SUPPLEMENTAL MATERIAL

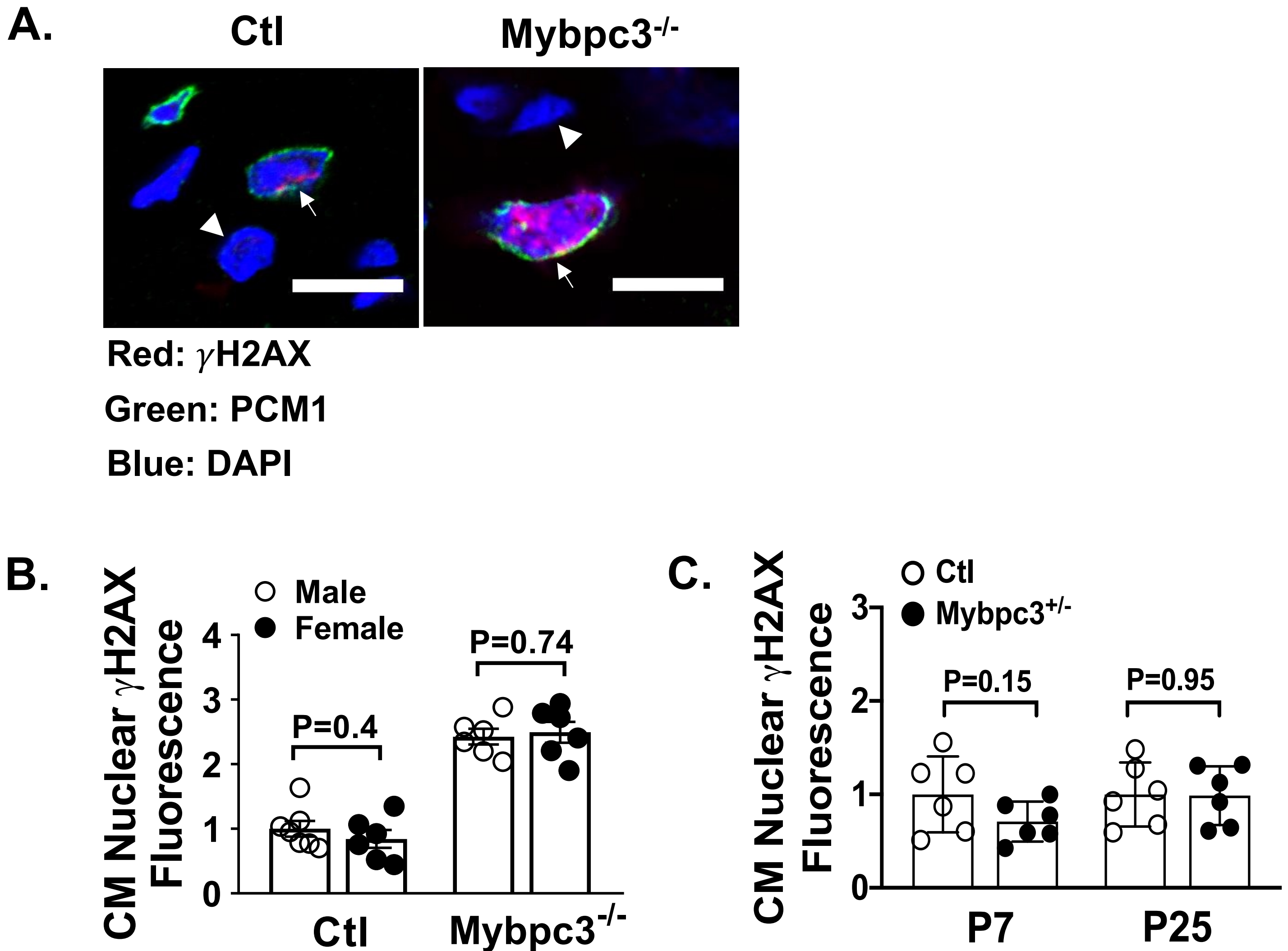
Table S1. Oligonucleotide primer sequences used for qRT-PCR

Gene	Forward Sequence	Reverse Sequence
p53	TGTTATGTGCACGTACTCTCCTC	GCTCCCAGCTGGAGGTGT
Mdm2	GGAGATCCATTAGTGAGACAGAAGA	AGACCCAGGCTCGGATCA
Cdkn1a	GCAGACCAGCCTGACAGATT	CTGACCCACAGCAGAAGAGG
Gdf15	CGGATACTCAGTCCAGAGGTG	GTGCACGCGGTAGGCTTC
Gadd45a	GCTCAACGTAGACCCCGATA	CACGGATGAGGGTGAAATG
Rpl32	CACCAGTCAGACCGATATGTGAAAA	TGTTGTCAATGCCTCTGGGTTT

Table S2. Clinical characteristics of human non-HCM (control) and HCM left ventricular septal tissue samples

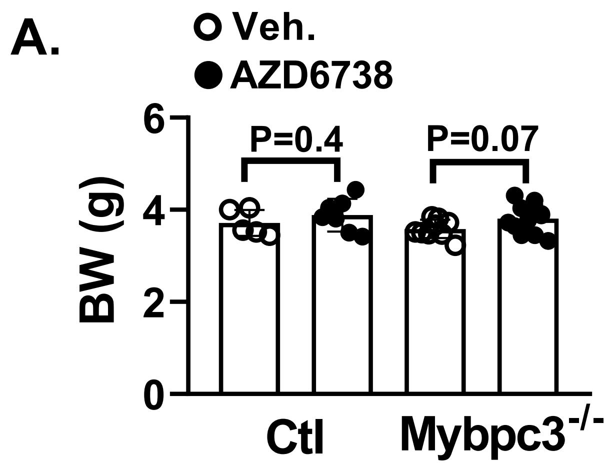
	Age (yrs)	Sex	Sarc Mut	Gene	cDNA	AA	IVSd (mm)	LVPWd (mm)	IVSd/ LVPWd	EF (%)	LVIDd (mm)
Non-HCM 1	47	M	N				9	9	1	73	54
Non-HCM 2	38	F	N				10	8	1.3	55	39
Non-HCM 3	51	F	N				8	8	1	50	37
Non-HCM 4	40	M	N				11	11	1	55	50
HCM 1	41	F	Y	MYBPC3	c.3330+2T>G		18	8	2.3	75	35
HCM 2	24	F	Y	MYBPC3	G2670A	W890X	27	7	3.9	88	35
HCM 3	28	M	Y	MYBPC3	c.927-9G>A		18	11	1.6	75	30
HCM 4	54	M	Y	MYBPC3	G1624C	E542Q	32	14	2.3	65	35
HCM 5	27	M	Y	MYH7	G1988A	R663H	33	11	3.0	69	42
HCM 6	34	F	Y	MYH7	G2770A	E924K	18	7	2.6	65	51

Figure S1. Comparison of DNA damage in cardiomyocytes vs non-cardiomyocytes and male vs female in Mybpc3 null mice.



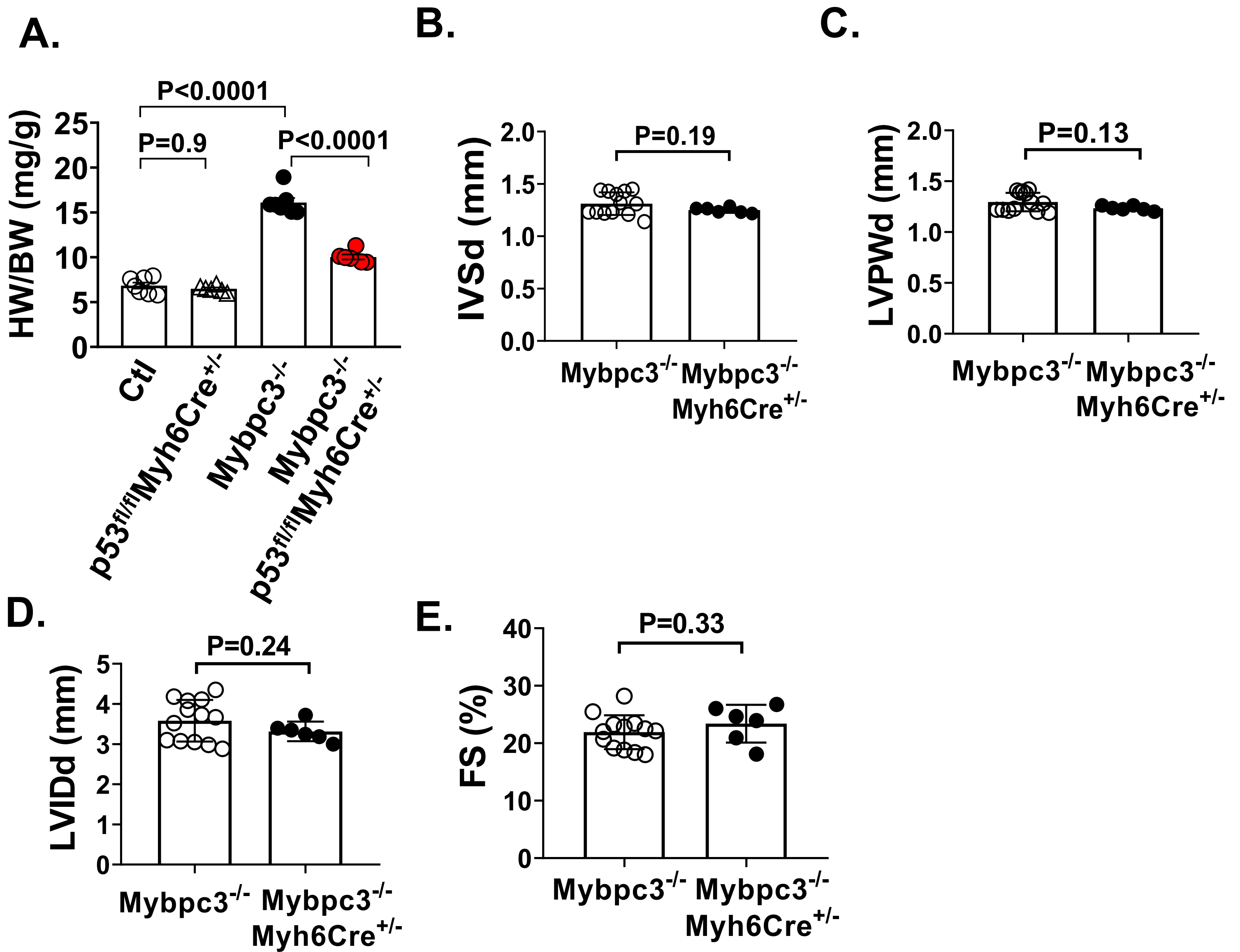
(**A**) Representative immunofluorescence staining of γH2AX (red) in Ctl and Mybpc3$^{-/-}$ myocardial tissue at postnatal day 7 (P7). Cardiomyocyte nuclei (arrow) were differentiated from non-cardiomyocyte nuclei (arrowhead) using PCM1 (pericentriolar material 1) (green) staining. Nuclei labeled with DAPI (blue). Scale bars, 10 µm. (**B**) Quantification of γH2AX fluorescence in cardiomyocyte nuclei in Ctl (n=6-7) and Mybpc3$^{-/-}$ (n=6) myocardial tissue from male and female mice at P7. Minimum 50 nuclei/sample. (**C**) Quantification of γH2AX fluorescence in cardiomyocyte nuclei in Ctl (n=6) and Mybpc3$^{+/-}$ (n=6) myocardial tissue at P7 and P25. Minimum 50 nuclei/sample. Results are shown as mean±SEM.

Figure S2. Effect of AZD6738 treatment on body weight in control and Mybpc3 null mice.



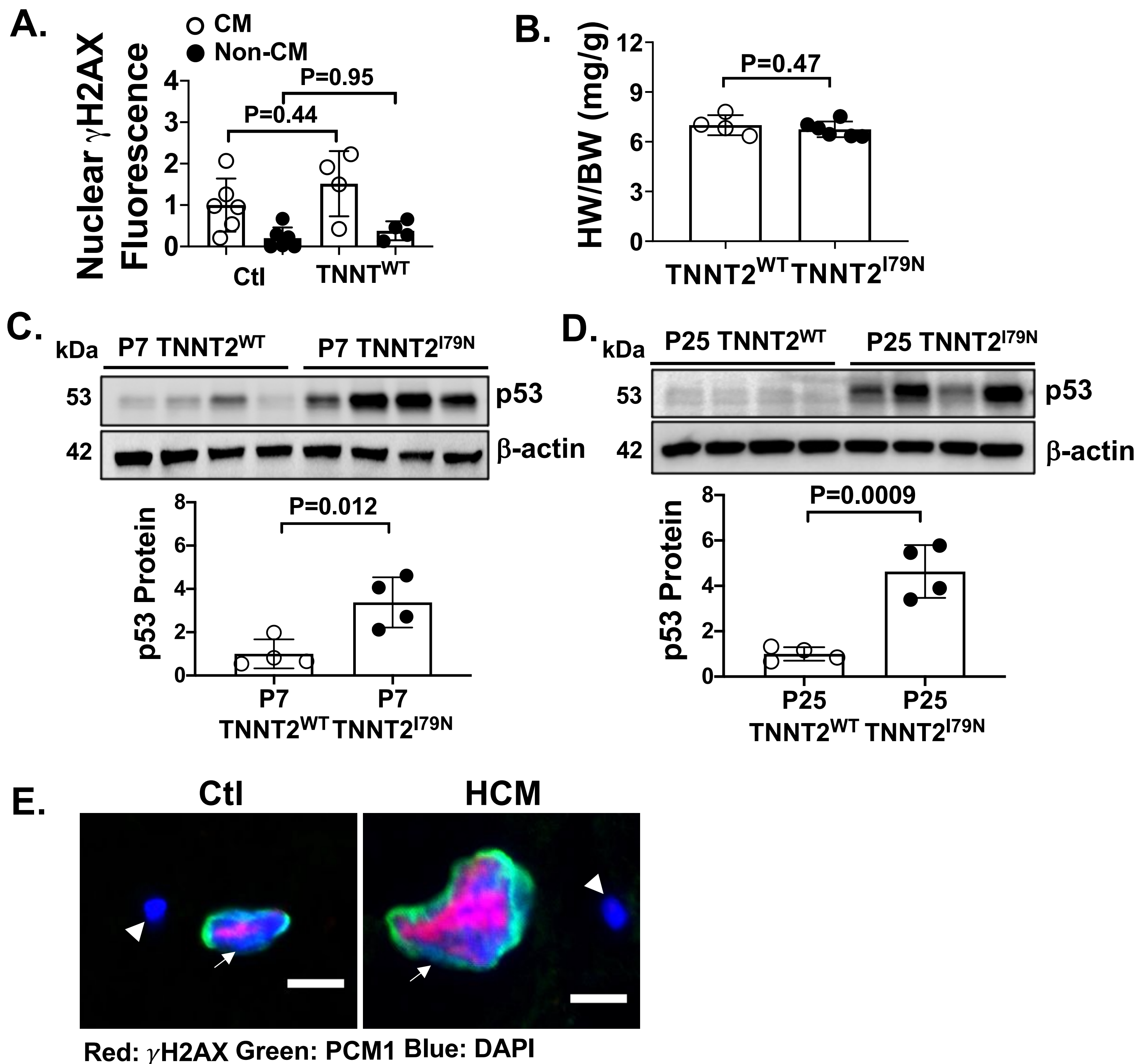
(**A**) Body weight (BW) in control (Ctl) (n=5-7) and $Mybpc3^{-/-}$ (n=9-12) mice either exposed to vehicle (Veh.) or 25 mg/kg/day AZD6738 at postnatal day 7. Results are shown as mean±SEM.

Figure S3. Comparison of echocardiography measurements between Mybpc3-/- and Mybpc3-/-/Myh6Cre+/- mice.



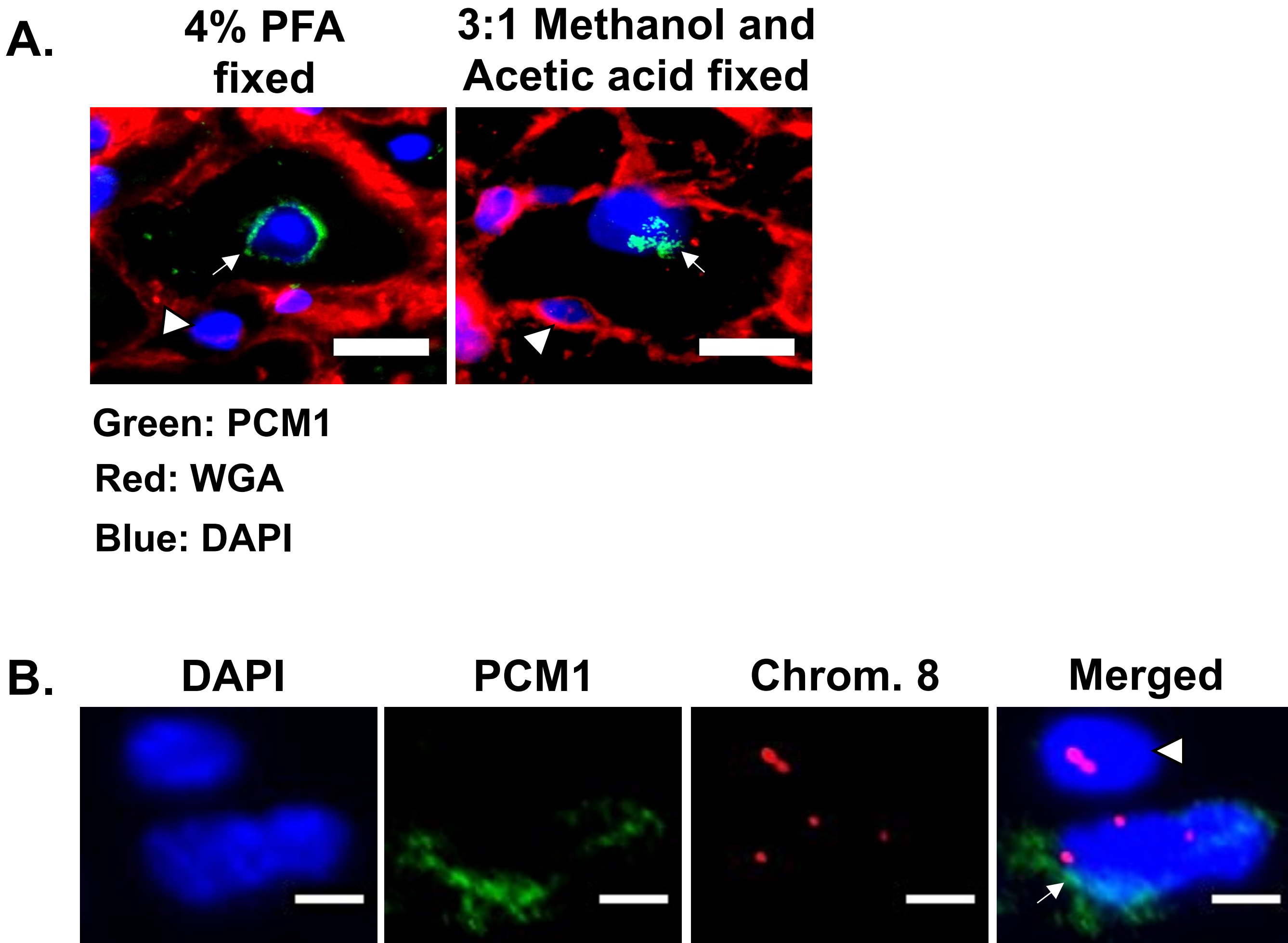
(**A**) Heart weight to body weight ratio (HW/BW) of control (Ctl) (n=7), p53$^{fl/fl}$Myh6Cre$^{+/-}$ (n=6), Mybpc3$^{-/-}$ (n=7), and Mybpc3$^{-/-}$/p53$^{fl/fl}$Myh6Cre$^{+/-}$ (n=6) mice at postnatal day 25 (P25). M-mode echocardiography assessment of (**B**) interventricular septal thickness at end diastole (IVSd), (**C**) left ventricular posterior wall thickness at end diastole (LVPWd), (**D**) left ventricular internal diameter at end diastole (LVIDd), and (**E**) fractional shortening (FS) in Mybpc3$^{-/-}$ (n=13) and Mybpc3$^{-/-}$/Myh6Cre$^{+/-}$ (n=6) mice at P25. Results are shown as mean±SEM.

Figure S4. DNA damage response in TNNT2 mice and human HCM.



(**A**) Quantification of γH2AX fluorescence in cardiomyocyte (CM) and non-cardiomyocyte (Non-CM) nuclei in control (Ctl) (n=6) and TNNT2^{WT} (n=4) myocardial tissue at postnatal day 7. Minimum 50 nuclei/sample. (**B**) Heart weight to body weight ratio (HW/BW) of TNNT2^{WT} (n=4) and TNNT2^{I79N} (n=6) mice at postnatal day 7. (**C**) Western blot of p53 in TNNT2^{WT} and TNNT2^{I79N} myocardial tissue at P7 (*top*). Relative quantification p53 in P7 TNNT2^{WT} (n=4) and TNNT2^{I79N} (n=4) myocardial tissue normalized to **β**-actin (*bottom*). (**D**) Western blot of p53 in TNNT2^{WT} and TNNT2^{I79N} myocardial tissue at P25 (*top*). Relative quantification p53 in P25 TNNT2^{WT} (n=4) and TNNT2^{I79N} (n=4) myocardial tissue normalized to **β**-actin (*bottom*). All results are shown as mean±SEM. (**E**) Representative immunofluorescence of γH2AX (red) in explanted non-HCM control (Ctl) or hypertrophic cardiomyopathy (HCM) patient LV septal tissue. CM nuclei (arrow) were differentiated from non-cardiomyocyte nuclei (arrowhead) using PCM1 (green) staining. Nuclei labeled with DAPI (blue). Scale bars, 10 μm.

Figure S5. Comparison of nuclear aneuploidy between cardiomyocytes and non-cardiomyocytes in Mybpc3 null mice.



(A) Representative immunofluorescence image of PCM1 staining (green) in Mybpc3$^{-/-}$ myocardial tissue either fixed with 4 % paraformaldehyde (4% PFA, used for γH2AX staining) or 3:1 methanol and acetic acid (used for chromosome FISH assay). Arrow and arrowhead represent cardiomyocyte and non-cardiomyocyte nuclei respectively. Cardiomyocyte area was marked with wheat-gram agglutinin (WGA, Red) staining. Nuclei labeled with DAPI (blue). Scale bars, 10 μm. **(B)** Representative immunofluorescence image of FISH assay with chromosome 8 centromeric probe (Chrom. 8) (red) from Mybpc3$^{-/-}$ myocardial tissue at postnatal day 25. Cardiomyocyte nuclei (arrow) were differentiated from non-cardiomyocyte nuclei (arrowhead) using PCM1 (pericentriolar material 1) (green) staining. Nuclei labeled with DAPI (blue). Scale bars, 5 μm.