IL-7R Blockade Reduces Post-Myocardial Infarction-Induced Atherosclerotic Plaque Inflammation in ApoE(-/-) Mice

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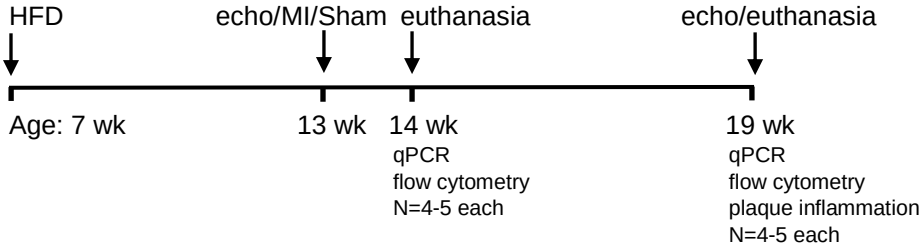
SUPPLEMENTARY MATERIAL

Antibodies Used

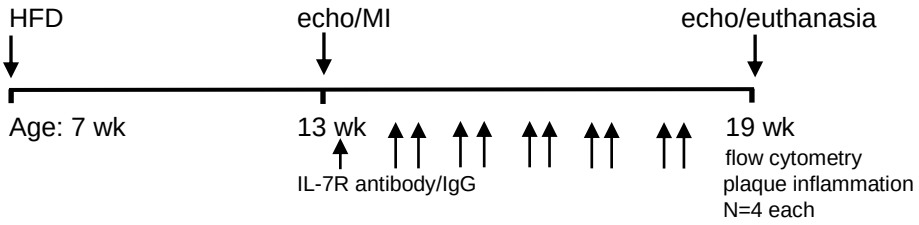
Antigen	Vendor or Source	Catalog #	Working concentration
MOMA-2	BioRad	MCA519G	1:50
CD3	Biolegend	100202	1:50
Ly6G	Biolegend	127602	1:50
CD3 BUV 395	BD Horizon	563565	1:100
CD11c AlexFluor 700	eBioscience	56-0114-80	1:100
CD11b APC-eF780	eBioscience	47-0112-82	1:100
F4/80 APC	Biolegend	123115	1:100
Ly6G FITC	eBioscience	53-5931-82	1:100
Ly6C eFlour 450	eBioscience	48-5932-80	1:100
CD127 PE	eBioscience	12-1271-81	1:100
IL-7	Boster Biological Technology	PA1467	1:50
IL-7Rα blocking antibody	BioXCell	BE0065	See Methods

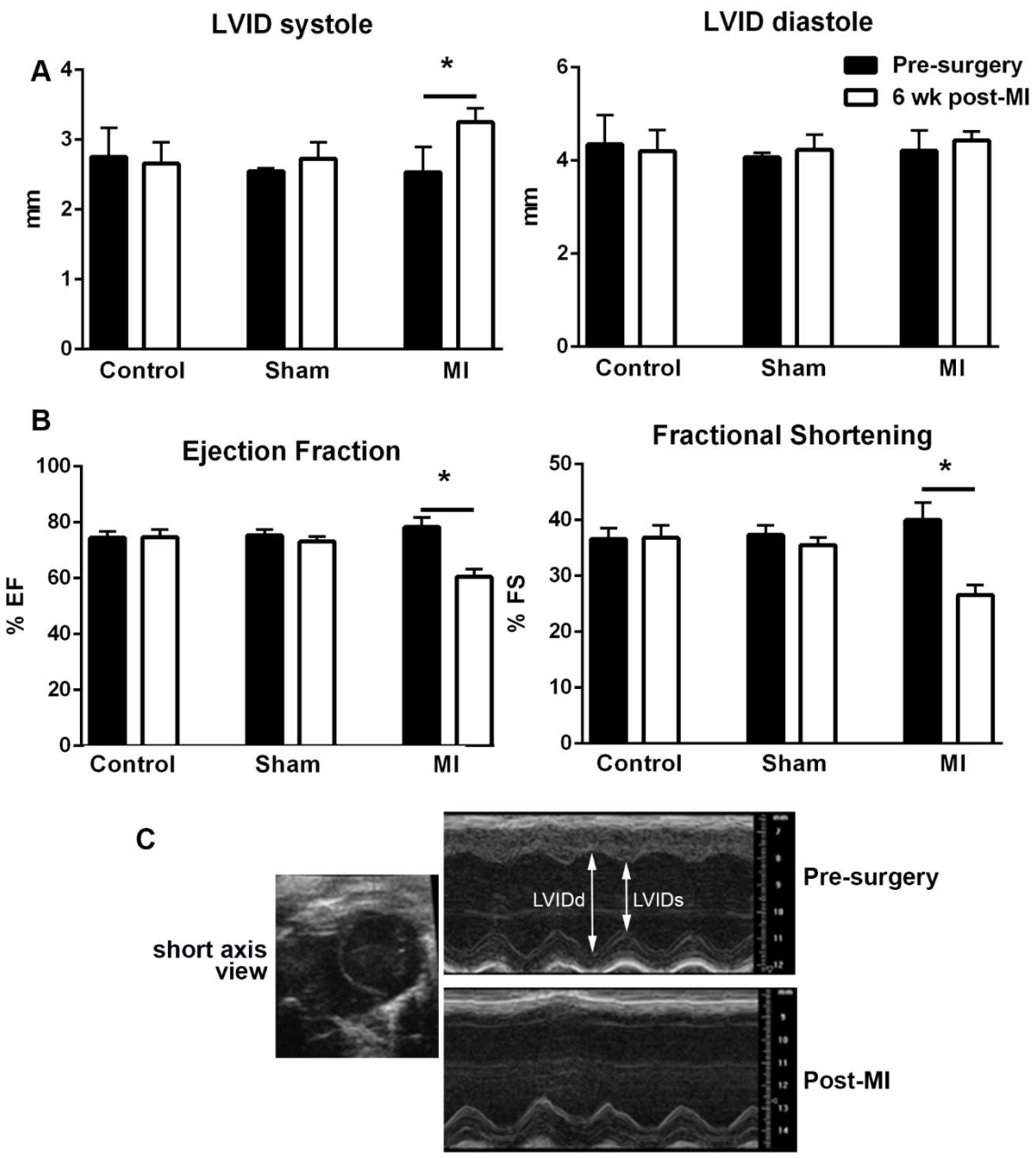
Study Design:

I.



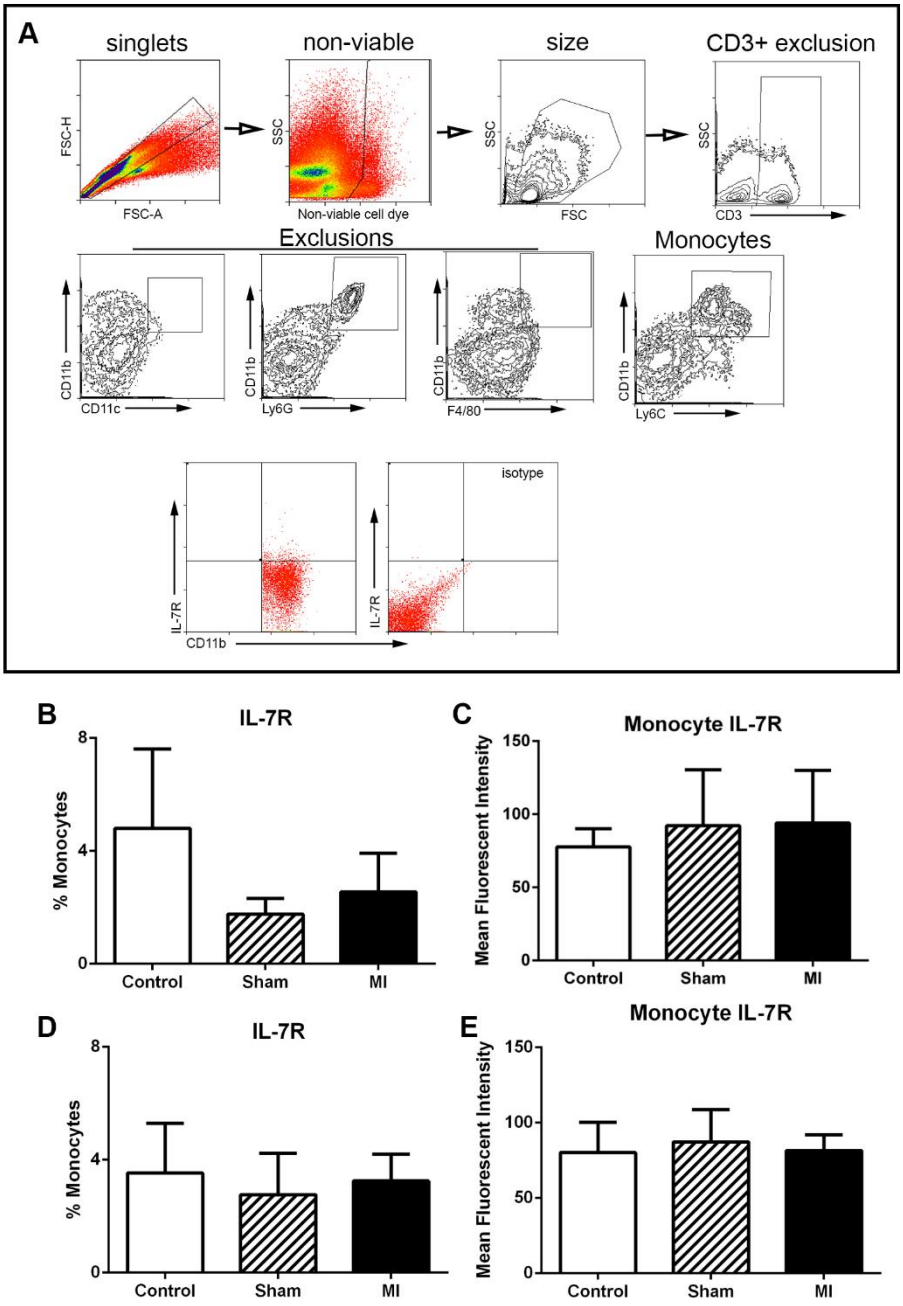
II.





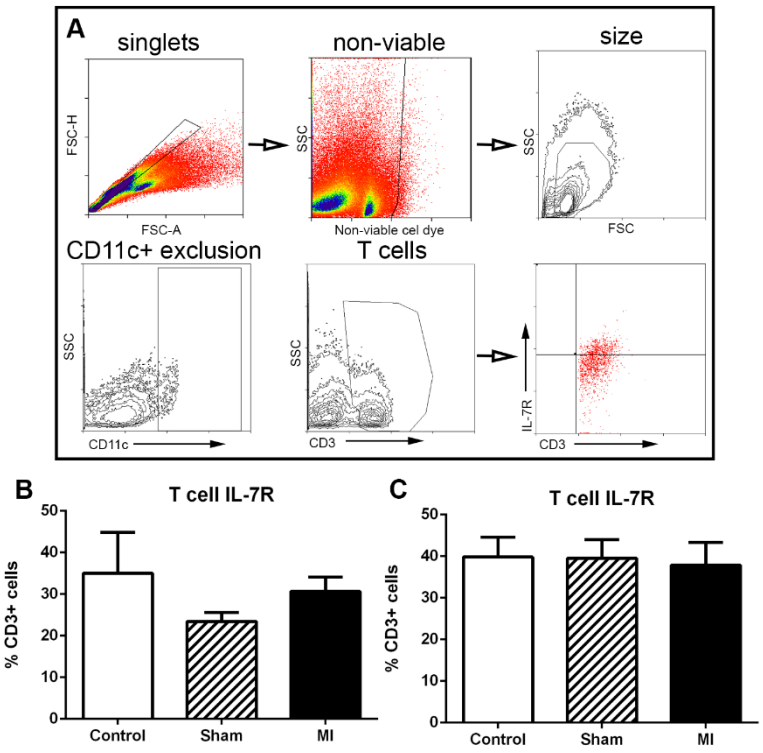
Supplemental Figure 1: Cardiac function of ApoE$^{-/-}$ mice subjected to MI.

ApoE$^{-/-}$ mice fed high fat diet for 6 weeks were subjected to surgical MI. Echocardiographic measurements of left ventricular internal dimension (LVID) at systole (A, left panel) and diastole (A, right panel). Ejection fraction (B, left panel) and fractional shortening (B, right panel) in mice. Representative echocardiography image (C) depicting measurement of LVID at diastole (LVIDd) and systole (LVIDs). Control and Sham N=4 each; MI N=5; LVID systole *p=0.01 t-test; ejection fraction *p<0.0001; fractional shortening *p<0.0001.



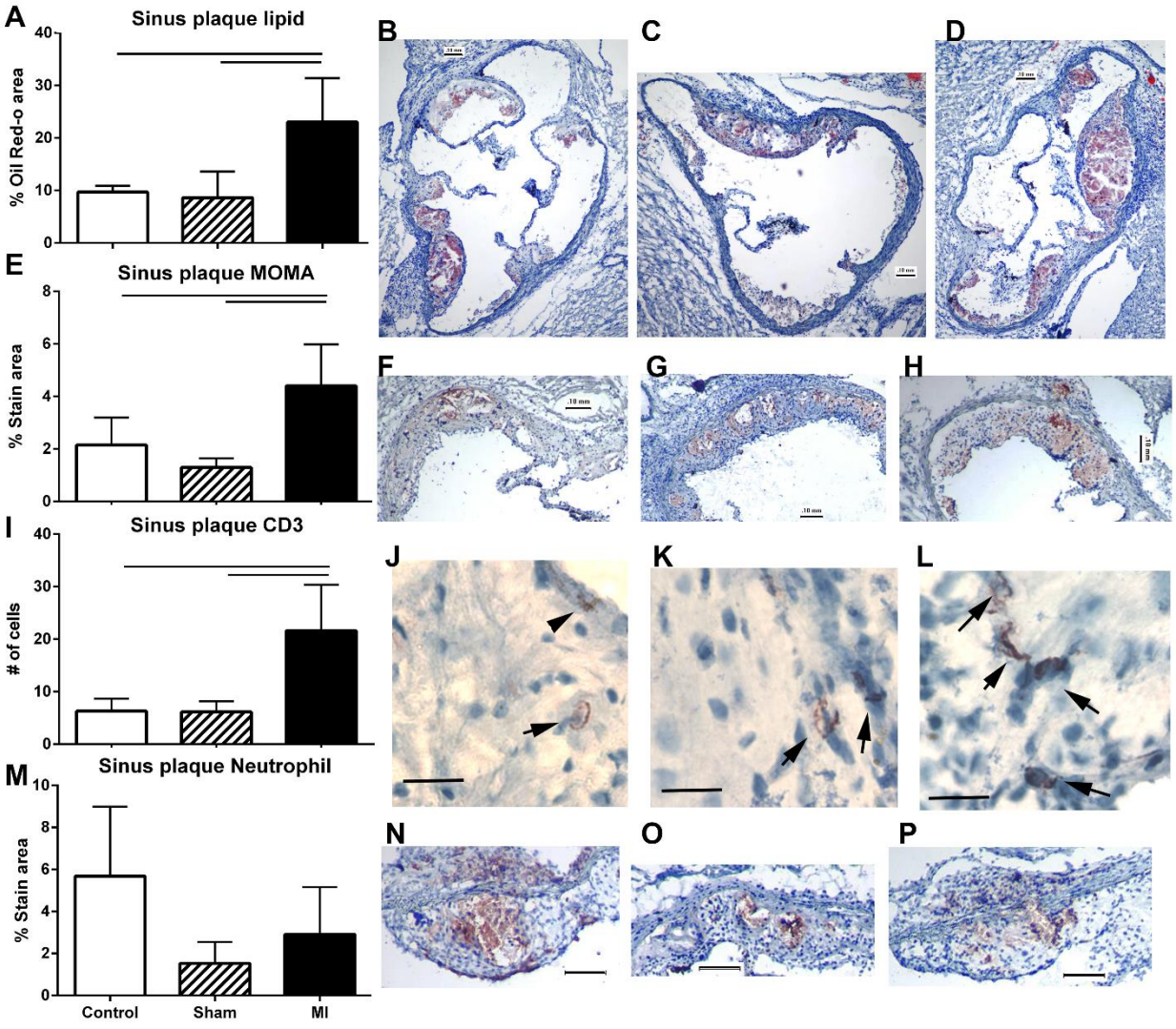
Supplemental Figure 2: Monocyte expression of IL-7R post-MI.

Gating strategy for analysis of monocytes from splenocytes (A). Cell singlets and viable cells were size gated and CD3+ T cells excluded. CD11b+Ly6C+ monocytes excluded CD11c, Ly6G, and F4/80 were analyzed for IL-7R expression. Splenocytes from mice 1 week (B and C) or 6 weeks (D and E) post-MI showing percentage of IL-7R+ cells as well as IL-7R expression level by mean fluorescent intensity. N=4-5 each group.



Supplemental Figure 3: T cell expression of IL-7R post-MI.

Gating strategy for analysis of T cells from splenocytes (A). Cell singlets and viable cells were size gated and CD3+ cells analyzed after excluding CD11c+ cells. Percentage of IL-7R+ T cells 1 week (B) and 6 weeks (C) post-MI. N=4-5 each.



Supplemental Figure 4: Plaque inflammation post-MI.

Aortic sinus plaque lipid (A-D), macrophage (E-H), T cells (I-L), and neutrophils (M-P) were stained using oil-red-o, MOMA-2 antibody, CD3 antibody, and Ly6G antibody, respectively. Representative photos from: Non-surgical control (B, F, J, N; N=4); sham (C, G, K, O; N=4), and MI mice (D, H, L, P; N=5). Scale bar=0.1mm, except J-L (scale bar=0.02mm). Bar over columns indicate statistical significance. Sinus plaque lipid: Control vs MI p=0.02; Sham vs MI p=0.01. Sinus plaque MOMA: Control vs MI p=0.03; Sham vs MI p=0.003. Sinus plaque CD3: Control vs MI p=0.007; Sham vs MI p=0.007.