

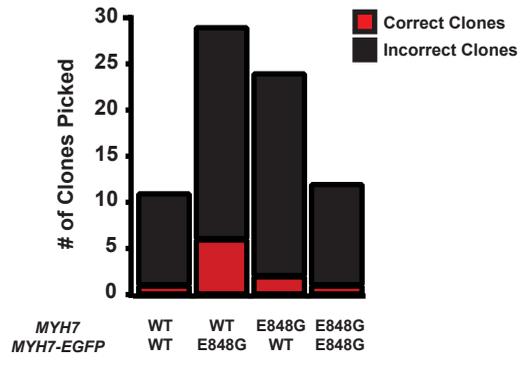
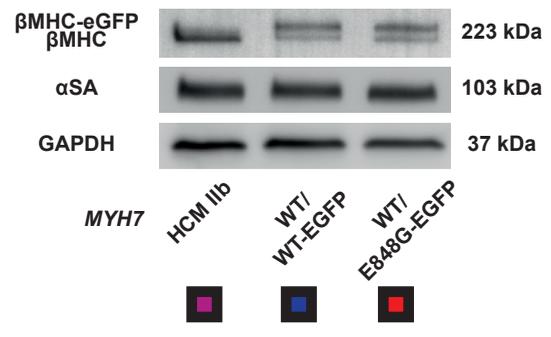
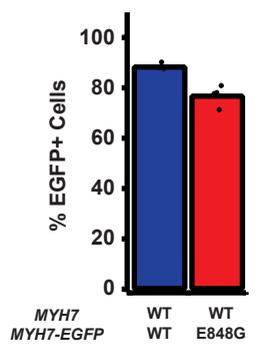
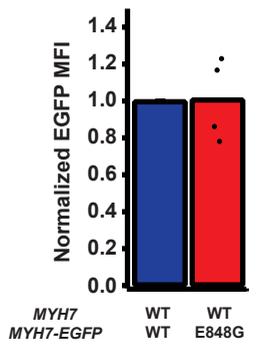
A**B****C****D****E****F**

Figure S1: Generation of isogenic β MHC-EGFP expressing hiPSC-CMs using CRISPR/Cas9 editing (A) Schematic of pJet-MYH7-EGFP-PGK-PuroR plasmid with E848 location highlighted. (B) Number of clones picked with correct or incorrect edited sequence for isogenic lines. (C) Repe-rentative western blot for MYH7 protein (β MHC), α -sarcomeric actinin (α SA), and GAPDH protein expression in patient-derived and isogenic differentiation day 40 hiPSC-CMs, 20 μ g total protein per lane. (D) Representative brightfield image of an engineered heart tissue attached to flexible (top) and stiff (bottom) posts. Scale bar 1 mm. (E) Total EGFP+ cells as percentage of total cells for dif-ferentiation day 25 hiPSC-CMs as measured by flow cytometry, n = 4 biological replicates. (F) EGFP mean fluorescent intensity in EGFP+ hiPSC-CMs for differentiation day 25 hiPSC-CMs as measured by flow cytometry, n = 4 biological replicates.

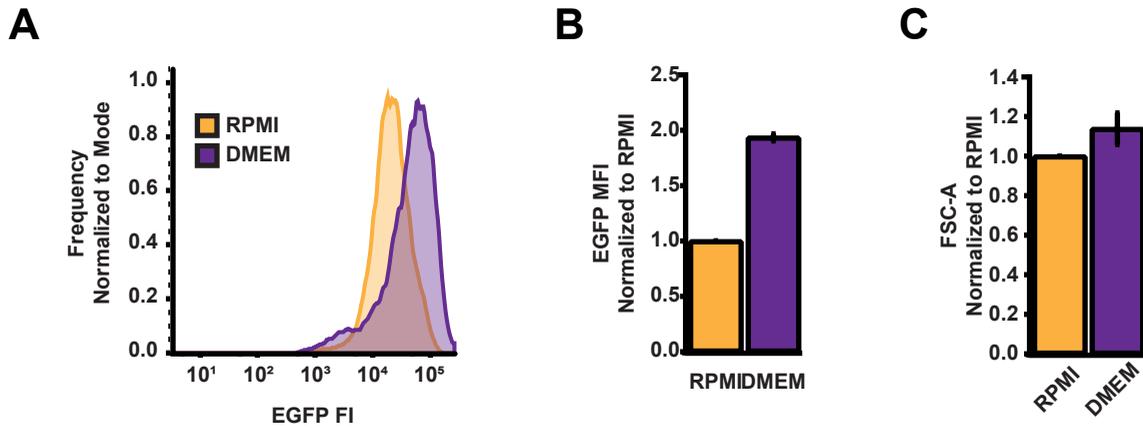


Figure S2: DMEM-based cardiomyocyte media accelerates maturation of hiPSC-CMs in monolayer culture

(A) Representative histogram of EGFP fluorescent intensity as measured by flow cytometry in MYH7WT/WT-EGFP hiPSC-CMs, differentiation day 40, after 10 days in RPMI-based and DMEM-based monolayer culture conditions. (B) Normalized EGFP geometric mean fluorescent intensity from (A). Mean \pm SEM, $n = 2$ biological replicates. (C) Normalized FSC-A as measured by flow cytometry of EGFP+ MYH7WT/WT-EGFP hiPSC-CMs, differentiation day 40, after 10 days in RPMI-based and DMEM-based monolayer culture conditions. Mean \pm SEM, $n = 2$ biological replicates. * in (B,C) indicates $p < 0.05$ significance calculated by Student's t-test.

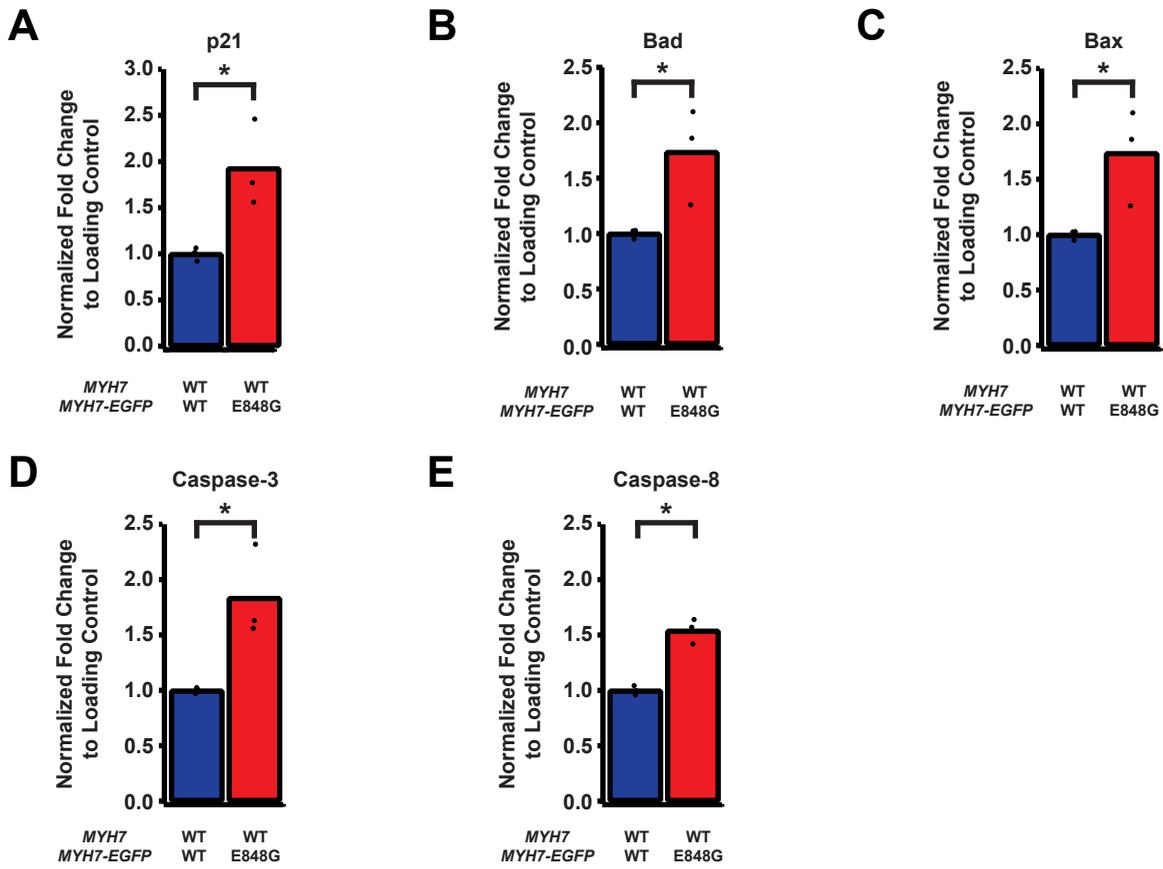


Figure S3: MYH7 E848G induces p53-associated intrinsic apoptosis in monolayer and EHT culture (A-E) Quantification of protein expression from Figure 3A normalized to loading control and MYH7WT/WT-EGFP. Mean and biological replicates shown, n = 3 technical replicates. * in (A-E) indicates p < 0.05 significance calculated by Student's t-test.

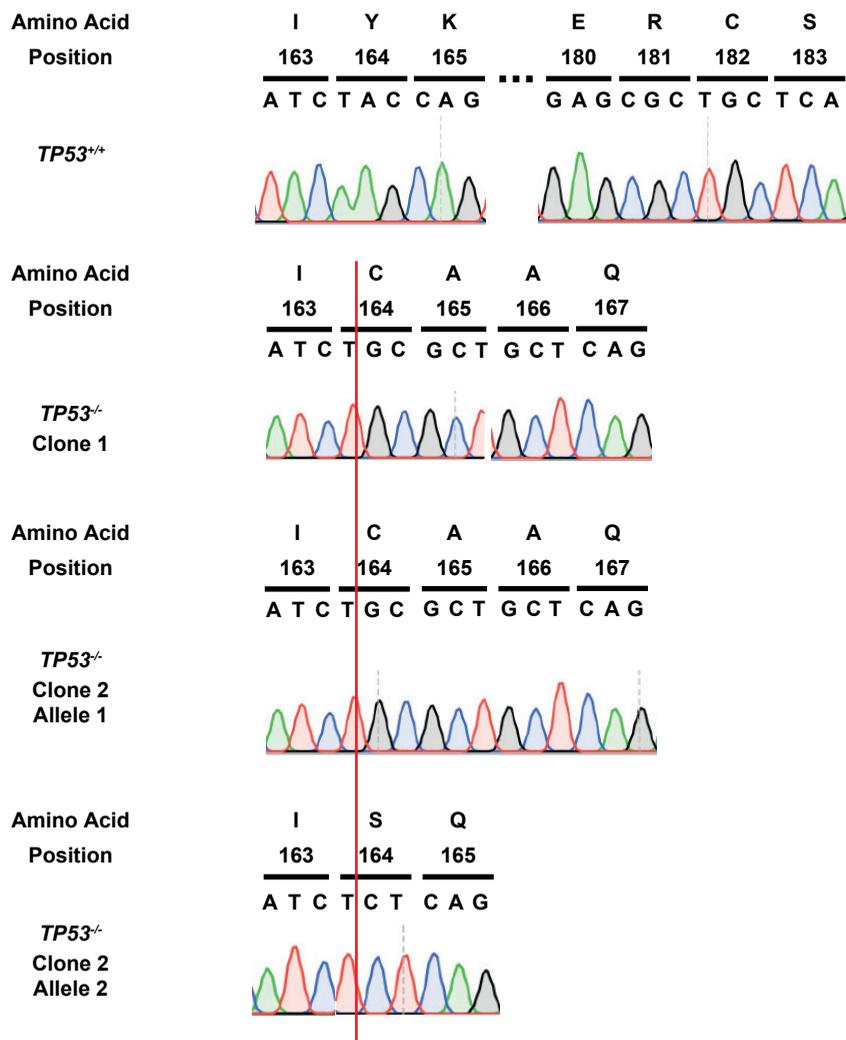


Figure S4: Sanger sequencing chromatograms of MYH7WT/E848G-EGFP *TP53*^{+/+} and MYH7WT/E848G-EGFP *TP53*^{-/-} clones.