steroid hormone stimulus	36	1.67E-04
GO:0051270~regulation of cell motion	36	1.85E-04
GO:0044093~positive regulation of molecular function	84	2.17E-04
GO:0042254~ribosome biogenesis	26	2.20E-04
GO:0031327~negative regulation of cellular biosynthetic process	81	2.24E-04
GO:0022613~ribonucleoprotein complex biogenesis	34	2.27E-04
GO:0030334~regulation of cell migration	32	3.43E-04
GO:0045860~positive regulation of protein kinase activity	39	3.72E-04
GO:0010629~negative regulation of gene expression	73	4.30E-04
GO:0010558~negative regulation of macromolecule biosynthetic process	78	4.31E-04
GO:0009890~negative regulation of biosynthetic process	81	4.37E-04
GO:0010604~positive regulation of macromolecule metabolic process	113	4.74E-04
GO:0010605~negative regulation of macromolecule metabolic process	99	5.10E-04
GO:0016310~phosphorylation	106	6.04E-04
GO:0031399~regulation of protein modification process	47	7.40E-04
GO:0033674~positive regulation of kinase activity	39	7.50E-04
GO:0008630~DNA damage response, signal transduction resulting in induction of apoptosis	11	7.90E-04
GO:0051272~positive regulation of cell motion	21	9.54E-04
GO:0006261~DNA-dependent DNA replication	15	9.92E-04
GO:0016481~negative regulation of transcription	66	0.001014
GO:0043067~regulation of programmed cell death	106	0.00103
GO:0043085~positive regulation of catalytic activity	73	0.001046
GO:0010941~regulation of cell death	106	0.001155
GO:0007049~cell cycle	101	0.001501
GO:0040012~regulation of locomotion	33	0.001517
GO:0042981~regulation of apoptosis	104	0.001526
GO:0051347~positive regulation of transferase activity	39	0.001549
GO:0051172~negative regulation of nitrogen compound metabolic process	72	0.001584
GO:0045934~negative regulation of nucleobase, nucleoside, nucleotide and nucleic acid metabolic process	71	0.001701
GO:0042771~DNA damage response, signal transduction by p53 class mediator resulting in induction of apoptosis	7	0.001861
GO:0030335~positive regulation of cell migration	19	0.001868
GO:0030330~DNA damage response, signal transduction by p53 class mediator	9	0.002361
GO:0040017~positive regulation of locomotion	20	0.002367
GO:0051491~positive regulation of filopodium assembly	5	0.002383
GO:0051489~regulation of filopodium assembly	5	0.002383

GO:0006350~transcription	240	0.002388
GO:0009891~positive regulation of biosynthetic process	90	0.003203
GO:0022604~regulation of cell morphogenesis	24	0.003389
GO:0008285~negative regulation of cell proliferation	52	0.003537
GO:0007243~protein kinase cascade	53	0.003614
GO:0042770~DNA damage response, signal transduction	17	0.003666
GO:0034470~ncRNA processing	31	0.003767
GO:0007178~transmembrane receptor protein serine/threonine kinase signaling pathway	20	0.004228
GO:0045892~negative regulation of transcription, DNA-dependent	51	0.004338
GO:0031328~positive regulation of cellular biosynthetic process	88	0.004562
GO:0000079~regulation of cyclin-dependent protein kinase activity	13	0.004641
GO:0042060~wound healing	31	0.005153
GO:0060491~regulation of cell projection assembly	7	0.00536
GO:0048701~embryonic cranial skeleton morphogenesis	7	0.00536
GO:0033993~response to lipid	7	0.00536
GO:0032268~regulation of cellular protein metabolic process	64	0.005412
GO:0043065~positive regulation of apoptosis	59	0.005449
GO:0032270~positive regulation of cellular protein metabolic process	36	0.005528
GO:0051253~negative regulation of RNA metabolic process	51	0.006008
GO:0045449~regulation of transcription	287	0.006014
GO:0035023~regulation of Rho protein signal transduction	19	0.006165
GO:0051247~positive regulation of protein metabolic process	37	0.00618
GO:0043068~positive regulation of programmed cell death	59	0.006408
GO:0006364~rRNA processing	18	0.006482
GO:0010942~positive regulation of cell death	59	0.006932
GO:0008286~insulin receptor signaling pathway	10	0.007312
GO:0010557~positive regulation of macromolecule biosynthetic process	83	0.007732
GO:0006357~regulation of transcription from RNA polymerase II promoter	91	0.007822
GO:0043069~negative regulation of programmed cell death	50	0.008095
GO:0001817~regulation of cytokine production	29	0.008262
GO:0055088~lipid homeostasis	12	0.00832
GO:0007167~enzyme linked receptor protein signaling pathway	48	0.008347
GO:0001932~regulation of protein amino acid phosphorylation	28	0.008357
GO:0070482~response to oxygen levels	24	0.008473
GO:0043405~regulation of MAP kinase activity	24	0.008473
GO:0060548~negative regulation of cell death	50	0.008507
GO:0042127~regulation of cell proliferation	97	0.009018
GO:0001666~response to hypoxia	23	0.009174
GO:0044087~regulation of cellular component biogenesis	24	0.009215
GO:0015031~protein transport	94	0.00973
GO:0016072~rRNA metabolic process	18	0.009941
GO:0006917~induction of apoptosis	45	0.01028
GO:0048534~hemopoietic or lymphoid organ development	38	0.010512
GO:0006974~response to DNA damage stimulus	51	0.010514
GO:0006730~one-carbon metabolic process	20	0.010547
GO:0012502~induction of programmed cell death	45	0.010842
GO:0007264~small GTPase mediated signal transduction	43	0.011676
GO:0070201~regulation of establishment of protein localization	21	0.011715
GO:0043255~regulation of carbohydrate biosynthetic process	7	0.01219
GO:0045184~establishment of protein localization	94	0.012321
GO:0016311~dephosphorylation	25	0.012609
GO:0033554~cellular response to stress	72	0.012681
GO:0031346~positive regulation of cell projection organization	11	0.012777
GO:0007005~mitochondrion organization	23	0.012802
GO:0032880~regulation of protein localization	23	0.012802
GO:0008629~induction of apoptosis by intracellular signals	12	0.012815
GO:0034660~ncRNA metabolic process	34	0.013434
GO:0051789~response to protein stimulus	19	0.01367

GO:0048703~embryonic viscerocranium morphogenesis	4	0.014357
GO:0010887~negative regulation of cholesterol storage	4	0.014357
GO:0043066~negative regulation of apoptosis	48	0.015118
GO:0007507~heart development	32	0.015177
GO:0002520~immune system development	39	0.015919
GO:0007010~cytoskeleton organization	57	0.016123
GO:0051271~negative regulation of cell motion	13	0.016151
GO:0006984~ER-nuclear signaling pathway	9	0.016328
GO:0019637~organophosphate metabolic process	30	0.017137
GO:0051301~cell division	41	0.017256
GO:0043406~positive regulation of MAP kinase activity	18	0.017716
GO:0043009~chordate embryonic development	45	0.017751
GO:0000165~MAPKKK cascade	28	0.018124
GO:0002474~antigen processing and presentation of peptide antigen via MHC class I	6	0.019187
GO:0045935~positive regulation of nucleobase, nucleoside, nucleotide and nucleic acid metabolic process	77	0.019429
GO:0014706~striated muscle tissue development	20	0.019435
GO:0051173~positive regulation of nitrogen compound metabolic process	79	0.020331
GO:0009792~embryonic development ending in birth or egg hatching	45	0.020606
GO:0006259~DNA metabolic process	64	0.020812
GO:0060627~regulation of vesicle-mediated transport	17	0.020987
GO:0031401~positive regulation of protein modification process	28	0.021955
GO:0030509~BMP signaling pathway	10	0.022663
GO:0031344~regulation of cell projection organization	16	0.022699
GO:0010888~negative regulation of lipid storage	5	0.022739
GO:0006264~mitochondrial DNA replication	4	0.023338
GO:0032365~intracellular lipid transport	4	0.023338
GO:0090003~regulation of establishment of protein localization to plasma membrane	4	0.023338
GO:0008380~RNA splicing	39	0.024033
GO:0051291~protein heterooligomerization	11	0.025195
GO:0006605~protein targeting	31	0.025197
GO:0051130~positive regulation of cellular component organization	27	0.025649
GO:0033630~positive regulation of cell adhesion mediated by integrin	3	0.026145
GO:0060317~cardiac epithelial to mesenchymal transition	3	0.026145
GO:0006644~phospholipid metabolic process	28	0.026391
GO:0006260~DNA replication	28	0.026391
GO:0007179~transforming growth factor beta receptor signaling pathway	12	0.027023
GO:0032869~cellular response to insulin stimulus	13	0.028287
GO:0045669~positive regulation of osteoblast differentiation	7	0.028551
GO:0030036~actin cytoskeleton organization	32	0.028654
GO:0032259~methylation	14	0.029105
GO:0008104~protein localization	103	0.029173
GO:0065003~macromolecular complex assembly	80	0.029583
GO:0045937~positive regulation of phosphate metabolic process	17	0.029718
GO:0010562~positive regulation of phosphorus metabolic process	17	0.029718
GO:0030100~regulation of endocytosis	12	0.030196
GO:0032870~cellular response to hormone stimulus	21	0.030268
GO:0006470~protein amino acid dephosphorylation	21	0.030268
GO:0017015~regulation of transforming growth factor beta receptor signaling pathway	9	0.030313
GO:0030097~hemopoiesis	33	0.030632
GO:0060537~muscle tissue development	20	0.03091
GO:0043414~biopolymer methylation	13	0.031345
GO:0051254~positive regulation of RNA metabolic process	60	0.031955
GO:0008360~regulation of cell shape	11	0.032018
GO:0042476~odontogenesis	11	0.032018
GO:0030099~myeloid cell differentiation	16	0.03243
GO:0035295~tube development	31	0.033169
GO:0030857~negative regulation of epithelial cell differentiation	4	0.034699
GO:0046321~positive regulation of fatty acid oxidation	4	0.034699

GO:0032042~mitochondrial DNA metabolic process	4	0.034699
GO:0046785~microtubule polymerization	4	0.034699
GO:0030278~regulation of ossification	14	0.035179
GO:0006195~purine nucleotide catabolic process	8	0.035189
GO:0051592~response to calcium ion	11	0.035875
GO:0032570~response to progesterone stimulus	6	0.037757
GO:0010627~regulation of protein kinase cascade	34	0.037804
GO:0006986~response to unfolded protein	13	0.038161
GO:0045893~positive regulation of transcription, DNA-dependent	59	0.03848
GO:0006869~lipid transport	22	0.03862
GO:0006461~protein complex assembly	62	0.03864
GO:0070271~protein complex biogenesis	62	0.03864
GO:0005979~regulation of glycogen biosynthetic process	5	0.039376
GO:0010962~regulation of glucan biosynthetic process	5	0.039376
GO:0032885~regulation of polysaccharide biosynthetic process	5	0.039376
GO:0008624~induction of apoptosis by extracellular signals	18	0.040106
GO:0010628~positive regulation of gene expression	70	0.040749
GO:0034976~response to endoplasmic reticulum stress	8	0.040753
GO:0051252~regulation of RNA metabolic process	197	0.041535
GO:0051094~positive regulation of developmental process	37	0.041934
GO:0042157~lipoprotein metabolic process	14	0.04211
GO:0045596~negative regulation of cell differentiation	30	0.042567
GO:0051056~regulation of small GTPase mediated signal transduction	34	0.043629
GO:0030336~negative regulation of cell migration	11	0.044531
GO:0042327~positive regulation of phosphorylation	16	0.044902
GO:0034620~cellular response to unfolded protein	6	0.045746
GO:0030968~endoplasmic reticulum unfolded protein response	6	0.045746
GO:0008283~cell proliferation	54	0.046341
GO:0017038~protein import	20	0.046812
GO:0051259~protein oligomerization	25	0.046869
GO:0032319~regulation of Rho GTPase activity	7	0.047465
GO:0010876~lipid localization	23	0.047768
GO:0044272~sulfur compound biosynthetic process	10	0.04778
GO:0010746~regulation of plasma membrane long-chain fatty acid transport	3	0.048958
GO:0010891~negative regulation of sequestering of triglyceride	3	0.048958
GO:0032367~intracellular cholesterol transport	3	0.048958
GO:0032366~intracellular sterol transport	3	0.048958
GO:0010748~negative regulation of plasma membrane long-chain fatty acid transport	3	0.048958
GO:0007032~endosome organization	5	0.049712
GO:0032881~regulation of polysaccharide metabolic process	5	0.049712

Supplementary Table 6. Biological processes enriched in the genes in the OPC_M25 module

Term	Count	PValue
GO:0009615~response to virus	36	1.91E-11
GO:0042127~regulation of cell proliferation	124	2.30E-09
GO:0042981~regulation of apoptosis	124	8.77E-09
GO:0010941~regulation of cell death	125	1.02E-08
GO:0043067~regulation of programmed cell death	124	1.50E-08
GO:0010033~response to organic substance	113	1.78E-08
GO:0008285~negative regulation of cell proliferation	64	5.08E-07
GO:0030335~positive regulation of cell migration	25	1.06E-06
GO:0016477~cell migration	52	1.10E-06
GO:0009891~positive regulation of biosynthetic process	103	1.29E-06
GO:0010942~positive regulation of cell death	72	1.31E-06
GO:0030334~regulation of cell migration	37	1.44E-06
GO:0043065~positive regulation of apoptosis	71	1.71E-06
GO:0040017~positive regulation of locomotion	26	1.95E-06
GO:0031328~positive regulation of cellular biosynthetic process	101	2.04E-06
GO:0043068~positive regulation of programmed cell death	71	2.22E-06
GO:0040012~regulation of locomotion	39	4.83E-06
GO:0051270~regulation of cell motion	39	5.49E-06
GO:0007242~intracellular signaling cascade	162	6.19E-06
GO:0034097~response to cytokine stimulus	22	6.33E-06
GO:0051272~positive regulation of cell motion	25	6.71E-06
GO:0010557~positive regulation of macromolecule biosynthetic process	95	8.19E-06
GO:0048660~regulation of smooth muscle cell proliferation	16	9.06E-06
GO:0048661~positive regulation of smooth muscle cell proliferation	13	9.72E-06
GO:0010604~positive regulation of macromolecule metabolic process	117	1.27E-05
GO:0051173~positive regulation of nitrogen compound metabolic process	92	2.25E-05
GO:0051674~localization of cell	52	2.49E-05
GO:0048870~cell motility	52	2.49E-05
GO:0001525~angiogenesis	31	2.91E-05
GO:0001666~response to hypoxia	29	2.99E-05
GO:0009725~response to hormone stimulus	59	3.22E-05
GO:0010324~membrane invagination	40	5.17E-05
GO:0006897~endocytosis	40	5.17E-05
GO:0046697~decidualization	8	5.86E-05
GO:0010628~positive regulation of gene expression	83	5.94E-05
GO:0007167~enzyme linked receptor protein signaling pathway	55	6.05E-05
GO:0009611~response to wounding	77	6.55E-05
GO:0033273~response to vitamin	18	7.32E-05
GO:0070482~response to oxygen levels	29	7.77E-05
GO:0002237~response to molecule of bacterial origin	21	8.46E-05
GO:0009719~response to endogenous stimulus	62	8.56E-05
GO:0045935~positive regulation of nucleobase, nucleoside, nucleotide and nucleic acid metabolic process	87	9.19E-05
GO:0006928~cell motion	70	9.48E-05
GO:0051789~response to protein stimulus	24	9.49E-05
GO:0045941~positive regulation of transcription	80	1.05E-04
GO:0045893~positive regulation of transcription, DNA-dependent	70	1.09E-04
GO:0006955~immune response	94	1.12E-04
GO:0042330~taxis	31	1.31E-04
GO:0006935~chemotaxis	31	1.31E-04
GO:0051254~positive regulation of RNA metabolic process	70	1.40E-04
GO:0048545~response to steroid hormone stimulus	35	1.56E-04
GO:0016265~death	97	1.59E-04
GO:0008284~positive regulation of cell proliferation	62	1.60E-04
GO:0001893~maternal placenta development	8	1.86E-04
GO:0008219~cell death	96	1.92E-04

GO:0012501~programmed cell death	84	1.95E-04
GO:0048514~blood vessel morphogenesis	37	2.18E-04
GO:0031349~positive regulation of defense response	18	2.77E-04
GO:0006917~induction of apoptosis	50	2.79E-04
GO:0042254~ribosome biogenesis	25	2.85E-04
GO:0006357~regulation of transcription from RNA polymerase II promoter	96	2.91E-04
GO:0012502~induction of programmed cell death	50	3.01E-04
GO:0016044~membrane organization	57	3.13E-04
GO:0010629~negative regulation of gene expression	71	3.28E-04
GO:0006915~apoptosis	82	3.29E-04
GO:0007626~locomotory behavior	44	3.79E-04
GO:0007584~response to nutrient	27	4.19E-04
GO:0048584~positive regulation of response to stimulus	39	4.87E-04
GO:0032496~response to lipopolysaccharide	18	5.40E-04
GO:0003006~reproductive developmental process	42	5.44E-04
GO:0032103~positive regulation of response to external stimulus	16	5.79E-04
GO:0032101~regulation of response to external stimulus	29	6.27E-04
GO:0051384~response to glucocorticoid stimulus	18	6.32E-04
GO:0031960~response to corticosteroid stimulus	19	6.38E-04
GO:0006796~phosphate metabolic process	120	7.37E-04
GO:0006793~phosphorus metabolic process	120	7.37E-04
GO:0006351~transcription, DNA-dependent	45	7.85E-04
GO:0045785~positive regulation of cell adhesion	15	9.15E-04
GO:0045071~negative regulation of viral genome replication	5	9.31E-04
GO:0001568~blood vessel development	39	0.00101
GO:0032774~RNA biosynthetic process	45	0.001046
GO:0007178~transmembrane receptor protein serine/threonine kinase signaling pathway	21	0.001051
GO:0034341~response to interferon-gamma	6	0.001129
GO:0033189~response to vitamin A	12	0.001159
GO:0042493~response to drug	35	0.001405
GO:0045944~positive regulation of transcription from RNA polymerase II promoter	53	0.001505
GO:0045765~regulation of angiogenesis	15	0.001519
GO:0006366~transcription from RNA polymerase II promoter	37	0.001565
GO:0043066~negative regulation of apoptosis	51	0.001569
GO:0001944~vasculature development	39	0.001588
GO:0030155~regulation of cell adhesion	25	0.001597
GO:0043434~response to peptide hormone stimulus	27	0.001809
GO:0007264~small GTPase mediated signal transduction	45	0.001909
GO:0006468~protein amino acid phosphorylation	85	0.001932
GO:0048525~negative regulation of viral reproduction	5	0.002014
GO:0050792~regulation of viral reproduction	8	0.002049
GO:0010605~negative regulation of macromolecule metabolic process	92	0.002057
GO:0043069~negative regulation of programmed cell death	51	0.002103
GO:0060548~negative regulation of cell death	51	0.002215
GO:0031667~response to nutrient levels	32	0.002239
GO:0006916~anti-apoptosis	33	0.002369
GO:0031327~negative regulation of cellular biosynthetic process	73	0.002438
GO:0009890~negative regulation of biosynthetic process	74	0.002777
GO:0016310~phosphorylation	98	0.002925
GO:0070555~response to interleukin-1	7	0.003101
GO:0051094~positive regulation of developmental process	41	0.003135
GO:0030031~cell projection assembly	17	0.003456
GO:0009628~response to abiotic stimulus	51	0.003506
GO:0030030~cell projection organization	51	0.003506
GO:0050727~regulation of inflammatory response	16	0.003612
GO:0006984~ER-nuclear signaling pathway	10	0.00367
GO:0002831~regulation of response to biotic stimulus	9	0.003798
GO:0045087~innate immune response	24	0.003806
GO:0009612~response to mechanical stimulus	13	0.004421

GO:0010558~negative regulation of macromolecule biosynthetic process	70	0.004527
GO:0030278~regulation of ossification	16	0.004667
GO:0050729~positive regulation of inflammatory response	9	0.004767
GO:0007169~transmembrane receptor protein tyrosine kinase signaling pathway	34	0.004788
GO:0045892~negative regulation of transcription, DNA-dependent	49	0.004875
GO:0009967~positive regulation of signal transduction	42	0.005214
GO:0043281~regulation of caspase activity	16	0.005282
GO:0051726~regulation of cell cycle	46	0.005364
GO:0016481~negative regulation of transcription	60	0.005545
GO:0048732~gland development	23	0.006024
GO:0007243~protein kinase cascade	50	0.006287
GO:0009991~response to extracellular stimulus	33	0.006546
GO:0045069~regulation of viral genome replication	6	0.006551
GO:0051253~negative regulation of RNA metabolic process	49	0.006649
GO:0002230~positive regulation of defense response to virus by host	4	0.006808
GO:0043280~positive regulation of caspase activity	13	0.006856
GO:0010952~positive regulation of peptidase activity	13	0.006856
GO:0016192~vesicle-mediated transport	72	0.006932
GO:0045859~regulation of protein kinase activity	47	0.007029
GO:0001936~regulation of endothelial cell proliferation	9	0.007262
GO:0051101~regulation of DNA binding	21	0.007325
GO:0006952~defense response	76	0.007337
GO:0052548~regulation of endopeptidase activity	16	0.007528
GO:0045934~negative regulation of nucleobase, nucleoside, nucleotide and nucleic acid metabolic process	65	0.007533
GO:0000122~negative regulation of transcription from RNA polymerase II promoter	38	0.007588
GO:0045766~positive regulation of angiogenesis	8	0.007705
GO:0030330~DNA damage response, signal transduction by p53 class mediator	8	0.007705
GO:0007179~transforming growth factor beta receptor signaling pathway	13	0.007869
GO:0010647~positive regulation of cell communication	45	0.007875
GO:0022613~ribonucleoprotein complex biogenesis	28	0.007951
GO:0030522~intracellular receptor-mediated signaling pathway	15	0.008057
GO:0001816~cytokine production	11	0.008116
GO:0043549~regulation of kinase activity	48	0.008177
GO:0070271~protein complex biogenesis	64	0.008266
GO:0006461~protein complex assembly	64	0.008266
GO:0002683~negative regulation of immune system process	16	0.008426
GO:0060348~bone development	21	0.008775
GO:0032526~response to retinoic acid	9	0.008827
GO:0001503~ossification	20	0.008849
GO:0042771~DNA damage response, signal transduction by p53 class mediator resulting in induction of apoptosis	6	0.00909
GO:0001938~positive regulation of endothelial cell proliferation	6	0.00909
GO:0032355~response to estradiol stimulus	12	0.009325
GO:0051240~positive regulation of multicellular organismal process	35	0.009918
GO:0051172~negative regulation of nitrogen compound metabolic process	65	0.009978
GO:0006350~transcription	224	0.010431
GO:0030099~myeloid cell differentiation	17	0.010656
GO:0002697~regulation of immune effector process	18	0.010723
GO:0009617~response to bacterium	29	0.010849
GO:0043242~negative regulation of protein complex disassembly	10	0.011032
GO:0052547~regulation of peptidase activity	16	0.01164
GO:0045449~regulation of transcription	272	0.011682
GO:0007565~female pregnancy	19	0.011724
GO:0022407~regulation of cell-cell adhesion	7	0.012532
GO:0051797~regulation of hair follicle development	4	0.012684
GO:0002755~MyD88-dependent toll-like receptor signaling pathway	4	0.012684
GO:0042634~regulation of hair cycle	4	0.012684
GO:0016126~sterol biosynthetic process	9	0.012693
GO:0007610~behavior	59	0.01296

GO:0051098~regulation of binding	24	0.013228
GO:0050921~positive regulation of chemotaxis	8	0.014333
GO:0008610~lipid biosynthetic process	43	0.014546
GO:0010038~response to metal ion	21	0.014557
GO:0043407~negative regulation of MAP kinase activity	9	0.015035
GO:0009057~macromolecule catabolic process	91	0.015501
GO:0006576~biogenic amine metabolic process	17	0.015692
GO:0002221~pattern recognition receptor signaling pathway	6	0.016039
GO:0051338~regulation of transferase activity	48	0.016637
GO:0019220~regulation of phosphate metabolic process	60	0.016772
GO:0051174~regulation of phosphorus metabolic process	60	0.016772
GO:0042325~regulation of phosphorylation	58	0.016791
GO:0030595~leukocyte chemotaxis	9	0.017673
GO:0051336~regulation of hydrolase activity	44	0.018236
GO:0051222~positive regulation of protein transport	13	0.018594
GO:0001501~skeletal system development	42	0.018648
GO:0043405~regulation of MAP kinase activity	22	0.019115
GO:0045778~positive regulation of ossification	7	0.019291
GO:0010740~positive regulation of protein kinase cascade	25	0.019339
GO:0008354~germ cell migration	5	0.019565
GO:0006809~nitric oxide biosynthetic process	5	0.019565
GO:0014910~regulation of smooth muscle cell migration	5	0.019565
GO:0032870~cellular response to hormone stimulus	21	0.019844
GO:0009743~response to carbohydrate stimulus	12	0.02012
GO:0050920~regulation of chemotaxis	8	0.020549
GO:0030501~positive regulation of bone mineralization	6	0.020558
GO:0002645~positive regulation of tolerance induction	4	0.020686
GO:0002643~regulation of tolerance induction	4	0.020686
GO:0044265~cellular macromolecule catabolic process	84	0.022458
GO:0050866~negative regulation of cell activation	12	0.022567
GO:0002274~myeloid leukocyte activation	10	0.022926
GO:0045669~positive regulation of osteoblast differentiation	7	0.023454
GO:0043331~response to dsRNA	7	0.023454
GO:0051345~positive regulation of hydrolase activity	26	0.023805
GO:0060326~cell chemotaxis	9	0.023918
GO:0007182~common-partner SMAD protein phosphorylation	3	0.023984
GO:0006970~response to osmotic stress	8	0.024269
GO:0006919~activation of caspase activity	11	0.02439
GO:0045088~regulation of innate immune response	11	0.02439
GO:0006954~inflammatory response	42	0.02461
GO:0043388~positive regulation of DNA binding	13	0.025592
GO:0045637~regulation of myeloid cell differentiation	13	0.025592
GO:0051099~positive regulation of binding	14	0.02561
GO:0045184~establishment of protein localization	88	0.025752
GO:0050688~regulation of defense response to virus	6	0.025832
GO:0048008~platelet-derived growth factor receptor signaling pathway	6	0.025832
GO:0002758~innate immune response-activating signal transduction	6	0.025832
GO:0070169~positive regulation of biomineral formation	6	0.025832
GO:0002218~activation of innate immune response	6	0.025832
GO:0006525~arginine metabolic process	5	0.026237
GO:0046209~nitric oxide metabolic process	5	0.026237
GO:0006730~one-carbon metabolic process	18	0.027758
GO:0006469~negative regulation of protein kinase activity	15	0.027761
GO:0015031~protein transport	87	0.027885
GO:0008630~DNA damage response, signal transduction resulting in induction of apoptosis	8	0.028427
GO:0030097~hemopoiesis	32	0.02845
GO:0042060~wound healing	27	0.028671
GO:0009259~ribonucleotide metabolic process	22	0.029012
GO:0018212~peptidyl-tyrosine modification	10	0.029597

GO:0040007~growth	26	0.030268
GO:0031958~corticosteroid receptor signaling pathway	4	0.030857
GO:0007184~SMAD protein nuclear translocation	4	0.030857
GO:0043627~response to estrogen stimulus	17	0.031072
GO:0051092~positive regulation of NF-kappaB transcription factor activity	9	0.031564
GO:0050714~positive regulation of protein secretion	9	0.031564
GO:0051607~defense response to virus	6	0.031898
GO:0032570~response to progesterone stimulus	6	0.031898
GO:0043123~positive regulation of I-kappaB kinase/NF-kappaB cascade	16	0.032106
GO:0000060~protein import into nucleus, translocation	8	0.03304
GO:0048520~positive regulation of behavior	8	0.03304
GO:0001952~regulation of cell-matrix adhesion	7	0.033491
GO:0045428~regulation of nitric oxide biosynthetic process	7	0.033491
GO:0030728~ovulation	5	0.034112
GO:0002224~toll-like receptor signaling pathway	5	0.034112
GO:0050900~leukocyte migration	11	0.0343
GO:0048754~branching morphogenesis of a tube	12	0.034553
GO:0031589~cell-substrate adhesion	16	0.0348
GO:0042592~homeostatic process	85	0.035657
GO:0033673~negative regulation of kinase activity	15	0.035927
GO:0051591~response to cAMP	9	0.035951
GO:0043122~regulation of I-kappaB kinase/NF-kappaB cascade	17	0.036237
GO:0051129~negative regulation of cellular component organization	21	0.037073
GO:0048534~hemopoietic or lymphoid organ development	34	0.03746
GO:0035023~regulation of Rho protein signal transduction	16	0.037654
GO:0001763~morphogenesis of a branching structure	13	0.037681
GO:0010035~response to inorganic substance	28	0.037719
GO:0045884~regulation of survival gene product expression	6	0.038781
GO:0001569~patterning of blood vessels	6	0.038781
GO:0045429~positive regulation of nitric oxide biosynthetic process	6	0.038781
GO:0002252~immune effector process	20	0.038997
GO:0007507~heart development	29	0.039464
GO:0048872~homeostasis of number of cells	16	0.040673
GO:0043244~regulation of protein complex disassembly	10	0.04197
GO:0043392~negative regulation of DNA binding	10	0.04197
GO:0051100~negative regulation of binding	11	0.042289
GO:0009199~ribonucleoside triphosphate metabolic process	18	0.043028
GO:0022408~negative regulation of cell-cell adhesion	4	0.043168
GO:0052173~response to defenses of other organism during symbiotic interaction	4	0.043168
GO:0060416~response to growth hormone stimulus	4	0.043168
GO:0050691~regulation of defense response to virus by host	4	0.043168
GO:0009125~nucleoside monophosphate catabolic process	4	0.043168
GO:0045684~positive regulation of epidermis development	4	0.043168
GO:0052200~response to host defenses	4	0.043168
GO:0075136~response to host	4	0.043168
GO:0021700~developmental maturation	16	0.04386
GO:0016311~dephosphorylation	22	0.044962
GO:0051798~positive regulation of hair follicle development	3	0.045043
GO:0010544~negative regulation of platelet activation	3	0.045043
GO:0045091~regulation of retroviral genome replication	3	0.045043
GO:0045896~regulation of transcription, mitotic	3	0.045043
GO:0006926~virus-infected cell apoptosis	3	0.045043
GO:0070498~interleukin-1-mediated signaling pathway	3	0.045043
GO:0001757~somite specification	3	0.045043
GO:0007568~aging	17	0.045114
GO:0030509~BMP signaling pathway	9	0.045914
GO:0016568~chromatin modification	35	0.046203
GO:0010212~response to ionizing radiation	11	0.046723
GO:0051091~positive regulation of transcription factor activity	11	0.046723

GO:0051241~negative regulation of multicellular organismal process	23	0.047682
GO:0046700~heterocycle catabolic process	13	0.049058

Supplementary Table 7. Overrepresented Gene Ontology categories regulated by IL-32 α in mouse carotid endothelium *in vivo*.

Molecular and Cellular Functions	Number of Changed Genes			
	Non-Tg_LCA vs. Non-Tg RCA	IL-32 α -Tg LCA vs. IL-32 α -Tg RCA	IL-32 α -Tg RCA vs. Non-Tg RCA	IL-32 α -Tg LCA vs. Non-Tg LCA
Transport	639	344	49	47
Translation	136	108	10	0
Response to stress	463	280	57	82
Cell cycle	254	135	20	35
Cell adhesion	131	89	28	32
Signal transduction	614	270	81	103
Cell proliferation	121	70	12	21
Homeostasis	208	103	22	28
Cell differentiation	473	237	59	48
Immune response	136	84	18	47
Inflammatory response	86	50	6	33
Behavior	73	36	12	9
Apoptotic process	235	109	24	25
Total	4251	2195	547	379

Supplementary Table 8. Changed genes that are involved in inflammation and cell growth and proliferation that regulated by IL-32 α in mouse carotid endothelium *in vivo*.

Gene Symbol						hIL-32 α -Tg LCA vs. Non-Tg LCA						
Inflammation												
Down-regulated			Fold-Changes	P value	Up-regulated			Fold-Changes	P value			
Ptafr			0.24	<0.001	Saa3			5.44	<0.001			
BC013712			0.25	<0.001								
Slc11a1			0.30	<0.001								
Fcer1g			0.32	<0.001								
Il1b			0.32	<0.001								
Clec7a			0.33	<0.001								
Ccr1			0.34	0.008								
Tlr13			0.34	0.002								
Il1rn			0.35	0.001								
Csflr			0.36	<0.001								
Cd14			0.38	0.006								
Ccl22			0.38	0.005								
Kdm6b			0.38	<0.001								
Pik3cg			0.39	0.002								
Mefv			0.39	0.001								
Fcgr3			0.39	<0.001								
Itgb2			0.39	<0.001								
Pla2g7			0.40	0.003								
Ccl3			0.40	0.003								
Hck			0.42	0.001								
Syk			0.42	<0.001								
Tlr8			0.43	0.004								
Serpib9			0.43	<0.001								
Nlrp3			0.45	0.002								
Adam8			0.46	0.001								
Lyn			0.47	0.002								
Tgfb1			0.48	<0.001								
Alox5ap			0.48	<0.001								
Tlr2			0.48	0.015								
Myd88			0.49	0.001								
Tnfrsf1b			0.49	0.005								
Osm			0.50	0.024								

Cell Cycle and Cell Proliferation												
Down-regulated			Fold-Changes	P value	Up-regulated			Fold-Changes	P value			
Ccnb2			0.12	<0.001	None							
Itgax			0.18	<0.001								
Igfbp3			0.21	0.002								
Kif20a			0.28	0.001								
Cep55			0.29	0.001								
Slc11a1			0.30	<0.001								
Ptprc			0.31	<0.001								
Il1b			0.32	<0.001								
Plk1			0.32	0.002								
Cdkn1a			0.33	<0.001								
Cks2			0.34	0.016								

Dusp1	0.34	<0.001
Mki67	0.34	<0.001
Ptpn6	0.35	<0.001
Ccna2	0.36	<0.001
Malt1	0.36	0.003
Cxcr4	0.36	<0.001
Cd74	0.36	<0.001
Prkcd	0.36	0.004
Ndc80	0.37	0.027
Kif23	0.37	0.001
Tpx2	0.37	<0.001
Anln	0.38	0.002
Itgam	0.39	0.001
C5ar1	0.39	0.028
Slfn1	0.39	<0.001
Top2a	0.40	0.010
Ect2	0.40	0.011
Rgs2	0.42	<0.001
Itgal	0.43	0.005
Tet2	0.44	<0.001
Nusap1	0.45	0.004
Rasgrp4	0.45	0.014
Kif11	0.45	<0.001
Dock2	0.45	<0.001
Stat4	0.46	0.049
Kif2c	0.46	<0.001
Nuf2	0.47	<0.001
Txnip	0.48	<0.001
D2Erttd750e	0.48	0.001
Sesn2	0.48	0.006
Racgap1	0.49	0.014
Gpr132	0.49	0.022
Rassf2	0.49	0.001

Supplementary Table 9. List and sequences of qPCR primers for mRNA expression.

Primer Name		Sequences
18s	For	5'-AGGAATTGACGGAAGGGCACCA-3'
	Rev	5'-GTGCAGCCCCGGACATCTAAG-3'
VCAM-1	For	5'-TCTTGGGAGCCTCAACGGTA-3'
	Rev	5'-CAAGTGAGGGCCATGGAGTC-3'
ICAM-1	For	5'-GGACTCACCTGCTGGTCTCT-3'
	Rev	5'-CCCCTGCGATCTAGGAATTT-3'
TIMP3	For	5'-CACGGAAGCCTCTGAAAGTC-3'
	Rev	5'-TCCCACCTCTCCACAAAGTT-3'
RECK	For	5'-ATAGGCTGCAAACCCATCAT-3'
	Rev	5'- CCGTAACATCCCAGCACATA-3'
IL-1 β	For	5'-AGAGCCTGTGTTTTCTCCT-3'
	Rev	5'-GAGAGTGCTGCCTAATGTCC-3'
eNOS	For	5'-ATCCAGTGCCCTGCTTCA-3'
	Rev	5'-GCAGGGCAAGTTAGGATCAG-3'
MMP9	For	5'-ACGACATAGACGGCATCCA-3'
	Rev	5'-GCTGTGGTTCAGTTGTGGTG-3'
DGCR8	For	5'-GGGGTTCCTTACTACGCATGT-3'
	Rev	5'-CACACTCTTGTCAGAGGTCTCCT-3'
DROSHA	For	5'- ATGCAAGGCAATACGTGTCAT -3'
	Rev	5'- TTTTGGGGTCTGAAAGCTGGT-3'
DICER1	For	5'-AGATGGAGGCGGAGTTCAG-3'
	Rev	5'-CAATGAGCAGGTTGGTCTCA-3'
RPRD2	For	5'-GCAGCTTCCCATAATGCCAG-3'
	Rev	5'-TTGGATTCGTGGATGTTTGTGA-3'
DDX5	For	5'-GCACCGCACAAGAGGTAGAT-3'
	Rev	5'-ATCCACTGAGAGCAACTGGC-3'
KHSRP	For	5'-GCTGGAAGATGGAGACCAGC-3'
	Rev	5'-GGGTCCCAGTTGAGAACCAA-3'
XRN1	For	5'-GGTCTTTATGCTTGGAACCTCCT-3'
	Rev	5'-AATCAAGTCTCCTGAATCCTGAA-3'