Supplemental Table 1. Antibodies

Primary Antibodies (Supplier Cat#)	**Secondary Antibodies (Supplier Cat#)**
CD31-Mouse- (Dianova-DIA310) CD31-Human- (Dako GA610) CD4 (BD Pharmingen 550280) CD8a (eBioscience 14-0808-82) CD3 & CD45 (Biolegend 100201 & 103101) Apelin (Santa Cruz sc33469) Mac-2 (Cedarlane: CL8942AP) Ym1 (StemCell: 60130) APJ (Phoenix H-001-79) ESM-1 (MyBiosource MBS2006250) EGFL7 (BIOMATIK CAU21428) F4/80 (Biolegend 123102). CD14 & CD16 (STEMCELL 60004AZ & 60041PE). T-AKT, P-AKT^{473}, T-eNOS, P-eNOS^{1177} and Actin-b (Cell signalling 4691, 4060, 5880, 9571 and 4970 / 3700) Rabbit Anti-ESM1 polyclonal antibody (Causabio; CSB-PA007825LA01HU) Anti-VE-Cadherin Antibody (Sinobiological 50192-T56) Anti-CD34 Monoclonal Antibody	Biotinylated Goat anti Rat / Rabbit & Mouse IgG (H+L) (Vector Labs BA-9401, BA-1000 & BA-9200 respectively). Goat anti Rat (H+L) Cross-Adsorbed Alexa Fluor 488 (ThermoFisher A-11006). Goat anti Rabbit (H+L) Cross-Adsorbed, Alexa Fluor 568 (ThermoFisher A-11036). Donkey anti-Goat IgG HL Cross-Adsorbed Secondary Antibody Alexa Fluor 568 (ThermoFisher A-11057) Donkey anti-Rat IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor 488 (ThermoFisher A-21208).

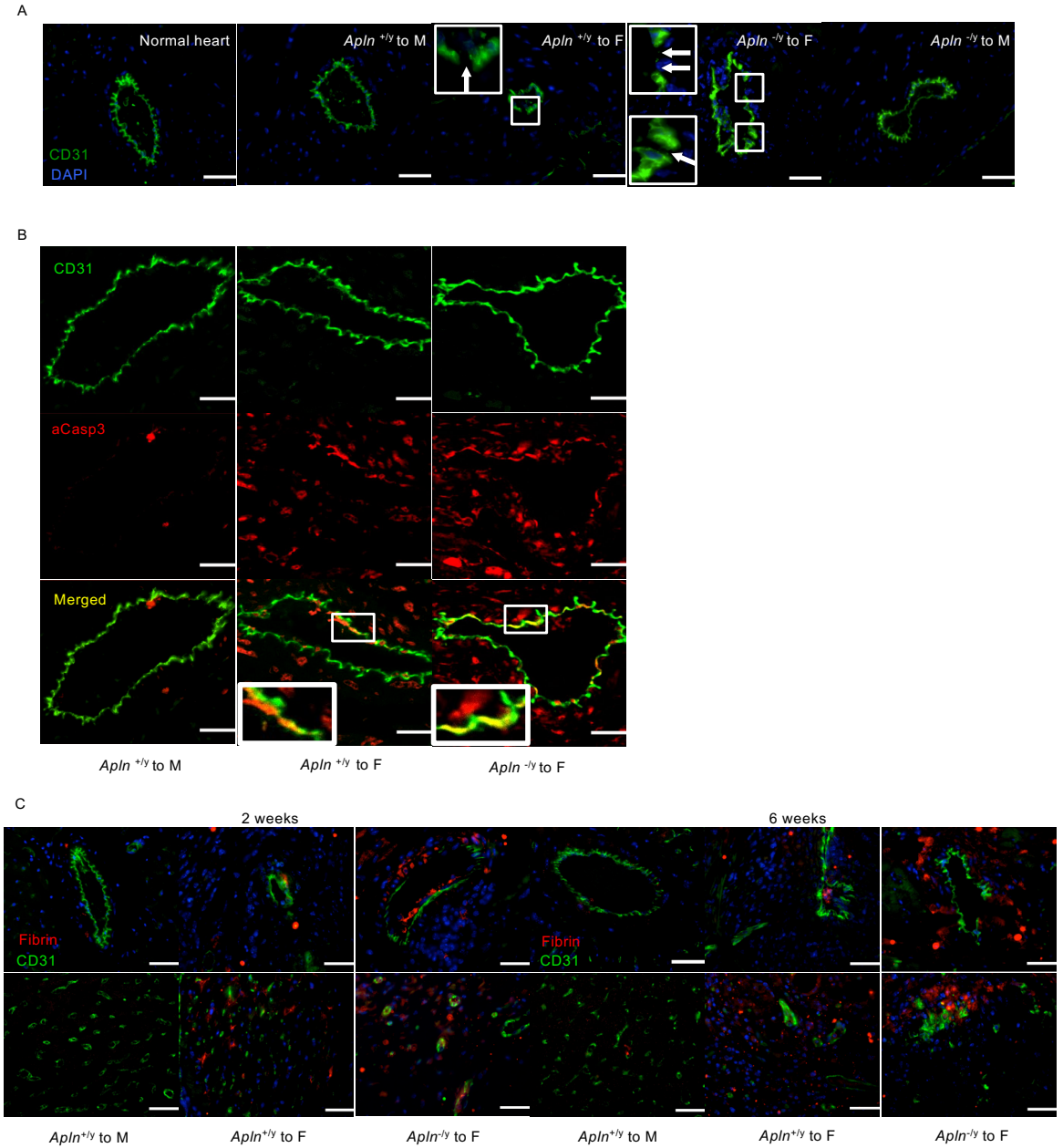
(Thermofischer; QBEND/10, MA1-10202) Anti human Apelin polyclonal antibody (Abcam; ab59469) Anti-fibrinogen antibody (DAKO; A008002-2)	

Supplemental Table 2. Mouse primers for qRT-PCR

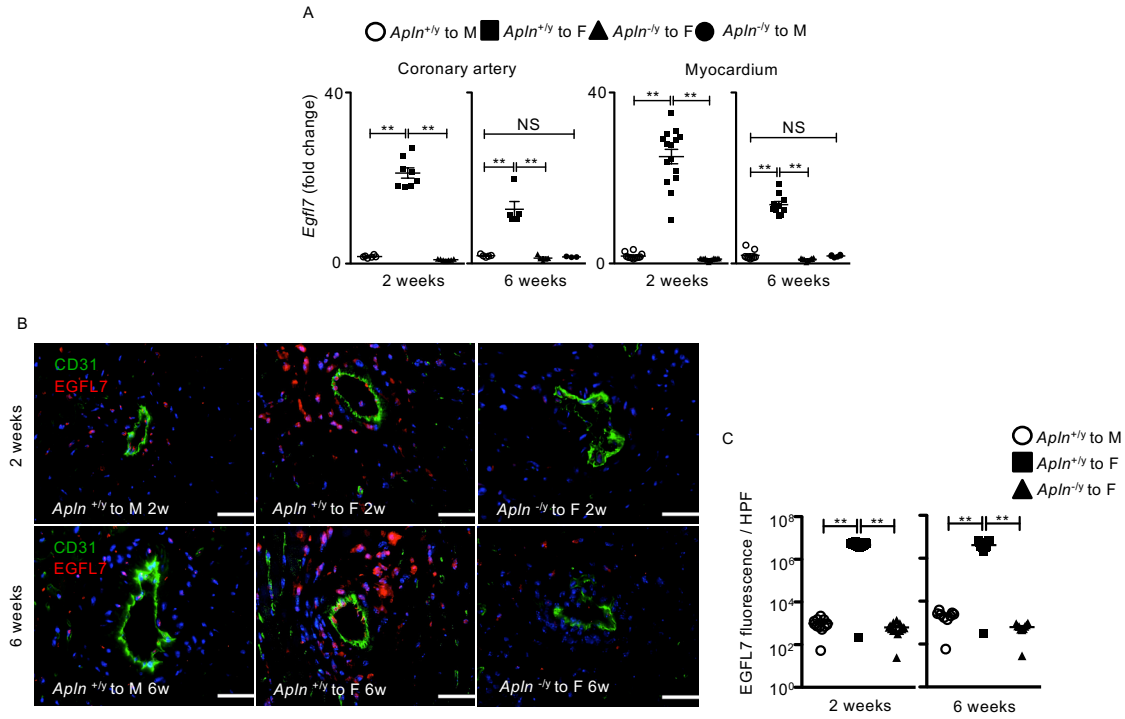
Esm1	F: 5' AGCGAGGAGGATGATTTTGGT 3' R: 5' TGCATTCCATCCCGAAGGT 3'
Apln	F: 5' TAGCCCCTGACACTGGTTGTC 3' R: 5' TTCTCCATCCCCCAAAAGC 3'
Pdgfb	F: 5' CCCTCGGCCTGTGACTAGAA 3' R: 5' AATGGTCACCCGAGCTTGAG 3'
Pecam1	F: 5' AGGACGATGCGATGGTGTATAA 3' R: 5' AAGACCCGAGCCTGAGGAA 3'
Tnfa	F: 5' ATGATCCGCGACGTGGAA 3' R: 5' TAGGCACCGCCTGGAGTTC 3'
Vegfa	F: 5' GCAGGCTGCTGTAACGATGA 3' R: 5' TCCGCATGATCTGCATGGT 3'
Cxcl11	F: 5' GGGCCGATGCAAAGACA 3' R: 5' GAGATGAACAGGAAGGTCACAG 3'

Supplemental Table 3. Human primers for qRT-PCR

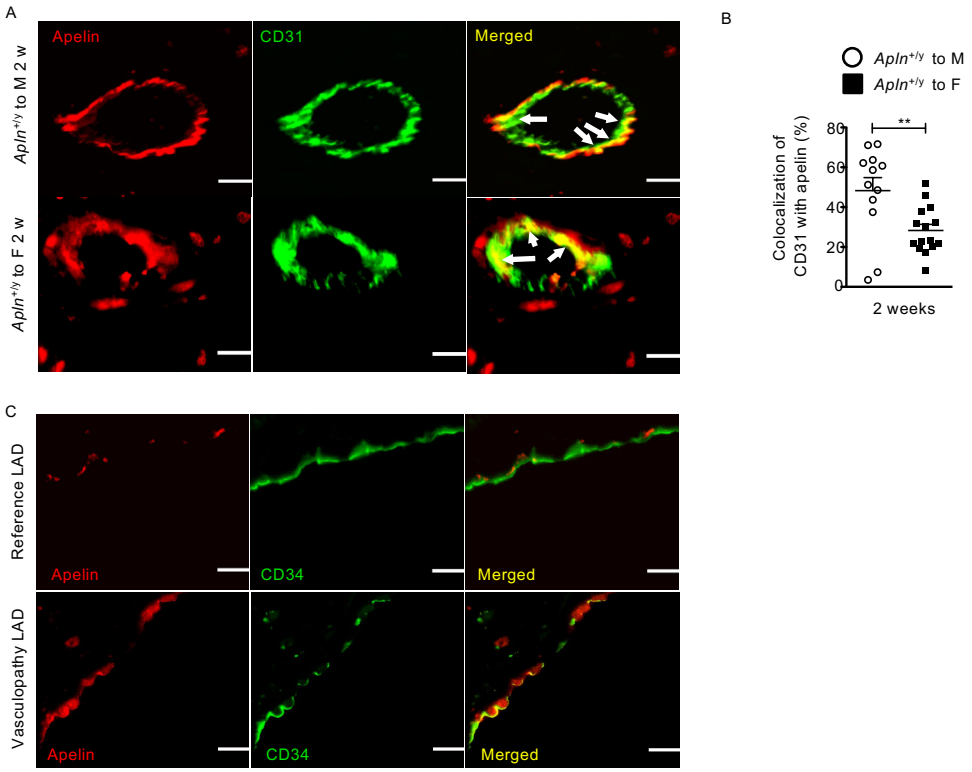
APLN	F: 5’ CCCATGCCCACATATTGCA 3’ R: 5’ TCAGTTTGAGGCCACTTGACCTA 3’
PECAM	F: 5’ AGTGGAGTCCAGCCGCATAT 3’ R: 5’ CAGTTCGGGCTTGGAAAATAGT 3’
PDGFB	F: 5’ AGATCGAGATTGTGCGGAAGA 3’ R: 5’ GCTGCCACTGTCTCACACTTG 3’
GAPDH	F: 5’ GATTCCACCCATGGCAAATT 3’ R: 5 TGATGGGATTTGCATTGATGAC 3’
ESM1	F: 5’ GGTGGACTGCCCTCAACACT 3’ R: 5’ GTCGTCGAGCACTGTCCTCTT 3’



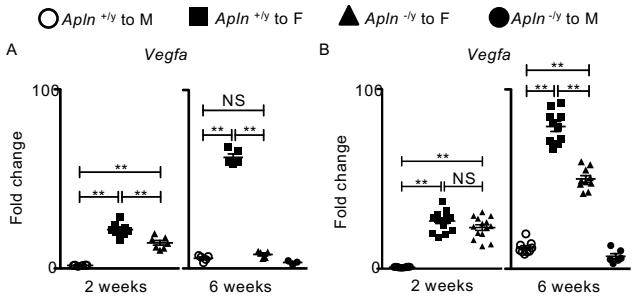
Supplemental Fig 1: Graft coronary arterial injury post-transplantation. A, Loss of continuity of the arterial endothelium in hearts at 2 weeks posttransplantation. Photomicrographs show immunofluorescent staining of the endothelial marker Pecam1 (CD31; green) with DAPI nuclear staining (blue). Arrows indicate areas of endothelial loss / gaps in endothelium (quantitation is in figure 1B). B, Arterial endothelial apoptosis in hearts at 2 weeks post- transplantation. Confocal photomicrographs show double immunofluorescent staining of the endothelial marker Pecam1 (CD31; green) with cleaved caspase 3 (red). Insets show caspase 3+ endothelial cells. Colocalization is quantitated, and is shown in Figure 1B (right panel). C, Fibrin associated with the graft arterial (upper panels) and microvascular (lower panels) endothelium post-transplantation. Photomicrographs show double immunofluorescent staining of the endothelial cell marker Pecam1(CD31; green) and fibrin (red). n=4-15. Scale bar = 50μm.



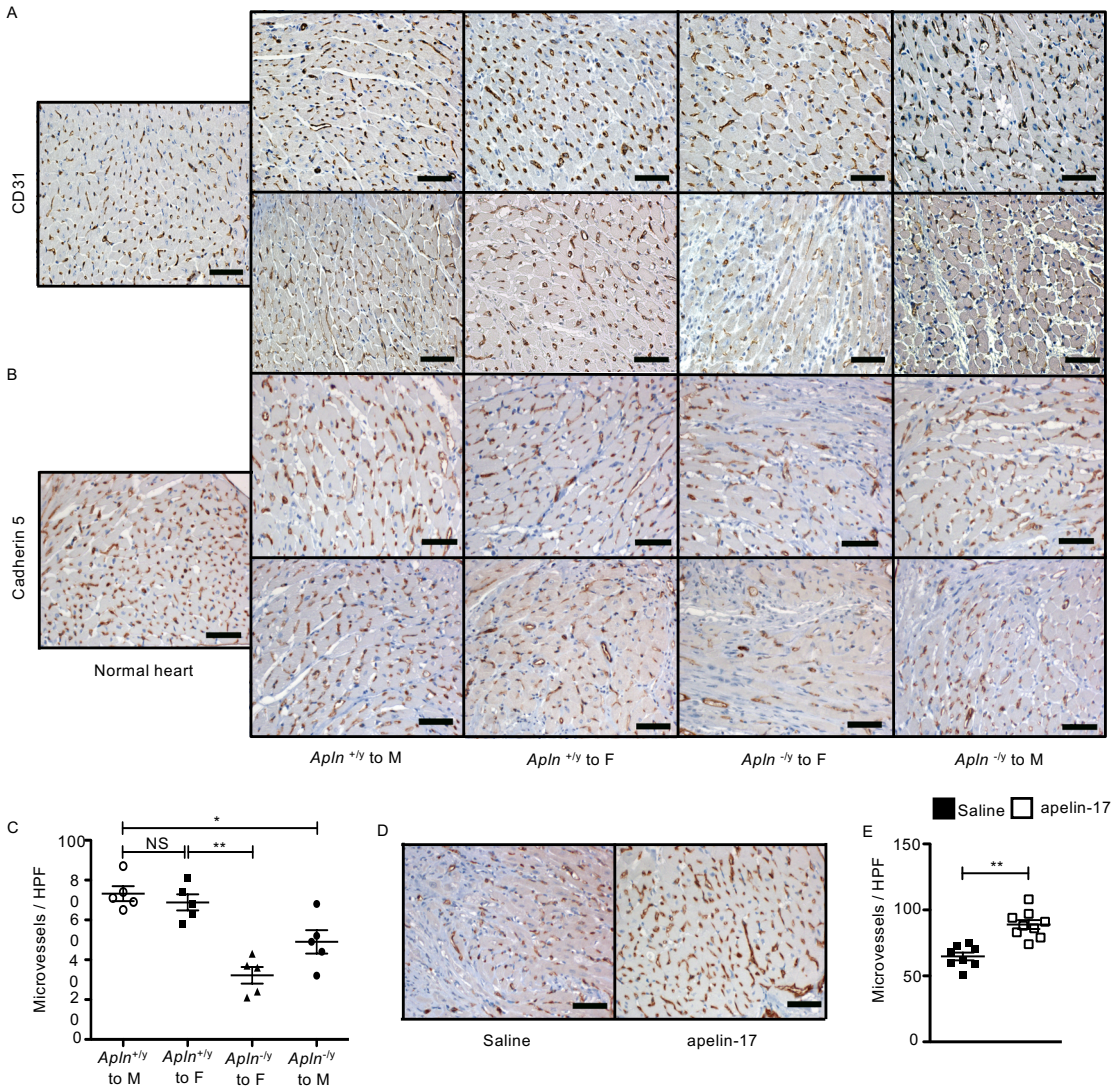
Supplemental figure 2: *Egfl7* gene expression in graft coronary artery or myocardium post-transplantation. A, Consecutive pairs of coronary artery samples were pooled for qRT-PCR analysis (n=3-8 pairs); individual myocardium samples are shown (n=6-15 biological replicates), relative to normal hearts. Samples of heart grafts were obtained at 2 weeks and 6 weeks post-transplant as indicated. Mean ± SEM; **P<0.01 and NS=non significant by one way ANOVA with Bonferroni's post-hoc test. B, Double immunofluorescent staining of CD31 (green) and EGFL7 (red) in the heart grafts. C, Quantitation of the total corrected EGFL7 fluorescence/ HPF from (C). n=9-15 biological replicate hearts/ group. Scale bar = 50 μm. Mean ± SEM; **P<0.01 by one way ANOVA with Bonferroni's post-hoc test.



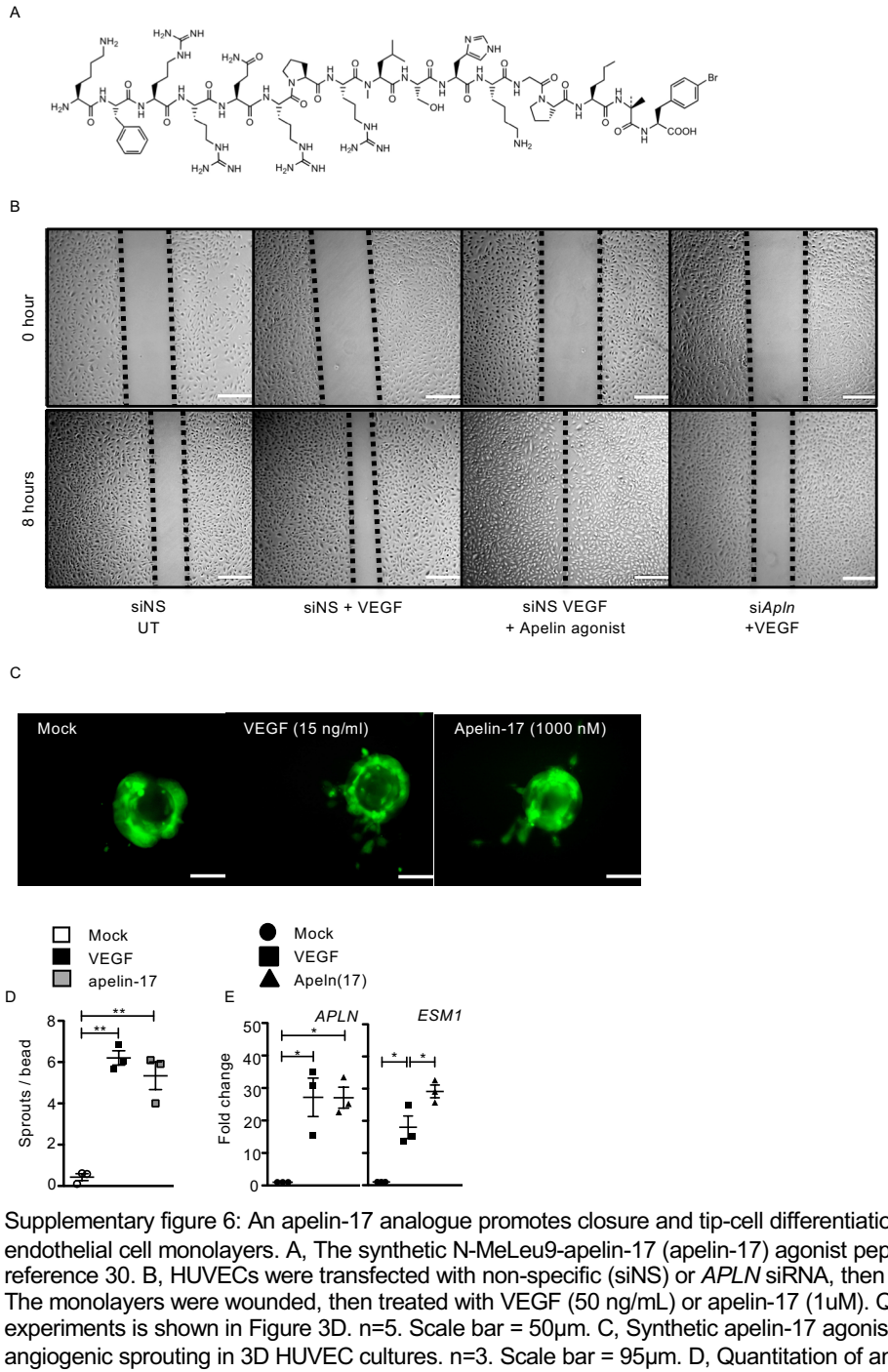
Supplemental figure 3: Apelin expression in heart grafts. A, Confocal photomicrographs show double immunofluorescent staining of apelin (red) and endothelial CD31 (green) in heart grafts at 2 weeks post-transplant (isograft hearts with reperfusion injury are represented in the top panel *vs* heart allografts with immune and reperfusion injury are in the lower panel). Apelin knockout (*Apln*$^{-/y}$) heart grafts show no apelin staining. Scale bar = 50 µm. B, Quantitation of endothelial cell co-localization with apelin (n=12-15 biological replicate hearts/ group). Mean ± SEM; **P<0.01 by Student's t-test. C, Photomicrographs of reference human left anterior descending artery (LAD samples; upper panel) and human LADs with vasculopathy (n=4, lower panel) double immunofluorescence stained for the human endothelial marker CD34 (green) and apelin (red). Scale bar = 50 µm.



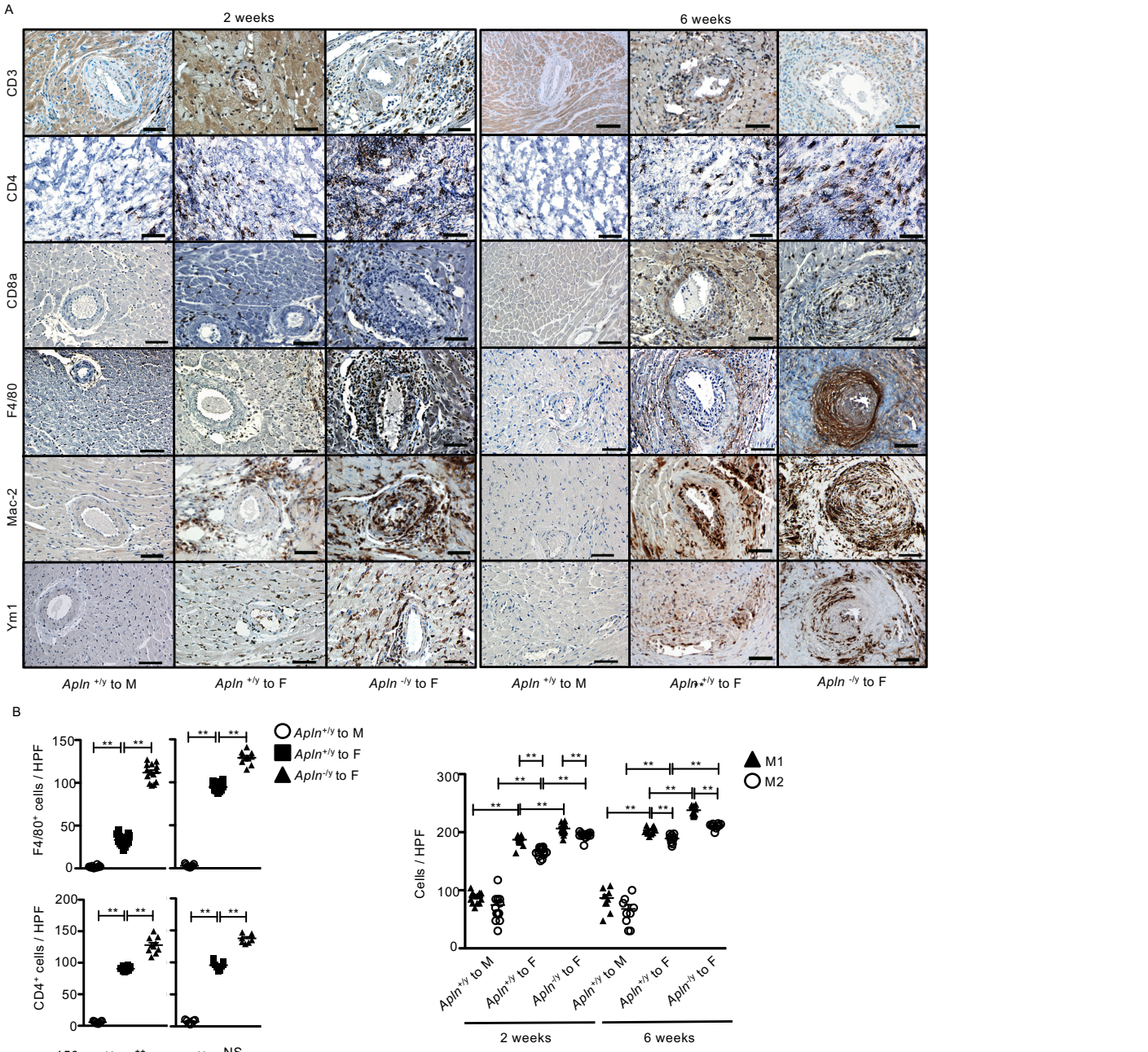
Supplemental figure 4: *Vegfa* expression in mouse heart allografts. A, Isolated coronary arteries (consecutive samples pooled in pairs for analysis (n=3-8 pairs)), and B, myocardium (n=6-15 biological replicates) at 2 and 6 weeks post-transplant, relative to normal hearts. Mean ± SEM; **P<0.01 and NS= non-significant by one way ANOVA with Bonferroni's post-hoc test.



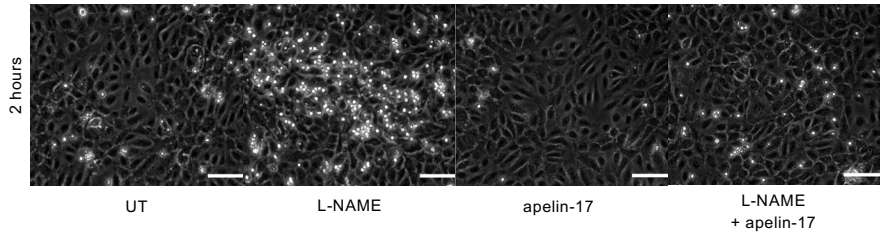
Supplemental figure 5: Microvessel density in mouse heart grafts. Immunohistochemical staining of endothelial marker A, CD31 (brown) or B, VE-Cadherin (brown) among mouse heart grafts at 2 and 6 weeks post-transplant. Quantitation is shown in Figure 1A and Figure 3C. C, Quantitation of VE-Cadherin^{+} microvessels at 6 weeks post-transplantation. n=6-10 biological replicate grafts/ group. Scale bar= 50µm. Mean ± SEM; NS= non significant, **P<0.01 by one way ANOVA with Bonferroni's post-hoc test. D, Photomicrographs show VE-Cadherin microvessel staining of hearts of allograft recipient mice treated with saline (n=8) or apelin-17-analogue (n=9) from week 2 through week 6 post transplantation. Quantitation of microvessel density is shown in E. **P<0.01 by Mann Whitney.



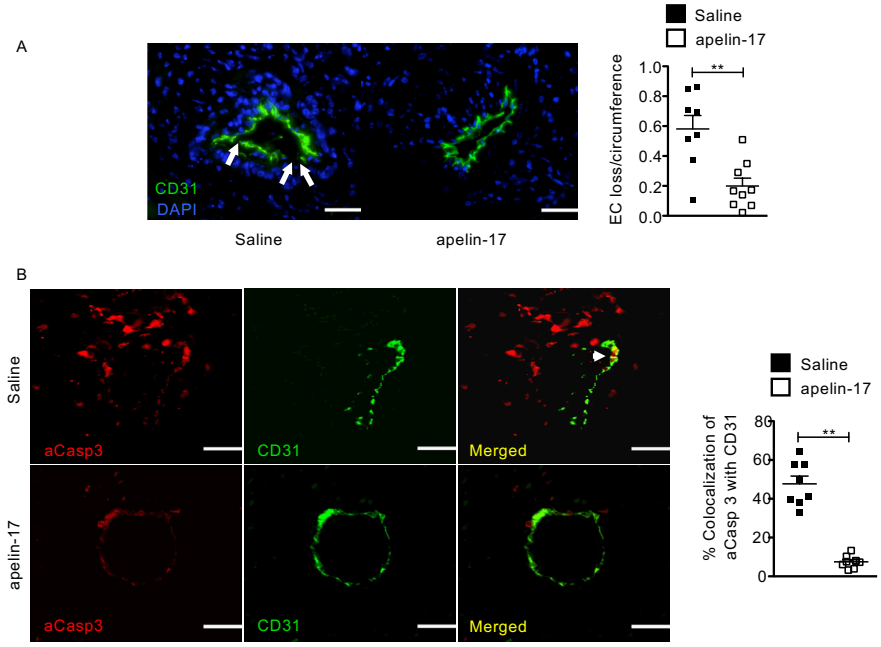
Supplementary figure 6: An apelin-17 analogue promotes closure and tip-cell differentiation in wounded endothelial cell monolayers. A, The synthetic N-MeLeu9-apelin-17 (apelin-17) agonist peptide, compound 11 from reference 30. B, HUVECs were transfected with non-specific (siNS) or *APLN* siRNA, then plated at confluence. The monolayers were wounded, then treated with VEGF (50 ng/mL) or apelin-17 (1uM). Quantitation of the experiments is shown in Figure 3D. n=5. Scale bar = 50µm. C, Synthetic apelin-17 agonist peptide induces angiogenic sprouting in 3D HUVEC cultures. n=3. Scale bar = 95µm. D, Quantitation of angiogenic sprouting in mock-, VEGF- (15 ng/ml), or apelin-17- (1µM) stimulated cultures. E, Tip cell gene expression in cultures from (D). n=3 biological replicates. Mean ± SEM; * $p<0.05$, ** $p< 0.01$ by one way ANOVA with Bonferroni's post-hoc test.



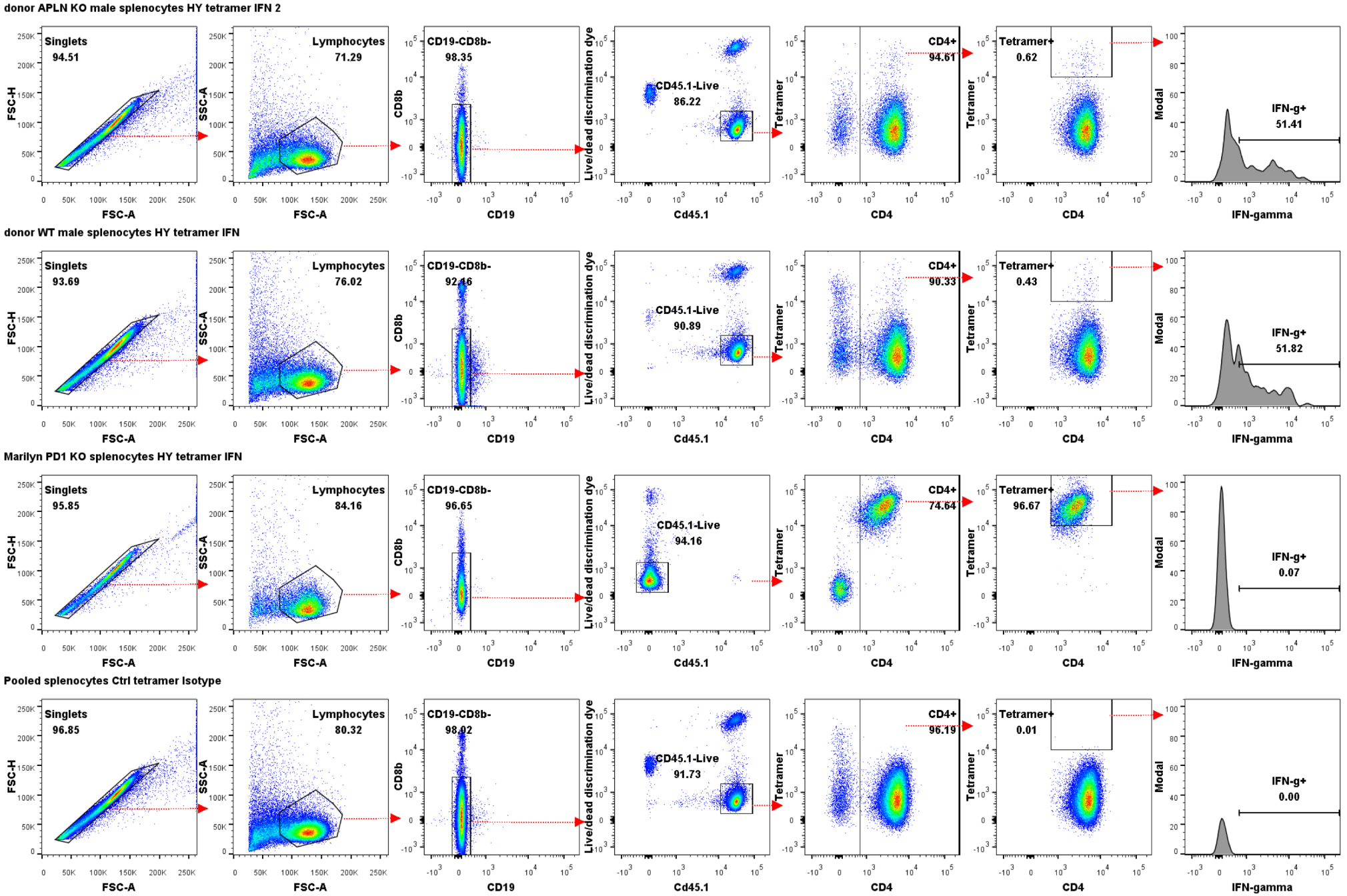
Supplemental figure 7: Characterization of leukocytes infiltrating mouse heart allografts. A) Photomicrographs showing immunohistochemical staining of CD3+ total, CD4+, or CD8a+ subsets of T lymphocytes; F4/80 macrophages, or Mac-2+ M1 , or YM1+ M2 macrophage subsets at 2 and 6 weeks post-transplant. CD3 and Mac-2 immunostains of week 2 grafts have been shown in Fig 4A. B) Quantitation of the inflammatory cellular infiltrate among groups. Scale bar = 50μm. n=9-15 biological replicate hearts. Mean ± SEM; **P<0.01 and NS= non significant by one way ANOVA with post hoc Bonferroni's post-hoc test.



Supplemental figure 8: apelin-17 inhibits monocyte adhesion to endothelial cell monolayers. Human umbilical vein endothelial cells were pretreated with TNFa (100 ng/mL) for 18 hours, then primary human monocytes were added to the co-culture for 2 hours in the presence of apelin-17 with or without the nitric oxide synthase inhibitor Nω-Nitro-L-arginine methyl ester hydrochloride (L-NAME), as indicated. The (phase-bright) monocytes adherent after gentle washes were photographed. The quantitation is shown in Figure 5D. Representative of n=5 biological replicates. Scale bar = 50 μm.



Supplemental figure 9. apelin-17 reduces endothelial cell loss and apoptosis. A, Photomicrographs show immunofluorescent staining of endothelial cell CD31 (green) in heart allografts from mice treated with with either saline (n=8) or apelin-17 (n=9) from week 2 through week 6 post-transplantation. Quantitation of arterial endothelial gaps (right panel). B, Confocal photomicrographs show immunofluorescent staining for cleaved caspase 3 (red) and endothelial cell CD31 (green; left panel). Quantitation of the cleaved caspase 3^+ co-localization with $CD31^+$ ECs by Mander's coefficient analysis (right panel). n=8-9 mice. Scale bar = 50 μm. Mean ± SEM; **P<0.01 Mann Whitney.



Supplementary figure 10: Gating strategy for male antigen specific CD4 T cells: A depiction showing the gating strategy used to identify live CD19-CD8β- CD45.1^{+}(host) IFN-γ^{+} HY-I-A^{b+} CD4^{+} cells from the spleens of the *Apln*$^{-/y}$ (1st row) or *Apln*$^{+/y}$ (2nd row) heart grafted mice, 2 weeks post-transplantation. Splenocytes from female Marilyn-*Rag2*$^{-/-}$-*pd1*$^{-/-}$ mice were used as a positive control cells (3rd row). Isotype control of anti-mouse IFN-γ antibodies (4th row) were used to exclude non-specific binding from the staining (n=4-7 biological replicates). The same gating strategy is used for determining the frequencies of CD44^{hi} cells.