

SUPPLEMENT MATERIAL

METHODS

Echocardiography and Hemodynamics Methods

Echocardiography (Vevo-770, Visualsonics Inc.) and invasive aortic micromanometry (SPR-671; Millar Instruments, Inc.) were performed at baseline to ensure there were no significant strain-related differences in hemodynamics and left ventricular function under anesthesia (n=8 for each group). A calibrated 1.4 French catheter-tip micromanometer (SPR-671; Millar Instruments, Inc.) was inserted into the carotid artery and advanced retrograde into the aorta. Pressure tracings were recorded at a sampling rate of 4,000 Hz (PowerLab 8SP; ADInstruments) to measure systolic and diastolic blood pressure (SBP, DBP). Echocardiography was performed in the parasternal long-axis and mid-ventricular short-axis planes using fundamental imaging at 40 MHz. Image sets were obtained by ECG gating of sequential M-mode lines for a post-acquisition 2-D frame rate of 1,000-1,100 Hz. Peak systolic (S') and early diastolic (E') radial velocities in the short-axis were measured by tissue Doppler imaging with a sample volume placed in the posterior endocardium. Velocity of circumferential fiber shortening (VcF) was measured by dividing fractional shortening by the ejection time.

Myocardial Blood Volume

For a blood pool agent, relative myocardial blood volume can be calculated by the ratio of signal intensity in the myocardium (A-value) during steady state to that in the blood pool (I_B), or A/I_B . To convert myocardial mass to volume, it is necessary to divide by the myocardial mass 1.06 g/cm^3 , or $A/(1.06 \times I_B)$. Since the (I_B) value was obtained from the LV cavity using a 10-fold lower infusion rate to avoid dynamic range saturation, and the sternum produces a 91%

reduction (or 1/1.1) in contrast signal intensity compared to limb skeletal muscle, MBV was calculated as $A/(1.06 \times I_B \times 10 \times 1.1)$.

SUPPLEMENT TABLES

Table I. Hemodynamic and Echocardiographic Data

	Wild-type (n=8)	db/db (n=8)
Heart Rate (min^{-1})	481±59	465±70
Systolic BP (mm Hg)	98±5	91±12
Thickening Fraction	0.50±0.11	0.48±0.15
Vcf (mm/s)	0.70±0.13	0.70±0.17
S' (mm Hg/s)	16.7±4.7	15.2±4.7
E' (mm/s)	13.5±4.8	13.6±3.9

Vcf, velocity of circumferential fiber shortening; S' and E', peak early systolic and diastolic endocardial velocity.

Table II. Parametric Contrast-enhanced Ultrasound Perfusion Imaging Data*

	Wild-type				db/db			
	Baseline	Day 1	Day 3	Day 21	Baseline	Day 1	Day 3	Day 21
Microvascular blood volume (ml/g)	6.0±1.3	2.2±1.2	2.8±1.9	4.0±1.7	4.1±1.1†	1.9±1.6	1.5±0.7	1.4±0.8†
Microvascular flux rate (s^{-1})	0.25±0.02	0.06±0.03	0.11±0.06	0.14±0.06	0.16±0.04†	0.07±0.06	0.06±0.02	0.08±0.04†

*p<0.05 for all post-ischemic values versus baseline values; † p<0.05 vs wild-type at the same time interval.

On-line Supplement on Number of Animals for each observation (Figures 1 and 3).**Table III Figure Number of animals for each data point for Figure 1A**

	Baseline	Day 1	Day 3	Day 21
Wild-type	5	12	8	12
db/db	8	12	8	8

Table IV Number of animals for each data point for Figure 3A

	Baseline	Day 1	Day 3
Wild-type	5	8	8
db/db	7	8	8
db/db + MCP-1	-	5	5