

Cardiac Calcineurin Deficiency

Supplemental Table 1. Sequences of mouse primers/probes used in real-time qPCR.

Calsequestrin (Casq2)	FWD primer RV primer Probe	5'- CATCCCTAACAAACCGTACACAGA-3' 5'- CGATGTGGATCCCATTCAAGT -3' 5'- AGCTTGTGGAGTTGTGAAGGAACATCAA -3'
Cytochrome C (Cycs)	FWD primer RV primer Probe	5'-ACCAAATCTCCACGGTCTGTT-3' 5'-GGATTCTCAAATACTCCATCAG-3' 5'-ACAGATGCCAACAAAGAACAAAGGCAT-3'
ATP Synthase Beta subunit (Atp5b)	FWD primer RV primer Probe	5'-CGTGAGGGCAATGATTATACCAT-3' 5'-TCCTGGTCTCTGAAGTATTCAAGCAA-3' 5'-ACCAACGCTACCTTGGAAAGTGGCATCT-3
Nppa (ANF)	FWD primer RV primer Probe	5'-AGTGCGGTGTCCAACACAGA-3' 5'-GACCTCATCTTCTACCGGCATCT-3' 5'-TCAAGAACCTGCTAGACCTGGAGGAG-3'
Nppb (BNP)	FWD primer RV primer Probe	5'-GCTGCTTGGGCACAAGATAG-3' 5'-GCAGCCAGGAGGTCTTCATA-3' 5'-CAGTGCCTTACAGCCCCAACGA-3'
Phospholamban (Pln)	FWD primer RV primer Probe	5'-CAGCTTCATGCTCTGCACTGT- 3' 5'-GCCAAATGTGAGCTGTCTTCT -3' 5'-ATCACCGAACGCAAGGTCTCCTAA -3'
Atp2a2 (SERCA2a)	FWD primer RV primer Probe	5'-GGAGATGCACCTGGAAGACT-3' 5'-CCACACAGCCGACGAAA-3' 5'-TTCATCAAATACGAGACCAACCTGACT-3'
L-type Calcium Channel (Cacna1c)	FWD primer RV primer	5'-GGAGGACAAGAGGGAGATCCA- 3' 5'-GGAGGCCCTTCGACCTAGAG-3'
Na-Ca Exchanger (Slc8a1)	FWD primer RV primer	5'-CCATCTTCCGAATGTCAATG- 3' 5'-GCCTTTTGGTCTTATTTC -3'
36B4	FWD primer RV primer	5'-ATCCCTGACGCACCGCCGTGA-3' 5'-TGCATCTGCTGGAGGCCACGTT-3

Probes are 5'-FAM and 3'-TAMRA modified

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Supplemental Table 2. Results of Echocardiographic analysis of csCnb1^{-/-} and mice at 1 and 4 months of age.

	ONE MONTH		FOUR MONTH	
	csCnb1 ^{+/+} (4)	csCnb1 ^{-/-} (5)	csCnb1 ^{+/+} (4)	csCnb1 ^{-/-} (4)
HR (bpm)	677 ± 30	726 ± 36	724 ± 50	576 ± 48
LVPWd (mm)	0.64 ± 0.01	0.59 ± 0.03	0.67 ± 0.02	0.62 ± 0.04
IVSd (mm)	0.69 ± 0.01	0.64 ± 0.02	0.79 ± 0.05	0.66 ± 0.04
LVIDd (mm)	3.11 ± 0.06	3.32 ± 0.09	2.98 ± 0.10	3.36 ± 0.32
LVPWs (mm)	1.33 ± 0.06	0.96 ± 0.08*	1.41 ± 0.02	0.96 ± 0.07*
IVSs (mm)	1.41 ± 0.08	1.04 ± 0.05*	1.59 ± 0.12	1.08 ± 0.05*
LVIDs (mm)	1.14 ± 0.09	2.05 ± 0.12*	1.07 ± 0.07	2.37 ± 0.24*
RWT	0.43 ± 0.01	0.37 ± 0.01*	0.49 ± 0.03	0.39 ± 0.05
FS (%)	63.3 ± 2.6	38.3 ± 2.1*	64.1 ± 1.5	29.5 ± 2.0*

Values represent mean ± SEM. Data were analyzed by ANOVA followed by the Tukey test. Abbreviations: HR, heart rate; LVPWd, left ventricle (LV) posterior wall thickness, diastole; IVSd, intraventricular septum thickness, diastole; LVIDd, LV internal dimension, diastole; LVPWs, LV posterior wall thickness, systole; IVSs, intraventricular septum thickness, systole; LVIDs, LV internal dimension, systole; RWT, relative wall thickness [(LVPWd+IVSd)/LVIDd]; and FS, fractional shortening [((LVIDd-LVIDs)/LVIDd) × 100]. *P<0.05 compared with age-matched controls.

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Supplemental Table 3. Doppler measurements of transmitral flow in 4 month old $\text{csCnb1}^{+/+}$ and $\text{csCnb1}^{-/-}$ mice.

	$\text{csCnb1}^{+/+}$ (n=4)	$\text{csCnb1}^{-/-}$ (n=4)
E	1.00 ± 0.04	1.02 ± 0.06
DT	25 ± 1.1	$12 \pm 1.1^*$
IVRT	13 ± 0.5	$17 \pm 0.7^*$
Ea	0.054 ± 0.002	$0.037 \pm 0.002^*$
E/Ea	18.6 ± 0.7	$27.2 \pm 0.2^*$
Sa	0.041 ± 0.01	$0.025 \pm 0.01^*$
PW (syst)	0.046 ± 0.001	$0.025 \pm 0.001^*$
PW (diast)	0.051 ± 0.002	$0.037 \pm 0.001^*$

Results of echocardiographic Doppler studies performed on $\text{csCnb1}^{+/+}$ and $\text{csCnb1}^{-/-}$ mice at 4 months of age. Values represent mean \pm SEM. Data were analyzed using the t-test. Abbreviations: E, Peak trans-mitral E velocity; DT, deceleration time of E; IVRT, Isovolumic relaxation time; Ea, Peak velocity of early diastolic mitral annular velocity; Sa, Peak velocity of systolic mitral annular velocity; PW (syst), Posterior wall rate of thickening; PW (diast), Posterior wall rate of thinning. *P<0.05 compared with age-matched controls.