Supplementary figure legends

Supplementary Figure 1

DS rats had congestive heart failure. (**A**) DR and DS rat hearts. (**B**) QRT-PCR analysis of *BNP* mRNA expression in DR and DS rat left ventricles (n = 5). (**C**) Plasma norepinephrine (NE) concentration in DR and DS rats (n = 5). (**D**) Tissue NE contents in DR and DS rat left ventricle (n = 5). (**E**) Triple immunofluorescence staining for α-actinin (purple), TH (red), and CHT (green) in DR and DS left ventricles. DS rat ventricles had fewer TH^+ nerves but more CHT^+ nerves than controls (low magnification view, compared with Figure 2). Boxed areas correspond to the high-power photomicrographs in the insets. LA, left atrium; LV, left ventricle. Representative data are shown in each panel. *, $P < 0.01$. Scale bars, 25 μm (**E**, insets); 100 μm (**E**).

Supplementary Figure 2

Cardiac nerve fibers in Dahl salt sensitive rat ventricles have both catecholaminergic and cholinergic activity. (**A**) Confocal images of double immunofluorescence staining for TH (green) and NF (red) in left ventricles at the epicardial sites in DR and DS rats. (**B**) Confocal images of double immunofluorescent staining for CHT (green) and NF (red) in left ventricles at the epicardial sites in DR and DS rats. Arrows indicate CHT^+ nerve fibers in the fasciculated sympathetic nerve bundles. (**C**) Confocal images of double immunofluorescent staining for calcitonin gene-related peptide (CGRP, green) and NF (red) in left ventricles at the epicardial sites in DR and DS rats. (**D**) Quantitative analysis of TH^+, CHT^+ and $CGRP^+$ nerve areas in left ventricles. *, $P < 0.01$; ns, not significant. Scale bars, 20 μm.

Supplementary Figure 3

DS rats have fewer dopamine beta-hydroxylase (DBH)-immunopositive neurons in the left ventricle and stellate ganglia than do DR rats. (**A**) Confocal images of double immunofluorescent staining for DBH (green) and NF (red) in the left ventricular epicardium of DR and DS rats. (**B**) Double immunofluorescent staining with DBH (red) and ChAT (green) of stellate ganglia in DR and DS rats. LV, left ventricle; SG, stellate ganglia. Scale bars, 20 μm (**A**); 100 μm (**B**).

Supplementary Figure 4

Cholinergic transdifferentiation occurs gradually in accordance with the progression of cardiac hypertrophy to heart failure in DS rats. (**A** and **B**) Quantitative analysis of TH^+/NF^+ and CHT^+/NF^+ area ratios in the LV epicardium of 6- (baseline), 11- (hypertrophy phase), 15- (heart failure phase) week-old rats (n = 5). (**C**) Quantitative analysis of $ChAT^+/TH^+$ ratio in stellate ganglia (n = 4). *, $P < 0.01$; **, $P < 0.05$; ns, not significant.

Supplementary Figure 5

Immunofluorescence staining of sympathetic ganglia neurons in DS rats. (**A**) Immunofluorescence staining for TH (red), ChAT (green), and Toto3 (blue) in DR and DS rat lumbar ganglia. (**B**) Quantitative analysis of $ChAT^+/TH^+$ ratio in lumbar ganglia (n = 4). Representative data are shown in each panel. ns: not significant. (**C**) Immunofluorescence staining for TH (red), ChAT (green) and Toto3 (blue) in DR and DS rat superior cervical ganglia (SCG), upper thoracic ganglia (TG) and lower TG. (**D**) Quantitative analysis of

$ChAT^+/TH^+$ ratio in SCG, upper TG and lower TG (n = 4). *, $P < 0.01$; ns, not significant. Scale bars, 100 μm.

Supplementary Figure 6

Anterograde labeling of cardiac sympathetic nerves with BDA. (**A**) Injected BDA is detected in the cardiac plexus and sympathetic nerve fibers in the left ventricle. (**B**) Representative transmission electron micrograph of nerve ending in DS rat ventricle that contains both SGV and SAGV. Black arrows indicate SGV (involving catecholamine) and white arrows indicate SAGV (involving acetylcholine). (**C**) Quantitative analysis of the (SGV)/(SGV+SAGV) ratio (%) in DR and DS rats. (**D**) Immunoelectron microphotographs of ventricles using gold particles labeled with anti-TH antibody and AF64A injection in DS rats. Note there are various patterns of damage in AF64A-treated DS rat sympathetic nerve endings. Red arrows indicate gold particles labeled with anti-TH antibody. Arrowhead indicates clumping of organelles. Asterisks indicate vacuolization. Star indicates degeneration of neurofilaments. epi, epicardium. Representative data are shown in each panel. Scale bars, 0.5 μm (**B** and **C**); 50 μm (**A**).

Supplementary Figure 7

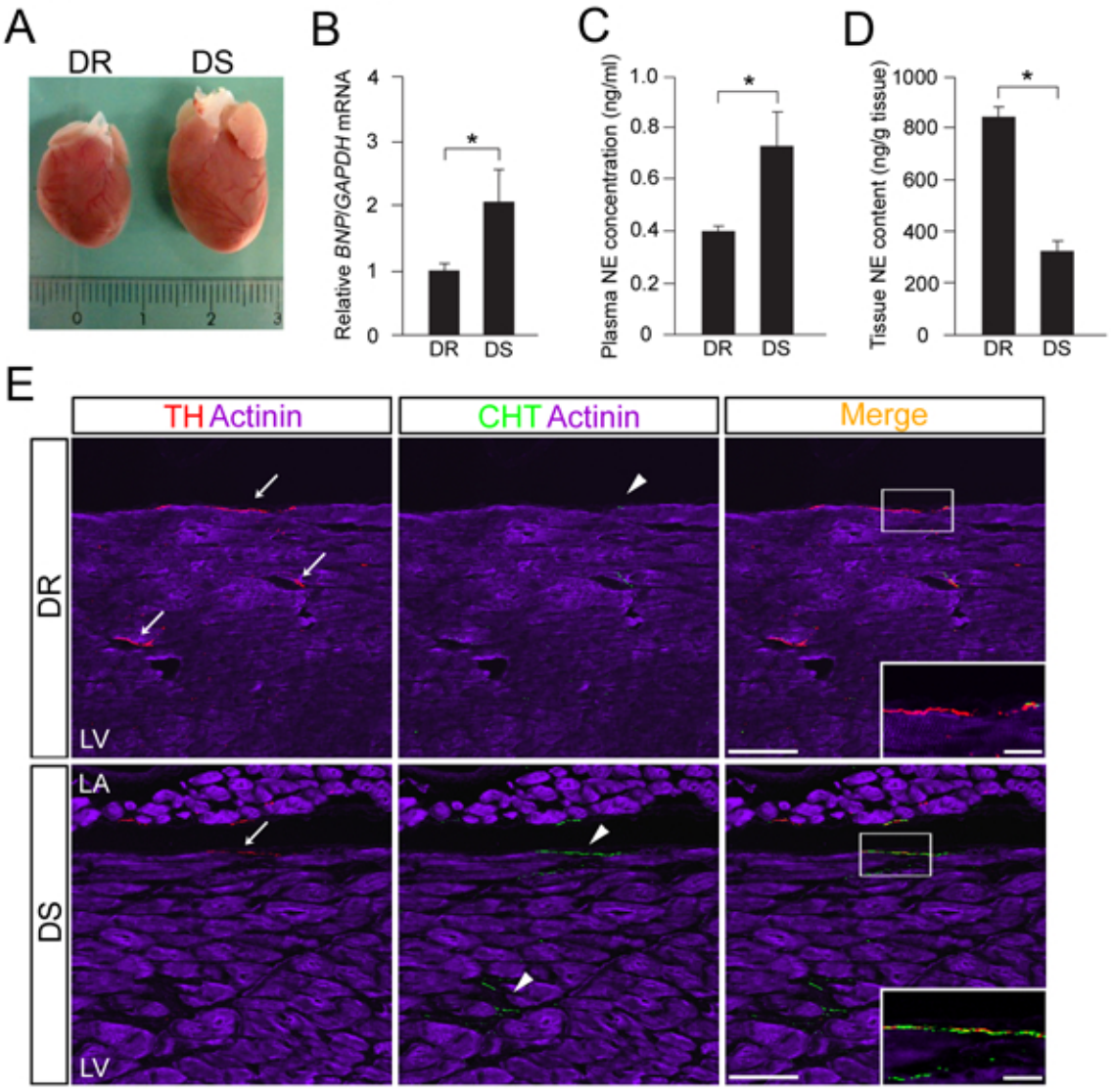
Evaluation of the mouse model of congestive heart failure. (**A–C**) Results from the hypoxia-induced pulmonary hypertension (PH) mouse model. (**A**) Representative transverse sections of the hearts in short axial view for control and PH mice at 8 weeks of age. (**B**) Right ventricular systolic pressure (RVSP) of the control and PH mice (n = 5). (**C**) Quantitative

RT-PCR analysis of BNP, LIF and CT-1 expression in the RV at 8 weeks of age (n = 5). (**D-F**) Transaortic constriction (TAC)-induced LV failure model mice. (**D**) Reduced fractional shortening (FS) in TAC mice at 4 weeks of age. (**E**) Measurement of the left ventricular end-diastolic diameter (LVEDD), LV end-systolic diameter (LVESD), and posterior wall thickness (PW) of TAC mice from echocardiograms. (**F**) Quantitative RT-PCR analysis of BNP, LIF and CT-1 expression in the LV of TAC mice at 4 weeks (n = 5). (G) Pulse wave Doppler echo of the control and the constricted flow distal to TAC. (H)The bar graph indicated the pressure gradient caused by TAC. *, $P < 0.01$; **, $P < 0.05$; ns, not significant. Scale bars, 1 mm (**A**).

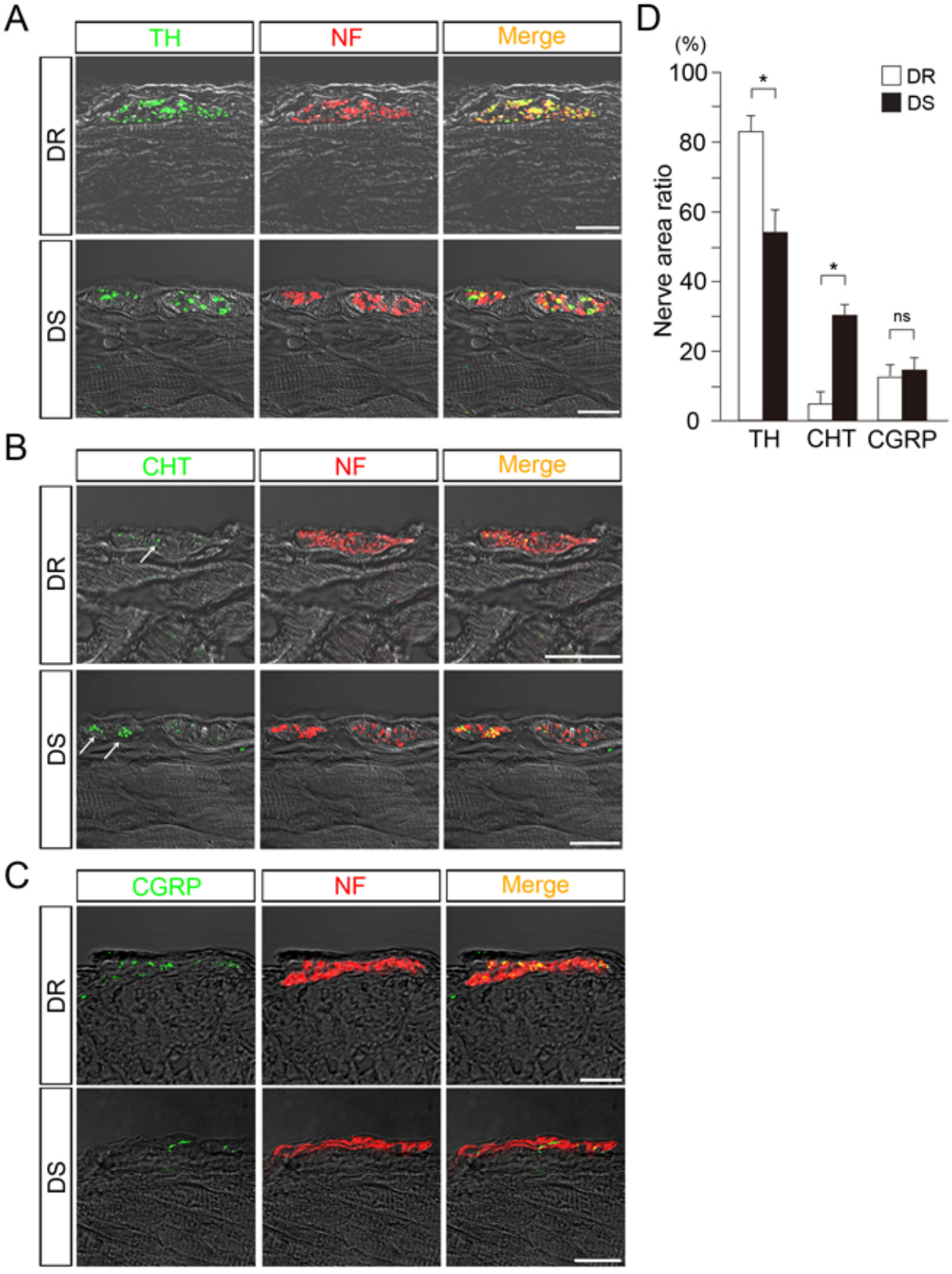
Supplementary Figure 8.

Measurement of the baseline cardiac hemodynamics and heart weight/body weight ratio in *gp130* $^{flox/flox}$ and *gp130* ^{DBHCre} mice. HW/BW: (heart weight)/(body weight) ratio, ns, not significant.

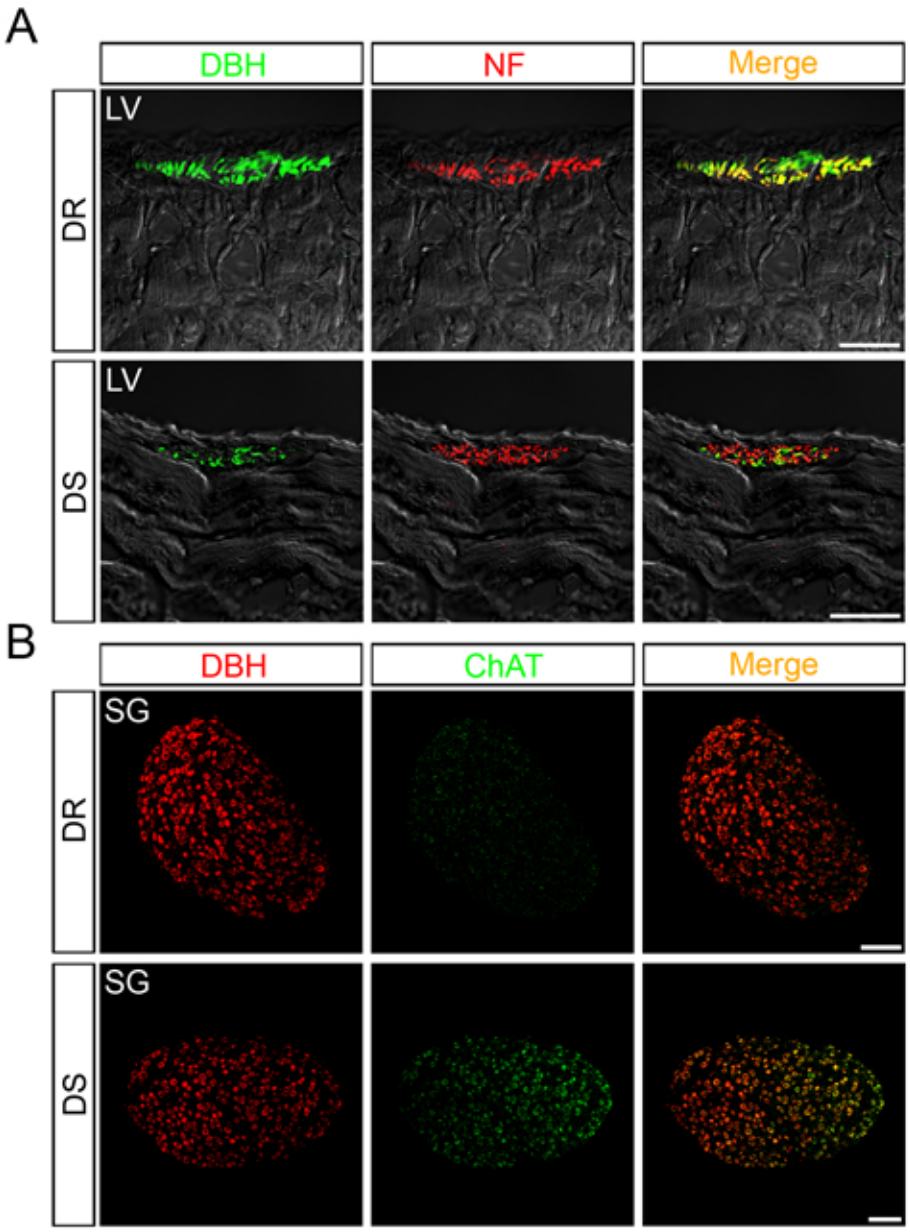
Supplementary Figure 1



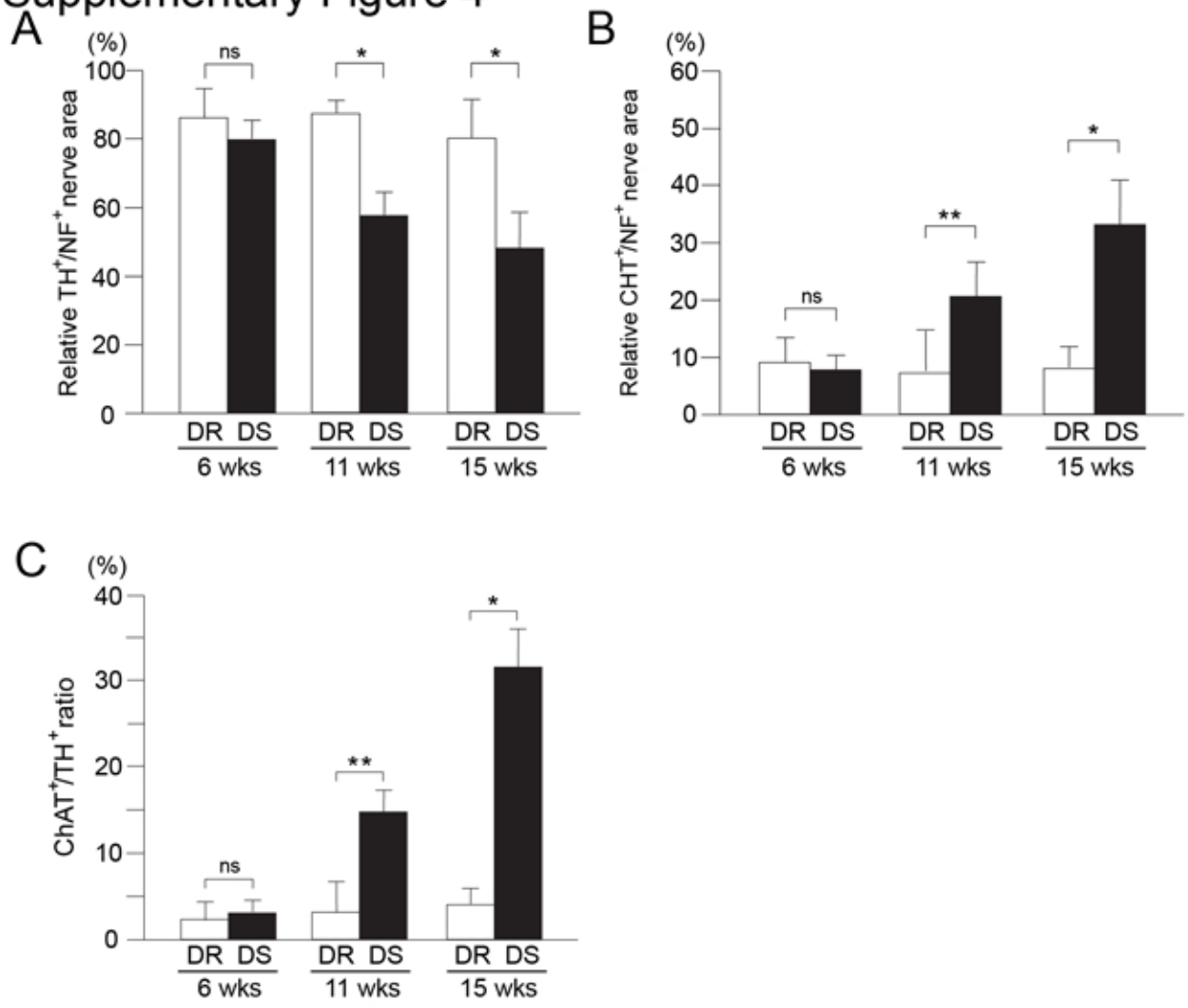
Supplementary Figure 2



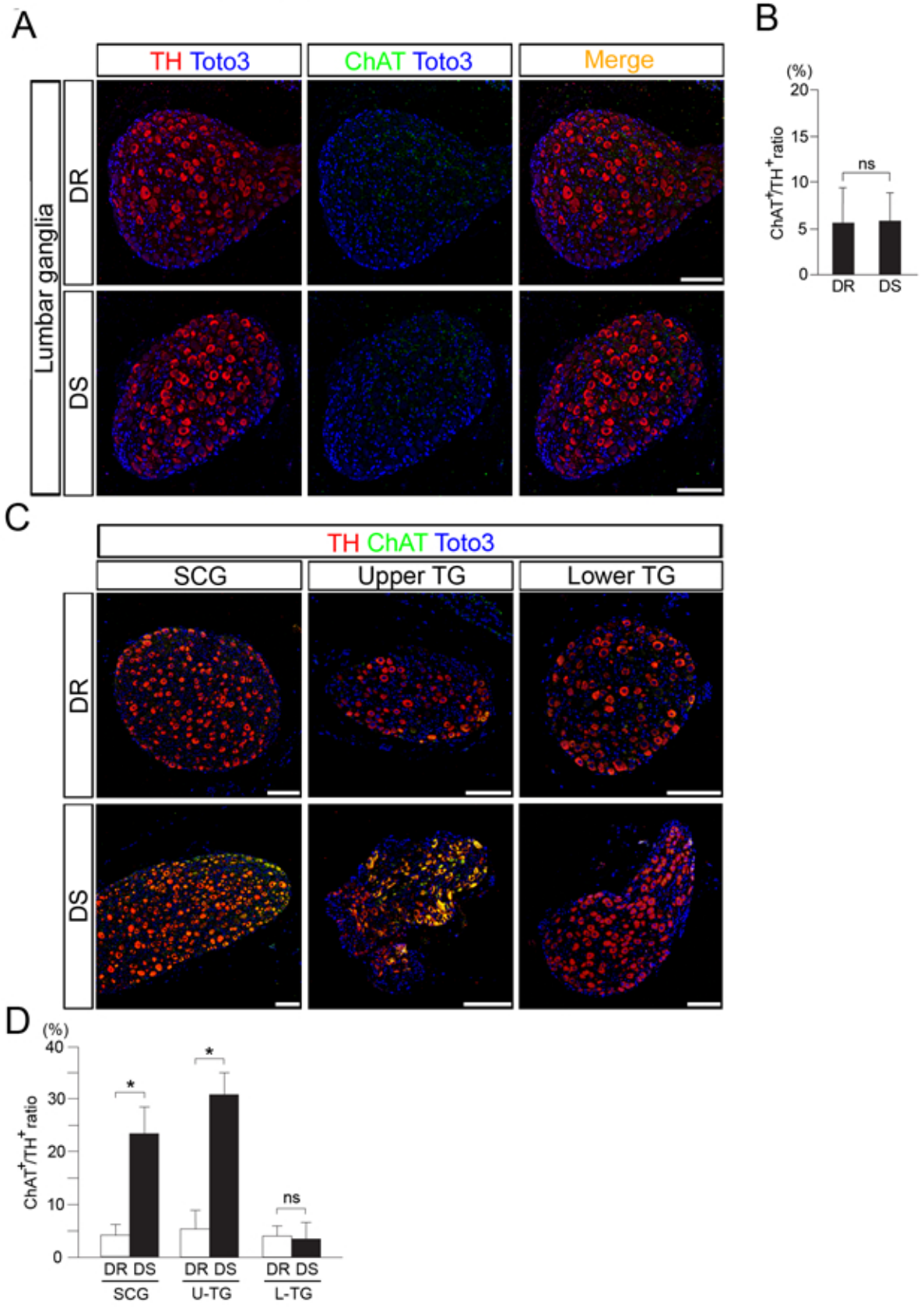
Supplementary Figure 3



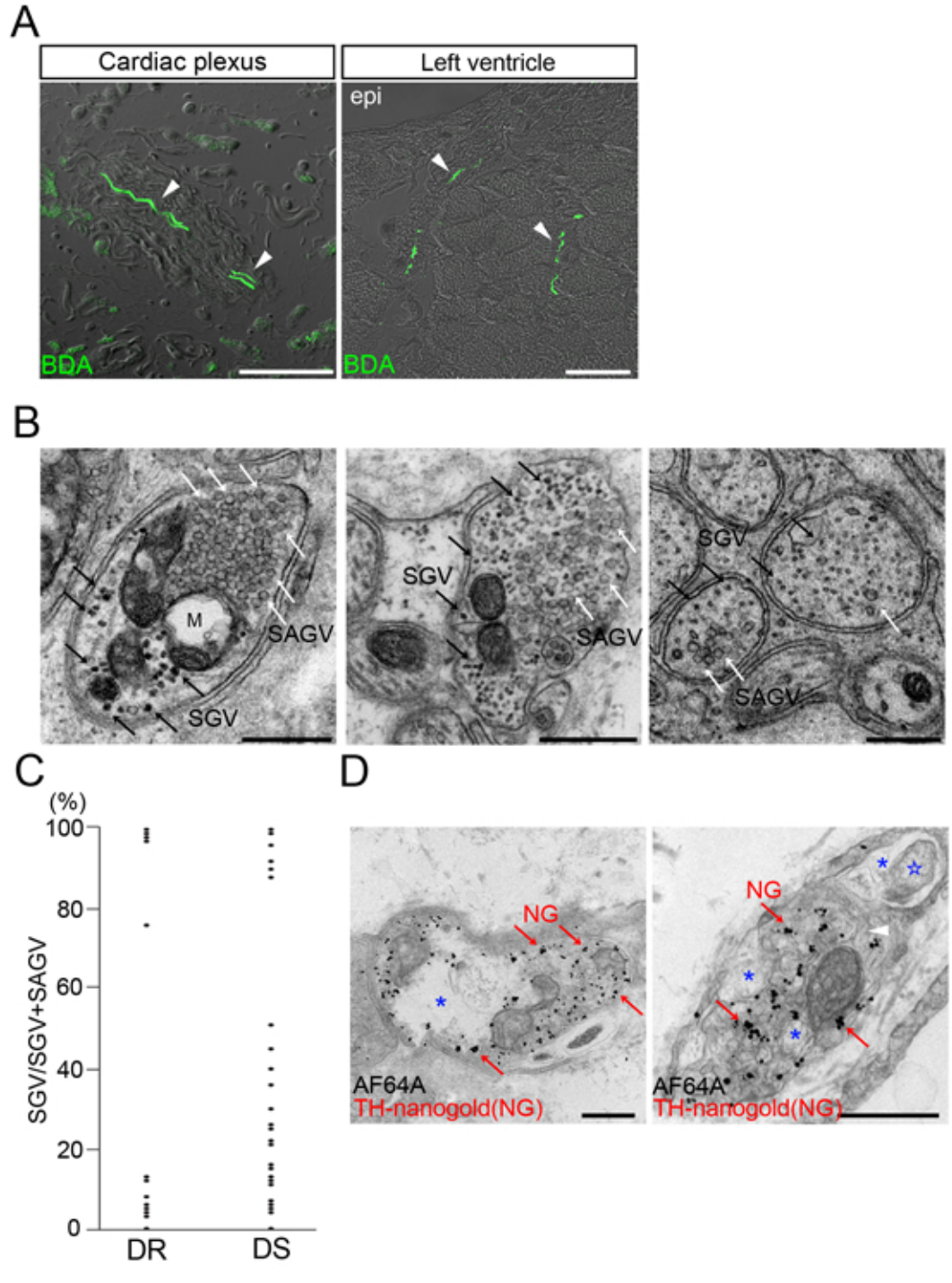
Supplementary Figure 4



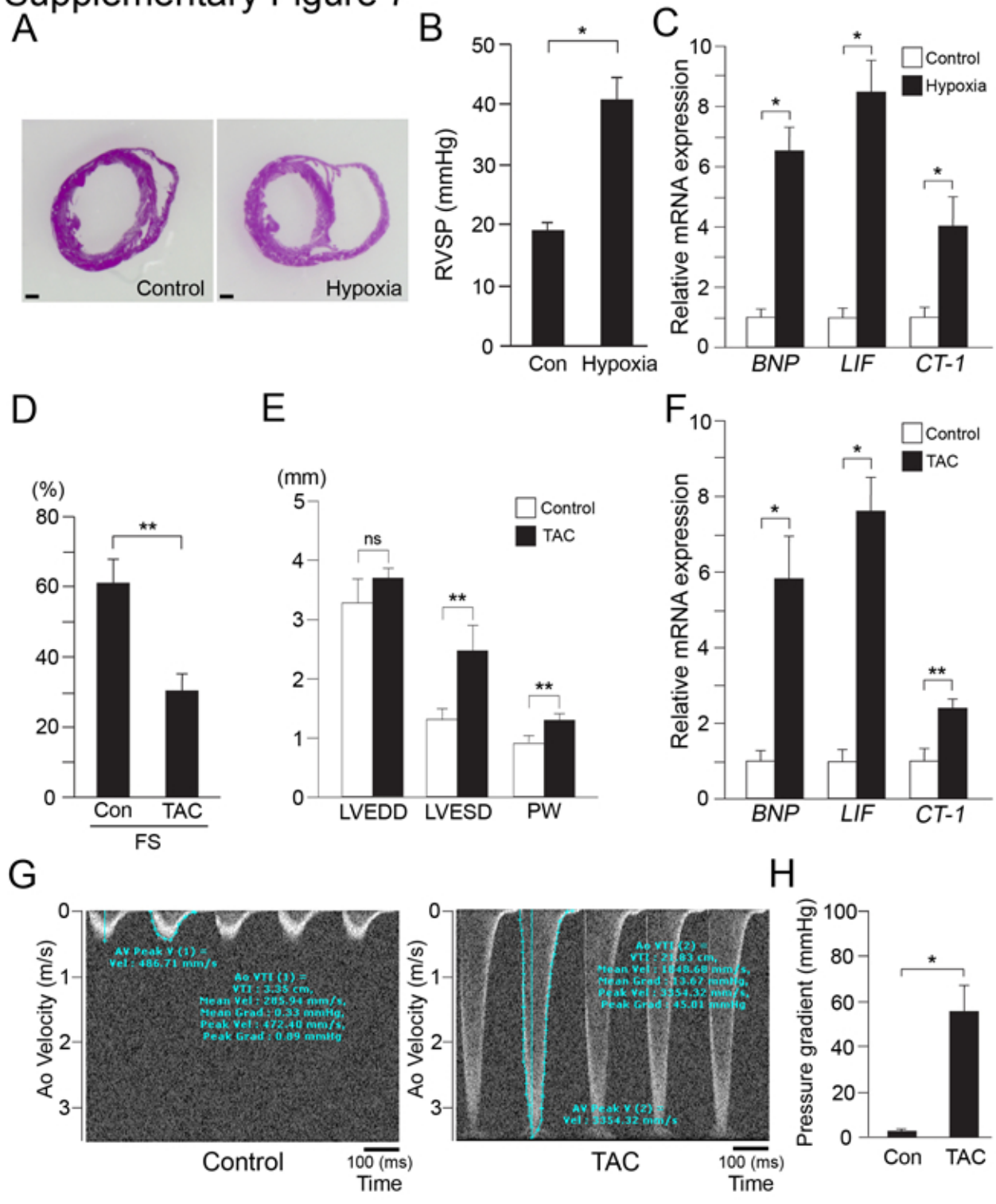
Supplementary Figure 5



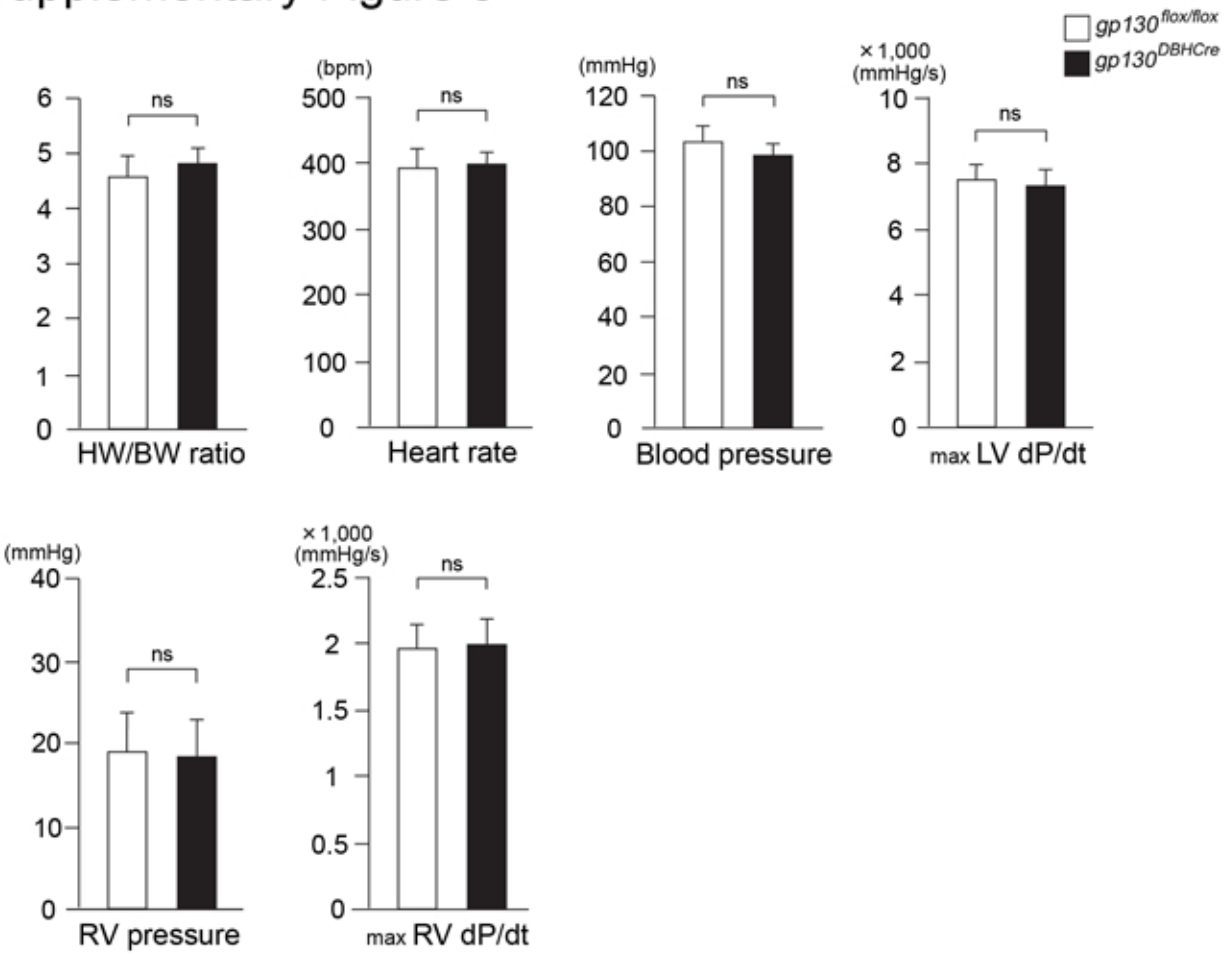
Supplementary Figure 6



Supplementary Figure 7



Supplementary Figure 8



Supplementary Table 1

Time course of changes in hemodynamics, body and heart weight, and detection of pleural effusion in DR and DS rats

	6 Weeks		11 Weeks		15 Weeks	
	DR	DS	DR	DS	DR	DS
SBP (mmHg)	94±1.7	101±5.6**	114±8.3*	202±14.8*†	128±6.7*	204±15.2*
HR (bpm)	400±15	411±25	405±30	428±41	410±35	421±32
BW (g)	155±3.6	168±3.5**	314±16†	311±38†	384±8.2‡	302±21*
HW (g)	0.65±0.01	0.75±0.05**	1.19±0.11†	1.78±0.25*†	1.25±0.05	1.69±0.08*
HW/BW (mg/g)	4.2±0.05	4.4±0.04	3.8±0.08	5.7±0.07*†	3.2±0.08‡‡	5.6±0.1*
Pleural effusion	−	−	−	−	−	++

Values are means ± SEM. SBP; systolic blood pressure, HR; heart rate, BW; body weight, HW; heart weight. *, $P < 0.01$ vs. age-matched DR rats. **, $P < 0.05$ vs. age-matched DR rats. †, $P < 0.01$ vs. 6-week-old rats of the same strain. ††, $P < 0.05$ vs. 6-week-old rats of the same strain. ‡, $P < 0.01$ vs. 11-week-old rats of the same strain. ‡‡, $P < 0.05$ vs. week-old rats of the same strain.

Supplementary Table 2

Time course of the echocardiographic findings in DR and DS rats

	6 Weeks		11 Weeks		15 Weeks	
	DR	DS	DR	DS	DR	DS
LVEDD (mm)	4.9±0.3	5.0±0.3	7.1±0.2†	6.9±0.2†	7.4±0.3	8.6±0.2*‡
LVESD (mm)	2.6±0.3	2.5±0.7	4.1±0.2†	3.2±0.1*†	4.4±0.2‡	5.9±0.2*‡
FS (%)	46±2.6	49±3.0	42±1.2††	54±0.7*††	39±1.8‡‡	32±1.0*‡
PWT (mm)	1.5±0.03	1.4±0.04*	1.4±0.03†	2.1±0.1*†	1.6±0.09‡	1.46±0.08**‡

Values are means ± SEM. LVEDD; left ventricular (LV) end-diastolic diameter, LVESD; end-systolic diameter, FS; fractional shortening, PWT; posterior wall thickness. *, $P < 0.01$ vs. age-matched DR rats. **, $P < 0.05$ vs. age-matched DR rats. †, $P < 0.01$ vs. 6-week-old rats of the same strain. ††, $P < 0.05$ vs. 6-week-old rats of the same strain. ‡, $P < 0.01$ vs. 11-week-old rats of the same strain. ‡‡, $P < 0.05$ vs. week-old rats of the same strain.

Supplementary Table 3

Measurements of hemodynamics, and body and heart weight, in WT, *αMHC-Cre*, *LIF-loxP*, and LIF transgenic mice (*Line 3 and Line 10*)

	WT	*αMHC-Cre*	*LIF-loxP*	*Line 3*	*Line 10*
SBP (mmHg)	109±10.8	113±11.4	111±8.3	118±10.2	121±5.2
HR (bpm)	420±12	416±15	425±10	430±15	423±13
BW (g)	12.9±1.1	12.5±0.9	12.7±0.7	12.8±1.8	9.2±1.1*††‡¶
HW (g)	0.059±0.004	0.058±0.004	0.059±0.005	0.087±0.01*‡¶	0.078±0.003*‡¶
HW/BW (mg/g)	4.55±0.28	4.65±0.12	4.59±0.18	6.87±1.47**‡‡¶¶	8.52±0.71*‡¶

Values are means ± SEM. SBP; systolic blood pressure, HR; heart rate, BW; body weight, HW; heart weight. *, $P < 0.01$ vs. age-matched *αMHC-Cre* mice. **, $P < 0.05$ vs. age-matched *αMHC-Cre* mice. ††, $P < 0.05$ vs. age-matched Line 3 mice. ‡, $P < 0.01$ vs. age-matched *LIF-loxP* mice. ‡‡, $P < 0.05$ vs. age-matched *LIF-loxP* mice. ¶, $P < 0.01$ vs. age-matched WT mice. ¶¶, $P < 0.05$ vs. age-matched WT mice.