**SUPPLEMENTAL METHODS**

***Cell cultures***

Mouse neonatal hearts were dissected and incubated with F-10 nutrient medium, containing 140 μg/ml of collagenase type-2 (Sigma) and 440 μg/ml pancreatin (Sigma) until cells were dissociated. The cells were then washed, resuspended in 1:1 mix of Dulbecco's minimal Eagle's medium (DMEM) and F10 nutrient mix with 5% each of equine and bovine calf serum. These cells were pre-plated in tissue culture dishes for 1 h to allow for attachment of nonmyocytes. The resulting non-adherent myocytes were plated on fibronectin-coated (Sigma) tissue culture dishes. Cells from one heart were plated in each 35 $mm^2$ petridish and cultured for 24 h, after which the medium was replaced with 1:1 mix of DMEM and F10 nutrient medium containing 1 μg/ml thyroxin, 5 μg/ml of transferrin, 1 μg/ml of insulin, 10 pM each of LiCl and selenite. A 0.1 mM bromodeoxyuridine concentration was used to check fibroblast growth.

***CaMKIIN mice***

The HA eptiope and membrane association domain (MEM), encoding a palmitoylation sequence, were added to the 5' of βCaMKIIN sequence by PCR using the 5' primer: gccattaagcttatgctctgctgtatgagaagaaccaaacaggttgaaaagaatgatgaggaccaaaagatcatgtccg-agatcctaccctacggc and the 3' primer ccgtatggatccctacactccggacggaggcttctc (Integrated DNA Technologies) with *Hin*dIII and *Bam*HI sites at the extreme ends. The DNA template for PCR was pSB3-His-CaMKIIN. CaMKIIN cDNA construct was a kind gift from Dr. Tom Soderling (Vollum Institute). The product (~360 base pairs) was purified from agarose gel (Quiagen gel extraction kit) and was digested with restriction enzymes, *Hin*dIII and *Bam*HI. The resulting DNA product was ligated into pCMV5-HA1 previously digested with *Hin*dIII and *Bam*HI. The HA-tagged MEM-CaMKIIN sequence was subcloned into pα-MHC promoter vector by PCR using the following primers: 5'-ccatgagtcgacgccgccaccatgtacccatacgacgtcccagactatgctgg taccacgcgtatcgataaactt and 3'-gccattaagcttctacactccggacggaggcttctc that incorporated *Sal*I and

kozak sites in the 5' region and a *Hin*dIII site in the 3' region. PCR products digested with *Sal*I and *HIn*dIII were ligated into the pαMHC-script-Hgh vector (9.2 kb). Mouse embryonic stem cells were injected with the linearized DNA (digested with *Not*I) in the Vanderbilt Transgenic Mouse Core Facility and implanted into B6D2 pseudo-pregnant females. Insertion of the transgene into the mouse genome was confirmed by PCR analysis (not shown) using the above primers.

***Quantitative RT-PCR***

For quantitative RT-PCR, 1 μl of reverse transcription reaction was mixed with 10 pmoles each specific primer and 12.5 μl SYBR PCR Master Mix (BioRad). The reaction was incubated in an iQ5 model thermocycler (BioRad) for 40 cycles consisting of denaturation at 95°C for 10 s and annealing/extension at 59.9°C for 1 min. The quality of the PCR product was routinely checked by thermal denaturation curve following the qPCR reactions. The threshold cycle ($C_T$) was determined by the iCycler software, and quantification of relative mRNA levels was performed by $\Delta\Delta C_T$ method. We thank Dr. Mollie Meffert (Johns Hopkins) for IκB-DN lentivirus expression construct. mCfbFor 5'-GAAACCCTGTCACTGTCATTC-3' and mCfbRev 5'-CCCCAAACACATACACA TCC-3'.

**SUPPLEMENTAL FIGURE LEGENDS**

***Suppl. Figure 1:***

Complement factor B expression in normal hearts. (**A**) Quantitative RT-PCR analyses of RNA from hearts (n=3) shows increased *Cfb* mRNA levels in AC3-I compared to WT. (**B**) Immunoblotting of heart extracts (n=3) shows similar amounts of CFB in both WT and AC3-I. (**C**) Immunoblot of WT and AC3-I heart extracts was probed with anti-CFB antibody (upper panel) as described in the Methods. Following the signal detection, the blot was stained with coomassie blue (lower panel).

***Suppl. Figure 2:***

*Cfb* expression in cardiomyocytes. (**A**) RT-PCR analysis of *Cfb* and *Hprt* performed on cultured neonatal (Neo) and isolated adult cardiomyocytes. The RT-PCR lanes representing reverse transcriptase reaction with and without reverse transcriptase enzyme are designated as '+' and '-', respectively. The far left and far right columns show molecular size markers marked in base pairs (bp). (**B**) Immunoblotting for CFB protein expression in neonatal and adult cardiomyocytes, and heart tissue. Equal amount of total protein from each sample was fractionated using NuPAGE gels. Antibody to CFB was used to detect the CFB protein band. Immunoblots of actin were used as a loading control.

***Suppl. Figure 3:***

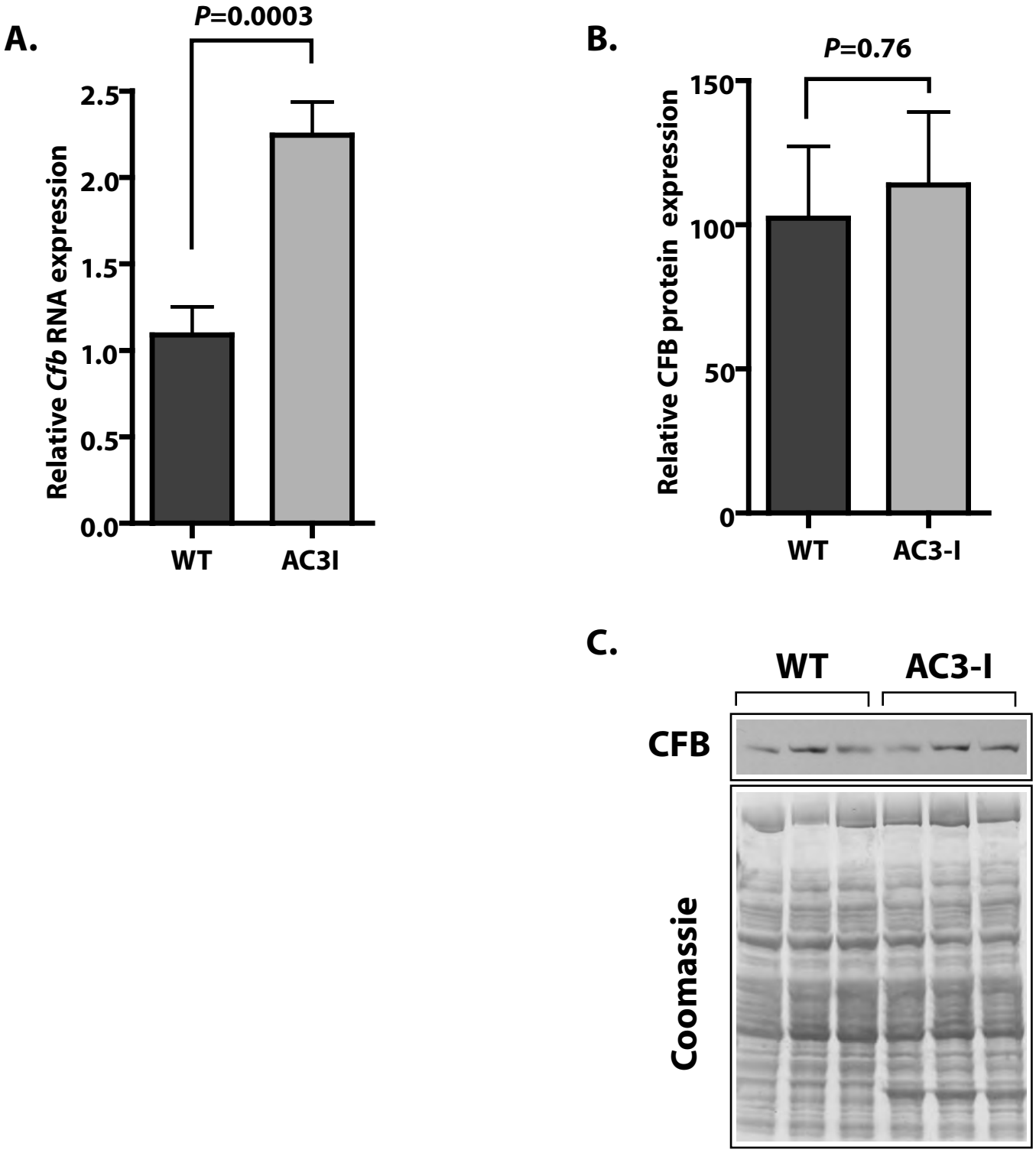
Death and apoptosis markers in neonatal cardiomyocytes treated with LPS. (**A**) Fraction of dead cardiomyocytes determined by flow cytometry using propidium iodide (PI) fluorescent vital dye (Molecular Probes). A total of 18,344 and 14,647 cells were sorted for control (serum alone) and treatment (serum and LPS), respectively. The significance of the results of PI-positive cells in each treatment was determined by Fisher's Exact test. (**B**) TUNEL staining data from mouse serum and LPS treated cultured cardiomyocytes. All data are from n>3 separate experiments.

***Suppl. Figure 4:***

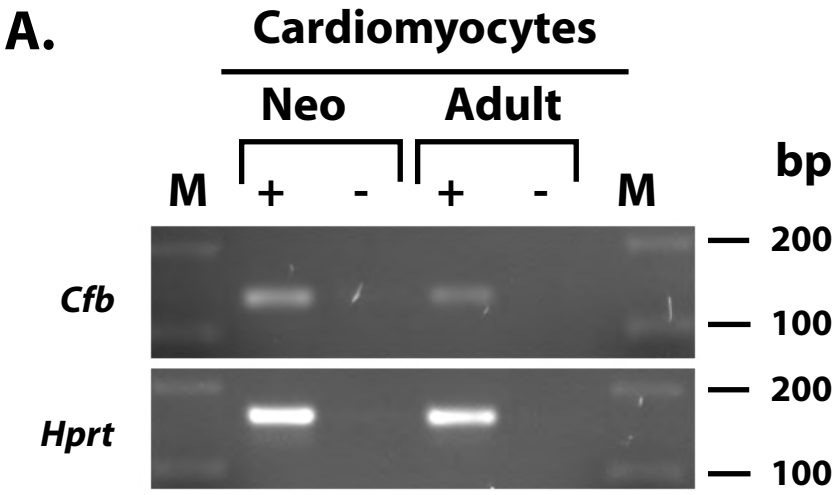
(**A**) Expression of βCaMKIIN in transgenic mouse heart and inhibition of CaMKII mediated phosphorylation of T17 residue of phospholamban (PLN). Heart extracts from wild type (WT) and βCaMKIIN transgenic mice (CaMKIIN) were fractionated on a NuPage gel (Invitrogen) and transferred onto a PVDF membrane (BioRad). Blots were probed with either a monoclonal

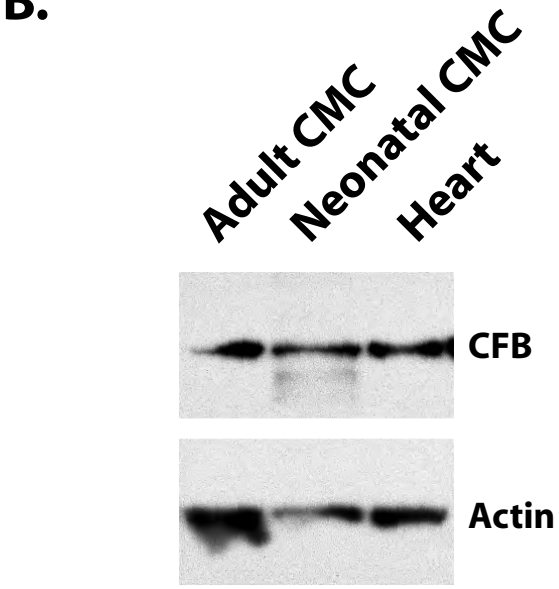
antibody to detect HA-epitope tagged cardiomyocyte-delimited expression of transgenic βCaMKIIN or antibodies to phospholamban (PLN). Specific antibodies to either total or phospho-specific epitope were used. For PLN, the same blot was stripped and reprobed in the following order: PLN-T17, PLN-S16, and PLN-total. (**B**) Membrane localization of βCaMKIIN. DNA binding fluorescent stain DAPI was used to visualize the nuclei. Images were obtained by BioRad multiphoton confocal microscopy and rendered pseudocolors (Red= anti-HA, Blue= DAPI). Scale bars = 20 μm.

# Suppl Figure-1

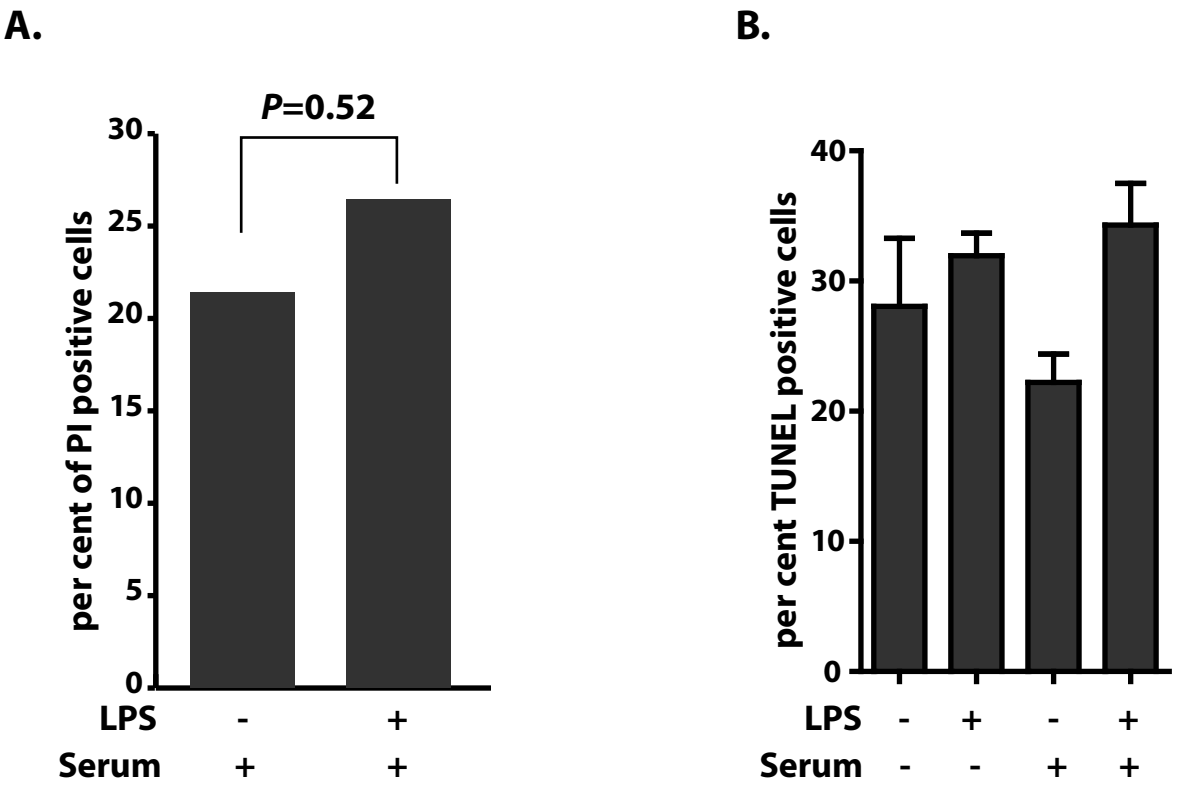


# Suppl Figure-2



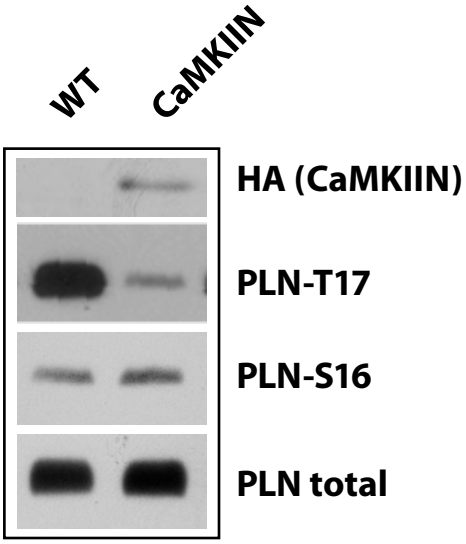



# Suppl Figure-3.

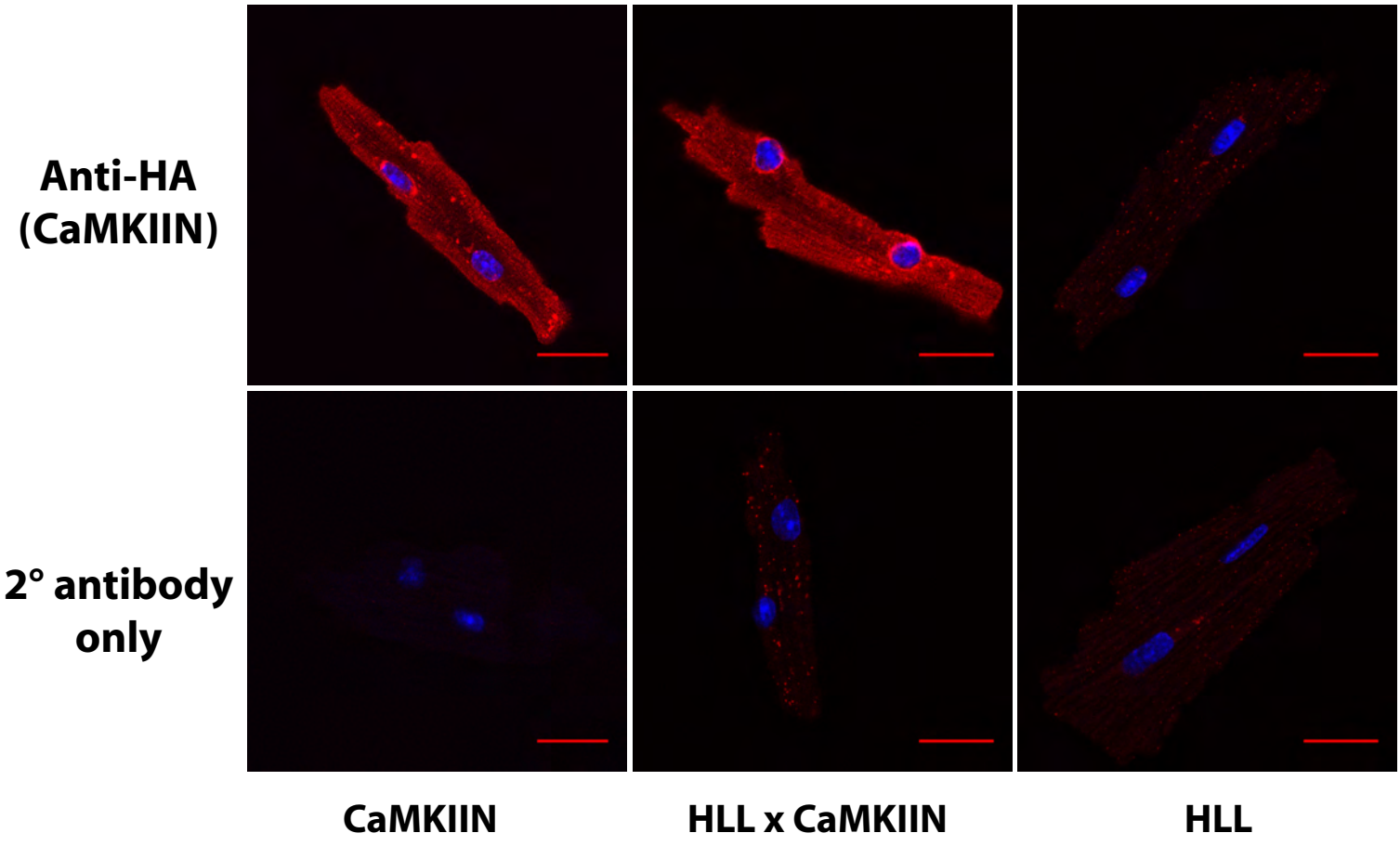


# Suppl Figure-4.

**A.**




**B.**



**Supplementary Table 1. List of genes upregulated in post-MI AC3-I hearts (1.8-fold increase cutoff)**

*1110002B05Rik*
AC124413.2
AC138640.7
*Adamts15*
*Adn*
AK024941.1
AL732556.7
*Ankrd1*
*Arntl*
*Atp13a1*
*ATXN1*
AY172581.1
*Btnl2*
BX883045.1
*C1s*
*C2*
*C4a*
*Ca3*
*Cacna2d1*
*Cald1*
*Cap1*
*Capzb*
*Cd2*
*Cd37*
*Cd74*
*Cdo1*
*Centg1*
*Cetn3*
*Cfb*
*Cfp*
*Cish*
*Clk4*
*Cma1*
*Col3a1*

*Col5a2*
*Cpne3*
*Cpxm1*
*Ctgf*
*Ctss*
*Cyp2e1*
*Cyp2f2*
*Cyp4f5*
*Dym*
*Efemp1*
*Eif3s10*
*Eln*
*Enpp2*
*Eprs*
*Fasn*
*Fn1*
*Fos*
*Gpc3*
*Gpnmb*
*Hbg1*
*Hmgn3*
*Hspa1a*
*Ifi30*
*IgG-2a*
*Igh-6*
*Il18*
*Itgam*
*Itih4*
*Klf9*
*Kras*
L22655
LOC64305
*Loxl1*
M21780.1
*Man2a1*
*Map2k6*
*Matr3*

*Mbnl2*
*Mgl1*
*Mgp*
*Mmp9*
*Msln*
*Myo1g*
*Ncl*
*Neb*
*Nfia*
*Nif3l1*
*Nppa*
*Nppb*
*Nr3c1*
*Plcb1*
*Plcb4*
*Plk2*
*Pnkd*
*Postn*
*Ppp4r2*
*Prei3*
*Prkcb1*
*Ptk2b*
*Rasa1*
*Rcan1*
*RGD1564450*
*Rock2*
*Rpl5*
*RT1-Aw2*
*Rtn4*
*S100a9*
*Saa*
*Sacm1l*
*Scd1*
*Sfrs12*
*Sorl1*
*Stat1*
*Stch*

*Stk25*
*Sv2b*
*Tbc1d5*
*Tbca*
*Tf*
*Tgfb2*
*Timp1*
*Tpp2*
*Ttc3*
*Ugt1a1*
*Uqcrfs1*
*Usp15*
*Vcp*
*Vps54*
X53054
X68312
XM_234581.2
XM_341386.1
*Zfp265*
*Zfp692*

**Supplementary Table 2. Genes downregulated in post-MI AC3-C hearts (1.8 fold decrease cutoff).**

*Angptl4*
AY172581.1
BX883045.1
*Camk2b*
*Cd164*
*Cd36*
*Cd63*
*Cish*
*Dbp*
*Dhrs7c*
*Dtx2*
*Eno3*
*Idh1*
*Irx3*
*Kbtbd10*
*Pam*
*Pdk4*
*Pfkfb1*
*Pim3*
*Ptgds*
*Ptp4a3*
*Pttg1*
*Rbp7*
*Slc9a3r1*
*Tcap*
*Uqcrbl*
XM_342364.1
XM_347242.1