

## Supplementary Figure Legends

### **Supplement Figure I TL1A decreases cholesterol efflux from THP-1 macrophages loaded with AcLDL and [4-14C]-cholesterol in the absence of 10 % (v/v) delipidated fetal calf serum**

24 hour PMA-differentiated THP-1 macrophages were converted into foam cells with 50 µg/ml AcLDL and 0.5 µCi/ml [4-14C]-cholesterol for 24 hours (black filled histogram) or 48 hours (black striped histogram) in media containing 0.2 % (v/v) fatty-acid free BSA (Sigma). Following this, cells were treated with 10 µg/ml apoA-I in the presence or absence of 100 ng/ml TL1A for 24 hours. Cholesterol efflux was then assayed. Control cells exhibited approximately 20 % efflux and were arbitrarily assigned as 1 from which fold change was calculated. Data is derived from four separate experiments (n=4) and represents mean±SD. Student's t-test was used ascertain the statistical significance of all results with \*\*  $P < 0.01$  and \*\*\*  $P < 0.001$ .

### **Supplement Figure II TL1A decreases apoE, ABCA-1 and ABCG-1 mRNA expression in AcLDL-converted foam cells**

Real-time quantitative PCR for apoE, ABCA-1 and ABCG-1 was performed on cDNA from 24 hour PMA-differentiated THP-1 macrophages incubated with 50 µg/ml AcLDL in the presence or absence of 100ng/ml TL1A for 24 hours. Relative expression levels were calculated using comparative Ct method and normalized to 28s rRNA levels with non-TL1A stimulated, AcLDL loaded cells given an arbitrary value of 1. Data is derived from four separate experiments (n=4) and represents mean±SD. Student's t-test was used to ascertain the statistical significance of the results with \*\*\*  $P < 0.001$

### **Supplement Figure III TL1A does not induce apoptosis in human macrophages**

Cell apoptosis was measured in 24 hour PMA-differentiated THP-1 macrophages and 7 day differentiated HMDMs by dual-color immunofluorescent staining with Annexin V-FITC and Propidium iodide (PI). Cells were either left untreated or stimulated with TL1A (100 ng/ml) for 24 hours. A representative flow cytometric analysis is shown in (A). The percentages in the upper right quadrants indicate the % Annexin V positive / PI positive cells, which represent late apoptotic cells. The percentages in the lower right quadrants indicate the % Annexin V positive / PI negative cells, which represent early apoptotic cells. The histograms in (B) correspond to data collected from both quadrants from four separate experiments (n=4) and represents mean $\pm$ SD. Black filled histograms represent data for THP-1 macrophages. Black striped histograms indicate data for HMDMs

### **Supplement Figure IV TL1A regulates the expression of key proteins implicated in the uptake and efflux of cholesterol in THP-1 macrophages and HMDMs**

24 hour PMA-differentiated THP-1 macrophages or 7 day differentiated HMDMs were incubated for 24 hours with or without 100 ng/ml TL1A. Equal levels of protein from total lysates were analyzed by Western blotting using antibodies against CD36, SR-A, SR-B1, apoE, ABCA-1, ABCG-1 &  $\beta$ -actin. Semi-quantitation of all Western blots was performed by densitometric analysis using the Gene Tools software (GRI) and the uptake/efflux protein: $\beta$ -actin ratio from untreated cells has been arbitrarily assigned as 1. For proteins with different forms (e.g. SR-A), all the immunoreactive bands on the blots were subjected to densitometric analysis. Black filled histogram indicates data for THP-1 macrophages and the black striped histogram for HMDMs. Data is derived from four separate experiments (n=4) and represents mean $\pm$ SD. Student's t-test was used to ascertain the statistical significance of the results with \*  $P < 0.05$ , \*\*  $P < 0.01$  and \*\*\*  $P < 0.001$ .

**Supplement Figure V TL1A up-regulates the mRNA expression of SR-PSOX in  
THP-1 macrophages and HMDMs**

Real-time quantitative PCR for SR-PSOX was performed on cDNA from 24 hour PMA-differentiated THP-1 macrophages or 7 day differentiated HMDMs. Relative mRNA expression levels were calculated using comparative Ct method and normalized to 28s rRNA levels with untreated cells given an arbitrary value of 1. Black filled histogram indicates data for THP-1 macrophages and the black striped histogram for HMDMs. Data is derived from four separate experiments (n=4) and represents mean±SD. Student's t-test was used to ascertain the statistical significance of the results with \* $P < 0.05$

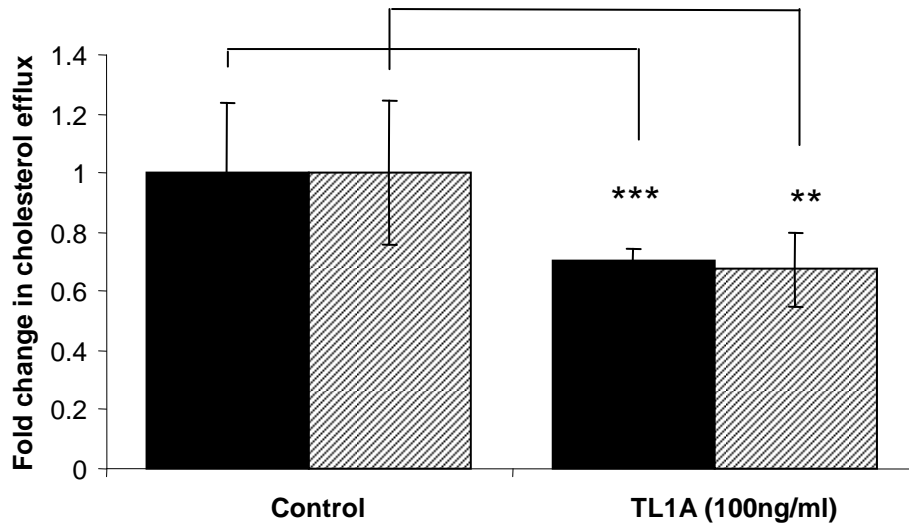
**Supplement Table I Primers used for real-time quantitative PCR**

<b>Gene</b>	<b>Forward Primer (5' - 3')</b>	<b>Reverse primer (5' - 3')</b>
Human SR-A	CCAGGGACATGGAATGCAA	CCAGTGGGACCTCGATCTCC
Human CD36	GAGAACTGTTATGGGGCTAT	TTCAACTGGAGAGGCAAAGG
Human SR-BI	TGATGATGGAGAATAAGCCCAT	TGACCGGGTGGATGTCCAGGAAC
Human LPL	GAGATTTCTCTGTATGGCACC	CTGCAAATGAGACACTTTCTC
Human SR-PSOX	ACTCAGCCAGGCAATGGCAAC	GGTATTAGAGTCAGGTGCCAC
Human ApoE	TTCTTGGCAGGATGCCAGGC	GGTCAGTTGTTCTCCAGTTC
Human ABCA1	TGTCCAGTCCAGTAATGGTTCTGT	AAGCGAGATATGGTCCGGATT
Human ABCG1	TGCAATCTTGTGCCATATTTGA	CCAGCCGACTGTTCTGATCA
Human 28S rRNA	TTAGACCGTCGTGAGACAGG	TTCAATAGATCGCAGCGAGG
Mouse SR-A	TGAACGAGAGGATGCTGACTG	GGAGGGGCCATTTTTAGTGC
Mouse CD36	GAACCACTGCTTTCAAAACTGG	TGCTGTTCTTTGCCACGTCA
Mouse SR-BI	TTTGGAGTGGTAGTAAAAAGGGC	TGACATCAGGGACTCAGAGTAG
Mouse ApoE	ACAGATCAGCTCGAGTGGCAA	ATCTTGCGCAGGTGTGTGGAGA
Mouse ABCA1	AGTGATAATCAAAGTCAAAGGCACAC	AGCAACTTGGCACTAGTAACTCTG
Mouse ABCG1	TTCATCGTCCTGGGCATCTT	CGGATTTTGTATCTGAGGACGAA
Mouse $\beta$ -actin	TGGAGAAGAGCTATGAGCTGCCTG	GTGCCACCAGACAGCACTGTGTTG

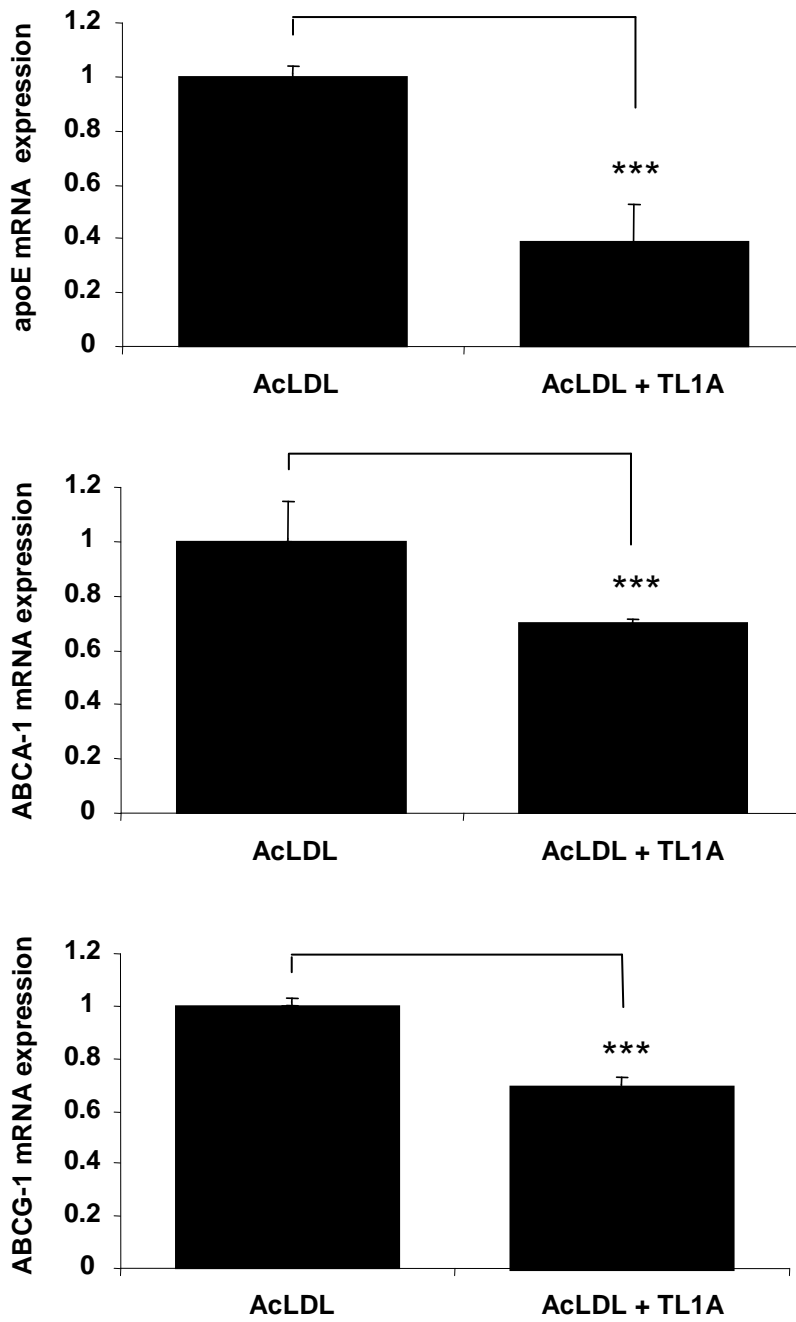
**Supplement Table II Forward and reverse oligonucleotide sequences used for generation of  
DR3 shRNA and scrambled shRNA adenoviruses**

<b>Oligonucleotide</b>	<b>Sequence</b>
<b>DR3 shRNA (F)</b>	AGCCTGGATCCCTGGAGGCTTGCTGAAGGCTGTATGCTGTTC TCACTGCTGTCAGGAGGTGTTTTGGCCACTGACTGACACC
<b>DR3 shRNA (R)</b>	ATTTGTTCCATGTGAGTGCTAGTAACAGGCCTTGTGTCCTGT TCTCACTGCTCAGGAGGTGTCAGTCAGTGGCCAAAACACC
<b>Scrambled shRNA (F)</b>	AGCCTGGATCCCTGGAGGCTTGCTGAAGGCTGTATGCTGTA TACGTTTGATTTGACCGCGTTTTGGCCACTGACTGACGCG
<b>Scrambled shRNA (R)</b>	ATTTGTTCCATGTGAGTGCTAGTAACAGGCCTTGTGTCCTGT ATACGTTTGATCGACCGCGTCAGTCAGTGGCCAAAACGCG

## Supplement Figure I

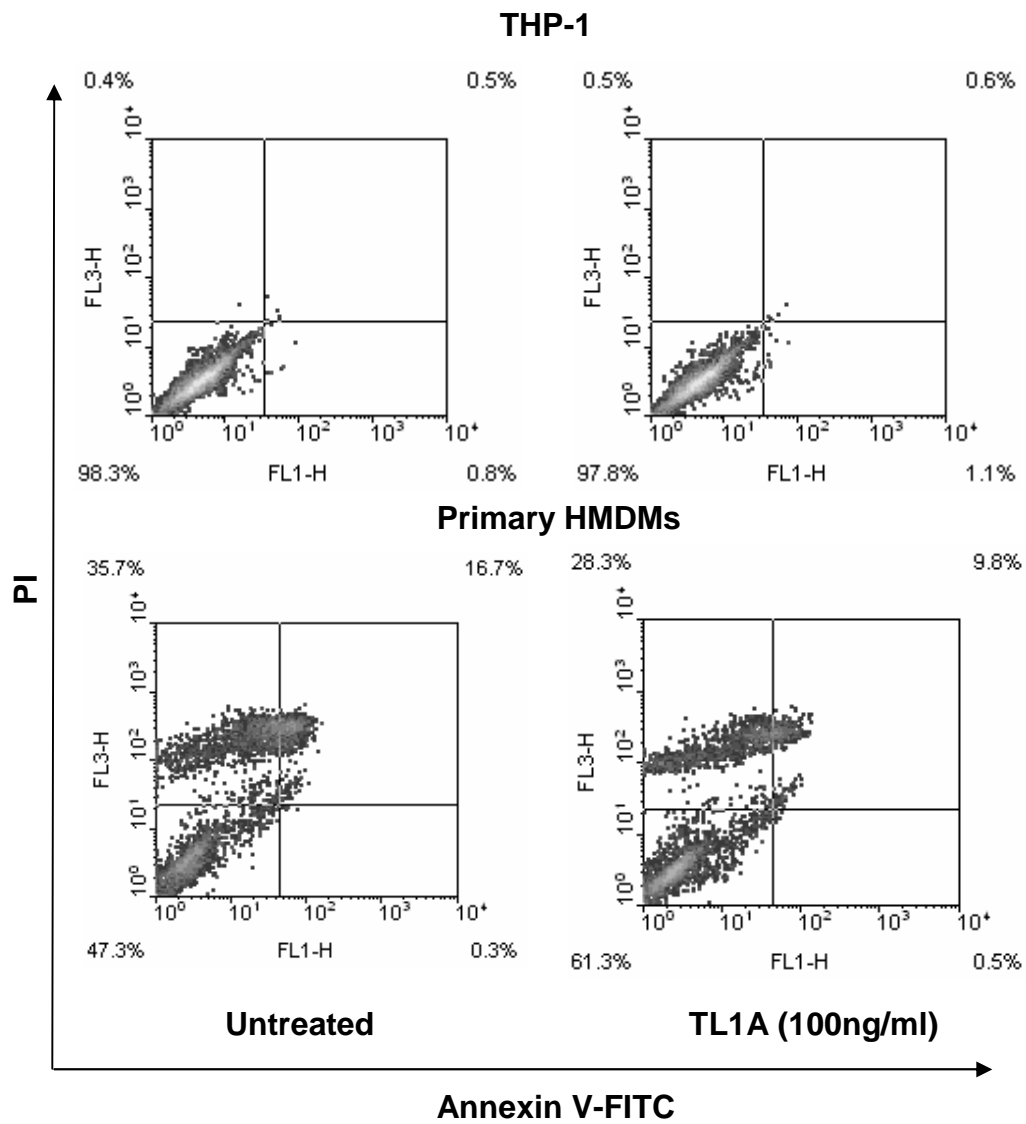


## Supplement Figure II

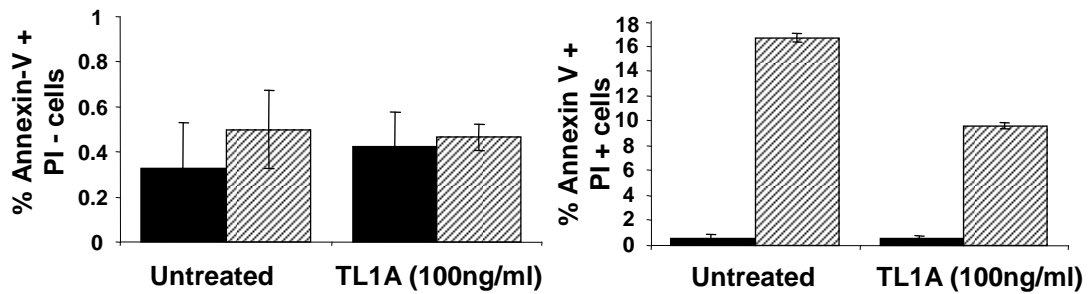


# Supplement Figure III

**A**

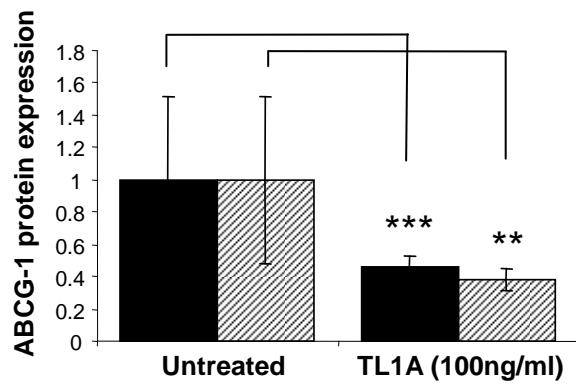
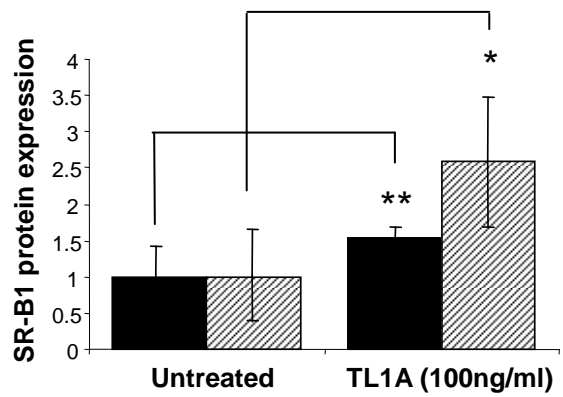
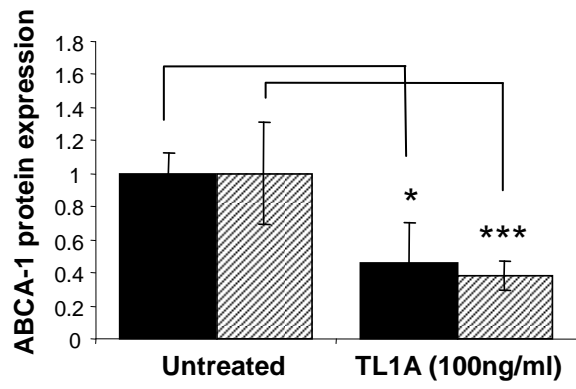
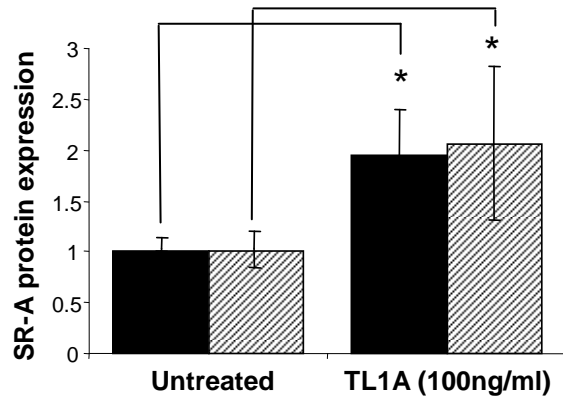
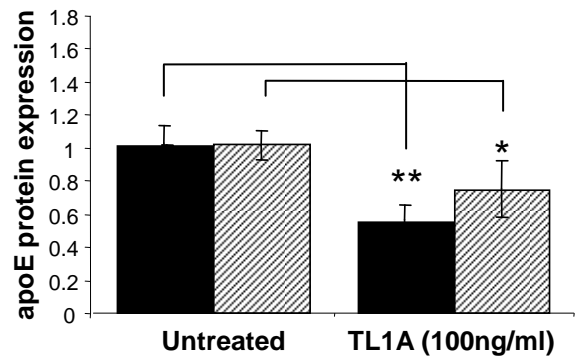
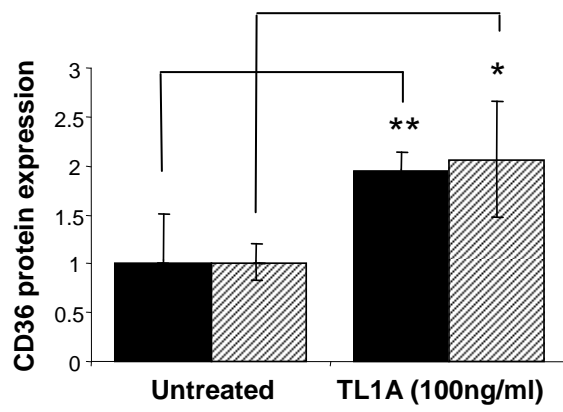


**B**





# Supplement Figure IV



## Supplement Figure V

