Analysis of Cardiac Function and Morphology in Male and Female GLUT4/Acrp30 Transgenic Mice

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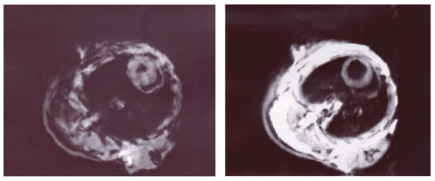
Introduction

Reduced expression of GLUT4 and Acrp30 (adiponectin) has been associated with obesity and type 2 diabetes, common endocrine diseases of aging that are nearly worldwide epidemics, especially in the United States, where obesity is becoming more prevalent (1,2,3). Diabeticss and obese patients are at risk (1,2,4) for developing cardiovascular disease (CVD). GLUT4 is a glucose transporter expressed in insulin sensitive tissues, including adipocytes and skeletal muscle (5-7). GLUT4 expression is reduced in adipose tissue early in the development of obesity and type 2 diabetes (6, 8-10). Acrp30 is synthesized and secreted by adipocytes and is correlated with maintaining glucose levels and insulin sensitivity (12-16). In obese and diabetic mice, the mRNA expression and plasma levels of Acrp30 were shown to be reduced (11,13). Mice with a partial lack of GLUT4 and/or Acrp30 might have altered heart chamber dimensions or cardiac morphology as a consequence of the CVD phenotype. In addition, only males might exhibit these changes because females may compensate with estrogen. NMR microimaging is used for the analysis of cardiac function and morphology in GLUT4/Acrp30 genetically engineered mice.

Methods and Results

For the magnetic resonance imaging (MRI) studies each mouse was anesthetized with Isoflorane inhalation anesthesia. Once anesthetized, a set of standard electrocardiographic (ECG) leads was attached to its limbs. The ECG signal was fed to a Gould ECG amplifier. The mouse was wrapped in a small blanket and positioned head up in a home built 35 mm (inner diameter) MRI coil in a GE OMEGA 400 MHz NMR instrument. Gating of the MRI data acquisition was accomplished by using the rising phase of the QRS complex to trigger a standard 5-V square-wave gating signal. This signal was fed from the Gould ECG amplifier to the cardiac gating box associated with the spectrometer. After locating the heart a series of transverse (short axis) images were acquired from the base to the apex of the heart. Image data were transferred to a PC for offline processing and analysis using home designed MRI analysis software running in MATLAB. Heart measurements include the left ventricular inner chamber diameter (LVID) and left ventricle wall thickness (LV wall). LVID represents an average of the septal-lateral and anterior-posterior diameters. For each mouse, these measurements are repeated seven times and an average value for these parameters is obtained for each slice. On average, a 10% uncertainty is obtained. A significantly smaller left ventricular inner diameter was found in male mice lacking adiponectin. Female mice were protected from this change. Female mice exhibit significanlty smaller wall thicknesses compared to males regardless of genotype. Female mice lacking adiponectin have the thinnest walls.



SYSTOLE DIASTOLE

Fig.1. Ejection fraction and fractional shortening (FS) are calculated by comparing systolic and diastolic chamber dimension.

Conclusions and Future work

Male GLUT4 +/+ ACRP30 nulls showed statistically significant ($p = 0.01$) smaller LVID with respect to WT using a two-sample, two-tail t-test/ANOVA. That the females did not show a decrease in LVID, lends credence to the idea that estrogen may impart protection from CVD in mice with complete KO of ACRP30 but with normal GLUT4 levels. Statistical comparisons were only made to WT, within a sex. No significant differences in LVID between sexes for any given genotype were found. KO of ACRP30 is the only correlate to a decrease in LVID and exclusively so in males.

Past findings in male mice have indicated cardio pathology in going from GLUT 4 +/+ to prediabetic, GLUT4 +/- N/H (normal glucose/high insulin), and to GLUT4 +/- H/H, where %FS goes to, respectively, 73±2, 64±3, and 47±4. We are measuring %FS for the genotypes in Table 1. We are looking at a group of OVX female mice and a group of OVX shams to see if GLUT4 +/+ ACRP30 null estrogen deficient females develop smaller LVID. It would be useful to correlate the MRI findings with ACRP30, GLUT4 and insulin levels and serum metabolite profiles.

GROUP	N M/F	Glut4	Acrp 30	Average ± SEM LVID (mm)		GROUP AVG AGE (weeks)		GROUP AVG BODY MASS (Grams)	
				Male	***Female***	*Male*	*Female*	*Male*	*Female*
1	3/3	+/+	+/+	**3.28 ± 0.09**	**2.96 ± 0.13**	81.1	81.1	42.1	36.3
2	3/2	+/+	+/-	**3.00 ± 0.23**	**2.97 ± 0.38**	81.3	78.3	44.5	38.3
3	3/3	+/+	-/-	**2.84 ± 0.06***	**3.10 ± 0.35**	79.2	79.5	54	43.8
4	5/3	+/-	+/+	**3.18 ± 0.15**	**3.15 ± 0.14**	79.8	79.9	44.7	36.5
5	3/2	+/-	+/-	**3.07 ± 0.14**	**3.30 ± 0.03**	79.6	79.7	47.2	41.4
6	3/3	+/-	-/-	**2.94 ± 0.22**	**3.09 ± 0.22**	79.8	80.1	42.8	31.3

Table 1. Mouse heart end-diastolic left ventricle inner diameter (LVID) for different GLUT4 & ACPR30 genotypes.
***** Statistically significant difference wrt WT using a two-sample, two- tail t-test/ANOVA
N=number of mice imaged from the cohort's group Male/Female

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