SUPPLEMENTAL MATERIAL

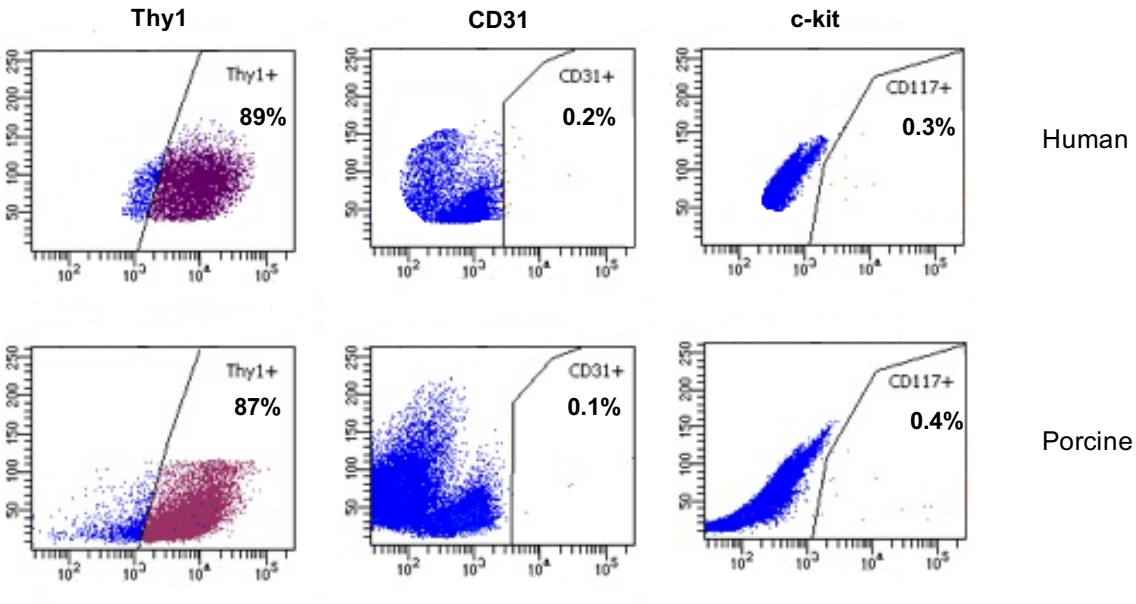

Figure S1. FACS analyses for Thy1, CD31 and CD117/c-kit in human and porcine cardiac fibroblasts. Representative FACS analyses of human cardiac fibroblasts (top) or porcine cardiac fibroblasts (bottom) showing majority of the cells were positive for the cardiac fibroblast specific surface marker Thy1 and negative for cardiac progenitor (c-kit/CD117) or endothelial cell (CD31) markers.

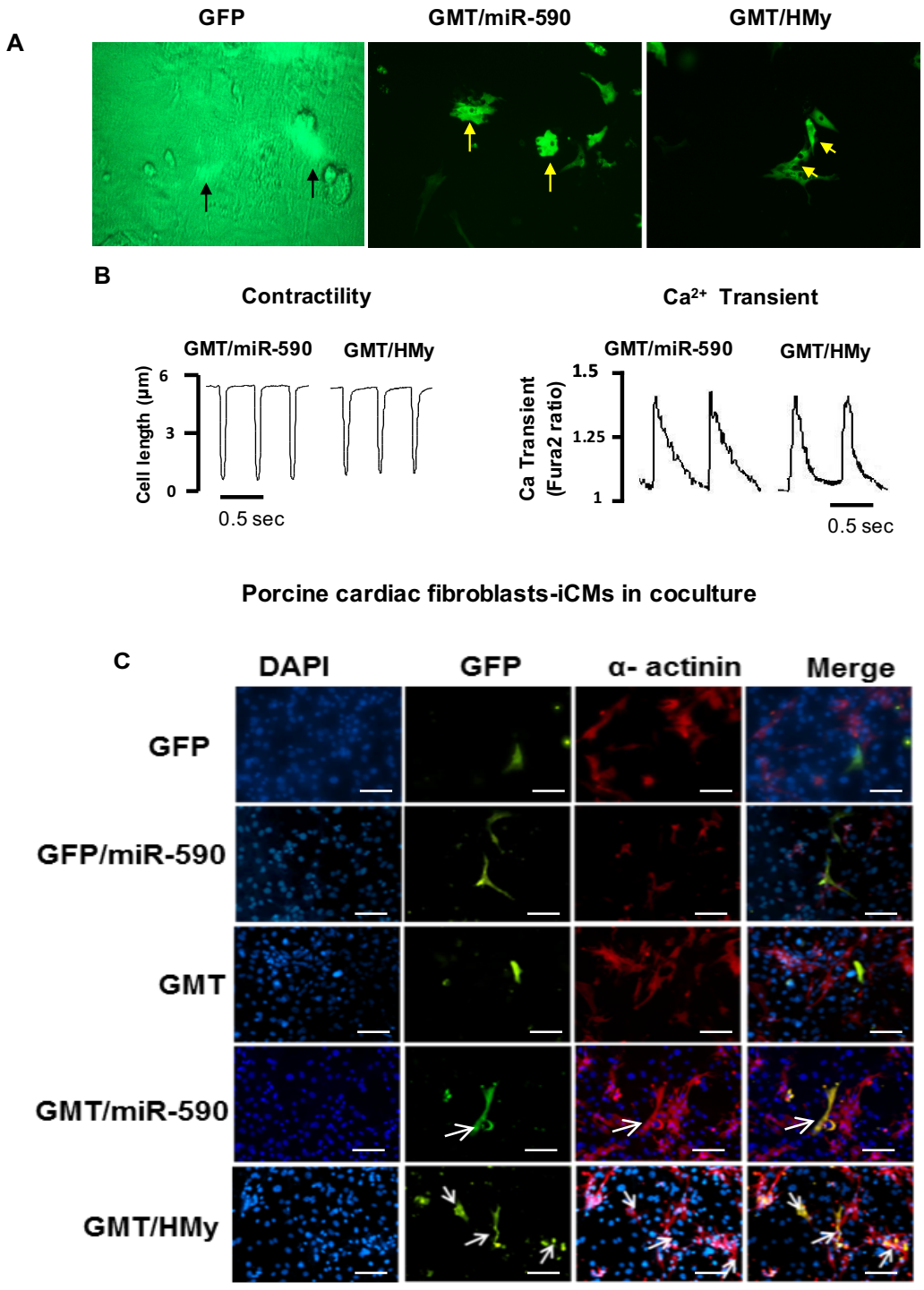

Figure S2. Reprogramming of porcine cardiac fibroblasts in co-culture with murine cardiomyocytes. Porcine cardiac fibroblasts were transduced with lentivirus expressing a GFP marker alone (negative control group; left); lentivirus vectors also expressing miR-590 as well as vectors expressing Gata4, Mef2c or Tbx5

("GMT plus miR-590" group; middle); or with lentivirus vectors also expressing Hand2 and Myocardin as well as vectors expressing Gata4, Mef2c or Tbx5 ("GMT plus H/My" group; right). Four weeks after initial transduction, these porcine cardiac fibroblasts were co-cultured with (untreated) neonatal murine cardiomyocytes (negative for GFP). A) Representative immunofluorescence staining demonstrating (green) GFP expression, which indicates lentivirus infection. Only (green fluorescent) cells that had been treated with GMT plus miR-590 or GMT plus H/My contracted synchronously with surrounding cardiomyocytes. Fibroblasts treated with a GFP-control vector only did not demonstrate contractions in these co-culture experiments (supplemental video S1-S3). B) Representative curves reflecting contraction (left) and Ca^{2+} transient trace (right) under basal conditions of (iCM) cells derived from porcine cardiac fibroblasts after co-culture with murine neonatal cardiomyocytes. For the calcium transients, cells were loaded with Fura-2 for 30 min, and then calcium transients were recorded (n=3) C) Representative immunofluorescence staining for DAPI ([blue] nuclear marker), GFP ([green] lentivirus infection marker) and the (red) cardiomyocyte marker α-actinin in porcine cardiac fibroblasts treated with lentivirus containing a GFP marker and GMT, GMT plus miR-590 or GMT plus H/My which were then co-cultured with neonatal murine cardiomyocytes. Porcine cardiac fibroblasts transduced with GFP, miR-590 or GMT alone did not express α-actinin in co-culture neonatal murine cardiomyocytes. Porcine cardiac fibroblasts transduced with GMT plus miR-590 or GMT plus H/My expressed the cardiomyocyte marker α-actinin (indicates by arrows) in co-culture with murine cardiomyocytes (scale bars: 100 μm). Black arrow indicates GFP positive non-beating cells, yellow arrow indicates GFP positive beating cells.

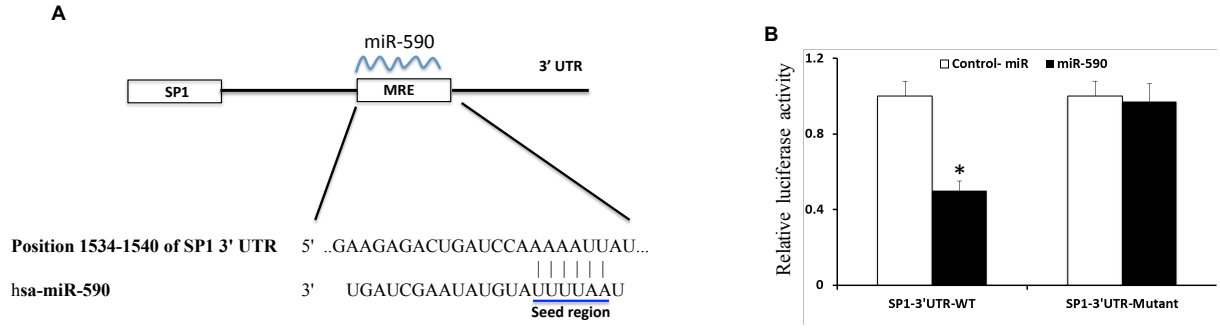

Figure S3. MiR-590 directly represses Sp1. A) Predicted interaction sites of miR-590 at the 3'-UTR of Sp1 (TargetScan: www.targetscan.org). B) HEK293 cells were co-transfected with (pGL_3-Sp1-3'UTR-WT) and mutated targeting sequence (pGL_3-Sp1-3'UTR-Mutant) and miRNA mimic-control/590 for 48 h, then luciferase activity was detected. MiR-590 directly repressed WT Sp1 3'UTR in luciferase assay, and the repression was abolished when binding site was mutated (n=3). Data are presented as mean ± SEM. *$p < 0.05$ vs. control-miR.

Video Legends:

Video S1. This video shows porcine fibroblasts treated with a GFP-control vector only did not demonstrate contractions in the co-culture experiments.

Video S2. This video shows porcine cardiac fibroblasts contracted synchronously with surrounding murine cardiomyocytes after transduction with GMT plus miR-590.

Video S3. This video shows porcine cardiac fibroblasts contracted synchronously with surrounding murine cardiomyocytes after transduction with GMT plus H/My.