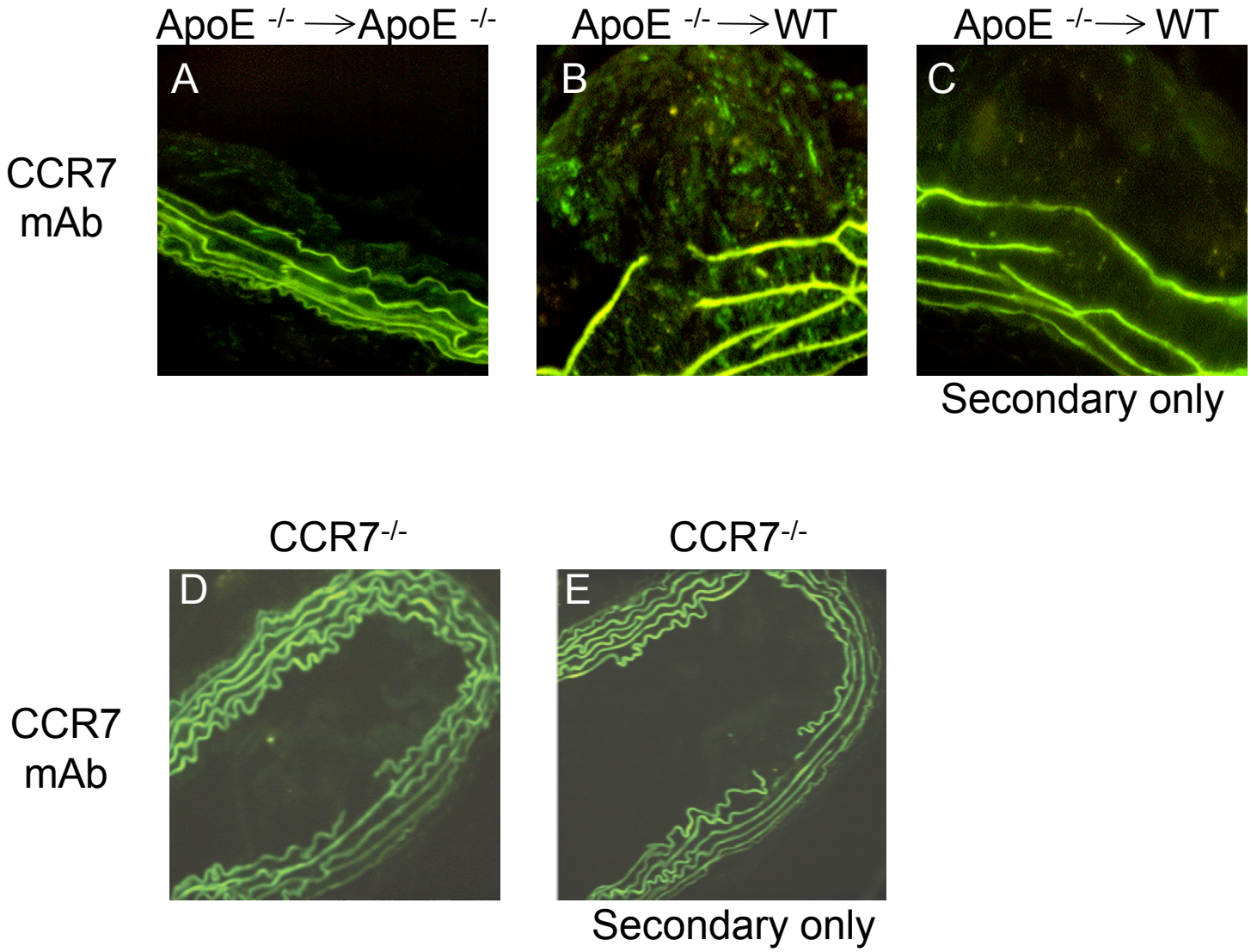
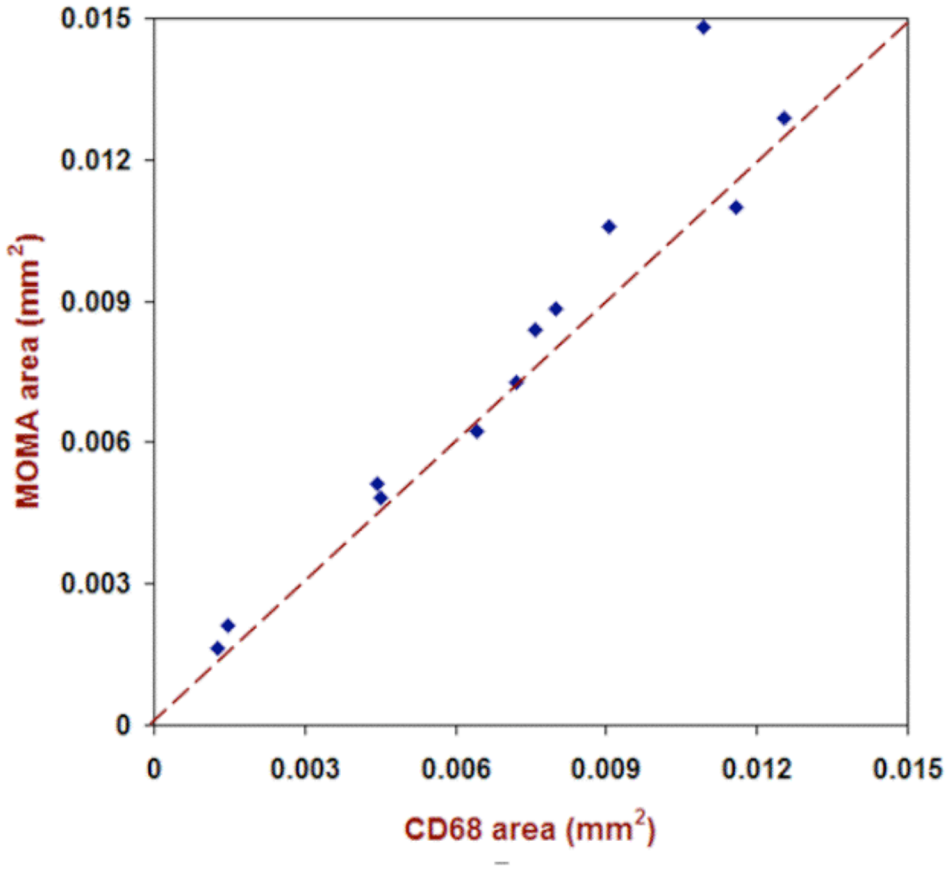
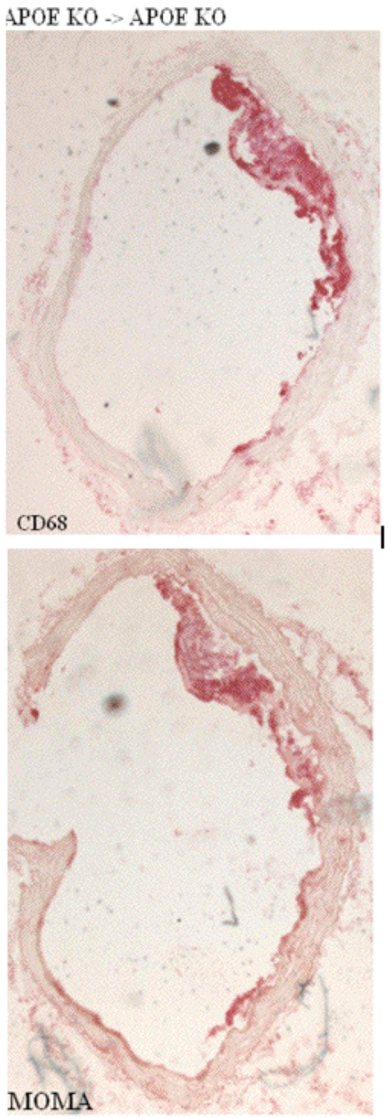


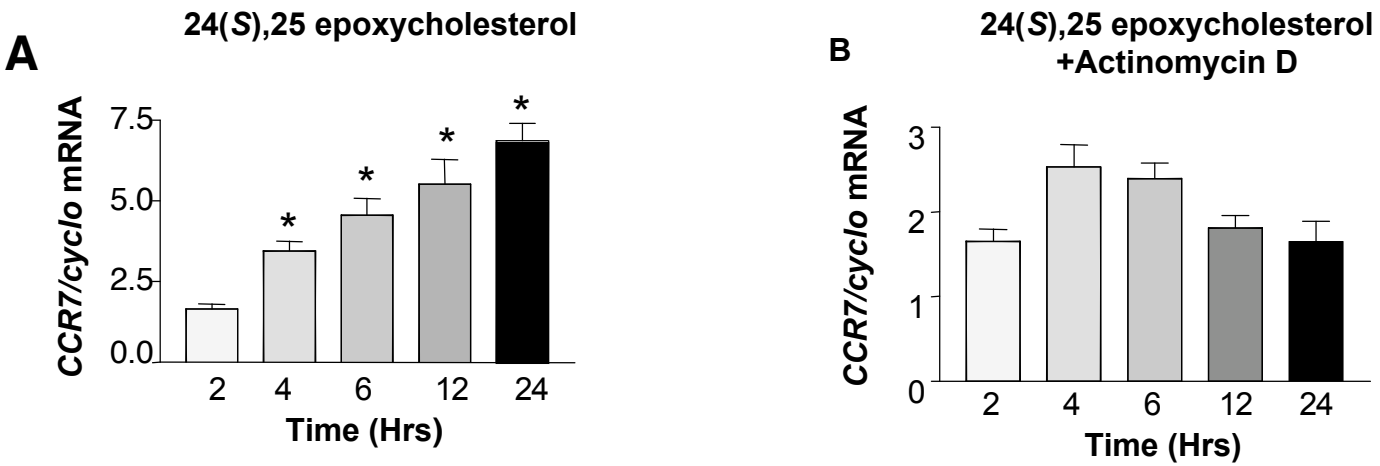
Supplemental Figure 1

F

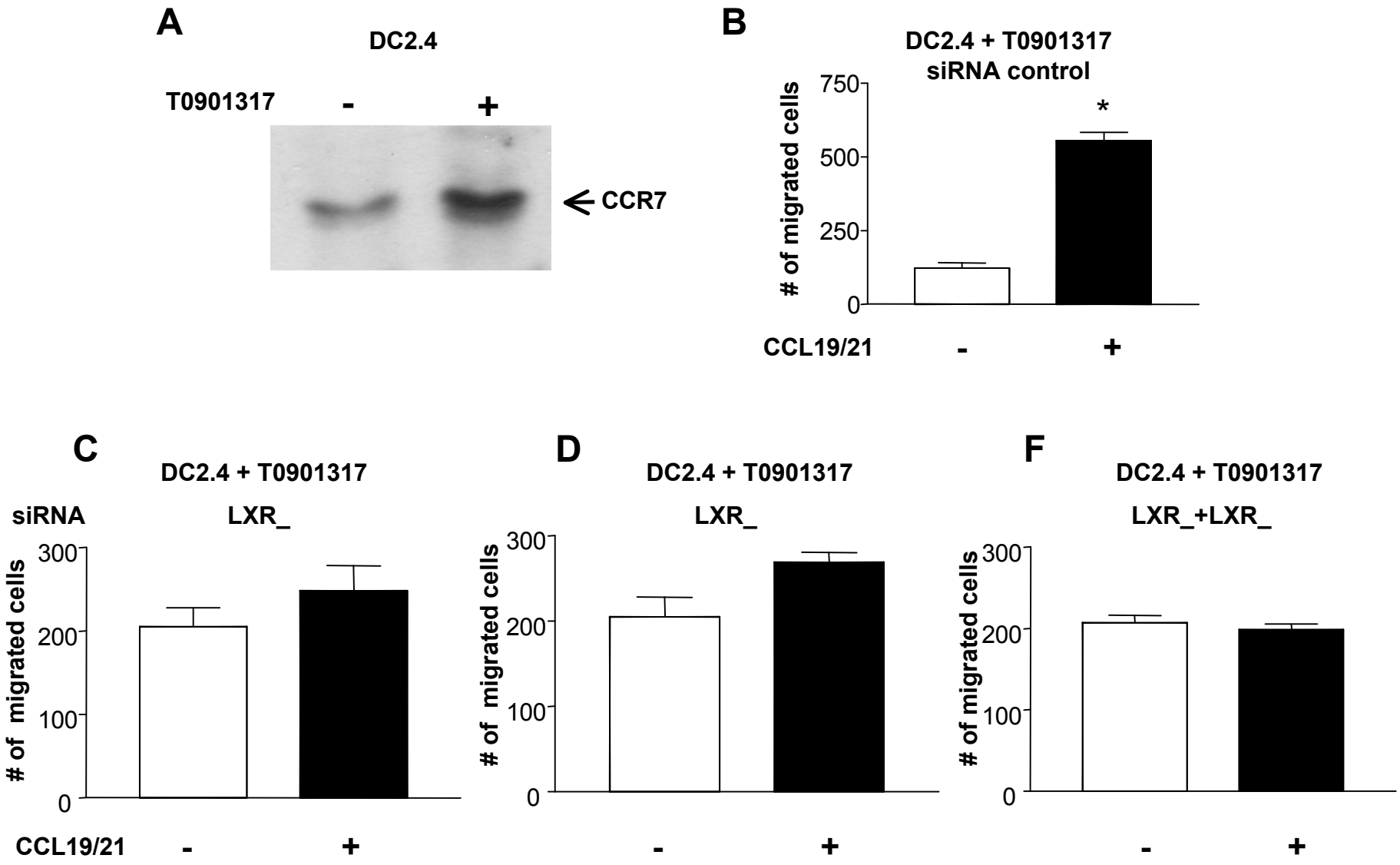




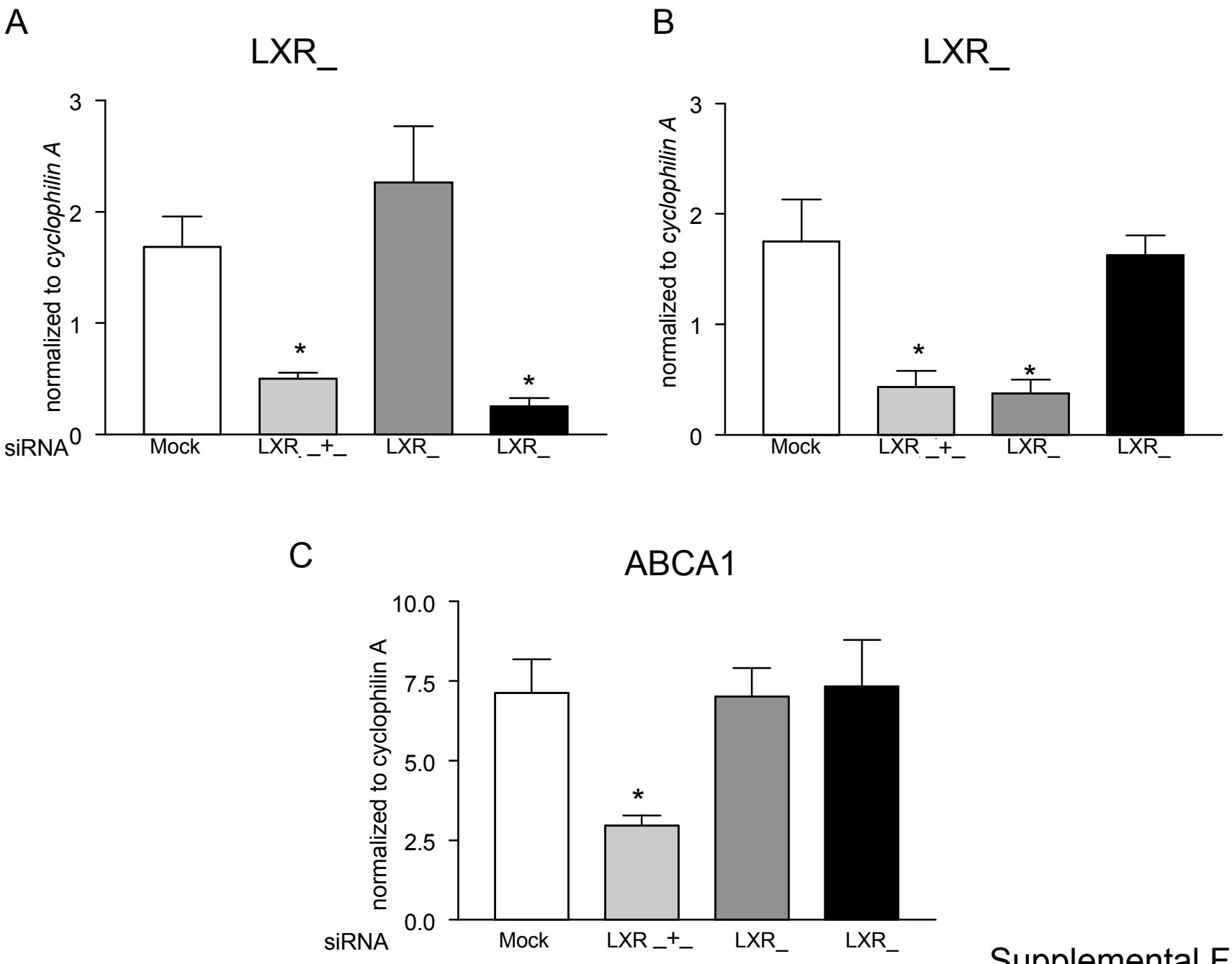
Supplemental Figure 1



Supplemental Figure 2

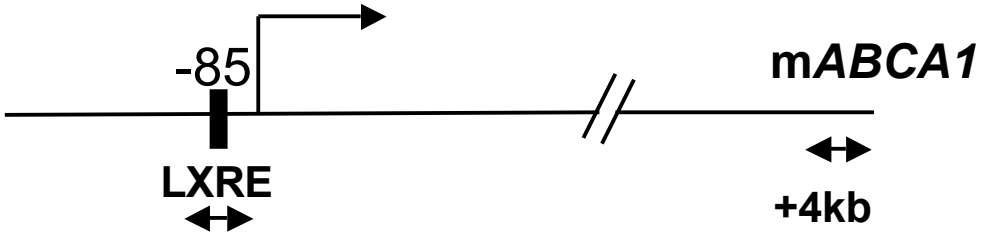


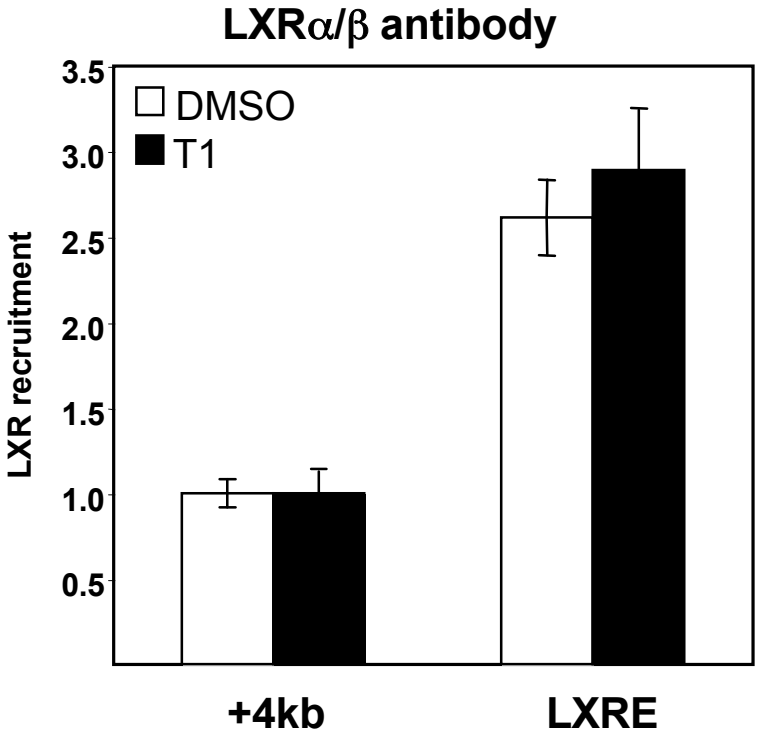
Supplemental Figure 3



Supplemental Figure 4

ChIP assays-mABCA1 LXRE in DC2.4 cells



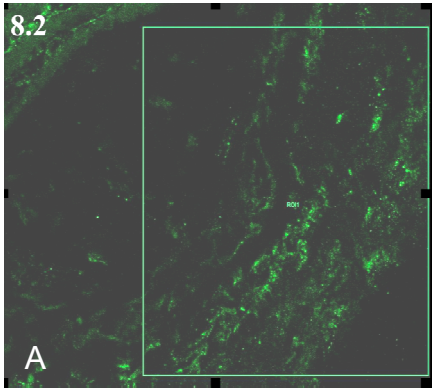


Supplemental Figure 5

Progression

ApoE$^{-/-}$ → *ApoE*$^{-/-}$

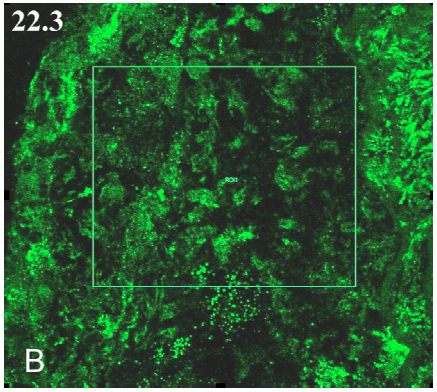
α LXR alpha



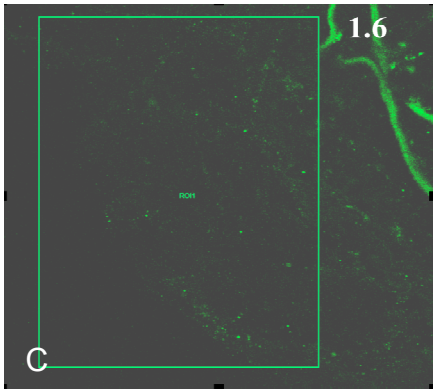
Regression

ApoE$^{-/-}$ → WT

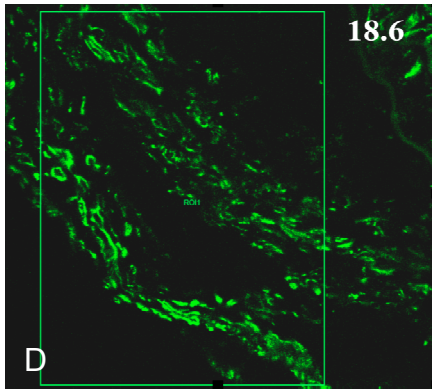
α LXR alpha



α CCR7



α CCR7



Supplemental Figure 6

Supplemental Figure 1. Specificity of CCR7 immunostaining and the correlation between macrophage markers CD68 and Moma-2 in atherosclerotic plaques. Frozen sections of aortic arches were prepared from donor apoE-/- mice 3 days after transplantation into wild type (WT) (A and C) or apoE-/- (B) recipients. Top panel: the staining of CRR7 using a CCR7 monoclonal antibody (mAb) (Abcam Cat # 32527) is shown. Bottom panel: D) Staining of aortic arches with CCR7 mAb from CCR7 knock out mice are shown. C and E) No staining above background is evident in the presence of the secondary antibody alone. The striped green background signal is due to autofluorescence from the internal elastic lamina from the medial layer of the artery. F) Congruency of CD68 and Moma-2 staining of macrophages in atherosclerotic plaques. The aortic arches from 20-week western diet fed donor apoE-/- were transplanted into apoE-/- recipients. At 3 days post-transplant the grafts were harvested. Serial aortic cryosections were immunostained for either CD68 or Moma-2. Lesional areas were determined and plotted against one another. The correlation coefficient between CD68 and Moma-2 immunostaining in the lesions is 0.96, consistent with CD68 expression reflecting macrophage content independent of LXR isoform specificity.

Supplemental Figure 2: A natural LXR agonist increases *CCR7* gene expression i*n vitro* at the transcriptional level. A) DC2.4 cells were treated with the natural LXR agonist 24(*S*), 25 epoxycholesterol for the indicated times. The Y-axes indicate the fold-change of *CCR7* mRNA relative to *cyclophilin A*. Panel B) The increases in *CCR7* mRNA in panel A were largely prevented by pre-incubation of the cells with actinomycin D, a transcriptional inhibitor. The symbol * corresponds to $p<0.05$, when compared to the initial 2 h.

Supplemental Figure 3: LXR agonist enhances CCR7 protein expression and CCR7-dependent chemotaxis to its ligands CCL19 and CCL21, with both LXR isoforms being required for maximal migration. A) DC2.4 cells were incubated with or without 5μM LXR pan-agonist T0901317 for 24h, and extracts were prepared and equal amounts of protein were loaded onto a 10% SDS-PAGE, blotted and probed with a monoclonal antibody to CCR7. B-E) DC2.4 cells were pre-treated with T0901317 and then transfected with either a control siRNA (mock), or siRNA specific to either of the LXR isoforms. Subsequently, 50,000 cells were placed in each well and after 2 h, the cells that migrated towards the CCR7 ligands, CCL19 and CCL21, were counted.
*indicates $p<0.05$ relative to the corresponding to no (-) CCL19/21.

Supplemental Figure 4: Degree of gene silencing with siRNAs to LXR isoforms. In order to ensure that differences in the *CCR7* gene expression related to LXR isoforms were not caused by variation in siRNA efficacy, we measured the mRNA level of each LXR species. As shown in panels A and B, siRNA to *LXR_* or *LXR_* mRNA led to ~75% reduction of either transcript in DC2.4 cells. * indicates $p<0.05$ relative to control siRNA. As expected, *ABCA1* expression was only significantly blunted when siRNAs to both LXR isoforms were used (Panel C), since it is known that either isoform can fully activate the expression of that gene.

Supplemental Figure 5. Occupancy of LXR at the *ABCA1* regulatory region. ChIP assays were employed to examine LXR binding to the *ABCA1* gene. DC2.4 cells were treated with 5 μM T0901317 or an equal amount of DMSO vehicle for 2 h and LXR was immunoprecipitated using an antibody that recognizes both LXRα and β, which was generated against a C-terminal epitope (Santa Cruz; SC-1000). Precipitated DNA was amplified by real-time PCR using primers encompassing the *ABCA1* LXRE (-85) or a control +4 kb intragenic region as indicated with double-headed arrows. LXR binding was normalized for input chromatin levels and reported as induction (*n*-fold) over the control region, which was set to 1. Samples were measured in triplicate, and the results are means ± standard deviations for a representative experiment.

Supplemental Figure 6. Confocal microscopic images of LXRα and CCR7 expression in progressing and regressing plaques. Aortic arches were transplanted from *apoE-/-* donors to *apoE-/-* (progression) or WT (regression) recipients. Aortic sections 3 days after transplantation were immunostained with LXRα or CCR7 primary antibodies and FITC-conjugated secondary antibody. Images were acquired under the same conditions (500-535 nm, gain 758 V, offset -0.3%, pinhole 168 μm) and signal intensity (SI), and quantified from an average of six sections. Boxes shown are representative of regions quantified. LXR_ in progression (mean SI = 8.2); CCR7 in progression (mean SI = 1.6); LXR_ in regression (mean SI = 22.3); CCR7 in regression (mean SI = 18.6).

.

Supplemental Table 1. Statistical Analysis of the Areas of the Atherosclerotic Lesions and of CD68+ Immunostaining[1]

Donor	Lesion Area in Wild Type Recipients
apoE-/-	0.051 ± 0.001
LXRα, apoE DKO	0.130 ± 0.006
LXRβ, apoE DKO	0.151 ± 0.003

ANOVA of Inequality of Means: P<0.0001

Boneferroni Test of Differences Between Means:

apoE-/- vs. *LXRα, apoE* DKO: P<0.001
apoE-/- vs. *LXRβ, apoE* DKO: P<0.001
LXRα, apoE DKO vs. *LXRβ, apoE* DKO: P<0.01

Donor	Area Immunostained for CD68 in Wild Type Recipients
apoE-/-	0.024 ± 0.001
LXRα, apoE DKO	0.049 ± 0.001
LXRβ, apoE DKO	0.055 ± 0.001

ANOVA of Inequality of Means: P<0.0001

Boneferroni Test of Differences Between Means:

apoE-/- vs. LXRα, apoE DKO: P<0.001
apoE-/- vs. LXRβ, apoE DKO: P<0.001
LXRα, apoE DKO vs. *LXRβ, apoE* DKO: P<0.001

[1]Units are mm^2; displayed are mean ± SEM; N=7-10; DKO= double knockout; ANOVA=analysis of variance