Online Supplement: Detailed Methods Section

Animal care compliance

All animal procedures were conducted according to the "Guide for the Care and Use of Laboratory Animals" (8th Edition. 2011), AVMA Guidelines for the Euthanasia of Animals: (2013 Edition) and were approved by the Institutional Animal Care and Use Committees at the University of Alabama, Birmingham, USA.

Mice and coronary ligation surgery

8-12 weeks C57BL/6 (wild type; WT) and 12/15 LOX null mice (12/15LOX$^{-/-}$) on the C57BL/6 genetic background were obtained from The Jackson Laboratory (Bar Harbor, Maine, USA) and were maintained under constant temperature (19.8-22.2°C). The mice were given free access to water and standard chow diet. To induce MI, WT and 12/15LOX$^{-/-}$ were subjected to the surgical ligation of the left anterior descending coronary artery, as described previously.[1, 2] In brief, the mice were anesthetized with 2% isoflurane and the left anterior descending coronary artery was permanently ligated using minimally invasive surgery. Prior to MI surgery, carprofen (5 mg/kg SQ) and buprenorphine (0.1 mg/kg SQ) were administered to reduce pain.[3]

Dosing for HO-1 inhibition

Tin protoporphyrin IX (1mg/kg/day; SQ) was injected post-MI 3 hr in 12/15LOX$^{-/-}$ mice till d1 or till d5 post-MI.

Autopsy and post-MI survival analysis

The mice were checked daily for 28 days post-MI survival analysis. At autopsy, cardiac rupture was confirmed if there were large blood clots in the thoracic cavity and the LV rupture site was seen.[4, 5]

Echocardiography

For the echocardiography analysis, mice were anesthetized using 0.8 –1.0% isoflurane in a 100% oxygen mix. Electrocardiograms and heart rates were monitored using a surface electrocardiogram. Images were acquired using the Vevo 770 *in vivo* imaging system (Visual Sonics) equipped with probes up to 40 MHz and a resolution of 30 µm. Short and long axis images were acquired at heart rates >400 beats/min to achieve physiologically relevant measurements. Measurements were taken from the two dimensional parasternal long-axis (B-mode) and short-axis (m mode) recordings from the mid-papillary region. Echocardiographic studies were performed before necropsy for day (d) 0 control mice and for d1, d5 and d28 post-MI mice. For each variable, three images from consecutive cardiac cycles were measured and averaged by operator blinded to genotype.

Necropsy and infarct area analysis

No-MI control day (d0), d1 or d5 post-MI, mice were anesthetized under isoflurane briefly, then mice were maintained under anesthesia using 2% isoflurane in 100% oxygen mix and heparin (4 IU/g) was injected. The blood was collected from carotid artery for plasma separation. The chest cavity was opened and the left ventricle was perfused with 2-3 ml cardioplegic solution; then heart, lung and spleen were removed. The spleen, lungs, LV and right ventricle were separated and weighed individually. The LV was divided into apex (infarcted area), mid-cavity, and base (remote area) under microscope. All three LV sections and the right ventricle were stained with 1% 2,3,5-triphenyltetrazolium chloride (TTC) (Sigma) and photographed for infarct area determination by using Adobe Photoshop CS5 (64 bit) software. Left ventricle infarct (LVI) and -remote (LVC) regions were separated under the microscope for precise separation of infarcted and remote area and were individually snap frozen and stored at -80°C. The mid-cavity

section was fixed in 10% zinc-formalin and paraffin-embedded for histological examination. The lung mass and tibia were removed, and the wet and dry weights of lungs (24 hr after necropsy) and the tibia length were determined.[5]

Mass spectrometric analysis of lipid mediators

LC-MS/MS (AB/Sciex API-4000 and 6500 Q TRAP mass spectrometer) was used to analyze extracted plasma lipids. Briefly, 100 µl aliquots of plasma were acidified by addition of 850 µL of 6 N HCl (pH 2.3) in water, vortexed and incubated for 15 min on ice. Deuterated standards for arachidonic acid, hydroxyoctadecadienoic (HODEs) and hydroperoxyeicosatetraenoic acids (HETEs; 20 ng of each, Cayman Chemical) were spiked into the samples prior to extraction. Samples were subsequently passed through solid phase extraction (SPE) columns (Oasis® HLB 30 mg 1 ml) using a light vacuum such that each sample took approximately 2 min to pass through. Then, the SPE columns were washed with 5% methanol and lipids were eluted using 1 ml 100% methanol, dried under argon and resuspended in 100 µl of 80% methanol. Samples (50 µl) were injected onto a Luna C18 (2)-HST reverse-phase LC column (2x100 mm, 2.5 µm ID, Phenomenex) using a Shimadzu auto-sampler with linear gradient elution at 0.3 ml/min (mobile phase A: 0.1% formic acid in H_2O, mobile phase B: 0.1% formic acid in acetonitrile). Gradient elution was as follows: from 0 min to 2 min hold at 50% B, from 2-3 min increase to 60% B, from 3-15 min an increase to 65% B, from 15-17 min hold at 65% B, from 17-19 min an increase to 100% B, from 19-21 min hold at 100% B, from 21-23 min decrease back to 50% B and a hold for the system to equilibrate to initial conditions until 27 min. A standard curve (0.1, 1, 2.5, 5, 7.5, 10, 25, 50, 75, 100, 250, 500, 750 and 1000 ng/ml) of a mixture of unlabeled arachidonic acid, HODEs and HETEs (13-HODE, 9-HODE, 13-HpODE, 5-HETE, 15-HETE, 12-HETE) were also run for quantification of individual HODE/HETE species. Each point of this standard curve was spiked with deuterated HODEs/HETEs standards exactly as described above for samples (20 ng of each deuterated HODE/HETE per standard curve point). Detail mass spectrometry conditions and data interpretation are described in the cited references. [6, 7]

Isolation of mononuclear LV cells for FACs

Single mononuclear cells were isolated from non-MI control day (d0) and at d5 post-MI from LV following the method of Ishmahil *et al.* with slight modification. LV of mouse hearts were excised and placed in cold HBSS (Gibco, Invitrogen) in order to remove blood. The heart was finely minced into ≈1 mm x 1 mm pieces and digested with collagenase II (600 U/mL; Worthington and DNAse (60 U/ml) in 20 ml HBSS for 10 min at 37°C. LV tissue was mechanically dissociated by pipetting up and down gently for 4-5 times to release cells. For digestion, collagenase solution was pipetted in 15 ml falcon, then placing it on ice in order to quench collagenase activity. Incubation was continued till the undissolved LVI tissue is dissociated into single cell suspension. The dissociated suspension was centrifuged at 250 x g for 10 min at 4°C. The supernatant was discarded and pellet was resuspended in 1 ml of HBSS. The final volume was made to 10 ml with HBSS. The supernatant was centrifuged at 250 x g for 10 min at 4°C and pellet was resuspended in 2 ml cold PEB buffer (MACS BSA solution and autoMACSTM in 1:19 dilution). Then, the cell suspension was passed through pre-separation filter 30 µm (Miltenyi Biotec) and the flow-through was collected. The flow-through was centrifuged at 250 x g for 10 min at 4°C and the pellet was resuspended in 2 ml PEB buffer. The single-cell suspension collected was layered on ficoll gradient solution and centrifuged at 2000 x g for 20 min. To reduce myocyte contamination in the mononuclear cell suspension, 75% of the total volume (excepting cell debris) was collected and washed with HBSS. The cell viability was measured by using trypan blue.[3]

Spleen cell preparation for FACs

Mice spleen from non-MI d0 control and d1 and d5 post-MI was removed aseptically and placed in a 35-mm tissue culture dish with ~5 ml HBSS (Gibco; Invitrogen). Spleen was teased and injected with 100 µg/ml collagenase D (Roche) solution with 26 gauge needles and incubated at 37°C in 5%CO_2 incubator for 20 min. After 20 min, in order to inactivate collagenase D, 1/10th volume of 100 mM EDTA/PBS without Ca^{2+} and Mg^{2+} (Gibco, Invitrogen) was added for 5 min. Then the cells were passed through nylon cell strainer, 70 µm (BD Falcon) with 10 ml of 10 mM EDTA/PBS solution and then with 30 ml PBS. The cells were centrifuged at 250 x g at 4°C for 5 min and resuspended in 3ml ACK lysis buffer (GIBCO, Invitrogen) for 5 min on ice. The ~10X volume of PBS was added to each falcon, and centrifuged at 250 x g for 5 min at 4°C. The cells were resuspended in FACS buffer (PBS+5% FCS). Total cell count of splenocytes was done using trypan blue.[5, 8]

Surface markers strategy for FACs

The cell count (LV mononuclear cell or splenocytes) was adjusted to ~1-2 million cells/stain. Isolated cell suspensions were finally suspended in 200 µl of 1:500 Fc block and incubated for 10 min on ice. A cocktail of fluorophore-labeled monoclonal antibodies in 2X concentration were added for 30 min on ice as appropriate for each study. We used CD11b-APC (BD Biosciences), F4/80-Percp (molecular probes), Ly6c-FITC (BD Biosciences), CD206-PE (Bio legend) cocktail. The single cell suspensions from freshly isolated LV or spleen were analyzed for CD11b expression using flow cytometry. The neutrophils were defined as Ly6G^{+} cells. Activated macrophages were defined as the cells dual expression CD11b (Mac-1) and F4/80^{+}surface marker. Further, F4/80^{+}macrophages classified as M1 (classically activated macrophages, CD206^{low} F4/80^{+}) and M2 (alternatively activated macrophages, CD206^{high}) based on CD206 polarity. The macrophages (F4/80^{+}) were also classified as M1 (classically activated macrophages) and M2 (alternatively activated macrophages) based on Ly6c^{high} and Ly6c^{low} respectively (Supplementary Figure 3). For the heart and spleen cells were normalized for total cell population. Data were acquired on BD FACSCalibur Flow Cytometer and analyzed with FlowJo software, version 7.6.3.[3]

Isolation of neutrophils and macrophages from LV infarct

The neutrophils (Ly6G^{+}) and macrophages (CD11b^{+}) cells were isolated from LV infarct at post-MI day1 (d1) and d5 from using magnetic beads, LVI of mouse hearts was flushed with cardiplegic solution and placed in cold HBSS (Gibco, Invitrogen). .The heart was finely minced into ≈1 mm x 1 mm pieces and digested with collagenase II (600 U/mL; Worthington and DNAse (60 U/ml) in 20 ml HBSS for 10 min at 37°C. LV tissue was mechanically dissociated by pipetting up and down gently for 4-5 times to release cells. For digestion, collagenase solution was pipetted in 15 ml falcon, then placing it on ice in order to quench collagenase activity. Incubation was continued till the undissolved LVI tissue is dissociated into single cell suspension. The dissociated suspension was centrifuged at 250 x g for 10 min at 4°C. The supernatant was discarded and pellet was resuspended in 1 ml of HBSS. The final volume was made to 10 ml with HBSS. The supernatant was centrifuged at 250 x g for 10 min at 4°C and pellet was resuspended in 2 ml cold PEB buffer (MACS BSA solution and autoMACSTM in 1:19 dilution). Then, the cell suspension was passed through pre-separation filter 30 µm (Miltenyi Biotec) and the flow-through was collected. The flow-through was centrifuged at 250 x g for 10 min at 4°C and the pellet was resuspended in 200 µl PEB buffer per 10^8 total cells. The cells were incubated with 50 µL of anti-Ly6G-Biotin per 10^8 total cells for 10 min at 4C. Then 150 µL of PEB buffer and 100 µL of anti-Biotin Microbeads per 108 total cells were added and incubate for 15 min at 4^{o}C. Ly6G^{+} cells were separated using MS columns (Miltenyi Biotec) in the magnetic field of the MiniMACS Separator on the MACS Multi-stand. The flow through effluent

left after separation of $Ly6G^+$ neutrophils well washed with PEB and incubated with $CD11b^+$ microbeads following similar procedure of $Ly6G^+$ cells. The effluent will contain $CD11b^+$ macrophages.[3]

LV histology and Immunohistochemistry

For immunohistochemistry (IHC), LV mid-cavity section was embedded in paraffin, sectioned at 5µm. For assessment of neutrophils, macrophages, COX-2, HO-1 and 5-LOX, paraffin-embedded sections were deparaffinized in citrisolv (Thermo Fisher Scientific) and rehydrated through graded ethanol. Heat-mediated antigen retrieval was performed to expose antigen epitopes (Target Retrieval Solution, Dako S1699) using pressure cooker (BioSB Tinto Retriever). Sections, blocked with normal rabbit or goat serum as per antibody, and were incubated with rat anti-mouse neutrophils (CL 8993AP, clone 7 1:50; Cedarlane), rat anti-Mac-3 monoclonal antibody (CL 8943AP, clone M3/84; 1:100 dilution; Cedarlane). For Neutrophils and macrophages staining were followed with the Vectastain Elite ABC kit (Vector) The slides were mounted using permount and allowed to dry for use in image analysis.[5, 8] For other markers the section were incubated with HO-1 (Enzo, 1:100), COX-2 (abcam, 1:250), and 5-LOX (abcam 1:50). The myofibroblast differentiation was detected by using α-SMA (smooth muscle actin) antibody (Sigma, 1:400) and followed by followed by respective conjugated secondary antibodies Alexa 555 and Alexa 488 - (Molecular probe), each for 60 min at room temperature. Nucleus was stained using DAPI (molecular probe).Cells were mounted using antifade mounting media (Invitrogen) and then visualized and photographed using Nikon A1 High Speed Laser Confocal microscope.[3, 9, 10]

Collagen measurements using picrosirius red staining

For PSR staining, paraffin-embedded unstained sections of LV tissue were deparaffinized in citrisolve (Fisher) and rehydrated through subsequent washes of ethanol. After a wash with water, phosphomolybdic acid (0.2% in water) was placed on the section. A subsequent wash with water was followed by addition of sirius red, 0.1% in saturated picric acid (26357-02) and then application of 0.01N hydrochloric acid. Then, slides were dehydrated and mounted using permount. The slides were allowed to dry for image acquisition and analysis.[3, 5, 10]

Image analysis for IHC and PSR staining

For each slide per mouse, a total of 5-7 images were captured focusing on infarct area and border zone using a microscope (BX43) with an attached camera (Olympus DP73). The images were captured using the cellSens Dimension program (Olympus version 1.9) and then analyzed for percent area stained using Image-Pro Premier 64-bit analyzer software. The percent area determined by the image analysis software (Image Pro Premier, Cybernatics) for the 5-7 images of each sample were recorded and averaged to determine the percent area stained for neutrophils, macrophages, and collagen (PSR) density in the infarcted area.[3, 8, 10]

RT^2profiler inflammatory and ECM PCR array

Frozen LV samples for d0 control and post-MI d1 or d5 remote (LVC) and infarcted (LVI) were processed separately for RNA extraction. LV tissue (4-8 mg) was homogenized with a sonic dismembrator (Fisher Scientific Inc. USA, amplitude between 10-100) and RNA was isolated using TRIzol (Invitrogen) as per manufacturer's instructions. RNA concentration was determined using the ND1000 nanodrop. cDNA synthesis was performed RT^2 first strand kit (Qiagen 330401) using 400ng RNA per sample. Each sample was prepared on a RT^2-PCR plate (Inflammatory Cytokine and Receptor by Qiagen PAMM-011E and Mouse ECM & adhesion

molecules PAMM-013E) and ran on an ABI 7900HT. Gene levels were normalized to (hypoxanthine phosphoribosyltransferase) Hprt-1 as the housekeeping gene control. The results were reported as $2^{-\Delta}$Ct (ΔΔCt) values.[3, 8, 10]

LV and spleen protein extraction for immunoblotting

For every mg of remote (LVC) or infarct (LVI) tissue 16 µl of reagent A (1xPBS from Invitrogen, without calcium and 1x proteinase inhibitor, Roche Diagnostics) was used. The tissues were dismembrated in short (5 sec) intervals with up to 100 amps (Fisher Scientific USA Inc.) using sonic dismembrator until completely homogenous. Homogenates were centrifuged at the maximum speed (14,000 rpm) for 5 min at 4˚C. The supernatant was transferred to a fresh tube and snap frozen and used as fraction A soluble protein. The pellet was washed 3 times with PBS, centrifuged at maximum speed after each wash and then resuspended in 16 □l reagent B (Reagent 4 from Sigma and 1xPI) per milligram of original tissue weight and the pellets were dismembrated using the same method as the A fraction. The new homogenous solution was snap frozen and used as fraction B insoluble protein.

Splenic lysates were prepared using radio-immunoprecipitation assay (RIPA) lysis buffer (Sigma) and protease inhibitor cocktail (Roche GmbH, Germany). Piece of spleen tissues was dismembrated in short (5 sec) intervals with up to 100 amps (Fisher Scientific USA Inc.) using sonic dismembrator until completely homogenous. Homogenates were centrifuged at the maximum speed (14,000 rpm) for 5 min at 4˚C. The supernatant was collected in fresh tubes and snap frozen until further use. Protein concentration in each sample was determined using 1X Bradford protein assay kit according to the manufacturer's instructions (Bio-Rad, Hercules, CA, USA) with bovine serum albumin (BSA) as a standard.[3, 10]

LV and splenic protein Immunoblotting

Immunoblotting was used to quantify the protein levels of homogenized LV samples. Each immunoblot was begun with the running of a criterion XT bis-tris 4-12% 18-gel (Bio-Rad Inc.) gel, MOPs Buffer (Bio-Rad). The kaleidoscope precision plus standard (Bio-Rad) was used to determine the molecular weight of the protein. Total 10ug of protein was loaded, run and transferred on nitrocellulose membrane (BioRad). The total protein stain was acquired using pierce reversible protein stain, nitrocellulose membranes (Thermo Scientific). After rinsing with water, the membrane was blocked for 1 hr at room temperature using 5% non-fat milk powder (Bio-Rad) dissolved in TPBS and probed with primary antibody (COX-2 1:1000, COX-1 1:1000, 5-LOX 1:1000, HO-1:2000, MMP9 1:1000, TIMP-1 1:1000, collagen VI 1:1000 , CYP2J 1:2000 and α-smooth muscle actin 1:10000) overnight at 4˚C followed by secondary antibody (Biorad). The proteins were detected using femto chemiluminescence detection system (Pierce Chemical, Rockford, IL, USA). Densitometry was performed using Image J software (NIH, USA).[3]

Measurements of macrophage phenotype using quantitative Real-Time PCR

For qPCR, reverse transcription was performed with 2.5 µg of total RNA using SuperScript® VILO cDNA Synthesis Kit (Invitrogen, CA, USA). Quantitative PCR for *ptgs-1, ptgs-2, hmox-1*, *Alox-12*, *Alox-5, tnf-α, IL-6, ccl2, IL-1β, CD163, Arg-1, Mrc-1 and Ym-1* genes was performed using taqman probes (Applied Biosystems, CA, USA) on master cycler ABI, 7900HT. The mRNA expression was normalized with the reference genes (β-Actin). Gene levels were normalized to Hprt-1 (hypoxanthine phosphoribosyltransferase) as the housekeeping control gene. The results were reported as $2^{-\Delta ct}$ (ΔΔCt) values. All the experiments were performed in duplicates with n=3-4/group.[5]

Venn diagram, hierarchical clustering of ECM and inflammatory array

Venn diagram, hierarchical cluster, and heat maps were generated for RT^2profiler inflammatory and ECM PCR array data using cluster 3.0 and java tree view software. The values of genes were normalized by taking geometric mean of genes for statistical significance.

Statistical analysis

Data are expressed as mean per group and SEM. Statistical analyses were performed using Graphpad Prism 5. Analysis of variance (ANOVA), followed by Newman–Keuls post-hoc test, was for multiple comparisons of post-MI-d1, d5 compared with d0 naïve control. We ensure that Bartlett's variation test passed while using Newman–Keuls post-hoc test or nonparametric Kruskal-Wallis ANOVA test followed by Dunn post-hoc test. Kaplan-Meier test and log-rank test was followed for survival analysis. All immunoblotting densitometry data were normalized to total protein. For 2 groups comparison, student-t test (unpaired) was applied and $p<0.05$ was considered as statistically significant.

References

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Supplementary figure legends

Figure 1. Schematic study design illustrating post-MI time points and output measurements in WT (n=93) and 12/15LOX$^{-/-}$ (n=97) mice.

Figure 2. **Gating strategy for immune cells measurements in the infarcted left ventricle (LVI) and spleen.** CD11b^{+} population contained CD11b^{+}/F4/80^{+} macrophages. Macrophages were further divided into classical and alternative macrophages based on the expression of CD206 and Ly6C expression. CD11b^{+} population contained CD11b^{+}/F4/80 population. CD11b^{+}/F4/80^{-} subpopulation were further classified into Ly6G^{+} (neutrophils) and Ly6G^{+}/Ly6C^{high} population.

Figure 3. 12/15LOX expression increases post-MI in WT mice. (A) Representative IHC images of LV showing 12/15LOX expression in WT and 12/15LOX$^{-/-}$ mice at d0, post-MI d1 and d5. **(B)** mRNA expression of *Alox12* and *Alox15* in LVI of WT and 12/15LOX$^{-/-}$ mice in no-MI and at post-MI d1 and d5**.** *p<0.05 vs d0 control, $p<0.05 vs MI-WT at respective day. Values are means ± SEM, n=4 mice/group.

Figure 4. Splenocytes representing macrophage phenotypes. Quantification of CD11b^{+}/F4/80^{+}, F4/80^{+}/Ly6C^{low}, F4/80^{+}/Ly6C^{high} F4/80^{+}/CD206^{high} and F4/80^{+}/CD206^{low} splenocytes at post-MI d5; n=4/ mice/group/time-point.

Figure 5. Gene expression of classical (M1) and alternative (M2) macrophages markers in the spleens of WT and 12/15LOX$^{-/-}$ mice. **(A)** M1-markers- *tnf-α, ccl2, IL-6* and *IL-1β* **(B)** M2-markers- *Mrc-1, Ym-1, Arg-1 and CD-163*. Gene expression is normalized with Hprt-1 as the housekeeping gene. *p<0.05 vs no-MI, $p<0.05 vs MI-WT at respective day. Values are means ± SEM, n=4 mice/group.

Figure 6. Protein expression levels of resolving enzymes in LV remote areas of WT and 12/15LOX$^{-/-}$ mice. Immunoblot representing **(A)** COX-1, COX-2, 5-LOX and HO-1 expression in WT and 12/15LOX$^{-/-}$ mice in No MI and at post-MI d1 and d5 in LVC fraction. **(B)** Densitometric analysis of COX-1, COX-2, 5-LOX and HO-1 expression. **(C)** LVC mRNA expression of *COX-1* (*ptgs-1)*, *COX-2* (*ptgs-2),* 5LOX*(Alox5)* and *HO-1 (hmox-1)* in WT and 12/15LOX$^{-/-}$ mice in no-MI

and at post-MI d1 and d5. *p<0.05 vs no-MI, $p<0.05 vs MI-WT at respective day. Values are means ± SEM, n=4 mice/group.

Figure 7. 12/15LOX deletion promoted early induction of COX-2 in infarcted LV tissues with post-MI splenic remodeling. (A) LV: Immunoblot representing LVI fraction COX-1 and COX-2 expression in WT and 12/15LOX$^{-/-}$ mice post-MI d1 and d5. **(B)** Densitometric analysis of COX-1 and COX-2 expression. **(C)** LVI mRNA expression of *COX-1* (*ptgs-1)* and *COX-2* (*ptgs-2)* in WT and 12/15LOX$^{-/-}$ mice at no-MI, post-MI d1 and d5. **(D)** Representative IHC images of LV COX-2 expression in WT and 12/15LOX$^{-/-}$ mice at post-MI d1. **(E)** Spleen: Immunoblot representing spleen COX-2, HO-1 and 5LOX expression in WT and 12/15LOX$^{-/-}$ mice at d0, post-MI d1 and d5. **(F)** Densitometric analysis representing spleen COX-2, HO-1 and 5LOX expression normalized to total protein (n=4 mice/group). *p<0.05 vs no-MI, $^{\$}$p<0.05 vs MI-WT at respective day. **(G)** Venn diagram representing changes post-MI inflammatory gene expression in LVI of 12/15LOX$^{-/-}$ mice at post-MI d5 compare with WT counterparts. Post-MI d5 gene expressions are normalized to respective no-MI control. Red indicates increased genes. Blue indicates decreased genes. Black indicates no change in genes. p<0.05 vs WT post-MI-d1 (n=4 mice/group).

Figure 8. Hierarchical cluster profiling of 84 gene Inflammatory and ECM array. Heat map representing gene expression changes **(A)** inflammatory array **(B)** ECM array in infarcted area (LVI) and remote area (LVC) of WT and 12/15LOX$^{-/-}$ mice in no-MI controls and at post-MI d1 and d5The color represents the expression level of the gene. Red represents high (↑) expression, while green (↓) represents low expression.

Supplementary table legends:

Table 1. Table representing total number of experiments and samples used in study.

Table 2. Lipid mediators and precursor/pathway/class in spleens of WT and 12/15LOX^{-}/- mice at d5 post-MI.

Table 3. Table describing early termination of inflammation promoting genes without altering inflammatory phase in 12/15LOX$^{-/-}$ mice.

Table 4. Table representing inflammatory cytokines and receptors gene expression in LVI and LVC of WT and 12/15LOX$^{-/-}$ mice.

Table 5. Table representing ECM and adhesion molecules gene expression in LVI and LVC of WT and 12/15LOX$^{-/-}$ mice.

Supplementary Figures

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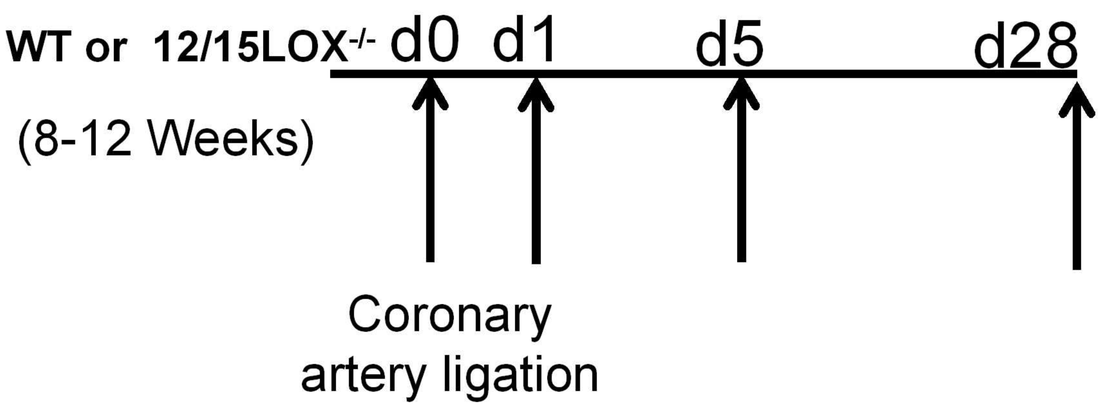
Supplementary fig. 1

Study Design

Invivo



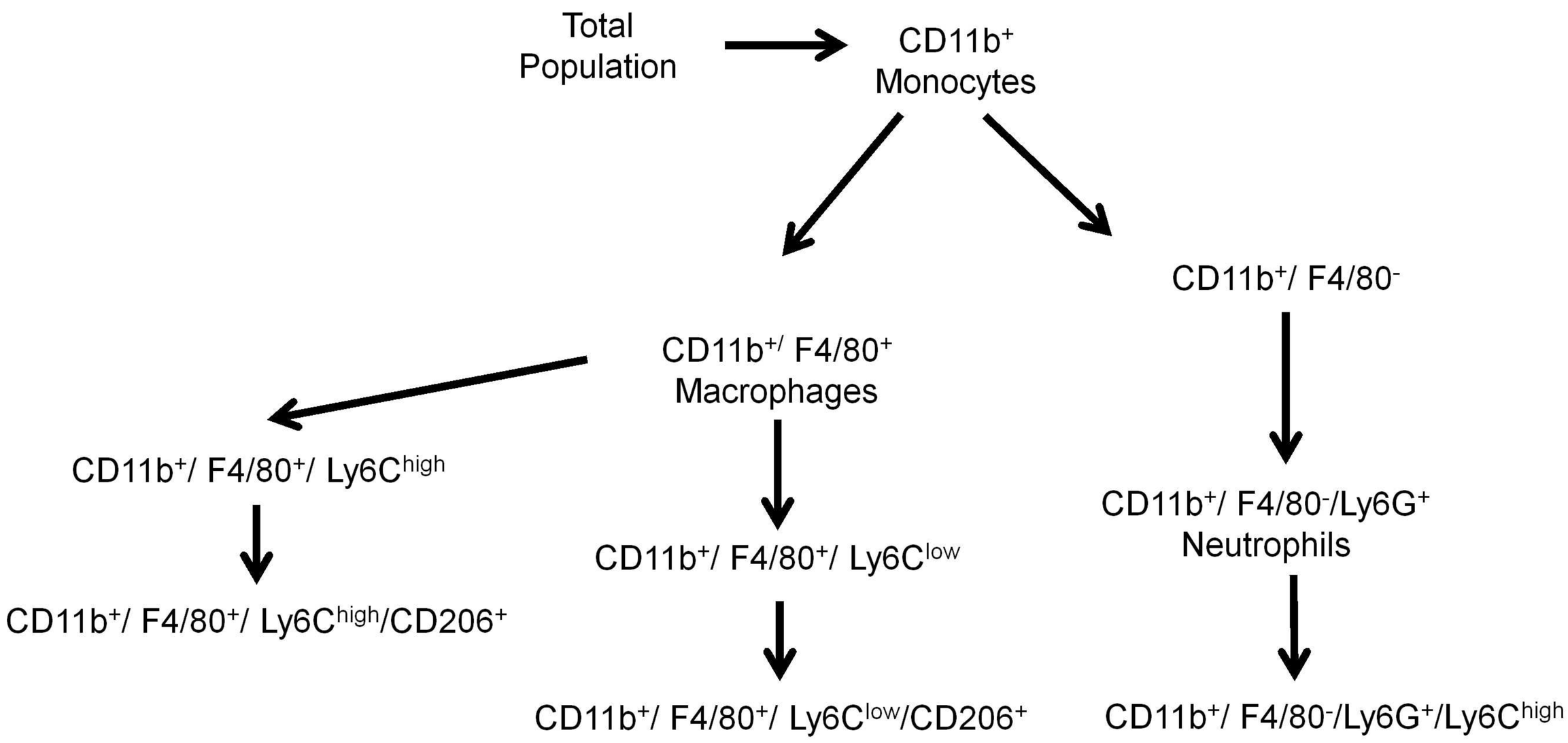
Output Measurement

Measurement at d0 naïve control and post-MI d1 and d5

- Echocardiography for left ventricle size, shape and function.
- Neutrophils, macrophages Isolation from LV.
- M1 and M2 markers by flow cytometry.
- N1 and N2 markers by qPCR.
- M1 and M2 markers by qPCR.
- Neutrophils, macrophages by IHC.
- Picrosirius red staining for collagen deposition.
- Inflammatory and ECM array.
- Immunoblot for HO-1, CYP2J, COX-1,COX-2 and 5-LOX, IHC for HO-1, 5-LOX, COX-2 and 12-LOX.
- RT-PCR for HO-1, 5-LOX, 12-LOX,15-LOX and COX-2.
- Immunofluorescence for SMA, DDR2.
- Post-MI spleen and plasma lipidomics analysis.

Supplementary fig. 2

FACs Gating Strategy



A

LV (Immunohistochemistry)

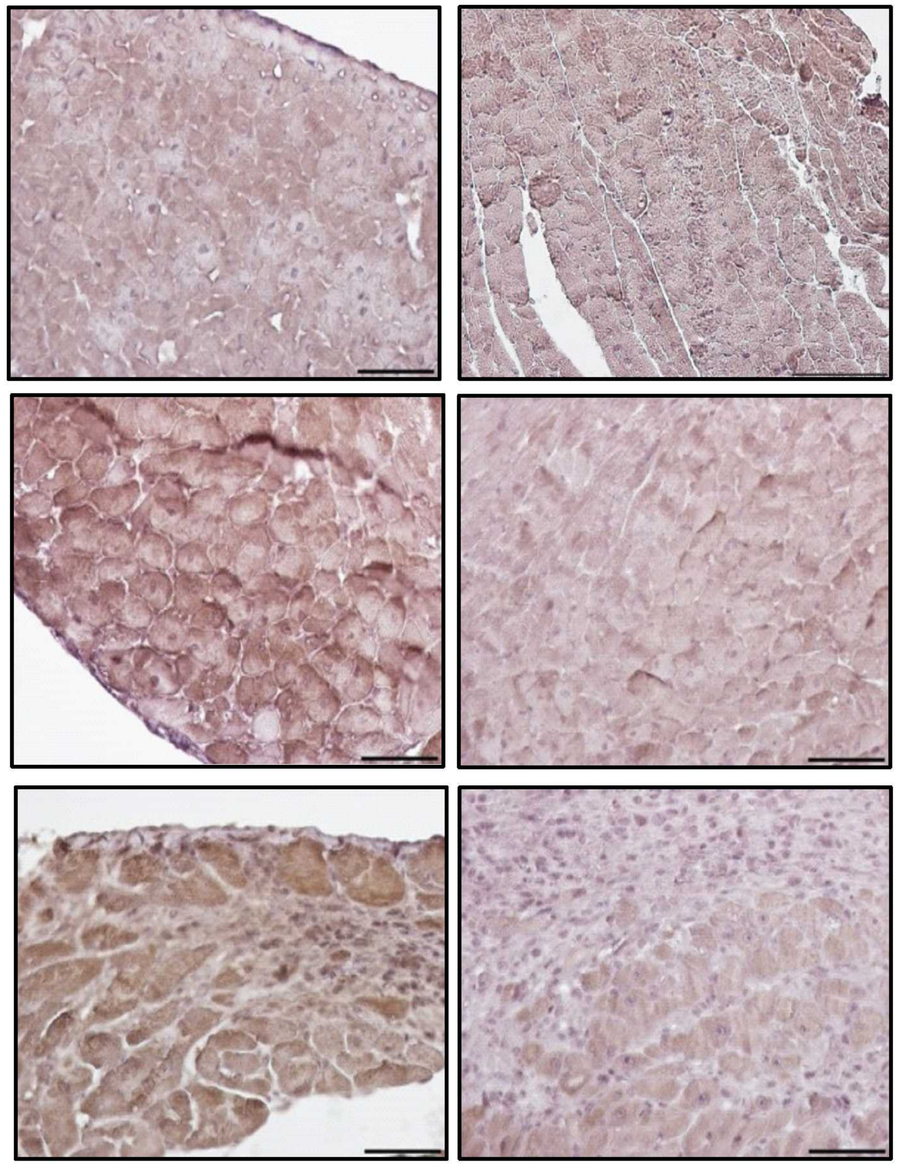
WT

12/15LOX$^{-/-}$

No-MI

MI-d1

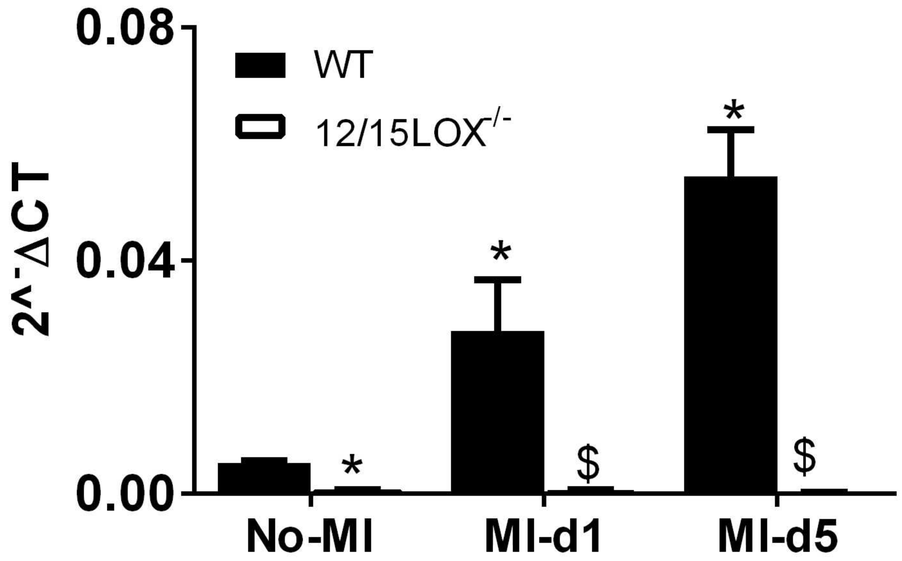
MI-d5



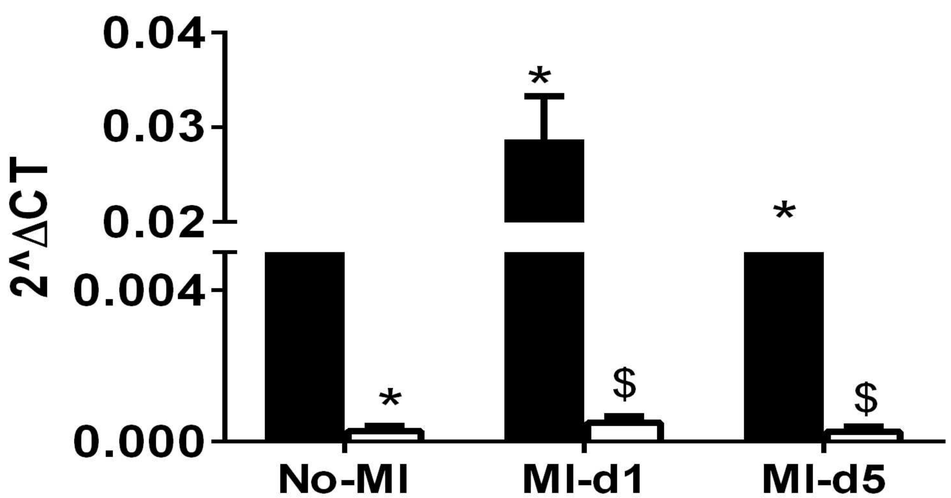
B

LV (mRNA)

Alox12

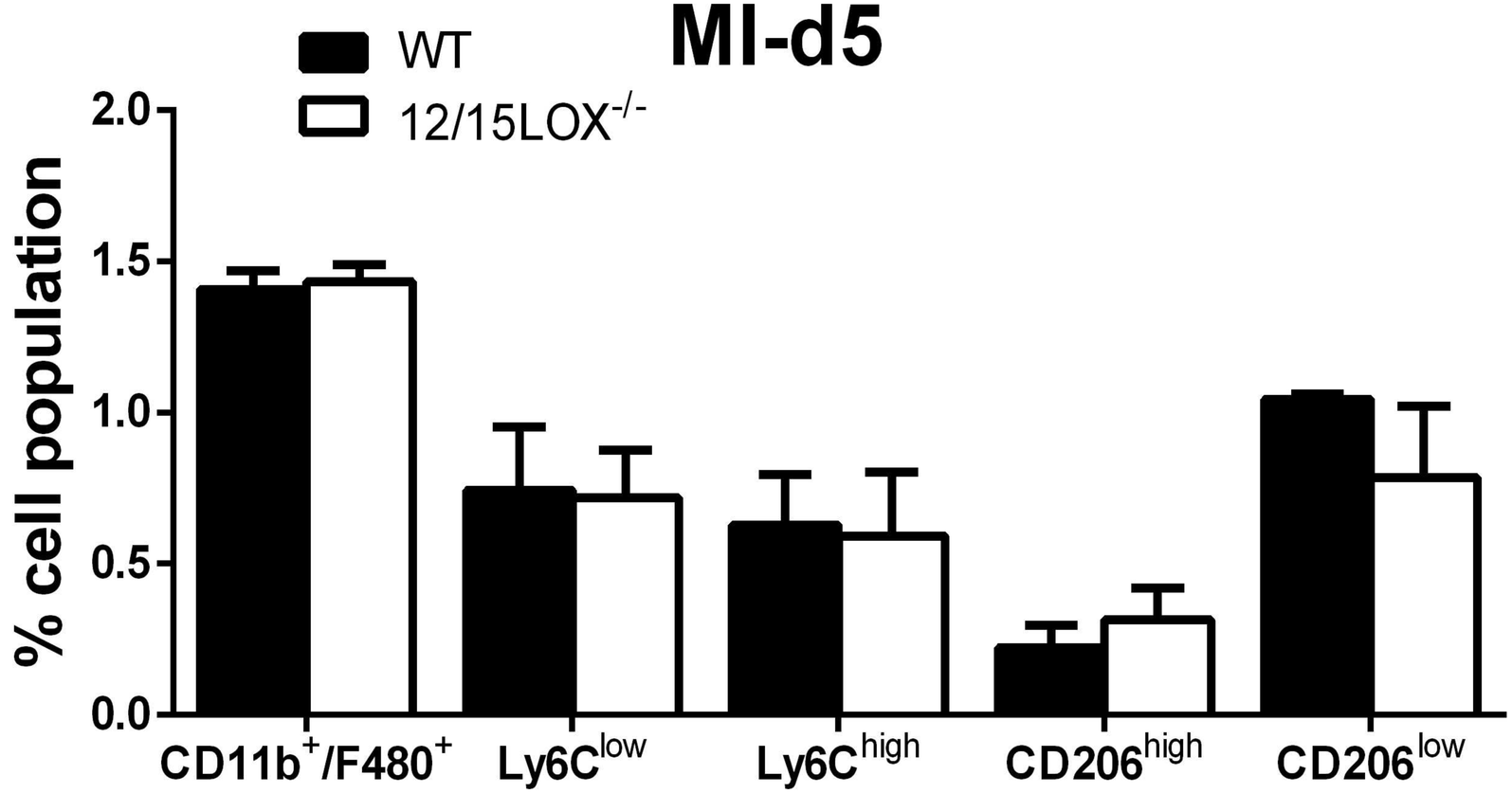


Alox15



Supplementary fig. 4

Spleen (Flow cytometry)

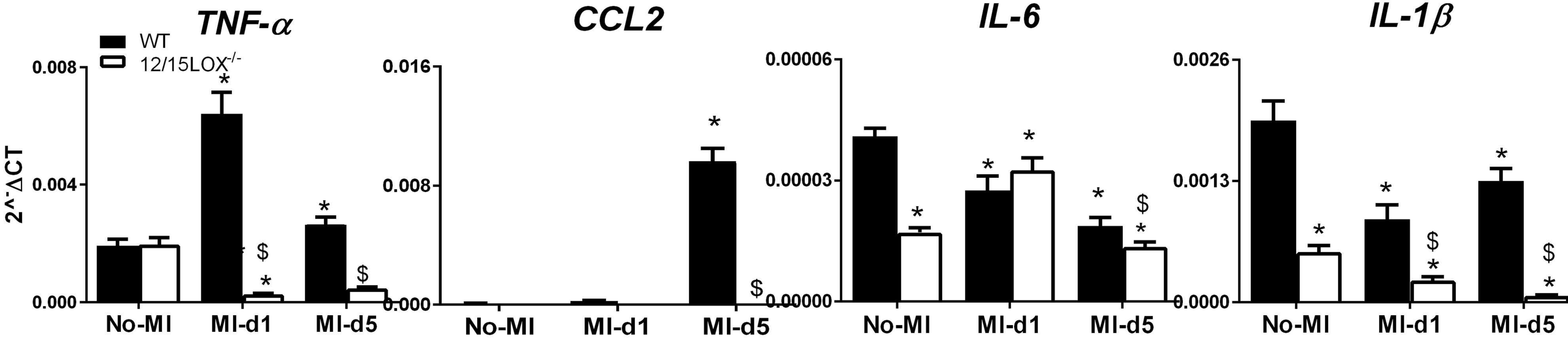


Supplementary fig. 5

Spleen

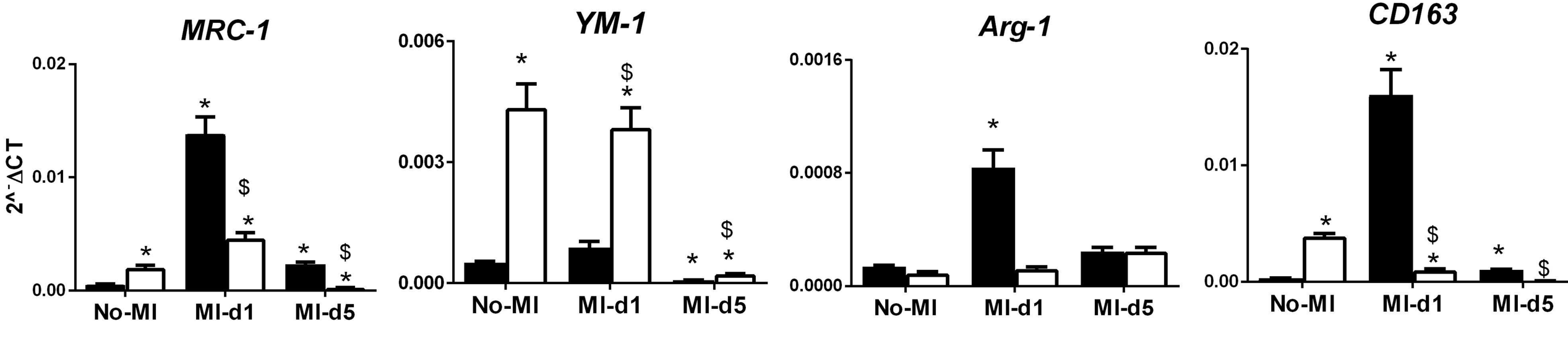
A

M1 macrophages markers



B

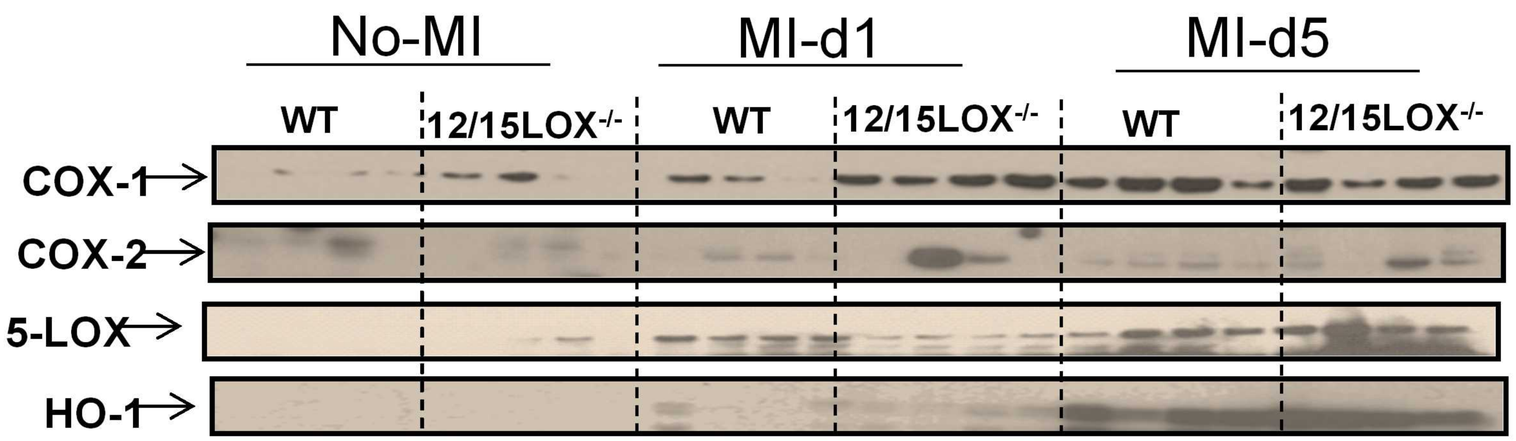
M2 macrophage markers



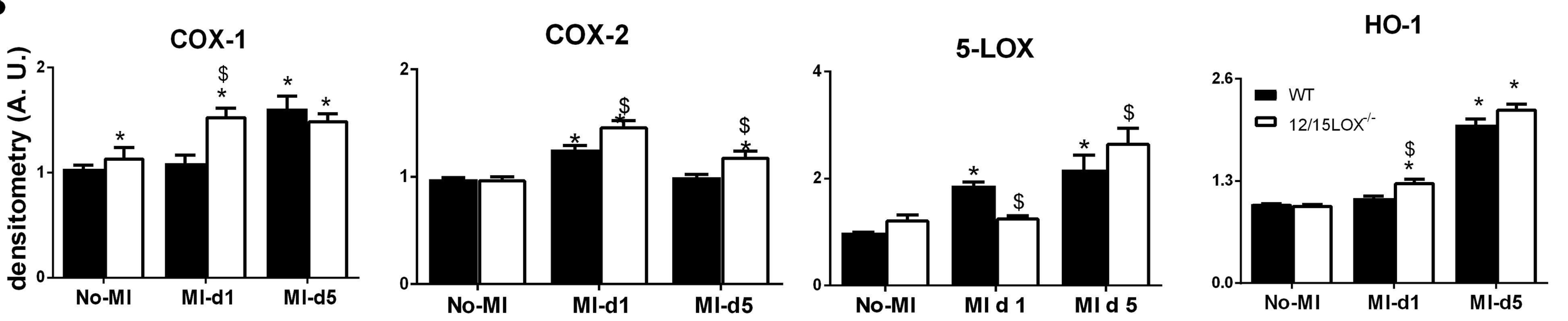
Supplementary fig. 6

A

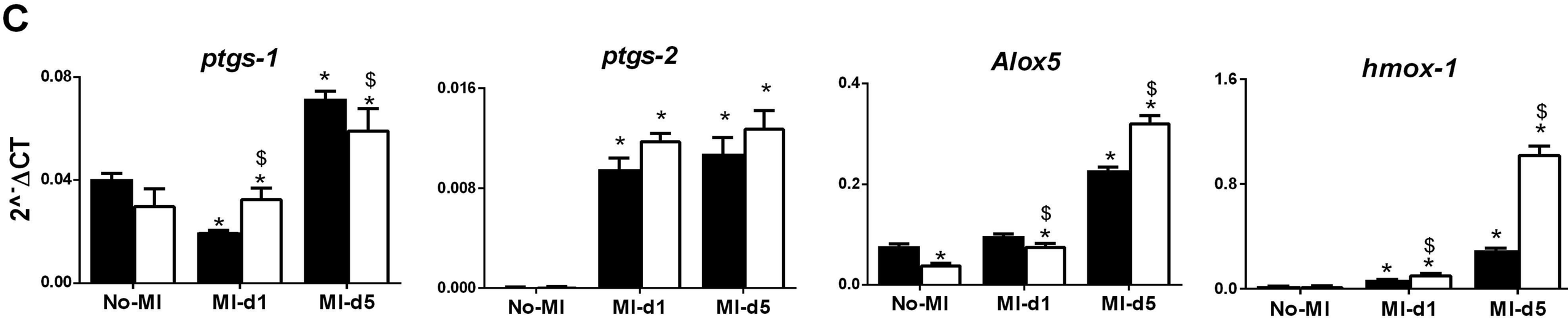
LV (Remote)



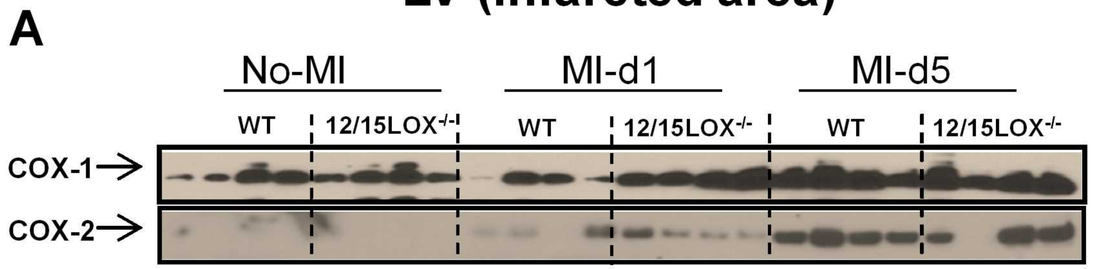
B

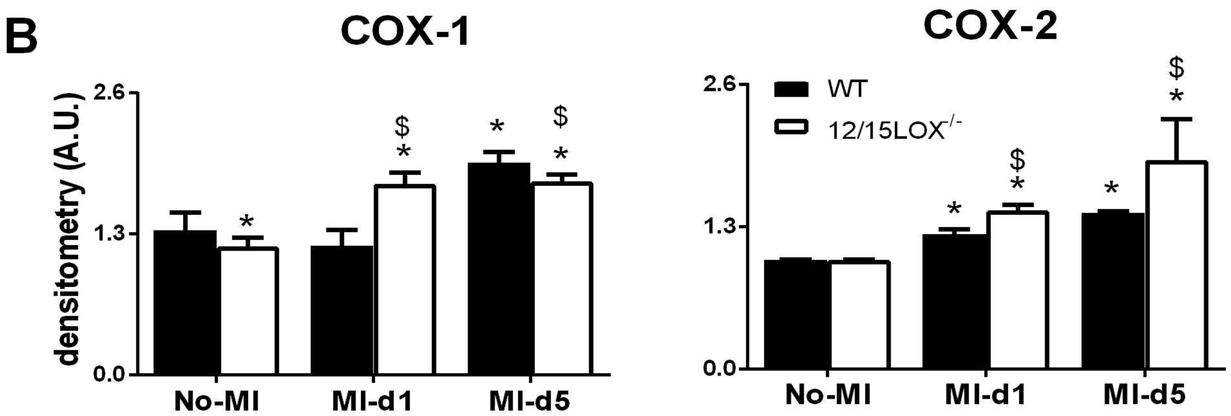


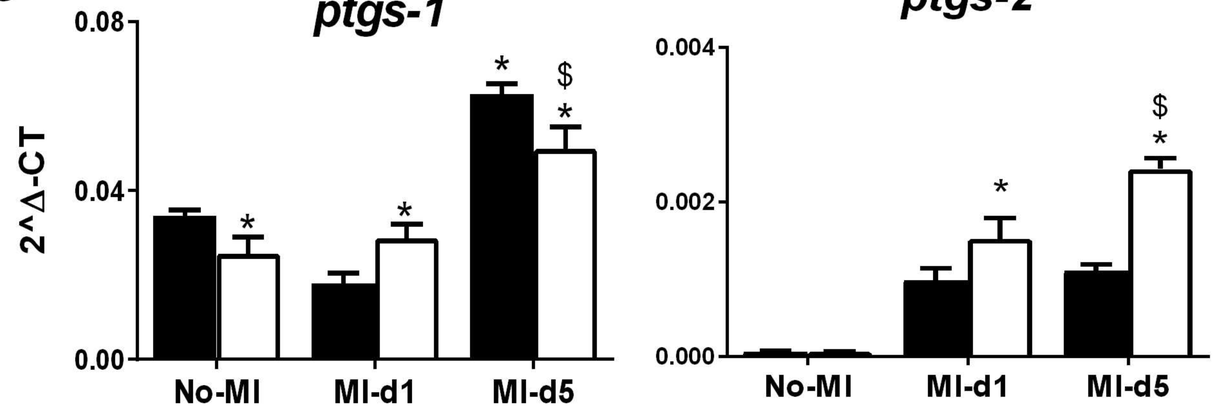
C

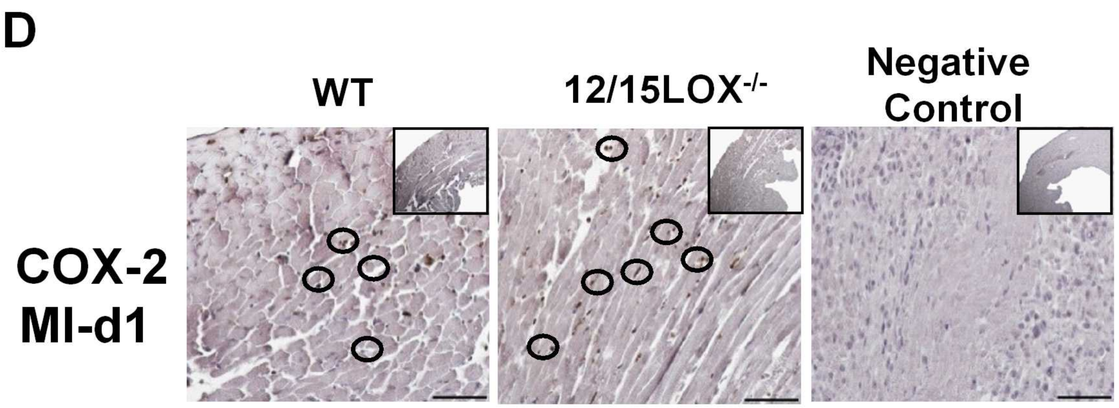


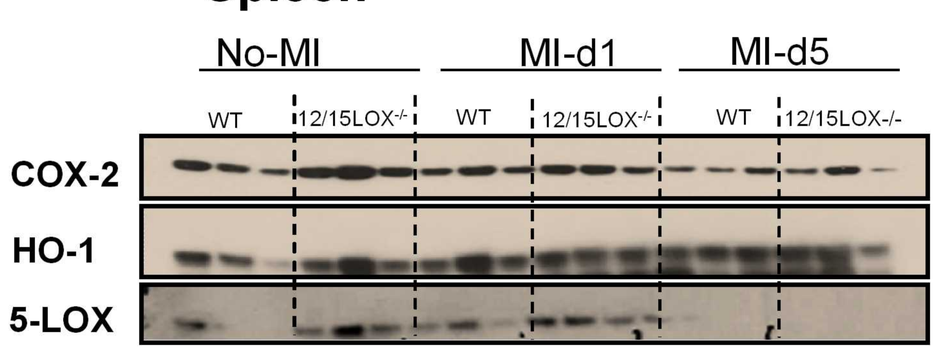
Supplementary fig. 7

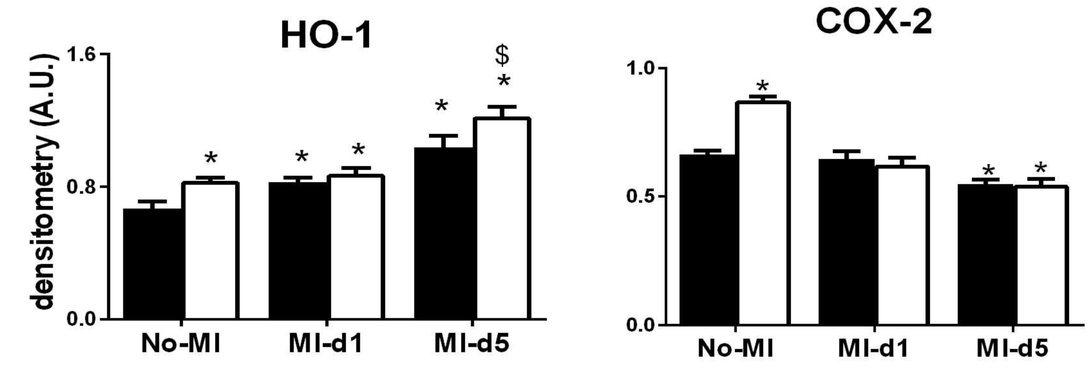


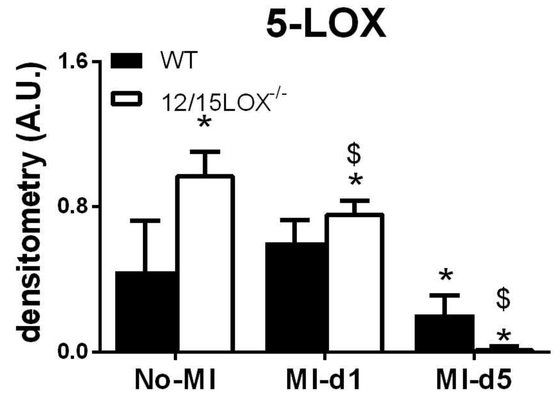


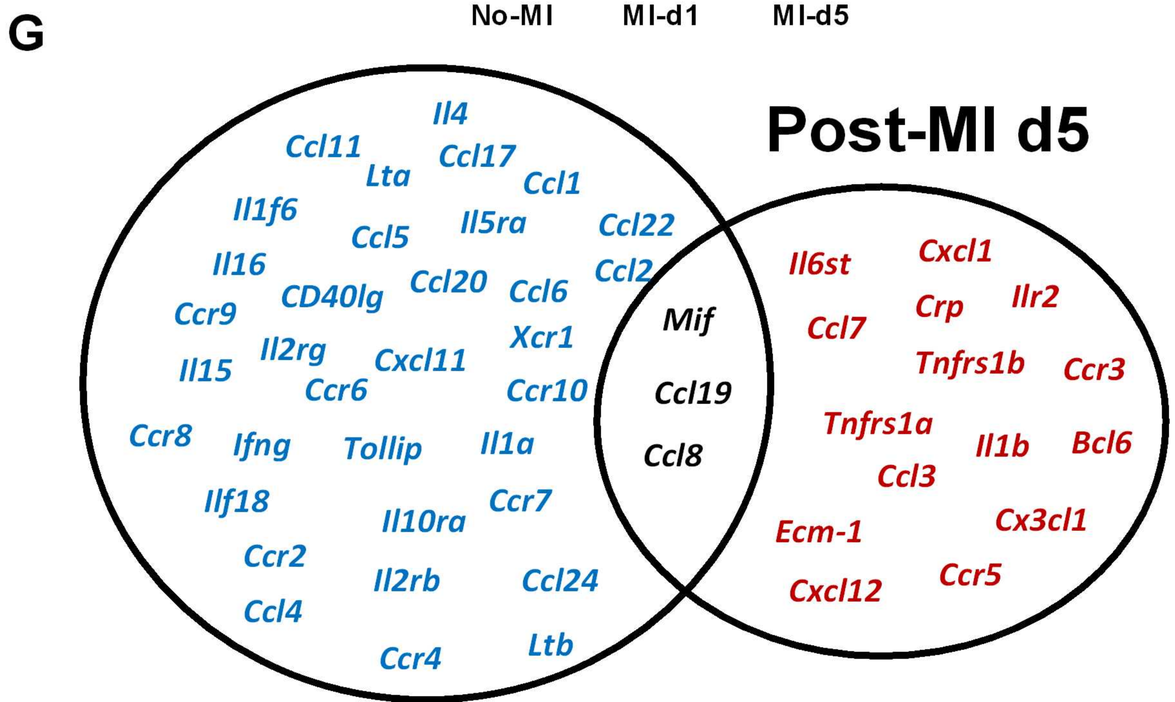








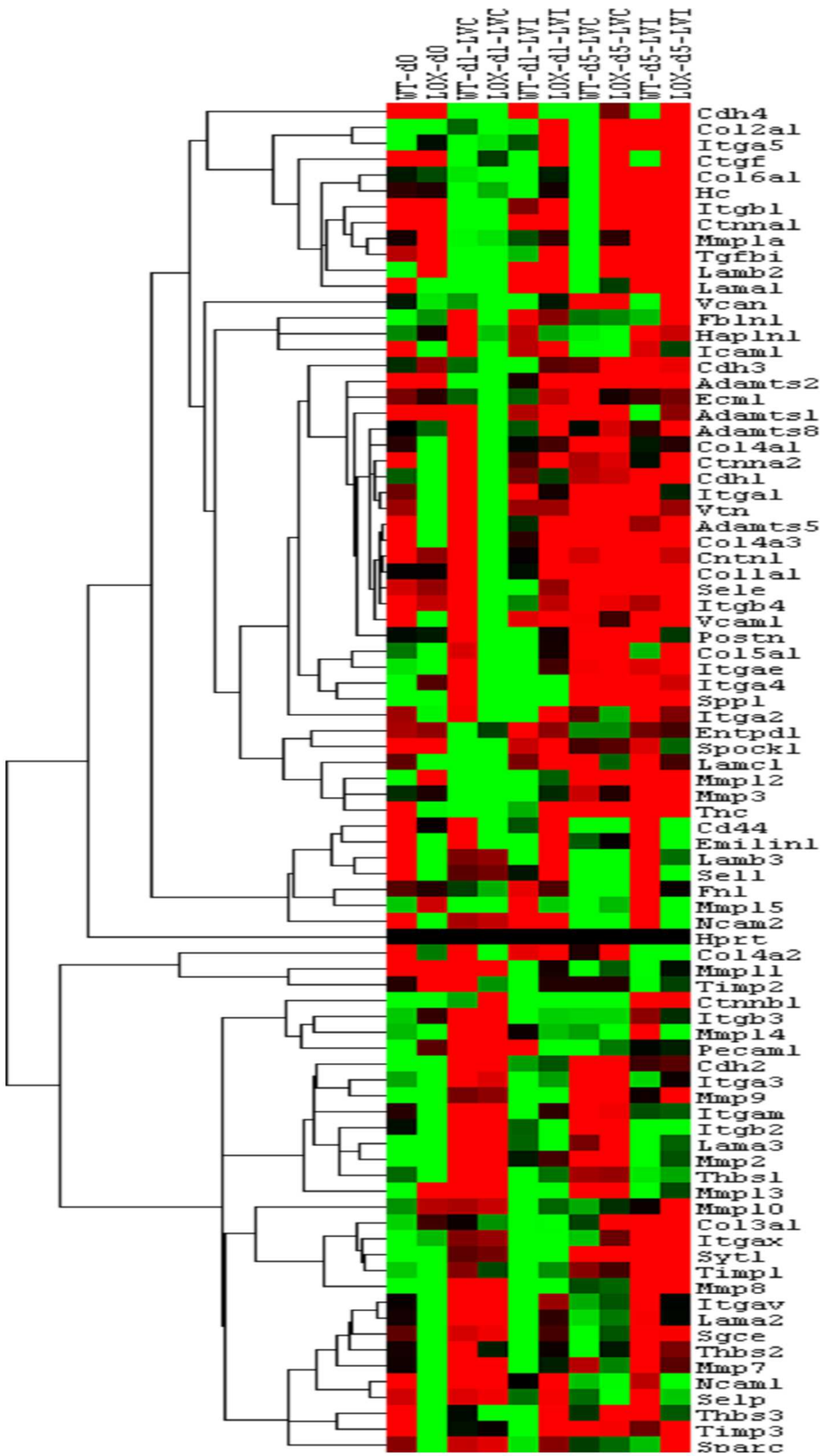




Supplementary fig. 8

A

Inflammatory array



B

Extracellular matrix array