INVENTORY OF SUPPLEMENTAL INFORMATION:

Figure S1. **Plasma cholesterol and lipoprotein levels in *Ldlr*-/- and *Apoe*-/- mice.** – This figure provides an important control for Figure 4 demonstrating that anti-atherosclerotic interventions (simvastatin or rosiglitazone) do not alter plasma cholesterol levels or lipoprotein profiles in mice.

Figure S2. **Effect of APOE knockdown on MSRN protein expression in *Ldlr*-/- macrophages.** – This figure provides an important control for Figure 5, showing that LDLR itself is not an important regulator of the MSRN. The results are directly comparable to a similar experiment conducted in macrophages isolated from wild-type mice (Fig. 5f).

Figure S3. **Co-regulation of MSRN protein expression in *Ldlr*-/- macrophages.** – This figure expands on the data presented in Figure 5, demonstrating that the MSRN constitutes a coherent network in which co-regulation of protein expression is mediated by a variety of nodes.

Figure S4. **Macrophage cholesterol levels in *Ldlr*-/- and *Apoe*-/- mice.** – This figure is an important control for Figure 6, showing that the inability of simvastatin to correct MSRN protein expression in *Apoe*-/- mice is not because this intervention fails to influence macrophage cholesterol levels.

Figure S5. **Assessment of APOE expression by macrophages in atherosclerotic lesions.** – This figure expands on the data presented in Figure 6. It displays additional images of the regulation of APOE expression in atherosclerotic lesions by rosiglitazone and simvastatin. In addition, it demonstrates this point using both immunohistochemistry and immunofluorescence.

Figure S6. Regulation of APOE expression in acetyl-LDL loaded peritoneal macrophages. – This figure is related to Figure 1 and provides an important control demonstrating that APOE is up-regulated in classically loaded peritoneal macrophages.

Table S1: Identification of sterol-responsive proteins in macrophages isolated from *Ldlr*-/- mice. – This table is related to Figures 1, 2, and 4. It presents the MS/MS data used for analysis.

Table S2: Macrophage proteins with established roles in atherogenesis. – This table is related to Figure 3. It provides a detailed, referenced list of all of the proteins identified in the macrophage conditioned medium (and MSRN) with documented atherosclerotic phenotypes in mice with genetic manipulations (e.g., targeted mutagenesis, transgenic overexpression) of proteins.

Table S3: MSRN proteomics in microvesicles isolated from *Ldlr*-/- macrophages. – This table is related to Figure 3. It provides the MS/MS data for the detection of MSRN proteins in the isolated microvesicle fraction.

Table S4: Sterol responsive proteins identified in *Apoe*-/- mice. – This table is related to Figure 5 and corresponds to a list of all of the sterol-responsive proteins identified in *Apoe*-/- macrophage foam cells.

Table S5: MSRN proteomics in macrophages isolated from *Apoe*-/- mice. – This table is related to Figure 5. It corresponds to the statistical analysis (*t*-test) used to determined whether or not MSRN proteins were regulated by genetic ablation of APOE.

SUPPLEMENTAL MATERIAL: FIGURES

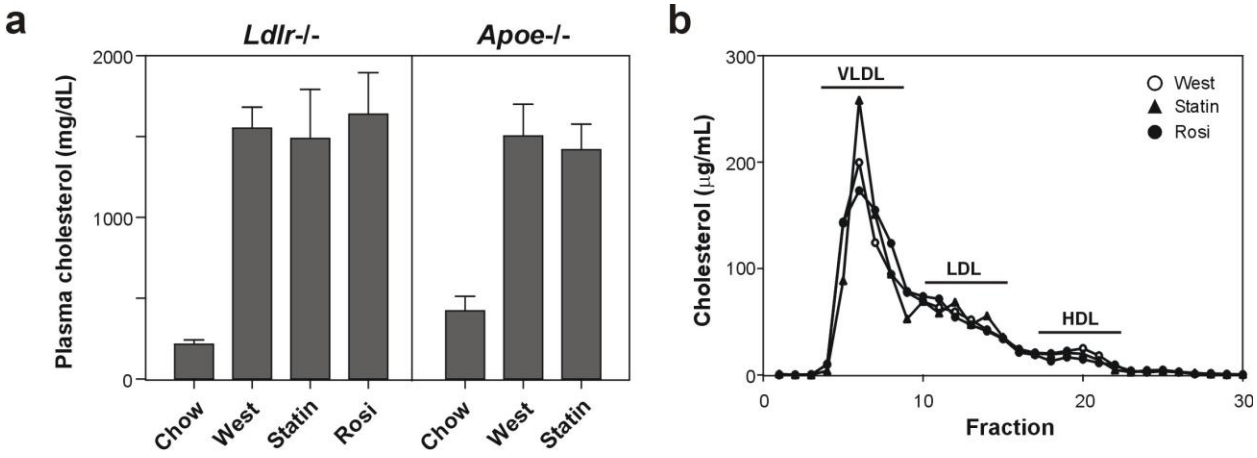

Figure S1. Plasma cholesterol and lipoprotein levels in *Ldlr*-/- and *Apoe*-/- mice, related to Figure 4. Panel A: Plasma cholesterol levels were measured in male *Ldlr*-/- or *Apoe*-/- mice fed a chow or Western (West) diet for 14 weeks. For rosiglitazone (Rosi) or simvastatin (Statin) therapy, mice fed a Western diet were treated with the interventions for the final two weeks of the regimen. Results are means and standard deviations. **Panel B:** Effect of simvastatin and rosiglitazone on plasma lipoprotein profiles in *Ldlr*-/- mice. Lipoproteins were separated by high resolution size exclusion chromatography with a Superose-6 column (GE Healthcare) and cholesterol in each fraction was measured. Plasma samples from 4 mice were pooled and analyzed in duplicate.

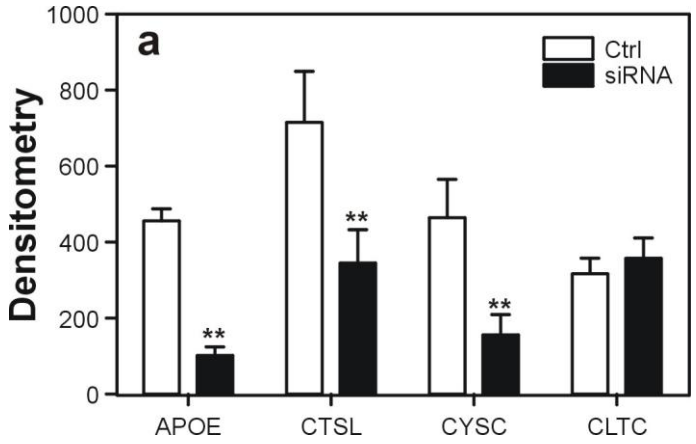

Figure S2. Effect of APOE knockdown on MSRN protein expression in *Ldlr*-/- macrophages, related to Figure 5. Elicited peritoneal macrophages isolated from *Ldlr*-/- mice were treated with control siRNA duplexes or siRNA duplexes specific for APOE. Proteins were quantified by immunoblot analysis and densitometry following a 24 h incubation in serum-free conditioned medium. Results represent means and standard deviations (N=4) and are representative of 2 independent experiments. **, $p < 0.05$ by two-tailed Student's t-test.

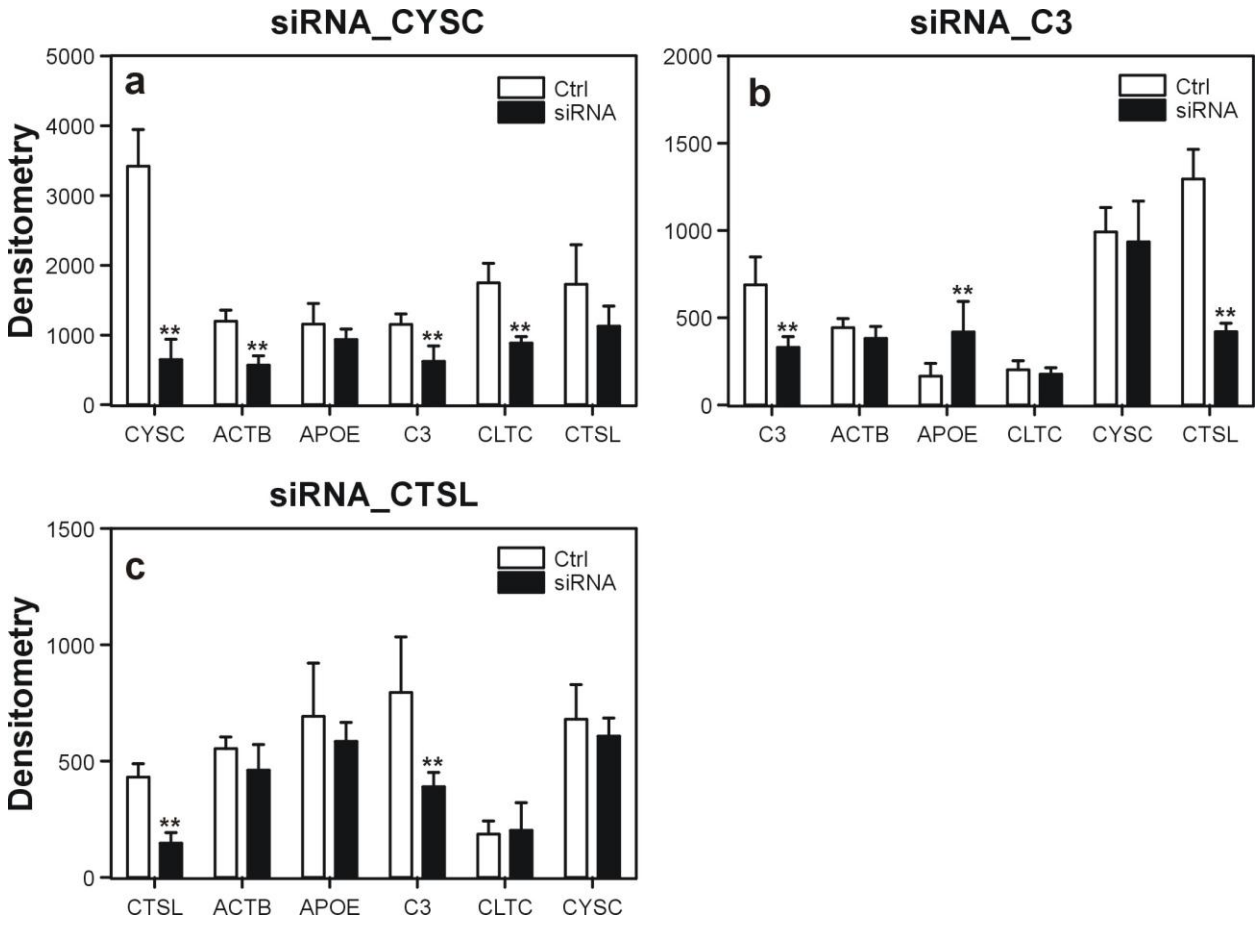

Figure S3. Co-regulation of MSRN protein expression in *Ldlr*-/- macrophages, related to Figure 5. Elicited peritoneal macrophages isolated from *Ldlr*-/- mice were treated with control siRNA duplexes or siRNA duplexes specific for CYSC (Panel A), C3 (Panel B), or CTSL (Panel C). Proteins in serum-free macrophage conditioned medium were quantified by immunoblot analysis and densitometry following a 24 h incubation. Results represent means and standard deviations (N=4) and are representative of 2 independent experiments. **, $p < 0.05$ by two-tailed Student's t-test.

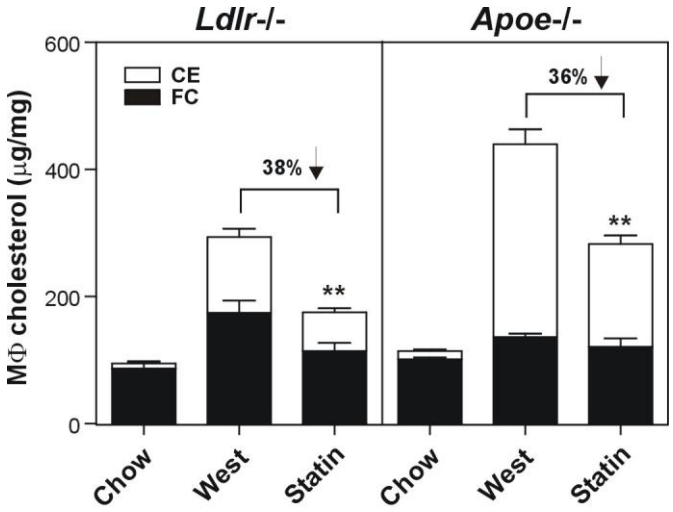

Figure S4. Macrophage cholesterol levels in *Ldlr*-/- and *Apoe*-/- mice, related to Figure 6. Cholesterol levels were measured in macrophages (MΦ) isolated from male *Ldlr-/-* or *Apoe*-/- mice fed a chow (low fat) or Western diet for 14 weeks. For rosiglitazone (Rosi) or simvastatin (Statin) therapy, mice fed a Western diet were treated with the interventions for the final two weeks of the regimen. Results are means and standard deviations. FC, free cholesterol; CE, cholesteryl ester. **, $p < 0.05$ by two-tailed Student's t-test.

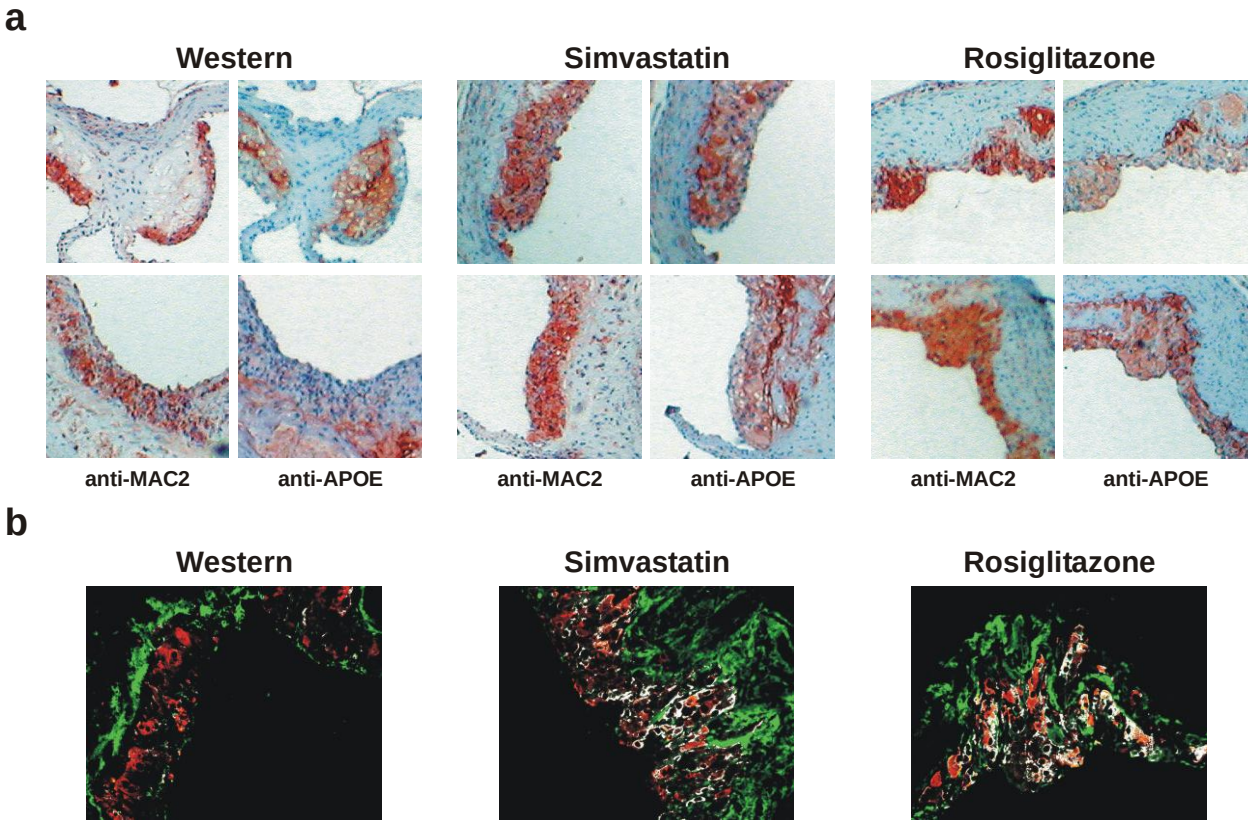

Figure S5. Assessment of APOE expression by macrophages in atherosclerotic lesions, related to Figure 6. *Ldlr-/-* mice were fed a Western diet for 14 weeks or a Western diet plus simvastatin or rosiglitazone for the final 2 weeks of the regimen. **Panel A**: Immunohistochemical staining of aortic sinus sections isolated from *Ldlr-/-* mice on the different regimens. Adjacent sections were immunostained with antibodies to APOE or MAC2 (a macrophage marker). **Panel B**: Aortic sinus sections were stained with antibodies to APOE (green channel) or MAC2 (red channel). Areas of colocalization (white) were identified using Image J software.

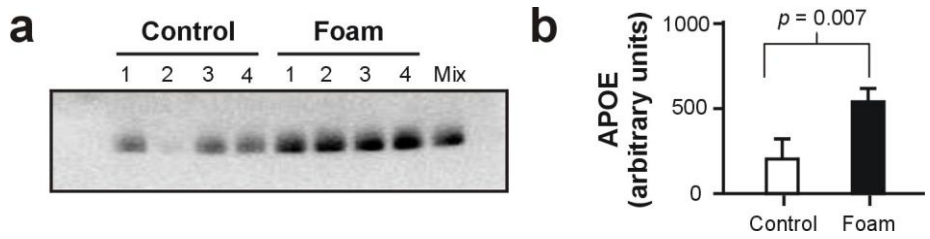

Figure S6. Regulation of APOE expression in acetyl-LDL loaded peritoneal macrophages, related to Figure 1. Elicited peritoneal macrophages were cultured in DMEM containing 10% FBS and 50 μg/mL LDL (Control) or 50 μg/mL acetylated-LDL (Foam) for 3 days with daily medium changes (Brown and Goldstein, 1983). Cells were washed with PBS and incubated in serum-free DMEM for 6 h. **Panel A.** Equal amounts of protein from medium conditioned by control (N=4) or foam (N=4) cells were subjected to SDS-PAGE using 4%–12% gradient gels, transferred to PVDF membranes, and subjected to immunoblot analysis with an antibody against murine APOE. Mix: a mixture of equal amounts of protein from all 8 biological samples. **Panel B:** Protein quantification. Immunoblots were quantified by densitometry (means and standard deviations). Statistical significance was assessed using a two-tailed Student's t-test.