Fig. S1. Analysis of mitochondrial genes during heart development. (A) Heatmap. Three clusters of genes with distinct temporal expression patterns were categorized. **(B)** GO enrichment analysis.
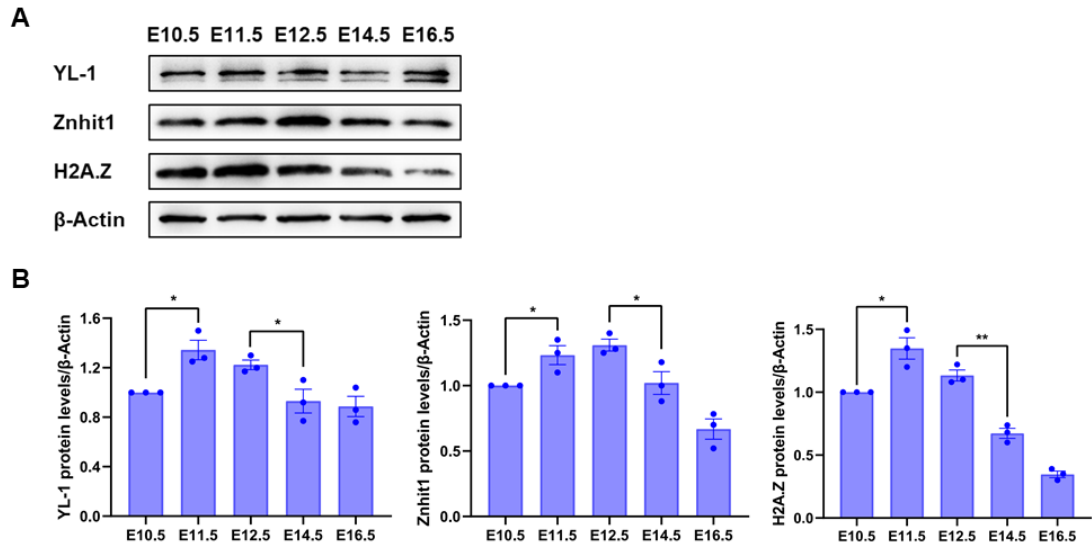
Fig. S2. The temporal expression pattern of the core subunits of SRCAP complex.
(A) Western blotting analysis. **(B)** Quantification of **(A)**. Experiments were repeated independently for three times, which produced similar results. Data are mean ± s.e.m. Student's t test: *$P < 0.05$, **$P < 0.01$.
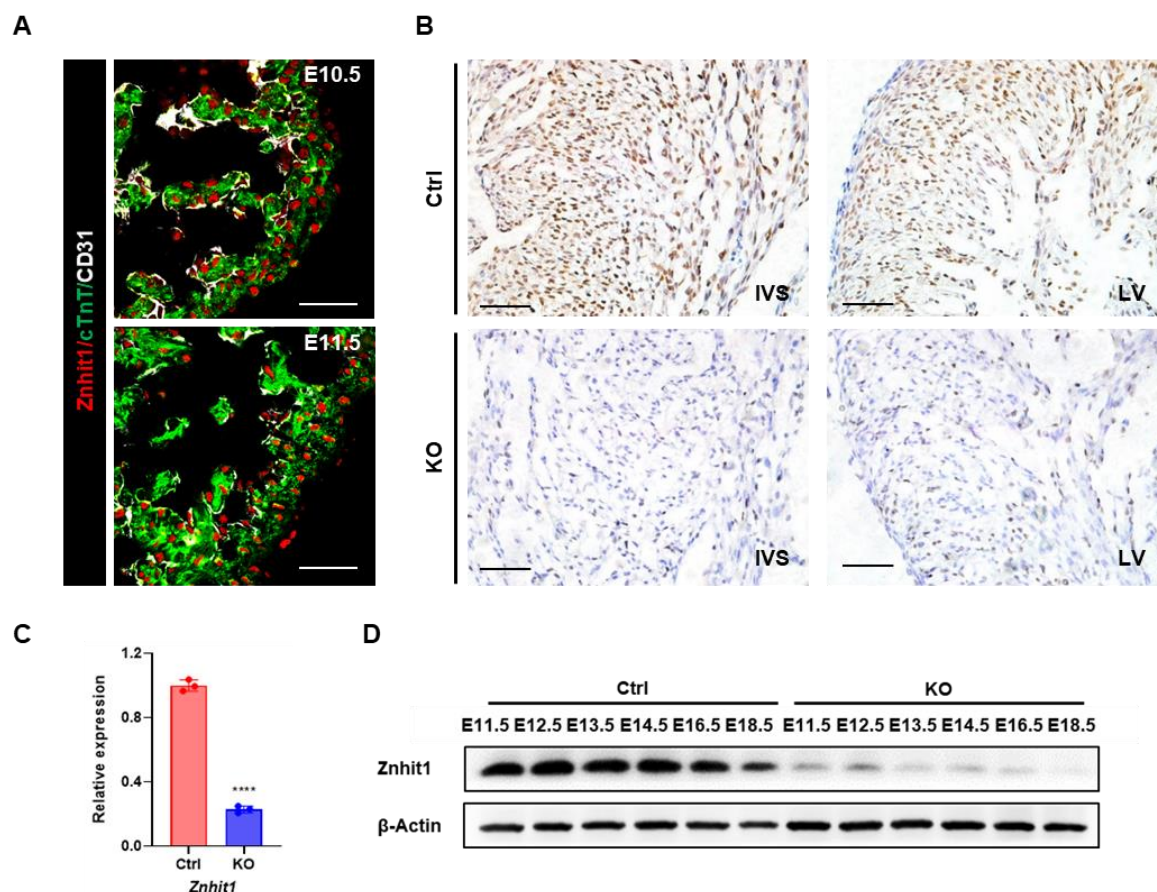
Fig. S3. Verification of *Znhit1* deletion in the heart tissues. (A) Immuno-fluoresence staining. Znhit1 was localized in the nuclei of cardiomyocytes at both E10.5 and E11.5. **(B-D)** Specific knock out *Znhit1* in cardiomyocytes by *Tnnt2*-Cre. **(B)** Immunohistochemical analysis of Znhit1 in the heart. IVS, interventricular septum; LV, left ventricle. Scale bar=50μm. **(C)** qRT-PCR analysis of *Znhit1* expression in the heart at E11.5 with normalization to *Gapdh* levels. Data are mean ± s.e.m. Student's t test: ****$P < 0.0001$. **(D)** Western blot analysis of Znhit1. β-Actin serves as loading control. Ctrl was control heart and KO was *Znhit1* deletion heart (*Tnnt2*-Cre mediated deletion).
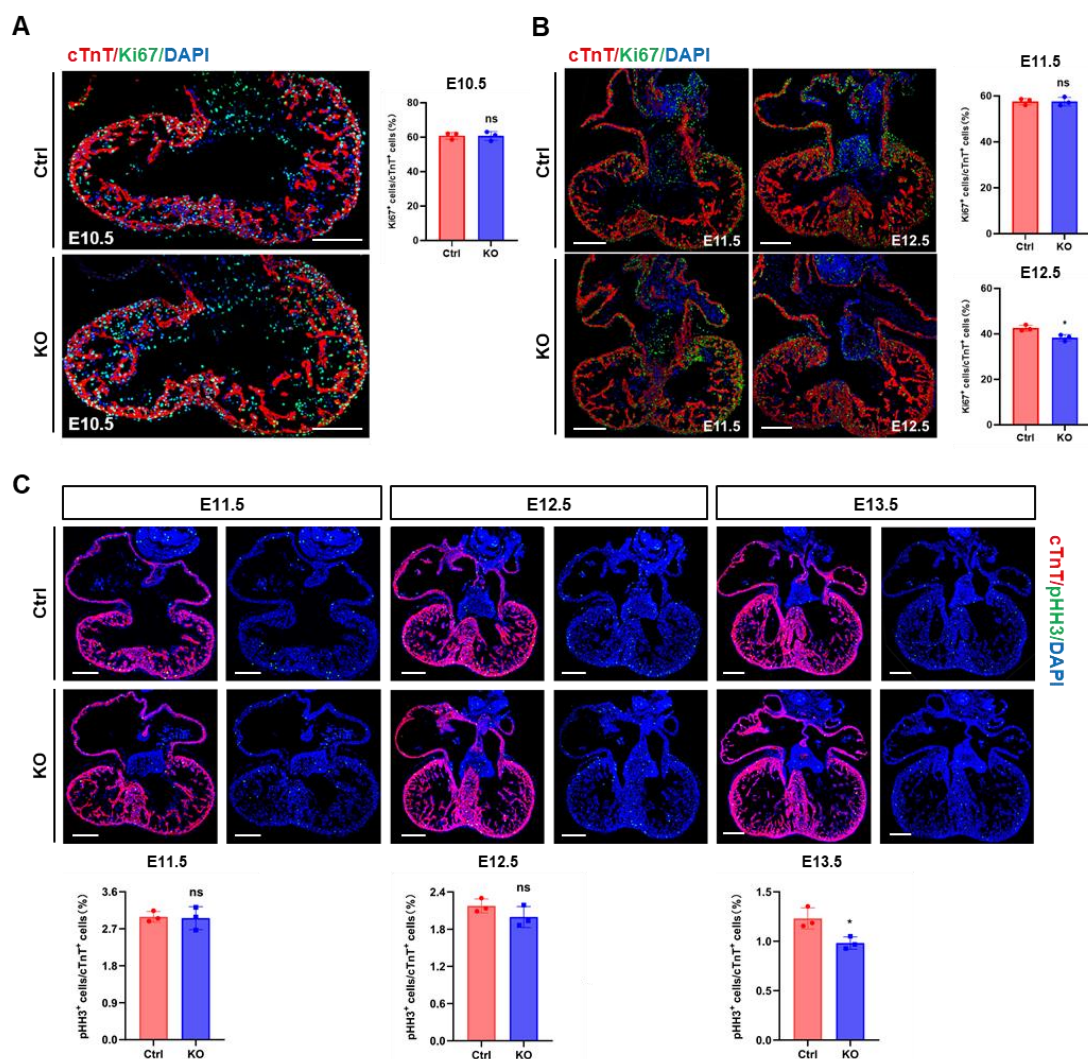
Fig. S4. Decreased cell proliferation of cardiomyocytes in the heart of *Znhit1* deletion mice. Ctrl was control heart and KO was *Znhit1* deletion heart (*Tnnt2*-Cre mediated deletion). **(A** and **B)** Ki67 immunostaining and quantification of proliferating cardiomyocytes (Ki67$^+$cTnT$^+$ cells). **(C)** pHH3 immuno-staining and quantification of proliferating cardiomyocytes (pHH3$^+$cTnT$^+$ cells). Data are mean ± s.e.m. Student's t test: *$P < 0.05$. ns, not significant. Scale bar= 200μm.
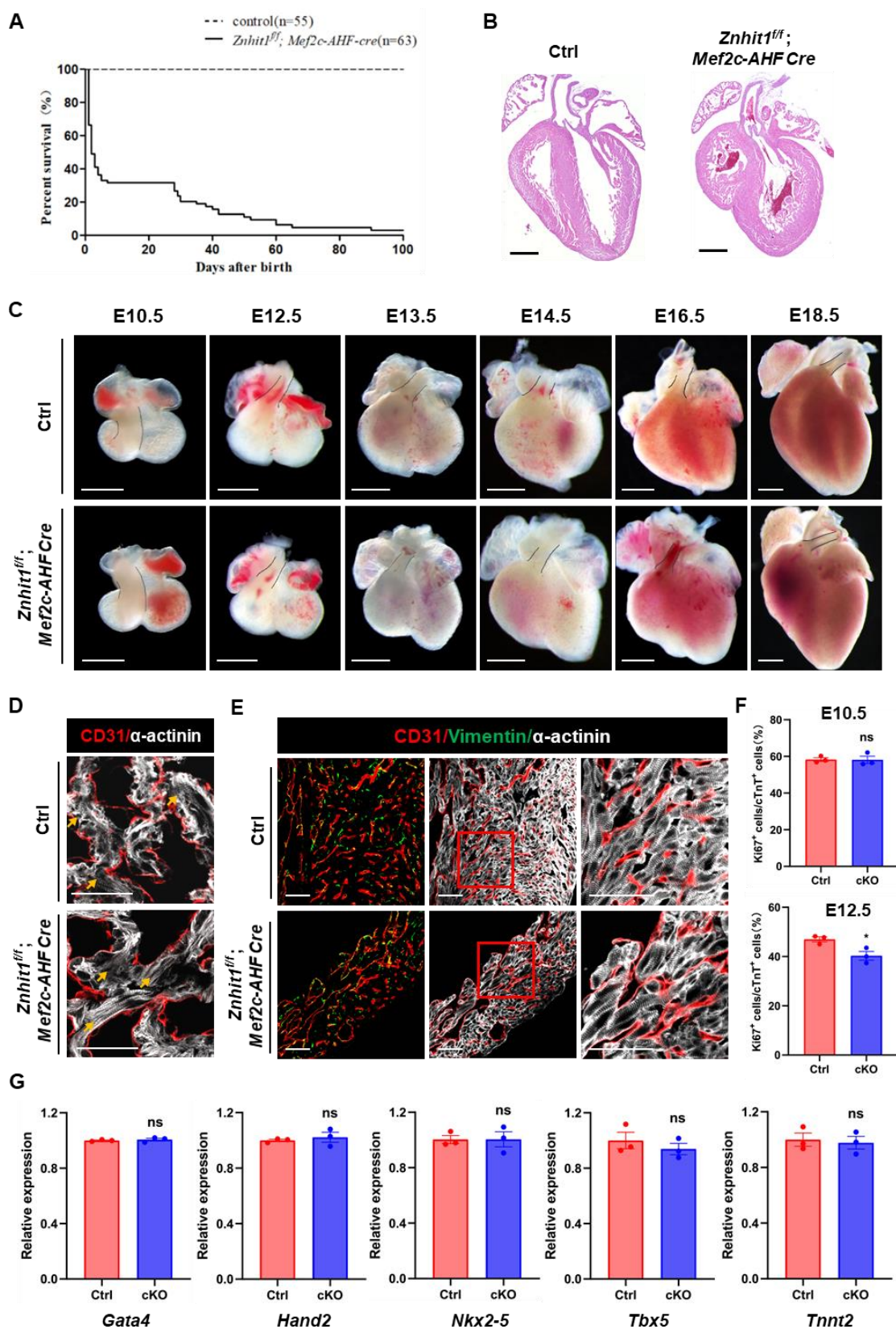
Fig. S5. Disruption of the SRCAP complex did not affect the early development of AHF-progenitor cells. (A) Survival curves. (B) H&E staining of P1 heart sections. Scale bars: 500μm. (C) Gross analysis of the hearts. Black lines depicted the outflow tract or pulmonary artery (PA). Slight PA stenosis was observed in the KO heart from

E13.5. Scale bars: 500µm. **(D** and **E)** Fluorescence staining of the right ventricle at E12.5 **(D)** and E18.5 **(E)** α-actinin (in white) labeled sarcomere, and CD31 (in red) indicated endocardium and coronary endothelial cells. Yellow arrows pointed to the sarcomere in **(D)**. **(E)** The rightmost panels were higher magnification of the boxer area in the adjacent left panels. Scale bars: 50µm. No big difference was found between the mice of the two groups. **(F)** Quantification of proliferating cardiomyocytes. **(G)** Quantitative analysis of mRNA expression level in the right ventricle at E11.5. Data are mean ± s.e.m. Student's t test: *$P < 0.05$. ns, not significant.
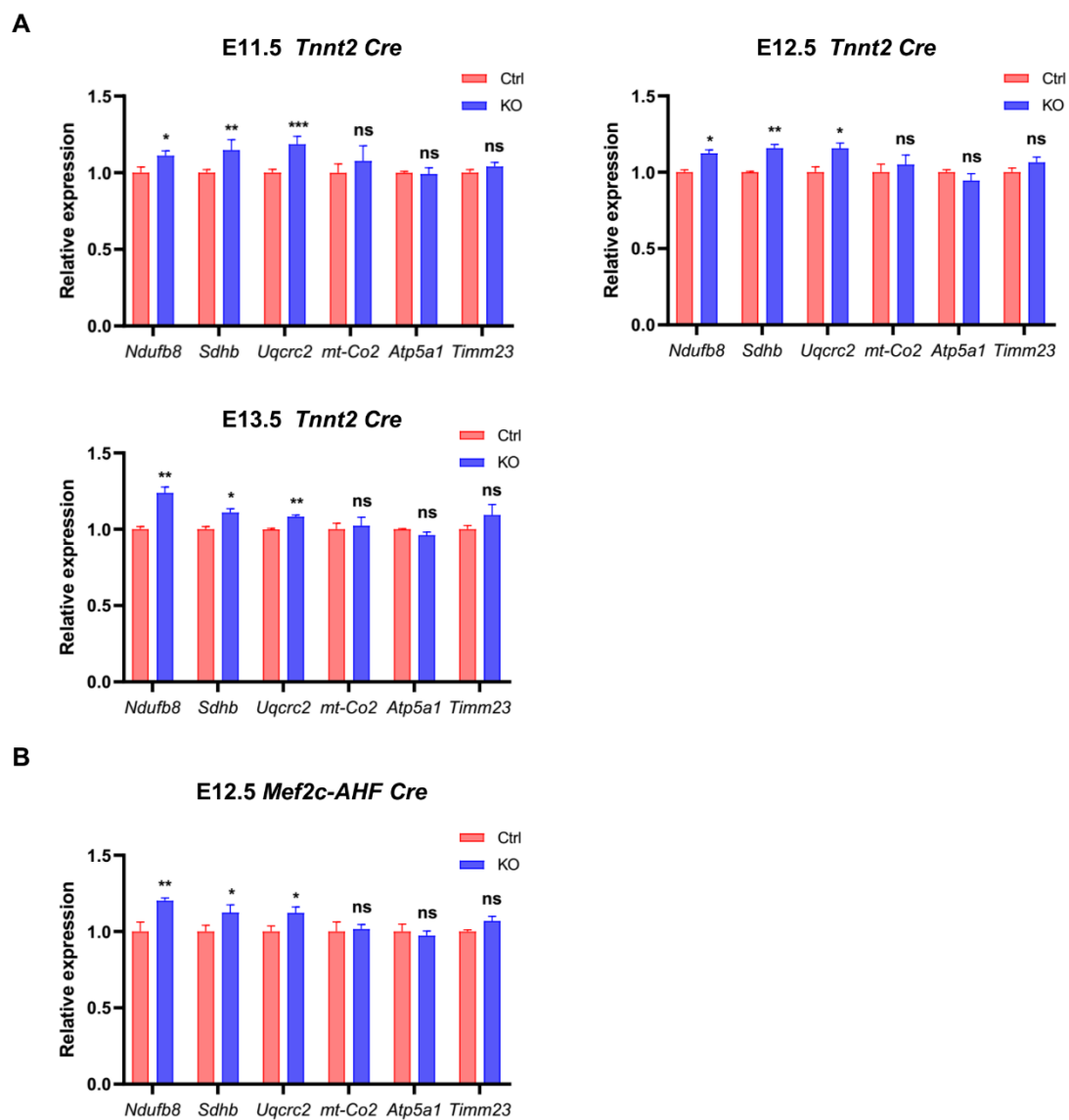
Fig. S6. Analysis of mRNA levels by qRT-PCR in the heart tissues. (A) qRT-PCR analysis of E11.5, E12.5 and E13.5 hearts. **(B)** qRT-PCR analysis of E12.5 right ventricles. Data are mean ± s.e.m. Two-way ANOVA: $*P < 0.05$, $**P < 0.01$, $***P < 0.001$. ns, not significant.
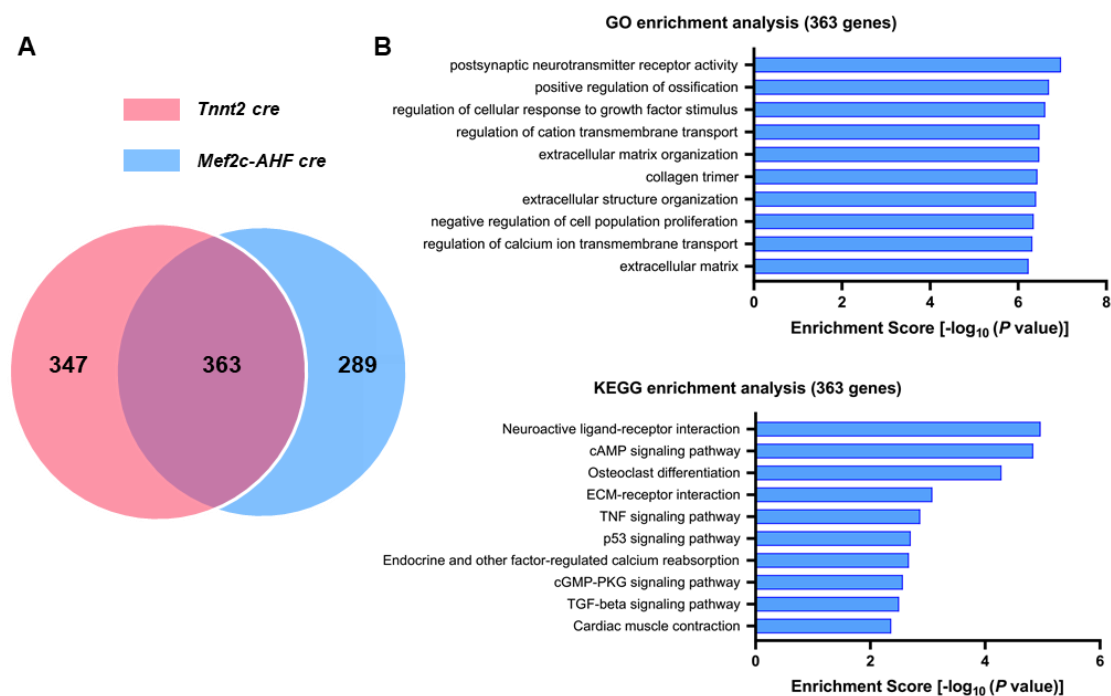
Fig. S7. Analysis of the up-regulated genes in the *Znhit1* mutant mice. (A) Venn diagram depicting the overlapping of the up-regulated genes in *Tnnt2* Cre (in red) and *Mef2c-AHF* Cre (in blue) mediated *Znhit1* deletion mice. **(B)** GO and KEGG enrichment analysis of the 363 overlapping genes in **(A)**.
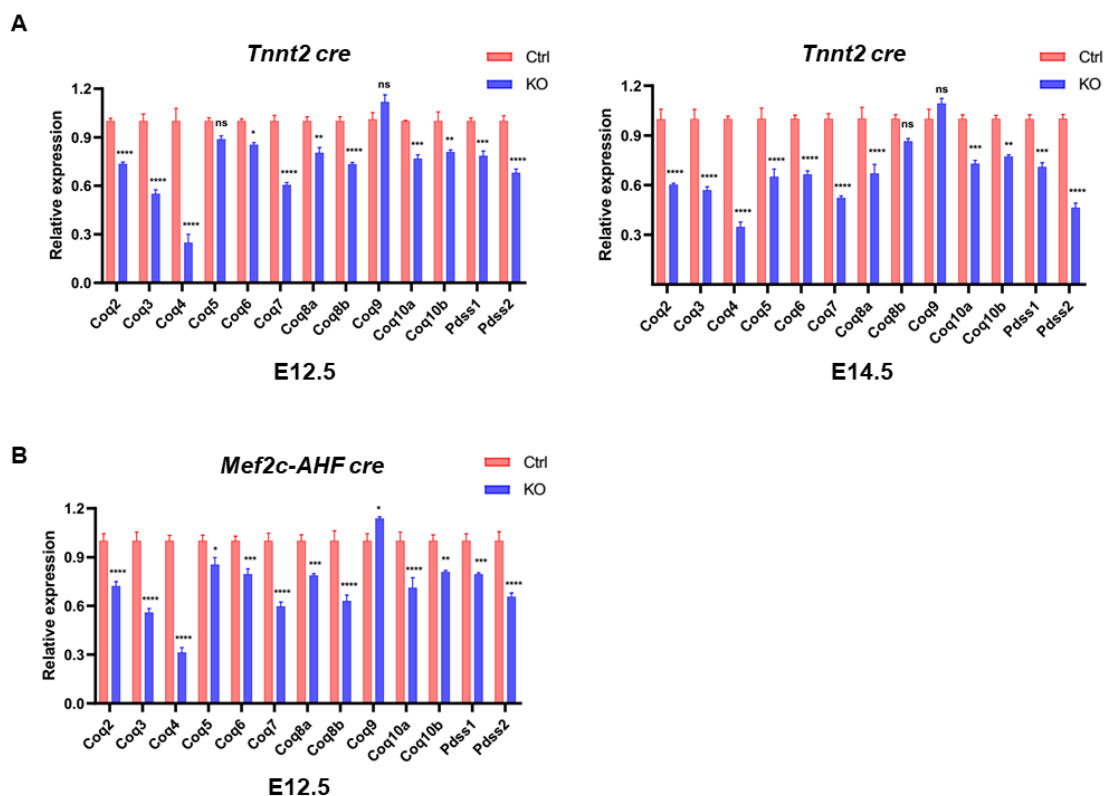
Fig. S8. 12 genes out of the 13 genes required for biosynthesis of CoQ showed markedly reduced expression level in *Znhit1* mutant mice. (A) qRT-PCR analysis of E12.5 and E14.5 hearts. **(B)** qRT-PCR analysis of E12.5 right ventricles. Data are mean ± s.e.m. Two-way ANOVA: *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$, ****$P < 0.0001$. ns, not significant.
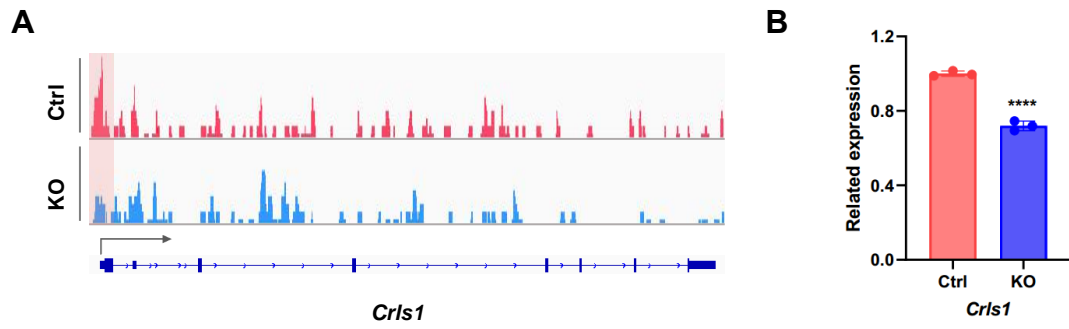
Fig. S9. *Crls1* was a direct regulatory target of the SRCAP complex. (A) CUT & Tag assay of H2A.Z-enriched regions in the heart at E13.5. The peak at the promoter region of *Crls1* was reduced in the KO. (B) qRT-PCR examination of *Crls1* expression level at E13.5. Data are mean ± s.e.m. Student's t test: ****$P < 0.0001$.

Table S1. Primary antibodies

Antibodies	Applications	Dilution	Species	Source	Catalog number
Ndufb8	WB	1:1000	Rabblt	Proteintech	14794-1-AP
SDHB	WB	1:1000	Rabblt	abcam	ab178423
Uqcrc2	WB	1:1000	Rabblt	Proteintech	14742-1-AP
COXII	WB	1:2000	Rabblt	Proteintech	55070-1-AP
COXIV	WB	1:10000	Rabblt	Proteintech	11242-1-AP
Atp5A	WB	1:1000	Rabblt	Proteintech	14676-1-AP
Tim23	WB	1:2000	Mouse	BD	611223
AMPKα	WB	1:1000	Rabblt	CST	#2532
Phospho-AMPKα (Thr172)	WB	1:1000	Rabblt	CST	#2535
β-Actin	WB	1:10000	Mouse	Bioworld	BS6007M
YL-1	WB	1:1000	Rabblt	Proteintech	15143-1-AP
Histone H2A.Z	WB	1:1000	Rabblt	abcam	ab4174
Histone H2A.Z	WB	1:1000	Rabblt	abcam	ab150402
Znhit1	WB	1:1000	Rabblt	abcam	ab238125
ZNHIT1	WB	1:1000	Rabblt	Proteintech	16595-1-AP
Coq3	WB	1:1000	Rabblt	Proteintech	28051-1-AP
Coq4	WB	1:1000	Rabblt	Proteintech	16654-1-AP
Ogdhl	WB	1:1000	Rabblt	Proteintech	17110-1-AP
Echs1	WB	1:1000	Rabblt	Proteintech	11305-1-AP
Cpt2	WB	1:1000	Rabblt	Proteintech	26555-1-AP
Cardiac Troponin T	IF	1:500	Mouse	Life	MA5-12960
Vimentin	IF	1:200	Rabbit	Santa Cruz	sc-5565
CD31	IF	1:200	Rat	BD	550274
a-Actinin	IF	1:200	Mouse	Sigma	A7811
Ki67	IF	1:2000	Rabbit	abcam	ab15580
Phospho-Histone H3	IF	1:200	Rabbit	CST	#9701

Table S2. Primers for qRT-PCR.

Genes	Primer sequence (5' to 3')	Genes	Primer sequence (5' to 3')
Znhit1	F: CAGACGGCGAGACAAGTTC R: CAAACTGAGGTAGCCTCTTGC	*Cpt2*	F: CAGCACAGCATCGTACCCA R: TCCCAATGCCGTTCTCAAAAT
Gata4	F: CCCTACCCAGCCTACATGG R: ACATATCGAGATTGGGGTGTCT	*Echs1*	F: TTGTGAACTTGCCATGATGTGT R: TGCTCGGGTGAGTCTCTGAG
Hand2	F: GAGAACCCCTACTTCCACGG R: GACAGGGCCATACTGTAGTCG	*Ogdhl*	F: AGCGGAGTCAGCTCCAGTTAT R: GGATCTGGTAGGCCCGGAT
Tbx5	F: ATGGCCGATACAGATGAGGG R: TTCGTGGAACTTCAGCCACAG	*Coq2*	F: ACAAGCCCATAGGAACCTGG R: CTCCACGCATCAGAATAGCTC
Nkx2-5	F: GACAAAGCCGAGACGGATGG R: CTGTCGCTTGCACTTGTAGC	*Coq3*	F: CTCGTGGGGTTCGTCTCCT R: GAGCTGCGTCCCTGAGTAAG
Tnnt2	F: TCTTCTGGTGCTACTCGAAGC R: CTCCATCGGGGATCTTGGGT	*Coq4*	F: TGTACCCGGACCACATCCC R: AACCATGTCGTGGCGATAGG
Mef2c	F: ATCCCGATGCAGACGATTCAG R: AACAGCACACAATCTTTGCCT	*Coq5*	F: CCCAGGTGCTGCGTTCTATG R: GTCTCAAACCCGAAGTGCG
Acta2	F: GTCCCAGACATCAGGGAGTAA R: TCGGATACTTCAGCGTCAGGA	*Coq6*	F: CTCAGCAGTTTTGGTGCATGG R: TGTCCCTGTCGAACATTATCAAG
Ndufb8	F: TGTTGCCGGGGTCATATCCTA R: AGCATCGGGTAGTCGCCATA	*Coq7*	F: CTCATCATCAGGTGTCACAGTTC R: GGTTTGCTCCATATTCACCAGC
Sdhb	F: AATTTGCCATTTACCGATGGGA R: AGCATCCAACACCATAGGTCC	*Coq8a*	F: GCAGAGCGCATTGTGAGTACA R: GCCAGGTGAGGGTTGATGAAG
Uqcrc2	F: AAAGTTGCCCCGAAGGTTAAA R: GAGCATAGTTTTCCAGAGAAGCA	*Coq8b*	F: GAGAGGATCGTGCAGACCTTA R: TAAAGTCGGCACTCTGTCGGA
mt-Co2	F: AACCATAGGGCACCAATGATAC R: GGATGGCATCAGTTTTAAGTCC	*Coq9*	F: GTGGGGTTCCGGTCTTCAG R: GGGGTGGACGGGAAAACTC
Atp5a1	F: TCTCCATGCCTCTAACACTCG R: CCAGGTCAACAGACGTGTCAG	*Coq10a*	F: CCAACGTCCAGGAGTACCG R: GGTGGAAACCCAACCTCCAAT
Timm23	F: GAAGGTGGCGGAAGAAGTAGC R: GGGGGTTCATACCAGTCAGC	*Coq10b*	F: GGAGACTATTTGGCGTTTTAGCC R: AAGAACAGAGTAGCGAGCTGA
Gapdh	F: AACTTTGGCATTGTGGAAGG R: ACACATTGGGGGTAGGAACA	*Pdss1*	F: ACACCAGCAATGTGCAGTTG R: ACAGACCTTTCAAGTCTCTCCAG
Actb	F: GGCTGTATTCCCCTCCATCG R: CCAGTTGGTAACAATGCCATGT	*Pdss2*	F: CGCTTGTCCGGTTACCTCG R: GGGTAGCCCACGATCTTCTC