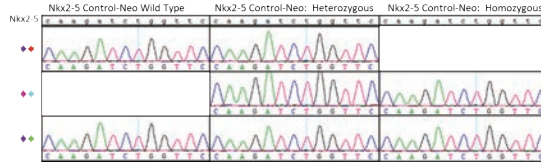
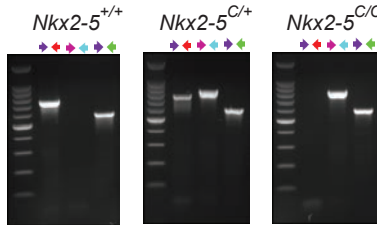
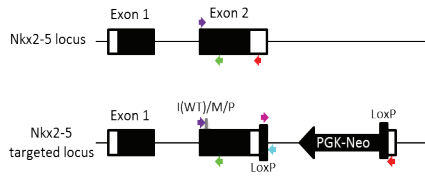
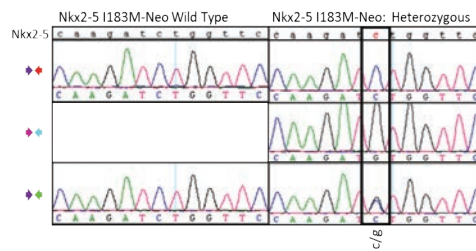
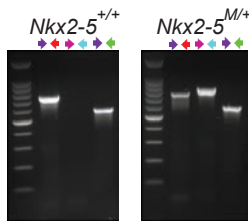
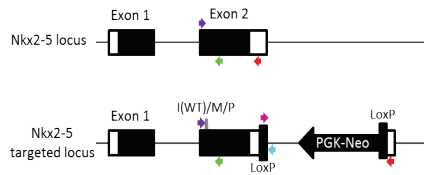


A



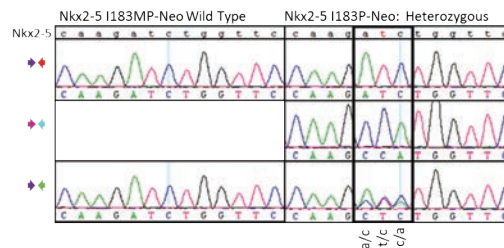
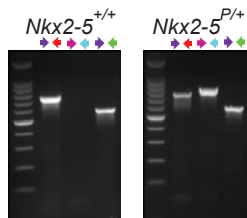
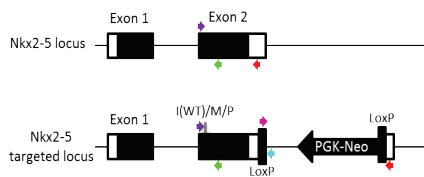
◆◆ Primer pair for WT allele
 ◆◆ Primer pair for 'knocked-in' allele
 ◆◆ Primer pair for both alleles

B



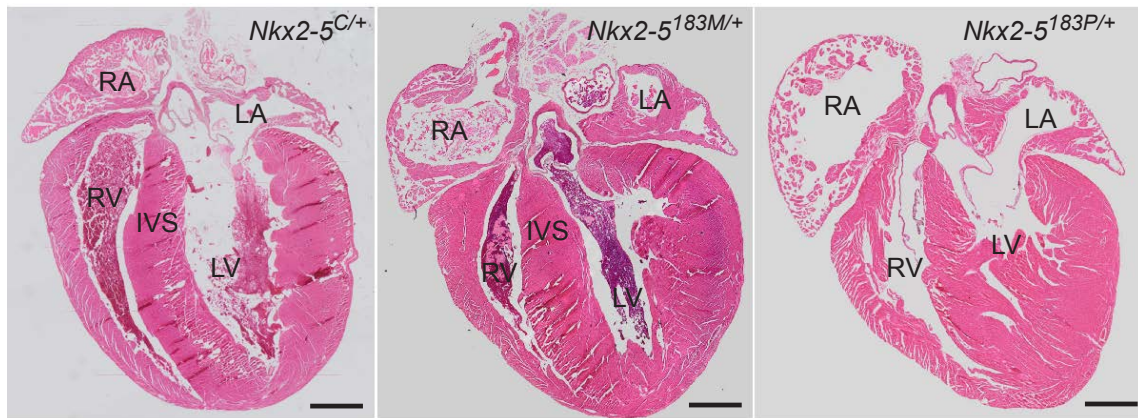
◆◆ Primer pair for WT allele
 ◆◆ Primer pair for 'knocked-in' allele
 ◆◆ Primer pair for both alleles

C



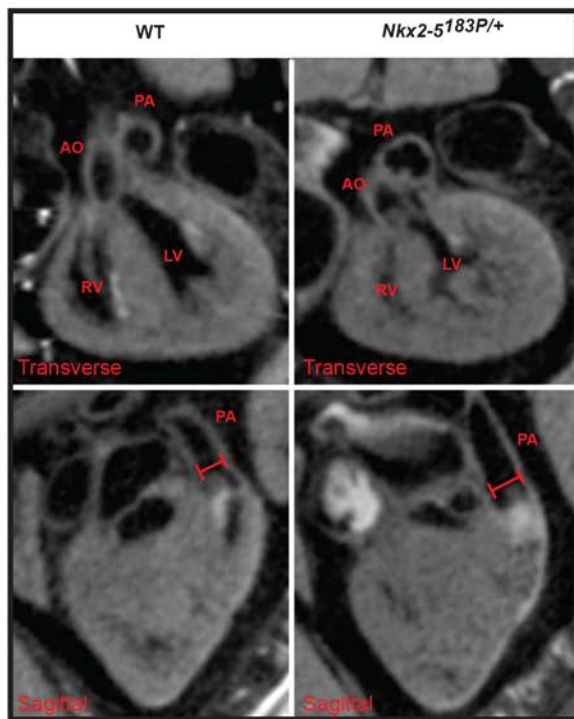
◆◆ Primer pair for WT allele
 ◆◆ Primer pair for 'knocked-in' allele
 ◆◆ Primer pair for both alleles

Supplementary Figure 1. Genotyping strategy to confirm mutated genes by PCR and sequencing.

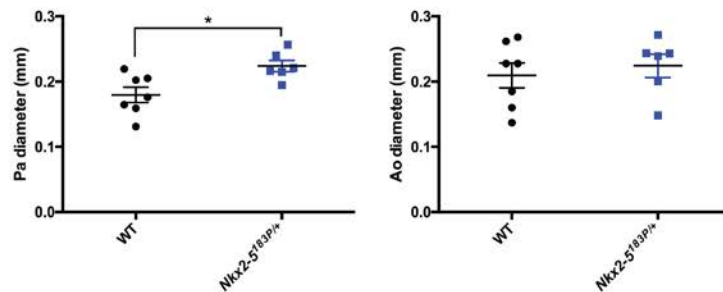


Supplementary Figure 2. Transverse sections of 15-week hearts from heterozygous *Nkx2-5* mice. Images show increased trabeculation, RA dilation and dysmorphic RV in mutant hearts. N=5.

A

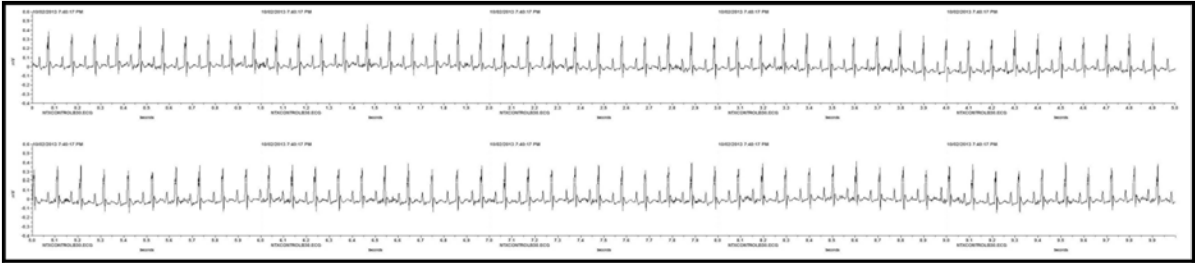


B

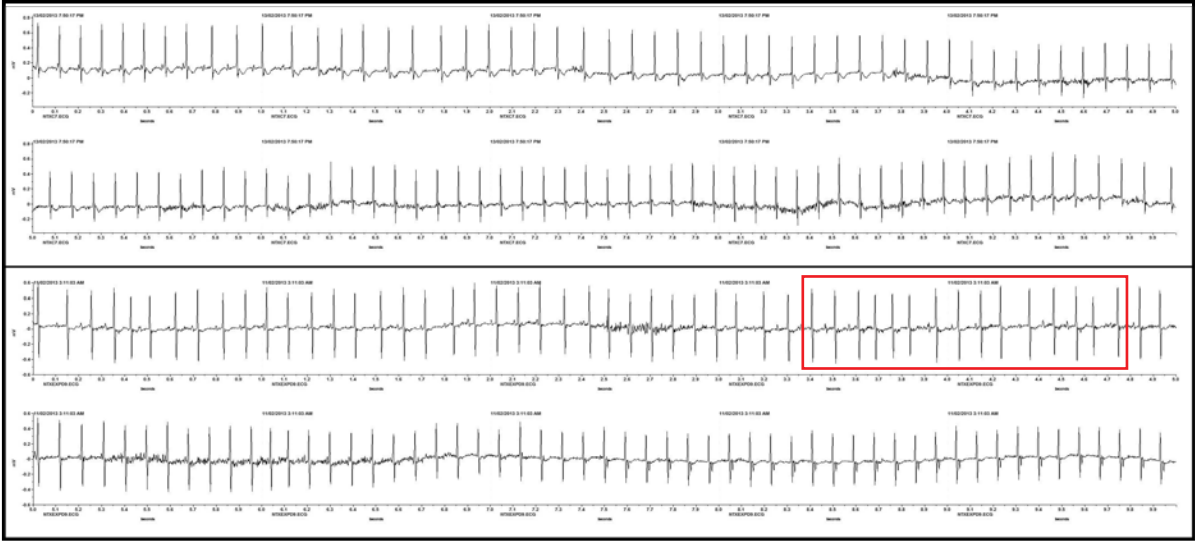


Supplementary Figure 3. MicroCT analysis of 18.5 dpc hearts. (A) Primary enlargement of pulmonary artery (PA) in *Nkx2-5^{183P/+}* embryos. No changes were observed in the aorta. (B) Quantifications performed of sagittal plan using ImageJ (n=6). **P<0.01, Student's t-test. Pa, pulmonary artery; Ao, aorta; LV, left ventricle; RV, right ventricle.

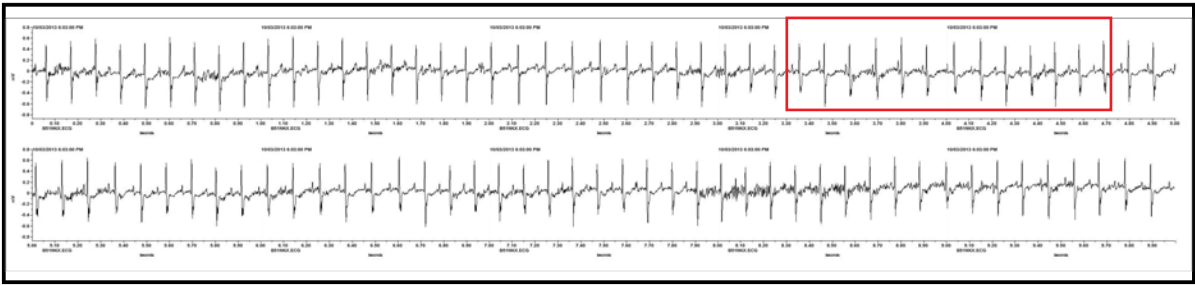
Nkx2-5^{C/+}



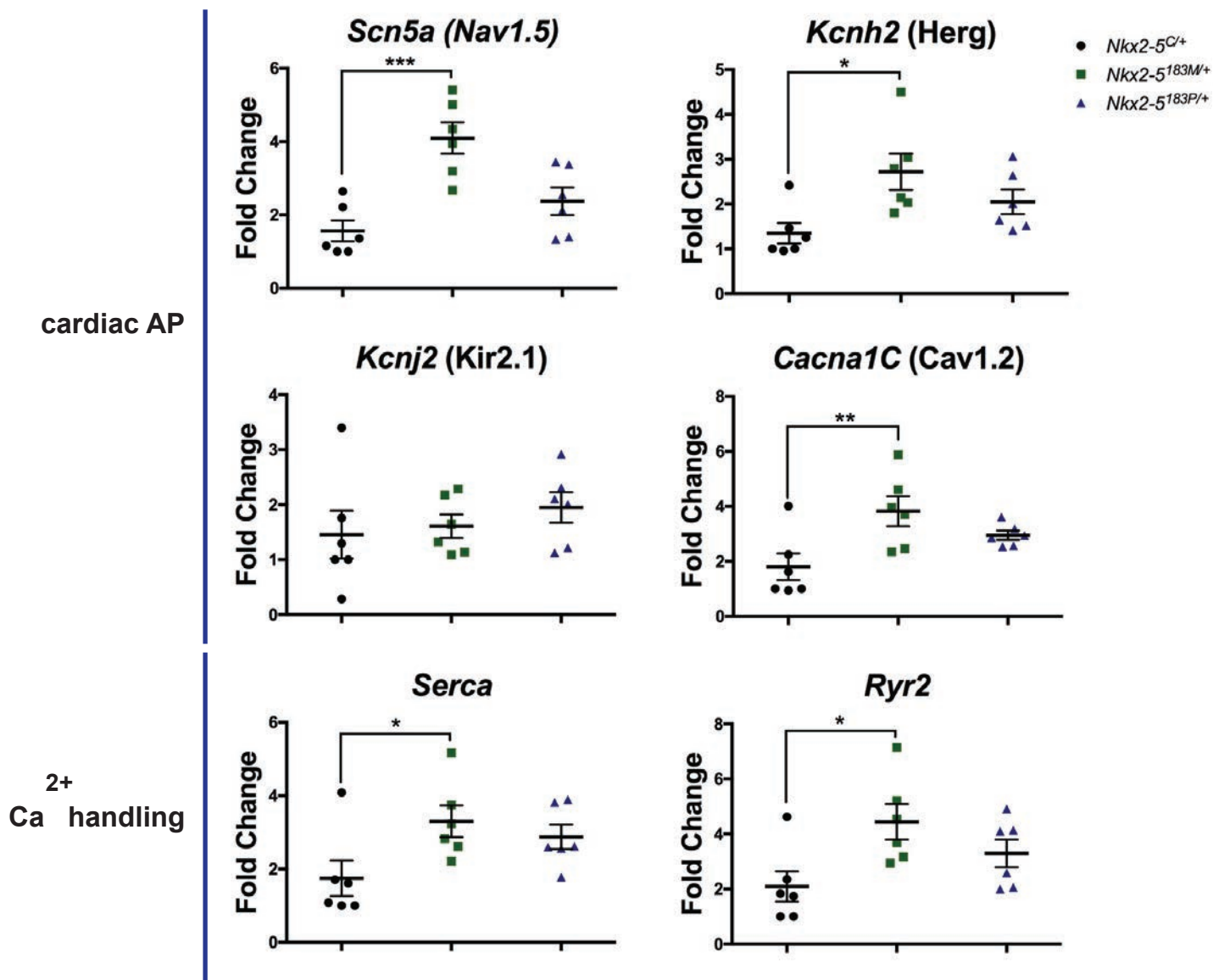
Nkx2-5^{183M/+}



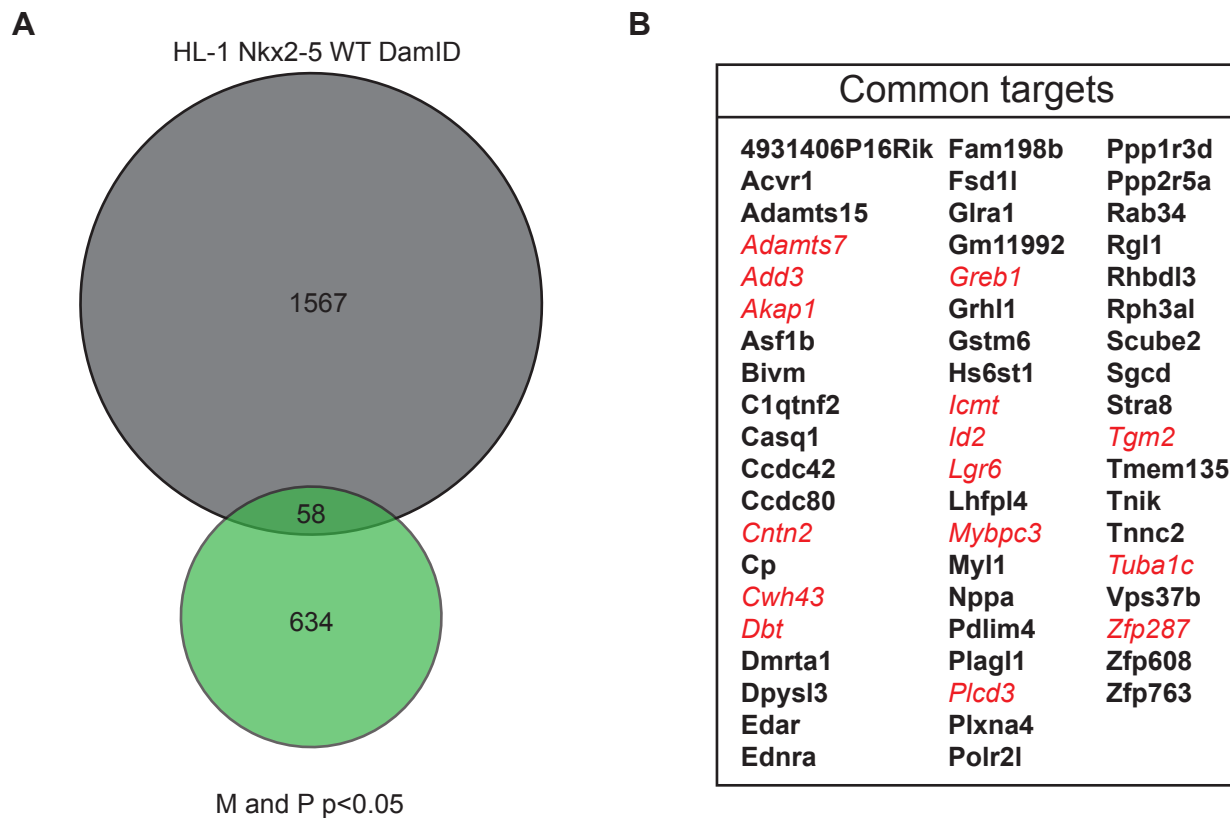
Nkx2-5^{183P/+}



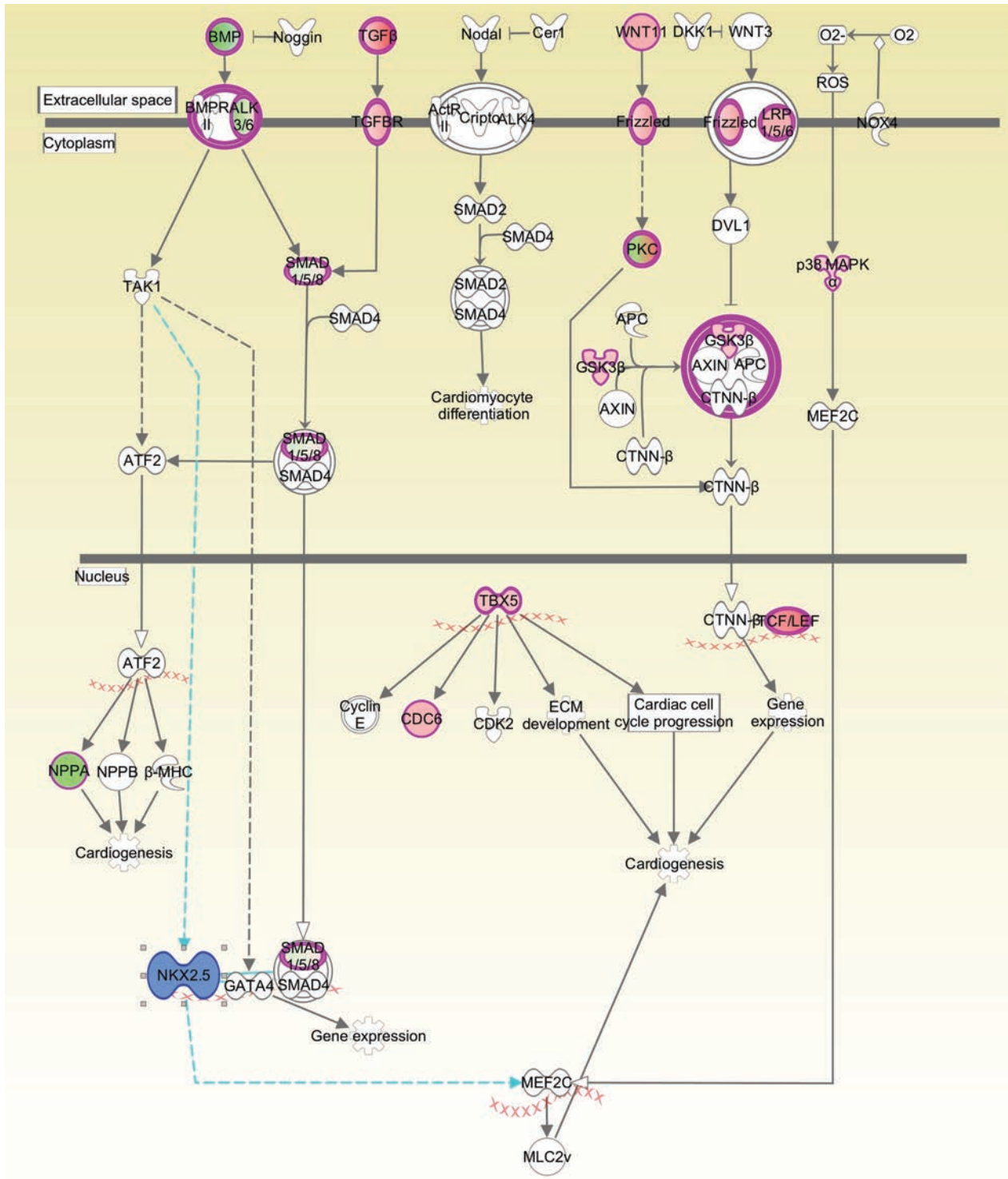
Supplementary Figure 4. Representative ECG traces obtained from telemetry of adult *Nkx2-5* lines showing presence of arrhythmic episodes in mutant hearts (highlighted regions).



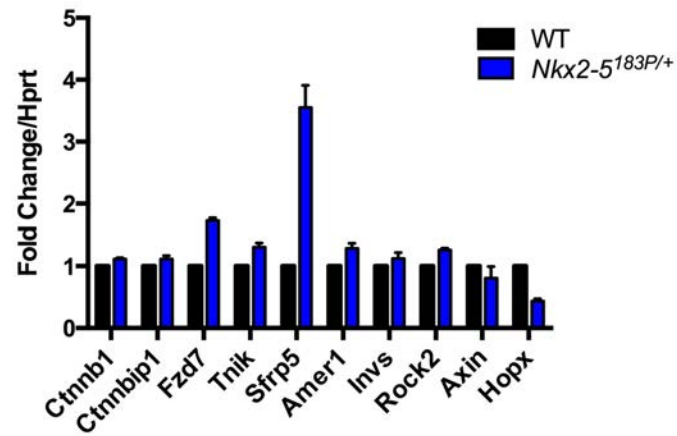
Supplementary Figure 5. qPCR analysis of ion channels, Ca²⁺ handling and sarcomere transcripts in adult heart. N=3, *P<0.05; **P<0.01; ***P<0.001, One-Way ANOVA.



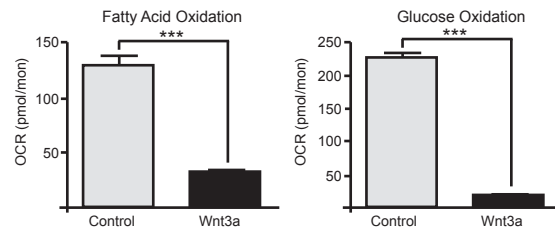
Supplementary Figure 6. Limited overlap of direct targets detected by DamID and transcriptional changes observed in adult microarray experiments. (A) Common and unique gene distributions are shown in Venn diagram. (B) Direct targets changed in heterozygous *Nkx2-5* mutants shown in bold. Novel putative "off target" genes (or co-regulated by *Nkx2-5* and other co-factors) shown in red italic.



Supplementary Figure 7. Cardiogenic pathways identified in *Nkx2-5* mutant hearts. IPA analysis (Qiagen) identified significant changes in several genes associated with BMP, TGFbeta and Wnt signalling pathways.



Supplementary Figure 8. qPCR validation of Wnt associated genes in neonatal cardiomyocytes show activation of several of key regulatory genes. N=3.



Supplementary Figure 9. Wnt3a impairs mitochondrial metabolism in *Nkx2-5*^{183P/+} neonatal cardiomyocytes. n=3; *P<0.001, Student's t-test.**

	τD_{slow} (ms)	α_{slow}	$\tau/4_{\text{slow}}$ ($\mu\text{m}^2\text{s}^{-1}$)	τD_{free} (μs)	D_{free} ($\mu\text{m}^2\text{s}^{-1}$)	%free	n
NKX2-5 WT	108±9.98	0.913±0.025	0.460±0.086	272.2±6.32	30.08±0.68	66.3±0.58	15
NKX2-5 183M	65.21±6.71****	0.826±0.027****	1.064±0.216****	267.3±5.89*	30.61±0.73*	69.98±0.78****	20
NKX2-5 183P	34.03±3.06****	0.841±0.017****	1.501±0.257****	242.0±6.34****	33.82±0.92****	67.56±0.56****	16
paGFP	-	-	-	190.9±9.11****	43.67±2.01****	95.79±0.59****	14

Supplementary Table 1. Summary of diffusion parameters for transfected proteins in the paFCS experiment. Data shown as mean \pm standard error of n measurements. τD_{slow} – dwell time of the slow-diffusing component; α_{slow} – anomalous parameter of the slow-diffusing component; $\tau/4_{\text{slow}}$ – transport coefficient for anomalous diffusion of the slow-diffusing component; τD_{free} – dwell time of the free-diffusing component; D_{free} – diffusion coefficient of the free-diffusing component; %free – percentage of the free-diffusing component. *P<0.05; ****P<0.0001. Student's t-test when compared to NKX2-5 WT.

Control		
	WT	C/+
	35	37
Chi ² P Value=0.81		
183M		
	WT	183M/+
	158	141
Chi ² P Value=0.33		
183P		
	WT	183P/+
	73	60
Chi ² P Value=0.26		

Supplementary Table 2. Heterozygous *Nkx2-5* mice are viable and do not display increased lethality compared to control mice. A non-significant shift in the proportion of born heterozygous mutants was detected indicative of possible decrease viability. Chi test shows expected mendelian ratios of mice generated from heterozygous *Nkx2-5* with WT crosses.

	LV-EF (%)	LV-ESV (μl)	LV-EDV (μl)	LV-SV (μl)	LV-mass (mg)	RV-EF (%)	RV-ESV (μl)	RV-EDV (μl)	RV-SV (μl)	n
<i>Nkx2-5</i>^{C/+}	74.57+/-1.22	18.33+/-1.73	74.29+/-3.69	54.50+/-2.57	87.43+/-3.75	64.21+/-1.08	17.07+/-1.60	44.36+/-3.58	27.21+/-2.13	14
<i>Nkx2-5</i>^{183M/+}	69.15+/-1.97*	17.30+/-1.61****	55.50+/-3.72**	38.30+/-2.65***	73.40+/-2.54*	46.40+/-3.24****	21.00+/-2.45	38.70+/-3.47	17.80+/-1.87*	10
<i>Nkx2-5</i>^{183P/+}	72.38+/-2.03	20.25+/-2.03****	72.88+/-5.08	52.5+/-3.80	89.50+/-4.57	49.38+/-2.29****	24.50+/-1.55*	48.88+/-3.80	24.50+/-2.72	8

Supplementary Table 3. Functional analysis of adult heterozygous *Nkx2-5* mice at 15 weeks of age. *P<0.05; **P<0.01; *P<0.001; ****P<0.0001. One-Way ANOVA.**

Supplementary Table 4. Differentially regulated genes between heterozygous mutant and control in 15 week adult hearts. P<0.001. Non-coding genes were excluded.

<i>Nkx2-5^{183M/+} x Nkx2-5^{C/+}</i>			<i>Nkx2-5^{183P/+} x Nkx2-5^{C/+}</i>			<i>Nkx2-5^{delta/+} x Nkx2-5^{C/+}</i>		
Probe ID	Genes	LogFC	Probe ID	Genes	LogFC	Probe ID	Genes	LogFC
A_51_P416858	Myl1	2.136	A_52_P566605	Hsd17b7	2.043	A_55_P2004551	Klra1	2.694
A_55_P2002893	Pfkfb1	1.572	A_55_P2007964	Cx3cr1	1.954	A_55_P1960738	Gm4470	2.430
A_55_P1997936	Hsd17b7	1.514	A_66_P125389	F830016B08Rik	1.890	A_55_P2370250	Syn3	2.182
A_55_P1987645	Unc13b	1.106	A_55_P2002893	Pfkfb1	1.880	A_55_P2180854	Mrgprg	2.061
A_51_P520650	Dlgap1	1.093	A_51_P416858	Myl1	1.829	A_55_P1983418	Amy1	1.962
A_55_P2108248	Art4	0.978	A_55_P2031781	Col22a1	1.826	A_66_P111689	Gm9372	1.940
A_66_P118699	Rorb	0.882	A_55_P2166049	Vmn1r65	1.806	A_55_P1996314	Amy2a5	1.901
A_55_P1991770	Pdlim4	0.872	A_55_P1983418	Amy1	1.738	A_55_P2109922	Gm2921	1.893
A_55_P1988699	Cacna2d1	0.858	A_55_P2133195	Gm4951	1.631	A_55_P1959430	Chat	1.890
A_55_P2134004	Gstm2	0.793	A_55_P2050390	Cdh22	1.609	A_65_P20683	Km1070	1.867
A_55_P1953400	Rbfox1	0.622	A_51_P229655	Acsm5	1.504	A_55_P2173857	Orly	1.797
A_55_P1958758	Olf965	-0.513	A_55_P2006494	Apol10b	1.459	A_51_P326229	Ddx25	1.785
A_51_P212038	Atp6v0e2	-0.527	A_52_P53019	Rhd13	1.453	A_55_P2006950	Usp8	1.746
A_55_P2015753	Enho	-0.540	A_66_P112495	Scn4b	1.403	A_66_P133993	Gm5093	1.731
A_55_P1980556	Gm4316	-0.626	A_51_P268069	Six1	1.402	A_55_P2133195	Gm4951	1.676
A_66_P108685	Cideb	-0.662	A_51_P382369	Zfp608	1.379	A_51_P416858	Myl1	1.568
A_55_P2041966	Dnajc5b	-0.678	A_55_P1997936	Hsd17b7	1.327	A_55_P2144248	Zfp790	1.520
A_52_P233515	Tll13	-0.693	A_55_P2096144	Gnao1	1.250	A_55_P2004761	H2-Eb2	1.481
A_55_P2162747	Accn4	-0.722	A_52_P434306	Col22a1	1.191	A_55_P2007964	Cx3cr1	1.478
A_55_P1955009	C130079G13Rik	-0.742	A_52_P308507	Chd6	1.184	A_55_P2019004	Ncoa7	1.435
A_55_P1991209	Lair1	-0.763	A_52_P111031	Pcdh17	1.129	A_51_P516728	Hap1	1.392
A_55_P2020449	Vmn1r31	-0.875	A_55_P1996314	Amy2a5	1.093	A_55_P2006494	Apol10b	1.314
A_55_P2123057	Olf853	-0.878	A_55_P1956418	Efr3b	1.080	A_52_P518922	Itga1	1.294
A_55_P2119764	Krtap6-1	-0.889	A_51_P520650	Dlgap1	1.073	A_55_P2177910	Lepr	1.265
A_55_P2232988	Scube2	-0.905	A_55_P2014427	Il17re	1.069	A_55_P2018017	Tnfsf10	1.199
A_55_P2130256	Polr2l	-0.909	A_55_P1991770	Pdlim4	1.058	A_55_P2039622	Clec4a2	1.195
A_51_P116932	Lad1	-0.911	A_55_P1991214	Lair1	1.036	A_55_P2106394	D2Ertd750e	1.165
A_55_P2089422	Olig3	-0.966	A_55_P2018017	Tnfsf10	1.006	A_55_P1979818	Srp54b	1.162
A_55_P2017929	Clu	-0.976	A_51_P346641	Armcx4	1.003	A_55_P1995141	Cas21	1.152
A_52_P603038	Olig1	-0.978	A_52_P340073	Efnb2	0.995	A_66_P139387	Prlr	1.143
A_52_P599573	4930596D02Rik	-1.030	A_52_P599789	Qrich2	0.937	A_51_P458852	Ina	1.133
A_66_P102910	Rcvrn	-1.032	A_55_P2108248	Art4	0.915	A_55_P1986566	Gm19668	1.110
A_51_P434758	Fam83e	-1.053	A_51_P234788	Cx3c5	0.906	A_52_P434306	Col22a1	1.098
A_52_P329414	Calm5	-1.097	A_55_P2143025	Sema3c	0.900	A_55_P2120596	BC018473	1.094
A_55_P2041286	Gm3970	-1.100	A_55_P2228122	BC024137	0.896	A_55_P2043833	Srgap2	1.073
A_55_P2019483	Ndr4	-1.243	A_55_P1968370	Gprc5c	0.894	A_52_P308507	Chd6	1.034
A_52_P222026	Chnb2	-1.275	A_65_P11092	Tspan15	0.883	A_66_P112495	Scn4b	1.018
A_52_P156775	Scgb1c1	-3.829	A_55_P2137611	Irgm2	0.875	A_55_P2148688	Nipal3	0.992
			A_66_P118699	Rorb	0.841	A_65_P10913	Tgfb2	0.950
			A_52_P596592	Rassf9	0.812	A_55_P2076524	1700028I16Rik	0.944
			A_51_P169516	Ppp1r3d	0.769	A_55_P1956418	Efr3b	0.942
			A_55_P1953400	Rbfox1	0.722	A_66_P118699	Rorb	0.933
			A_55_P2060343	Zfp352	0.702	A_55_P2049553	Gm6003	0.925
			A_55_P2134004	Gstm2	0.696	A_55_P2161342	Pkib	0.922
			A_51_P483473	St3gal5	0.657	A_55_P2422243	2900072G11Rik	0.897
			A_51_P284486	Gstm2	0.629	A_52_P340073	Efnb2	0.890
			A_55_P2096043	Acot11	0.574	A_55_P1966438	Gstm2	0.885
			A_51_P458866	F11r	0.562	A_55_P2173353	Gm628	0.875
			A_52_P552665	Fzd7	0.523	A_51_P284486	Gstm2	0.850
			A_55_P1963712	Cyb5b	0.519	A_55_P2120993	Zfp282	0.842
			A_55_P1980556	Gm4316	-0.473	A_55_P2335648	D630045M09Rik	0.827
			A_51_P306047	Sec13	-0.555	A_55_P2134581	Papola	0.804
			A_55_P2147280	Myh1	-0.591	A_55_P2009427	Nrsn2	0.802
			A_52_P322941	Gm11992	-0.628	A_55_P2057321	Myh7b	0.797
			A_55_P2023001	Slc47a1	-0.657	A_55_P2005941	Plcx1	0.796
			A_55_P2110935	2310046A06Rik	-0.666	A_52_P596592	Rassf9	0.786
			A_55_P1959510	Oog3	-0.668	A_55_P2134004	Gstm2	0.768
			A_55_P1973868	Sema3b	-0.700	A_51_P485260	Rgl1	0.748
			A_51_P108489	Gtl3	-0.734	A_55_P1997614	Gm9693	0.731
			A_51_P403049	Heat5b	-0.767	A_55_P2131379	Ybx1	0.727
			A_66_P106661	Slc7a1	-0.804	A_55_P2016851	Lce1a2	0.724
			A_66_P114543	Jmjd6	-0.821	A_55_P2088331	Ctdsp2	0.717
			A_55_P1970126	Heat5b	-0.824	A_55_P2072656	Ckmt1	0.708
			A_55_P2315366	D630022N01Rik	-0.836	A_55_P2156785	Ppfia3	0.701
			A_51_P116932	Lad1	-0.873	A_51_P505493	Elovl5	0.677
			A_52_P112110	Tmem82	-0.908	A_55_P1992049	Gucy1a3	0.623
			A_52_P368306	Tmem100	-0.944	A_55_P2009576	Rnu12	0.617
			A_52_P319438	Ankrd37	-0.988	A_55_P2096535	Icmt	0.615
			A_55_P2109877	Gm4718	-1.019	A_55_P2164998	LOC546711	0.590
			A_55_P2041286	Gm3970	-1.052	A_55_P1956179	Git2	0.579
			A_51_P253984	Pcp4	-1.210	A_55_P1955244	LOC677113	0.572
			A_51_P434758	Fam83e	-1.232	A_55_P1979705	Spop	0.568
			A_51_P254425	Ahrr	-1.306	A_55_P2068451	Gm7634	0.565
			A_55_P2019483	Ndr4	-1.634	A_65_P09177	Strn3	0.530
			A_51_P308844	Nrn1	-1.795	A_55_P2106351	Pde8a	0.526

A_66_P108685	Cideb	-1.832	A_55_P2161829	Hmgb1-rs17	0.509
A_51_P386899	Mfsd7c	-1.977	A_55_P2049256	Mapkapk5	-0.513
A_52_P156775	Scgb1c1	-3.764	A_51_P306047	Sec13	-0.528
			A_55_P2186558	5730437N04Rik	-0.549
			A_51_P259902	Arl6ip5	-0.559
			A_55_P2093949	Zfp428	-0.595
			A_66_P111666	Nprl3	-0.614
			A_55_P2089820	Adk	-0.615
			A_51_P427828	2310046A06Rik	-0.639
			A_55_P2140288	Phka2	-0.659
			A_55_P2041966	Dnajc5b	-0.664
			A_55_P1973868	Sema3b	-0.665
			A_55_P2064243	Rhot2	-0.678
			A_55_P2047305	Adcy5	-0.685
			A_55_P2160686	Tsc22d1	-0.714
			A_55_P1970126	Heatr5b	-0.721
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			A_51_P173285	Nkx2-5	-0.729
			A_55_P2058947	Gpr113	-0.737
			A_55_P2059323	Gm13315	-0.759
			A_51_P139030	Slc38a3	-0.786
			A_55_P2028942	Ncam1	-0.789
			A_55_P2141878	Ldha	-0.801
			A_51_P416137	Slc31a2	-0.801
			A_52_P444785	N4bp2l2	-0.813
			A_52_P319438	Ankrd37	-0.822
			A_55_P2017929	Clu	-0.828
			A_55_P1977926	Itgam	-0.829
			A_51_P207892	Pla2g5	-0.834
			A_55_P1991475	Sesn1	-0.835
			A_66_P106661	Slc7a1	-0.848
			A_55_P2029517	Wnk2	-0.863
			A_55_P2415372	Mta1	-0.890
			A_55_P2198648	5830420C07Rik	-0.925
			A_55_P2152962	Mtr	-0.954
			A_55_P2001048	Rom1	-0.958
			A_51_P308844	Nrn1	-0.990
			A_51_P264527	Fam69b	-1.043
			A_51_P366672	Slc36a2	-1.050
			A_51_P403049	Heatr5b	-1.083
			A_51_P441687	Lrrc10	-1.099
			A_52_P322941	Gm11992	-1.139
			A_55_P2243828	LOC552901	-1.185
			A_51_P116932	Lad1	-1.197
			A_66_P108685	Cideb	-1.198
			A_55_P2221647	Al605517	-1.238
			A_52_P112110	Tmem82	-1.261
			A_51_P253984	Pcp4	-1.379
			A_51_P386899	Mfsd7c	-1.617
			A_51_P102257	Tns1	-1.624
			A_55_P2091005	Emilin3	-1.726
			A_55_P2019483	Ndrgr4	-1.765
			A_51_P416059	Dirc2	-3.131
			A_52_P156775	Scgb1c1	-3.339
			A_55_P2042778	Pde5a	-3.912

Supplementary Table 5: Nuclear mitochondrial genes altered in Nkx2-5 mutant hearts , p<0.05

Unique genes in the dataset are highlighted.

<i>Nkx2-5^{C/+} vs Nkx2-5^{183M/+}</i>		
Gene	LogFC	P
1110001A16Rik	-0.2735286	0.03047379
Abat	-0.666291	0.04384559
Acadl	0.30546113	0.03183362
Acsm2	-0.545393	0.02471223
Acsm5	1.29224592	0.00154268
Agxt2	-0.6908551	0.01612945
Akap1	-0.5103126	0.04614019
Amt	-0.6748733	0.00285085
Casp8	0.76276356	0.02175548
Cbr3	0.72490057	0.03283243
Cdc25c	-0.5441091	0.03934748
Chdh	0.42025026	0.03729079
Cox4i2	0.53064339	0.02645452
Cps1	-0.5487251	0.00546736
Dbt	0.48975997	0.02276071
Fasn	-0.8122031	0.0287357
Gcat	0.333234	0.04556823
Gcdh	0.31986668	0.04989917
Hadha	0.33235942	0.02684848
Hmgcs2	1.65147922	0.01235979
Ivd	0.31014115	0.02372705
Maob	0.45267599	0.01746585
Me2	0.41942569	0.04354536
Me3	-0.474724	0.02861808
Nags	0.91847019	0.04157501
Ndufab1	0.63786613	0.01936018
Nme6	-0.6824001	0.0414029
Ppm1k	0.509431	0.01581422
Ppm1m	-0.6462861	0.0220071
Prodh2	-0.595124	0.00823404
Ptrh2	0.43433368	0.0457698
Rmnd1	0.34720346	0.00925468
Rps15a	-0.4576475	0.03456073
Slc25a29	0.76007537	0.02892512
Timm22	-0.6810465	0.03081372
Trmt1	-0.8409265	0.03605288
Trmt1	-1.2295023	0.02465549
Tst	0.37721316	0.02695517

<i>Nkx2-5^{C/+} vs Nkx2-5^{183P/+}</i>		
Gene	LogFC	P
1110001A16Rik	-0.3684221	0.00624785
Abcb7	0.91775109	0.04919295
Acox3	0.43592058	0.04566804
Acs16	-0.7315617	0.039003
Acsm5	1.50401067	0.00047378
Adeck2	0.31050142	0.04922751
Agxt2	0.50324262	0.0359715
Akap1	-0.619753	0.01921539
Aldh1b1	0.52841921	0.03472067
Amt	-0.5074039	0.01596197
Arl2	-0.3680006	0.01445108
Bad	-0.5935721	0.02454758
Bak1	0.291371	0.02696098
Bcat1	-0.734577	0.03348201
Bdh1	-0.8739484	0.01357747
Bdh1	-1.0106383	0.00319729
Bdh1	-1.0802411	0.00209509
Cbr3	0.68455371	0.04193345
Chchd7	-0.2985351	0.04369802
Clpp	-0.2971245	0.02727664
Coq10a	-0.243407	0.03453831
Cox10	-1.2687552	0.00976674
Cox4i2	0.57062091	0.01858243
Cps1	-0.4221114	0.0233802
Cpt1a	0.46017903	0.01784874
Cyb5b	0.51932933	0.00086999
Cyb5r2	-0.9789595	0.04460805
Cycs	0.90661363	0.04133428
Dbt	0.65499598	0.0044012
Dmpk	0.25711383	0.04679421
Dnajc19	0.45284948	0.04996707
Ehhadh	1.09748815	0.00982639
Fasn	-1.1116536	0.00521318
Fdx11	-0.3002078	0.03123492
Fhit	-1.3132776	0.03443687
Fis1	-0.6122372	0.04754122
Fxn	-0.3078358	0.02543058
Glrx2	0.6607267	0.00996206
Gm2382	0.55726484	0.03866337
Grhpr	-0.6572423	0.00150302

Grpel2	-0.6036483	0.01068428
Guk1	-0.2825575	0.03369678
Hmgcs2	1.70759453	0.01025461
Hscb	-0.327761	0.02845506
Idh1	0.96962212	0.00238931
Ivd	0.34382811	0.01406349
Kmo	0.76216892	0.01264214
Letm1	-0.4711852	0.0114351
Maob	0.54301714	0.00625622
Mccc1	0.46395968	0.02345801
Me1	0.36044501	0.01239332
Me1	0.35266655	0.01995034
Me2	0.6410993	0.00475765
Nars2	0.9629256	0.03229298
Ndufa11	-0.3311165	0.01490096
Neu4	-1.0561795	0.04995186
Obscn	-0.4408223	0.0045021
Pdp2	0.80287553	0.00503707
Phb2	-0.4212033	0.03497485
Ppif	-0.4731227	0.00807968
Ppm1m	0.5112551	0.040396
Pptc7	1.08357813	0.03087387
Prdx6	-0.3656082	0.03964185
Ptcd1	-1.113848	0.03245393
Pthr2	0.59968795	0.00952199
Rab32	0.6130598	0.0479702
Rhot2	-0.4179646	0.01453174
Rmnd1	0.31438739	0.01598274
Sfxn1	0.58947106	0.01339047
Sfxn2	0.90694058	0.03452953
Sfxn5	1.21129972	0.0358932
Sfxn5	-1.381183	0.03627511
Slc25a22	0.75229823	0.00241471
Slc25a29	0.75346365	0.03009611
Slc25a42	0.3473958	0.04078559
Timm10	-0.3698313	0.0391626
Timm50	-0.4911362	0.01037723
Tomm40	-0.4879227	0.00273034
Tpi1	-0.3418447	0.01506538
Trmt1	-1.0016886	0.01565189
Tst	0.34673418	0.03913891
Tstd1	0.65750023	0.03182337
Txn2	0.68971734	0.03448759
Ucp3	0.92425566	0.00589386

<i>Nkx2-5^{C/+}</i> vs <i>Nkx2-5^{I83P/+}</i>		
Genes	Log FC	P-Value
<i>Smad1</i>	-0.3721	0.0333
<i>Smad5</i>	0.2589	0.0459
<i>Smad6</i>	0.8491	0.0375
<i>Bmp5</i>	-1.5434	0.0445
<i>Bmp7</i>	-0.8044	0.0020
<i>Bmp10</i>	-0.8628	0.0350
<i>Hopx</i>	0.4720	0.0332

Supplementary Table 6. Differential expression of Tgfb/Bmp associated genes in *Nkx2-5^{I83P/+}* mice.

Supplementary Table 7. Primers and antibodies used.

Gene	primer name	sequence 5'-3'	PCR fragm	ID
qPCR				
Hypoxanthine phosphorybosyl transferase	Hprt1 F Hprt1 R	GCGAGGGAGAGCGTTGGGCT CATCATCGCTAATCACGACGCTGGG	146 bp	NM_013556.2
NK2 transcription factor related, locus 5	Nkx2-5 F Nkx2-5 R	TCAAGCCCGAGGCCTACTCTGG TGGTCTCTCGGCGCCATCCG	248 bp	NM_008700.2
Secretoglobin, family 1C, member 1	Scgb1c1 F Scgb1c1 R	CTGCGTCTGTGGGCTGACTA CGTAGAGTTCTTCCGGGGTC	95 bp	NM_001099742.1
N-myc downstream regulated gene 4	Ndr4 F Ndr4 R	TTCGGCAAATCCCCTTCCTC CCGGATCACCACATGCAGAA	95 bp	NM_001195006.1
Jumonji domain containing 6	Jmjd6 F Jmjd6 R	GCACAAGACGGTAAGAGGGAG CTCAGGGTGCTCCTGTTTCA	73 bp	NM_033398.2
Sine oculis-related homeobox 1	Six1 F Six1 R	AGAACCGGAGGCAAAGAGAC CCCCTTCCAGAGGAGAGAGT	108 bp	NM_009189.3
Calcium channel, voltage-dependent, alpha2/delta subunit 1	Cacna2d1 F Cacna2d1 R	GTCATGGGTGGACAAGATGC ATTTCAACCAGTTGGCGTGC	141 bp	NM_001110843.1
Myosin, light polypeptide 1	Myl1 F Myl1 R	TTGGGAACCCAGCAATGAA ACGCAGACCCTCAACGAAAT	128 bp	NM_021285.3
Signal peptide, CUB domain, EGF-like 2	Scube2 F Scube2 R	TGTGACAACACACTCAACGGA TGTTCTCCAAGCATTCGTCCA	139 bp	NM_020052.2
Signal peptide, CUB domain, EGF-like 3	Scube3 F Scube3 R	CTGCTACGACGGATTTACCT AGCTGCCCATCATGTTGACACA	113 bp	NM_001004366.1
Catenin (cadherin associated protein), beta 1	Ctnnb1 F Ctnnb1 R	GAGCACATCAGGACACCCAA CCGAGCAAGGATGTGGAGAG	122 bp	NM_007614.3
Catenin beta interacting protein 1	Ctnnbip1 F Ctnnbip1 R	CACAGCACTCCATCGACCAG CGGTCTTCCGTCTCCGATCT	70 bp	NM_023465.4
Frizzled homolog 7	Fzd7 F Fzd7 R	ACCCTACTGCTCCCTACCTG AGAAGGGGAAAGACAAGCGG	84 bp	NM_008057.3
TRAF2 and NCK interacting kinase	Tnik F Tnik R	CCCATGAGCCTTCCAAGGTG GCTAATGCCGTCAGATCCTCA	106 bp	NM_026910.1
Secreted frizzled-related sequence protein 5	Sfrp5 F Sfrp5 R	CTGGACAACGACCTCTGCAT GCTGTGCTCCATCTCACACT	99 bp	NM_018780.3
APC membrane recruitment 1	Amer1 F	GCAGCGCAGCAGACAATAAA	102 bp	NM_175179.4

Inversin	Amer1 R Invs F Invs R	CTTTGGATTCCGGGCACACT AGCAGCGCCTGCTGATAAT CAAATGCAGAAGCCGAGACA	137 bp	NM_010569.4
Rho-associated coiled-coil containing protein kinase 2	Rock2 F Rock2 R	CTCATCCGAGACCCTCGCTC CAAGGACCAAGGAATTTAAGC	70 bp	NM_009072.2
Axin2	Axin2 F Axin2 R	ATAAGCAGCCGTTCCGCGATG GCAATCGGCTTGGTCTCTCT	135 bp	NM_015732.4
HOP homeobox	Hopx F Hopx R	AGGTGGAGATCCTGGAGTACA AGGCGCTGCTTAAACCATT	127 bp	NM_175606.3
Sodium channel, voltage-gated, type V, alpha	Nav1.5 F Nav1.5 R	GATGAGGAGAACAGCCTTGG CACAACCTTGGGATTCCTGCT	66 bp	NM_021544.3
Potassium voltage-gated channel, subfamily H, member 2	Kenh2 F Kenh2 R	GATCGCCTTCTACCGGAAA CATTCTTCACGGGTACCACA	68 bp	NM_013569.2
Calcium channel, voltage-dependent, L type, alpha 1C subunit	Cav1.2 F Cav1.2 R	CATGAAGCTCAACTCAACTGTTTC CGTGGGCTCCCATAGTTG	62 bp	NM_009781.3
Sarcoplasmic reticulum Ca ATPase	Serca F Serca R	TCGACCAGTCAATTCTTACAGG CAGGGACAGGGTCAGTATGC	63 bp	NM_009722.3
Ryanodine receptor 2 (cardiac)	Ryr2 F Ryr2 R	TTCAACACGCTCACGGAGTA TGCCAGGCTCTGCTGATT	60 bp	NM_023868.2
Genotyping				
	Nkx2-5 CO WT For Nkx2-5 CO WT Rev	CGTGAACCTTTGGCGTCGGGG ATAAATACGGGTGGGTGGG	398 bp WT 486 bp MUT	
Antibodies				
Nkx2-5, rabbit	Abcam	ab35842		
α -tubulin, mouse	St Cruz Bio.	sc-5286		
b-catenin, mouse	BD Biosciences	610154		
anti mouse IgG-488	Invitrogen	A11001		
anti rabbit IgG-568	Invitrogen	A11011		
anti rabbit IgG-680	Invitrogen	A10043		
anti mouse IgG-IR dye 800	Li-Cor	926-32214		