Left ventricular metabolism, function, and sympathetic innervation in men and women with type 1 diabetes

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Background. **In type I diabetes (T1DM), alterations in LV function may occur due to changes in innervation, metabolism, and efficiency.**

Objectives. **We evaluated the association between sympathetic nerve function, oxidative metabolism, resting blood flow, LV efficiency and function in healthy diabetics, and assessed gender differences.**

Methods. **Cross-sectional study of 45 subjects with T1DM, 60% females, age 34 ± 13 years, and 10 age-matched controls. Positron emission tomography (PET) imaging with [^{11}C]acetate and [^{11}C]*meta*-hydroxyephedrine was performed, in addition to cardiac magnetic resonance imaging.**

Results. **There were no significant differences in LV function, innervation, or oxidative metabolism between T1DM and controls. Cardiac oxidative metabolism was positively associated with higher levels of sympathetic activation, particularly in women. Diabetic women had significantly lower efficiency compared with diabetic men. Resting flow was significantly higher in diabetic women compared with diabetic men, and tended to be higher in female controls as well.**

Conclusions. **Measures of myocardial function, metabolism, blood flow, and sympathetic activation were preserved in young, otherwise healthy, T1DM patients. However, T1DM women presented with greater myocardial oxidative metabolism requirements than men. Ongoing studies are evaluating changes over time. (J Nucl Cardiol 2016;23:960–9.)**

Antecedentes. **En la Diabetes Mellitus tipo 1 (T1DM) los cambios en la función ventricular izquierda pueden deberse a cambios en la inervaciœn, metabolismo, y eficiencia.**

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Objetivos. **Evaluamos la asociación entre la inervación simpática, el metabolismo oxidativo, flujo sanguíneo en reposo, la eficiencia y función del ventrículo izquierdo en diabéticos sanos, de acuerdo a diferencia de género.**

Métodos. **Estudio transversal de 45 sujetos con DM tipo 1 : 60% mujeres (34años +−13) y 10 controles pareados por edad. Se realizó un estudio de PET / CT con ^{11}C acetato y ^{11}C meta hydroxyephedrina. Adicionalmente se obtuvieron imágenes con resonancia magnética.**

Resultados. **No se encontraron diferencias en la función, inervación y metabolismo del ventrículo izquierdo entre los pacientes con DM tipo 1 y el grupo control. El metabolismo oxidativo cardiaco tuvo una asociación positiva con niveles altos de activación simpática, particularmente en las mujeres. Las mujeres diabéticas tuvieron una eficiencia significativamente menor comparadas con los hombres diabéticos. El flujo en reposo fue significativamente mayor en mujeres diabéticas comparadas con los hombres diabéticos, la tendencia fue igualmente mayor en las mujeres del grupo mayor.**

Conclusiones. **Las medidas de función miocárdica, metabolismo, flujo miocárdico y activaciœn simpática estuvieron conservados en pacientes jóvenes con DM tipo 1 por lo demás sanos. Sin embargo las mujeres con DM tipo 1 tienen mayores requerimientos de metabolismo oxidativo miocárdico que los hombres. Estudios prospectivos están evaluando estos cambios en el transcurso del tiempo. (J Nucl Cardiol 2016;23:960–9.)**

背景. 在I型糖尿病**(T1DM)**患者中, 由于心脏自主神经支配、心脏代谢以及左心室射血效率的改变, 左心室功能也会发生相应的变化。

目的. 评估健康糖尿病患者心脏交感神经功能、心脏氧化代谢、静息血流量、左心室射血效率与左心室功能之间的关系, 并分析不同性别之间的差异。

方法. 本研究采用横断面法, 入选**45**例I型糖尿病患者, 年龄在 **34±13** 岁之间, 其中 **60%** 为女性, **10**例年龄相当的正常人作为对照组。除了心脏磁共振成像外, 本研究还使用了醋酸盐 **([^{11}C]acetate)**和元-羟基麻黄碱 **([^{11}C]meta-hydroxyephedrine)** 作为显影剂的正电子发射型计算机断层显像 **(PET)** 技术。

结果. 与对照组相比,**I**型糖尿病患者的左心室功能, 心脏神经支配或氧化代谢均无显著差异。心脏的氧化代谢与交感神经活性呈正相关, 女性尤其如此。女性糖尿病患者的左心室射血效率明显低于男性糖尿病患者。女性糖尿病患者静息血流量明显高于男性, 在对照组中女性静息血流量亦有偏高的趋势。

结论. 健康年轻人与健康的I型糖尿病患者左心室功能、心脏氧化代谢、静息血流量以及交感神经活性相同。然而, 女性**I**型糖尿病患者较男性有更高的心脏氧化代谢。正在开展的后续研究将进一步评估时间对上述指标的影响。 **(J Nucl Cardiol 2016;23:960–9.)**

Key Words: Diabetes Mellitus · cardiac autonomic neuropathy · positron emission tomography · female · myocardium

Abbreviations

CAN	Cardiac autonomic neuropathy
CMR	Cardiac magnetic resonance
LV	Left ventricular
Eff	Efficiency
PET	Positron emission tomography
[^{11}C]HED	[^{11}C]*Meta*-hydroxyephedrine
RI	Retention index
rMBF	Resting myocardial blood flow
SVI	Stroke volume index
SBP	Systolic blood pressure
T1DM	Type 1 Diabetes Mellitus
WMI	Work-metabolic index

BACKGROUND

Type 1 diabetes (T1DM) may lead to left ventricular (LV) dysfunction and cardiomyopathy in the absence of coronary artery disease (CAD) or hypertension.[1-3] Changes in both systolic and diastolic LV function have been demonstrated in otherwise healthy T1DM subjects, preceding the development of other clinically recognizable diabetic complications.[4-6] One proposed mechanism mediating these changes is the presence of cardiovascular autonomic neuropathy (CAN), and associated sympathetic/parasympathetic imbalance, which may promote earlier changes in LV function by critically influencing myocardial substrate

utilization,[7] impairing oxidative metabolism, and altering LV efficiency. The pathophysiology of diabetic cardiomyopathy, which includes increased myocardial fibrosis as well as accelerated apoptosis, may be partially related to these changes in myocardial substrate utilization and increased oxidative stress.[8-10]

Positron emission tomography (PET) can be used to assess myocardial sympathetic innervation with the norepinephrine analog [^{11}C]*meta*-hydroxyephedrine ([^{11}C]HED), as well as myocardial oxidative metabolism and blood flow via measurements of [^{11}C]acetate kinetics.[11,12] This technique also provides a non-invasive assessment of LV efficiency via calculation of LV work-metabolic index (WMI), an early indicator of LV dysfunction.[13]

Available evidence shows that intensive glycemic control is most important to prevent chronic diabetes complications, but findings based on more recent data have raised the question of whether glycemic variability may also influence the risk of cardiovascular complications in T1DM.[14] Furthermore, emerging evidence has shown an increased risk of cardiovascular disease complications and death in diabetic women compared with diabetic men.[15]

The long-term goals of this study are to identify potential mechanisms contributing to the development of myocardial dysfunction in T1DM, as well as the role of CAN in promoting these deficits, in a longitudinal study of men and women with T1DM of more than 5 years duration and without CAD. The overall hypothesis is that, in T1DM, sympathetic activation promotes downstream alterations in myocardial oxidative metabolism and efficiency, impaired myocardial blood flow regulation and LV function.

The objectives of this initial analysis were to evaluate associations between LV function, innervation, perfusion, and metabolism in these subjects at baseline compared with age-matched nondiabetic controls, and to evaluate potential effects of glucose control and glucose variability. Furthermore, we assessed whether gender differences in these parameters of LV function, perfusion, and metabolism may explain gender differences in cardiovascular risk in T1DM.

METHODS

Study Population and Design

The University of Michigan Institutional Review Board approved the study, and all subjects signed a written consent. Forty-five subjects with T1DM and ten age-matched healthy controls were enrolled in this study. All subjects had normal resting ECGs and normal exercise treadmill test results before enrolling in the study. Inclusion criteria for diabetic subjects were as follows: T1DM, age 18–65 years, with a minimum of 5 years and less than 20 years diabetes duration, and no signs of microvascular complications. Patients with a history of cardiovascular disease were excluded from the study. Demographic and anthropometric measures were collected through questionnaires and a physical examination; fasting blood and urine samples were obtained for the measurement of metabolic parameters including HbA1c, a lipid panel, and renal function tests.

Assessments of CAN

Assessment of CAN included cardiovascular reflex tests, heart rate (HR) variability studies, and PET studies with [^{11}C]HED. All CAN and PET evaluations were performed in a standardized fashion on all subjects after an overnight fast, and analyzed as previously described.[16-19]

PET Studies. All PET imaging was performed using a Siemens ECAT Exact HR+ PET Scanner (Siemens Molecular Imaging, Knoxville, TN). LV sympathetic innervation was assessed using PET measures of the regional myocardial retention of [^{11}C]HED, as previously described.[19] A 'retention index' (RI; mL blood min^{-1} mL^{-1} tissue) was generated for each sector by normalizing the measured tissue concentration of [^{11}C]HED in the final image frame to the time integral of the blood time-activity curve. The generated RI data were displayed in the standard cardiac 'polar map' format. A *z*-score analysis of the regional RI values was performed to obtain measures of the regional heterogeneity of [^{11}C]HED retention in the subjects.[19] All analyses were performed by a single investigator (DR) who was masked to subjects' clinical and laboratory data.

The myocardial kinetics of [^{11}C]acetate were used to assess LV oxidative metabolism and resting myocardial perfusion. LV measurements obtained from cardiac magnetic resonance (CMR) imaging were used to subsequently calculate LV efficiency.

The pre-specified primary measures for CAN were the global mean [^{11}C]HED RI and measures of HR variability.

[^{11}C]Acetate Analyses. Measures of global and regional LV oxidative metabolism were derived from the myocardial kinetics of [^{11}C]acetate using previously validated methods.[11] The kinetic data were analyzed by fitting the activity concentration data in the last 7 frames to a single exponential clearance term, with an exponent of $k_{mono} \cdot t$, where the clearance rate constant k_{mono} (min^{-1}) is related to the regional rate of oxidative metabolism in the myocardial tissue. Using the k_{mono} values, measured values of the stroke volume index (SVI) by CMR (described below), peak systolic blood pressure (SBP_{peak}) and HR during PET, global and regional estimates of the LV WMI, or LV Efficiency, were calculated as

$$WMI = \frac{SVI \times SBP_{peak} \times HR}{k_{mono}}. \quad (1)$$

Resting myocardial blood flow values were estimated from the myocardial kinetics of [^{11}C]acetate using previously reported methods.[20] Briefly, a reversible 1-tissue compartment model was used to provide estimates of the two rate constants K_1 (mL^{-1} min^{-1} mL) and k_2 (min^{-1}). In addition, a total blood

volume term (TBV; dimensionless) was included in the model to account for recovery and spillover effects, as previously validated for [^{13}N]ammonia.[21] A whole-blood time-activity curve $C_{wb}(t)$ was obtained by placing a small region-of-interest in the LV chamber at the base of the heart. The intact tracer in plasma curve, $C_p(t)$, used as the input function for compartmental modeling, was generated by applying an averaged metabolite correction to $C_{wb}(t)$, using a value of 5.3 min for $T_{1/2}$, as previously described:[22]

$$C_p(t) = (0.91)e^{-\ln(2)\times\left(t/T_{1/2}\right)} \times C_{wb}(t) \quad (2)$$

Nonlinear regression analysis with the input function $C_p(t)$ and [^{11}C]acetate kinetics provided estimates of K_1, k_2, and TBV for each region. The estimated rate constant K_1 is equal to the product $E \times F$, where E is the unidirectional extraction fraction (dimensionless) and F is blood flow (mL min^{-1} mL^{-1}). The equation describing the [^{11}C]acetate extraction fraction E as a function of blood flow derived by van den Hoff et al ($a = 0.64$, $b = 1.20$ mL min^{-1} mL^{-1}) was used to convert estimates of K_1 to resting myocardial perfusion values F (mL min^{-1} mL^{-1}) in each LV region.[20]

The pre-specified measure to assess efficiency was the work-metabolic index, and for oxidative metabolism it was the global k_{mono}.

CMR Studies. MRI examinations were obtained using a 3T Ingenia scanner (Philips Medical Systems, Best, The Netherlands) and a 16-channel phased-array coil. A retrospective ECG-gated steady state with free-precession cine images was acquired in the LV short- and long-axis planes to measure global LV volumes, mass, and ejection fraction (EF). All images were obtained during breath-hold.

CMR Data Analysis. Global LV end-diastolic volume (EDV), end-systolic volume (ESV), and mass (LVM) were quantified by a senior cardiac-trained MR physician masked to all clinical and laboratory data, using commercially available software (QMASS, version 7.5, Medis, Leiden, The Netherlands). LV volumes and mass were indexed to body surface area (Mosteller).

Assessment of glycemic variability (GV) was performed from data obtained with continuous glucose monitoring (CGM) sensors (iPro CGM System, Medtronic, Northridge, CA, USA) as previously described, and included low blood glucose index, high blood glucose index, area under the curve (AUC) for hypoglycemia, and dynamic stress factor (DySF).[16,23] Measures of glucose variability were exploratory.

Data Analysis. Data are presented as mean ± standard deviation (SD) or median (interquartile range) for quantitative variables depending on whether the distribution was symmetric or skewed. The two groups (T1DM and controls) were compared using the student's t test assuming unequal variances, or the Mann-Whitney two-sample test, respectively. With 45 T1DM and 10 control subjects there is 80% power for a two-tailed t test using a 5% level of significance to detect an effect size of 1.0; i.e., an expected difference of 1.0 SD between the two groups.

Using data only from the diabetic subjects, Spearman correlation coefficients (R) were calculated to estimate the associations between global mean [^{11}C]HED RI and LV Eff, oxidative metabolism, rMBF, LV size and function, HbA1c, and indices of glycemic variability. Since LV Eff, k_{mono}, rMBF, and [^{11}C]HED RI were found to be related to gender, the correlation coefficients were subsequently adjusted for gender. With 18 men and 27 women T1DM subjects, there is 80% power for a two-tailed t test using a 5% level of significance to detect an effect size of 0.9; i.e., an expected difference of 0.9 SD between the two groups. There is 80% power to detect a correlation of 0.4 using all 45 T1DM subjects, a correlation of 0.5 within the 27 women and a correlation of 0.6 within the 18 men. There were insufficient control samples to compare diabetics and controls within each gender; with only 5 control subjects of each gender, there would be 80% power for a two-tailed t test using a 5% level of significance to detect an effect size of 1.41 for women and 1.49 for men.

All statistical analyses were performed using SAS 9.4 (SAS Institute, Cary, NC, USA). The P value for each test is reported with no correction for multiple testing to enable the reader to assess the reproducibility of the result.

RESULTS

The clinical characteristics of the enrolled subjects are shown in Table 1. The T1DM subjects were relatively young, with a mean age of 34 ± 13 years, and diabetes duration of 14 ± 6 years. None had clinical evidence of chronic complications or CAD at baseline. No significant differences between T1DM subjects and the controls were noted, except for blood glucose and HbA1c, used to define the groups (Table 1). Cardiac parameters of systolic function (EF, LV ESV, LV EDV, and stroke volume) were within normal ranges in the T1DM subjects (Table 1).

The standardized cardiovascular reflex tests in these subjects were not different between groups.[24,25] Global sympathetic innervation, assessed by [^{11}C]HED retention, was homogenous throughout the LV in all subjects, and was not different in T1DM subjects compared to healthy controls. Similarly, no differences were observed in global rMBF between T1DM and control subjects.

Assessment of LV oxidative metabolism revealed uniform rates within each subject, which did not significantly differ between the diabetic and control subjects (k_{mono} = 0.063 ± 0.016 min^{-1} vs. 0.055 ± 0.011 min^{-1}, respectively; P = 0.10). LV efficiency, measured by a calculated WMI, was 6.0 (5.4–7.2) × 10^6 mmHg mL m^{-2} among diabetes subjects, as compared to 5.6 (4.8–6.8) × 10^6 mmHg mL m^{-2} among healthy controls (P = 0.30).

To understand potential differences in risk between T1DM men and women, we analyzed the associations between sympathetic innervation and oxidative metabolism in T1DM stratified by gender. As shown in Table 2,

Table 1. Baseline characteristics of study subjects and healthy controls.

	Type 1 diabetes ($n = 45$)	Healthy controls ($n = 10$)	p value
Age (y)	34 ± 13	34 ± 12	0.96
Gender female *n* (%)	27 (60%)	5 (50%)	0.56
Diabetes duration (y)	14 ± 6	N/A	
Baseline HbA1c (%)	8.0 ± 1.3	5.4 ± 0.3	<0.0001
Fasting glucose mmol L^{-1} (mg dL^{-1})	8.6 ± 4.3 (155 ± 77)	4.8 ± 0.8 (86 ± 14)	0.0015*
BMI (kg m^{-2})	26 ± 5	24 ± 3	0.050
Baseline systolic blood pressure (mm Hg)	117 ± 11	115 ± 9	0.69
Baseline diastolic blood pressure (mmHg)	72 ± 8	70 ± 9	0.38
Heart rate (bpm)	89 ± 14	77 ± 16	0.042
Total cholesterol mmol L^{-1} (mg dL^{-1})	4.30 ± 0.75 (166 ± 29)	4.20 ± 0.83 (162 ± 32)	0.77*
LDL-c mmol L^{-1} (mg dL^{-1})	2.33 ± 0.6 (90 ± 23)	2.23 ± 0.62 (86 ± 24)	0.66*
HDL-c mmol L^{-1} (mg dL^{-1})	1.63 ± 0.49 (63 ± 19)	1.50 ± 0.34 (58 ± 13)	0.55*
Triglycerides mmol L^{-1} (mg dL^{-1})	0.79 ± 0.36 (70 ± 32)	0.98 ± 0.40 (87 ± 35)	0.12*
Serum creatinine μmol L^{-1} (mg dL^{-1})	76 ± 15 (0.86 ± 0.17)	72 ± 9.7 (0.81 ± 0.11)	0.33
E/I ratio	1.23 ± 0.12	1.25 ± 0.13	0.73
Valsalva ratio	1.35 ± 0.28	1.38 ± 0.21	0.82
Stroke volume (mL)	90 ± 18	88 ± 16	0.74
LV ED volume (mL)	146 ± 29	146 ± 29	0.99
Stroke volume index (mL m^{-2})	47 ± 7	48 ± 9	0.76
LV mass/EDV (g mL^{-1})	0.66 ± 0.12	0.60 ± 0.13	0.23
Mean ^{11}C HED retention index	0.081 ± 0.013	0.082 ± 0.012	0.88
rMBF (mL g^{-1} min^{-1})	0.90 ± 0.25	0.83 ± 0.22	0.42
Kmono global (min^{-1})	0.063 ± 0.016	0.055 ± 0.011	0.10
Work-metabolic index global (mmHg × mL m^{-2})	6.0 (5.4,7.2) × 10^6	5.6 (4.8,6.8) × 10^6	0.30*

All data are shown as mean ±SD, median (interquartile range) or *n* (%). Laboratory values are given in SI units, with metric units shown in parentheses.
LDL-c, low-density lipoprotein cholesterol; *HDL-c* high-density lipoprotein cholesterol; *E/I ratio* expiration-to-inspiration ratio; *LV ED*, left ventricular end diastolic; *LV Mass/EDV*, left ventricular mass/ end diastolic volume.
* *P* value from Wilcoxon rank sum test, otherwise two-sample *t* test with unequal variances for continuous variables or chi-square test for gender and race.

T1DM men were taller, heavier and had higher systolic blood pressures, lower HDL-cholesterol levels and significantly lower mean [^{11}C]HED retention index compared with T1DM women. However, T1DM women presented with significantly higher k_{mono} values, indicative of a higher rate of oxidative metabolism compared with diabetic men. Consequently, the WMI was significantly lower in the diabetic women than the diabetic men [5.7(4.9,6.4) × 10^6 mmHg mL m^{-2} vs. 7.3 (6.3,8.3) × 10^6 mmHg mL m^{-2}, respectively; $P = 0.0006$], while no gender differences were seen in WMI in the non-diabetic controls ($P = 0.24$). Resting myocardial blood flow was significantly higher in diabetic women compared with diabetic men (1.3 ± 0.22 vs. 0.7 ± 0.14 mL g^{-1} min^{-1}; $P < 0.0001$), and tended to be higher in non-diabetic women, although this difference was not significant (0.95 ± 0.20 vs. 0.76 ± 0.21; $P = 0.19$).

Global [^{11}C]HED RI was positively related to the rate of LV oxidative metabolism assessed by k_{mono} ($r = 0.47$; $P = 0.0013$, adjusted for gender) (Figure 1) After adjustment for gender, the association between sympathetic innervation and LV efficiency was no longer significant (Figure 2) ($r = -0.24$, $P = 0.12$). The rMBF was strongly correlated with k_{mono} and [^{11}C] HED RI ($r = 0.57$, $P < 0.0001$) in T1DM, even after adjustment for gender. Since HR and blood pressure correlated positively with rMBF ($r = 0.58$, $P < 0.0001$ and $r = 0.48$, $P = 0.0016$, respectively), hemodynamics exerted a strong influence on rMBF. Stroke volume index was negatively correlated with HbA1c ($r = -0.48$; $P = 0.0009$), mainly due to findings in diabetic women.

Table 2. Baseline characteristics of diabetic subjects stratified by gender.

	Men ($n = 18$)	Women ($n = 27$)	*P* value
Age (y)	34 ± 13	34 ± 13	0.84
Duration (y)	15 ± 7	12 ± 5	0.13
Baseline HbA1c (%)	7.9 ± 1.3	8.1 ± 1.3	0.63
Fasting glucose mmol L^{-1} (mg dL^{-1})	9.99 ± 5.44 (180 ± 98)	7.66 ± 3.11 (138 ± 56)	0.16
Height (cm)	181 ± 9	165 ± 6	<0.0001
Weight (kg)	84 ± 15	72 ± 12	0.0086
BMI (kg m^{-2})	25 ± 3	27 ± 5	0.27
Systolic blood pressure (mmHg)	121 ± 9	114 ± 12	0.021
Diastolic blood pressure (mmHg)	73 ± 8	72 ± 8	0.96
Heart rate (bpm)	88 ± 17	89 ± 12	0.90
Total cholesterol mmol L^{-1} (mg dL^{-1})	4.07 ± 0.73 (157 ± 28)	4.45 ± 0.75 (172 ± 29)	0.08
LDL-c mmol L^{-1} (mg dL^{-1})	2.25 ± 0.47 (87 ± 18)	2.41 ± 0.65 (93 ± 25)	0.40
HDL-c mmol L^{-1} (mg dL^{-1})	1.42 ± 0.57 (55 ± 22)	1.76 ± 0.41 (68 ± 16)	0.0022
Triglycerides mmol L^{-1} (mg dL^{-1})	0.86 ± 0.41 (76 ± 36)	0.75 ± 0.33 (66 ± 29)	0.21
Serum creatinine μmol L^{-1} (mg dL^{-1})	86.6 ± 11.5 (0.98 ± 0.13)	68.0 ± 12.4 (0.77 ± 0.14)	<0.0001
Stroke volume (mL)	101 ± 15	83 ± 16	0.0005
LV ED volume (mL)	167 ± 28	132 ± 20	<0.0001
Stroke volume index (mL m^{-2})	49 ± 4	46 ± 8	0.14
LV mass/ EDV(g mL^{-1})	0.73 ± 0.11	0.61 ± 0.11	0.0004
Mean retention index	0.072 ± 0.011	0.087 ± 0.011	0.0001
rMBF (mL g^{-1} min^{-1})	0.70 ± 0.14	1.03 ± 0.22	<0.0001
Kmono global (min^{-1})	0.051 ± 0.012	0.071 ± 0.013	<0.0001
Work-metabolic index global (mmHg × mL^{-1} m^{-2})	7.3(6.3,8.3) × 10^6	5.7(4.9,6.4) × 10^6	0.0006

All data are shown as mean ±SD, median (interquartile range) or *n* (%). Laboratory values are given in SI units, with metric units shown in parentheses.
LDL-c, low-density lipoprotein cholesterol; *HDL-c,* high-density lipoprotein cholesterol; *LV ED,* left ventricular end diastolic; *LV Mass/EDV,* left ventricular mass/end diastolic volume.
**P* value from Wilcoxon rank sum test, otherwise two-sample *t* test with unequal variances for continuous variables or chi-square test for gender and race.

We found no relationship between the degree of glucose control at baseline as assessed by the HbA1c levels, or between indices of glycemic variability and measures of LV oxidative metabolism, efficiency, or LV function (data not shown).

DISCUSSION

In this cohort of subjects with T1DM without evidence of cardiovascular disease, we found that greater LV presynaptic sympathetic function, assessed by [^{11}C] HED RI, was associated with higher rates of oxidative metabolism and resting myocardial blood flow, as assessed by [^{11}C]acetate kinetics. Our results support the hypothesis that preclinical changes of CAN in T1DM are associated with abnormal myocardial energy metabolism, and suggest mediation of myocardial energy deficits by enhanced sympathetic function. Furthermore, we found significant gender differences in these indices, as diabetic women had significantly lower myocardial efficiency compared with diabetic men, which may imply earlier metabolic and energy deficits in the myocardium of diabetic women. We believe that this is the first study to analyze the relationship between changes in LV innervation and myocardial oxidative metabolism in patients with T1DM and no clinically evident heart disease. These findings may provide mechanisms for the known higher risk of cardiovascular disease in women with T1DM compared with T1DM men.[15]

The natural history and the mechanisms of myocardial dysfunction in T1DM are not well understood. Several studies have reported that in both T1DM and type 2 diabetes, LV dysfunction may occur in the absence of ischemic heart disease or hypertension.[1-3] Evidence from experimental and clinical studies suggests that there may be additional mechanisms for a deleterious synergy between diabetes and heart failure, including increased myocardial oxygen consumption, fatty acid uptake, greater oxidative stress and impaired myocardial glucose uptake in cardiomyocytes.[26-29] A

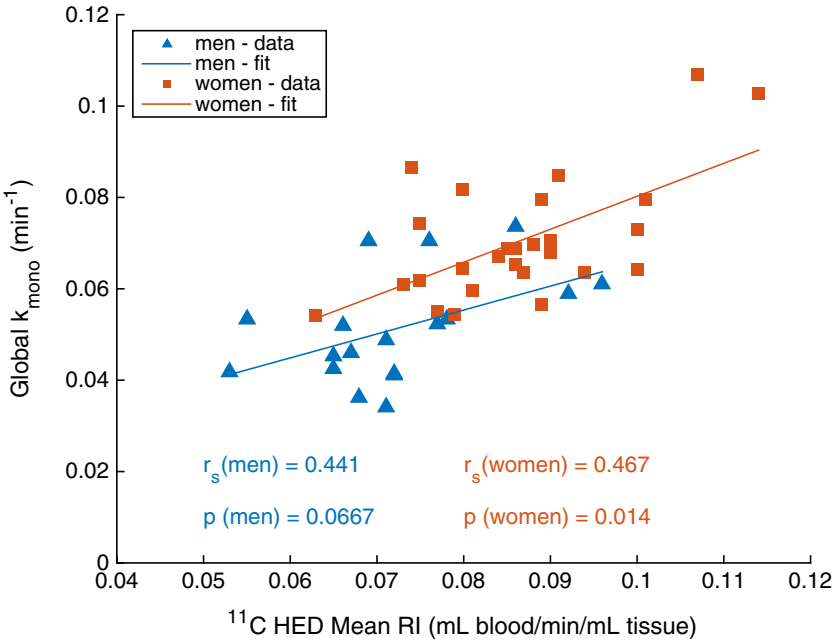

Figure 1. k_{mono} vs. [^{11}C]HED RI—Correlation between cardiac sympathetic innervation, assessed by [^{11}C]*meta*-hydroxyephedrine (^{11}C HED) as a global retention index (RI) and myocardial oxidative metabolism, assessed by the clearance rate of [^{11}C]acetate (k_{mono}), after adjustment for gender. Shown are the correlations for men in *blue*, and for women in *red*.

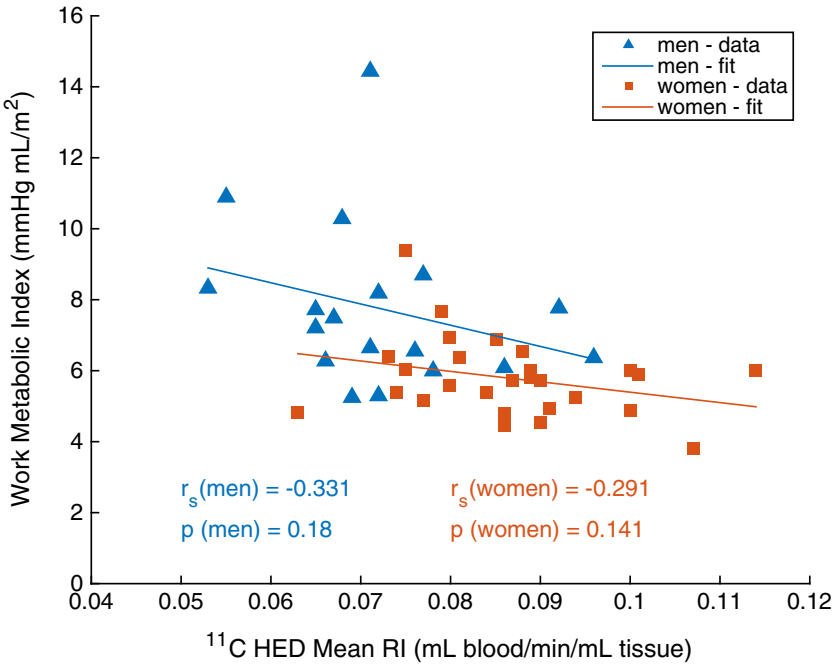

Figure 2. LV efficiency vs. [^{11}C]HED RI—Correlation between cardiac sympathetic innervation, assessed by [^{11}C]*meta*-hydroxyephedrine (^{11}C HED) as a global retention index, (RI) and myocardial efficiency, given as Work-Metabolic Index, after adjustment for gender. Shown are the correlations for men in *blue*, and for women in *red*.

role for sympathetic dysfunction and CAN in the pathogenesis of heart failure has been suggested by many studies, especially in patients with diabetes.[1-3] Our group reported prior associations between CAN and diastolic dysfunction in patients with T1DM and mild microangiopathy.[2] Furthermore, sympathetic hyperinnervation in proximal myocardial segments coupled with denervation in distal segments has been observed in prior studies of diabetics with demonstrated autonomic neuropathy, and has been postulated to result in vascular hyperreactivity, paradoxical vasoconstriction, and potential arrhythmogenesis.[19] Studies of myocardial oxidative metabolism and substrate utilization have reported increased oxygen consumption in T1DM subjects as compared with normal controls, although this difference was no longer significant after correction for the rate pressure product.[29,30] Earlier stages of diabetic CAN have been associated with sympathetic activation due to an initial predominant parasympathetic denervation.[31,32] This may affect heart metabolism through its impact on sympathovagal balance and baroreflexes. Increased myocardial oxidative stress known to occur in CAN[32] may directly impact myocardial glucose uptake and metabolism, with longer-term deleterious effects that may include progressive cardiac fibrosis and subsequent development of cardiomyopathy.

The present study is the first to concomitantly evaluate presynaptic sympathetic function and myocardial efficiency in patients with T1DM at much earlier stages of their disease, in an attempt to identify potential drivers of the higher cardiovascular risk in T1DM that could be targeted for interventions. We found a direct relationship between these indices in the absence of traditional risk factors, with higher [^{11}C]HED retention associated with higher substrate utilization and lower work-metabolic index, driven largely by the findings in diabetic women. Although women are smaller, we have corrected for BSA in our studies, therefore we do not believe that size differences between men and women are the reason for the lower efficiency observed.

Earlier PET studies have established alterations in myocardial substrate metabolism in diabetic subjects, even in the absence of coronary ischemia and overt heart failure. Rijzewijk et al. demonstrated increased fatty acid uptake and metabolism, and decreased myocardial glucose uptake in insulin-naïve type 2 diabetic men, compared with age-matched controls.[33] Elevated myocardial free fatty acid levels have been associated with increased triglyceride levels in the heart, higher rates of lipolysis, and decreased contractile reserve. As a consequence, the myocardium may demonstrate an abnormally high requirement for oxygen, and there may be intracellular accumulation of potentially toxic intermediates of fatty acid metabolism.[34]

Of specific interest are our findings of significant gender differences in LV efficiency in T1DM. Gender differences in oxidative as well as glucose metabolism have been demonstrated in healthy volunteers in past studies; women volunteers demonstrated higher oxidative metabolism and lower glucose utilization than men.[35] Furthermore, in T1DM women, administration of dobutamine to increase cardiac work has been shown to reduce glucose oxidation and to increase overdependence of the myocardium on fatty acid metabolism.[36] Several studies have described higher oxidative metabolism as well as higher myocardial blood flow in women than in men, independent of diabetes, and in obese as compared with non-obese subjects.[37,38] Evidence from observational and prospective studies has described that diabetic women have a greater relative risk of cardiovascular diseases than diabetic men, which is in contrast to the lower risk observed in age-matched non-diabetic women compared to men.[39]

The reasons for this excess risk in diabetic women are still unclear. Our analysis of rMBF showed normal resting perfusion in T1DM, but higher flow in women as compared with men. Our groups as well as others have previously shown that women have higher myocardial blood flow rates than men.[40,41] It has been postulated that gender differences in myocardial blood flow may be related to the vasodilatory effects of estrogen.[42] Postulated mechanisms of increased risk relating the contribution of estrogen to markers of inflammation such as C-reactive protein, which has been shown to predict subsequent development of metabolic syndrome and diabetes preferentially in women, may play a role as well.[43] Our findings also raise the possibility that mitochondrial metabolism (oxidative phosphorylation) is impaired to a greater degree in women, perhaps in part mediated by insulin resistance and oxidative stress. The mechanism by which altered sympathetic activity leads to diminished mitochondrial oxidative phosphorylation deserves exploration in future studies. As such, our findings of significant gender differences in LV efficiency in T1DM, with higher energy requirements and decreased efficiency in T1DM women who are otherwise healthy, are in line with the recent reports above, and merit discussion and further study. Our analysis showed that gender and HR were the strongest determinants of higher resting flow. This finding is consistent with that of many other investigators, and again leads to speculation that estrogen plays a crucial role in vascular function.

The strengths of this analysis lie in the sophisticated and sensitive measurements of myocardial function, innervation, and metabolism that we were able to perform. These tools have given us a window into the earliest stages of possible cardiac abnormalities, a time

point at which the heart still contracts and is perfused quite normally, even as subtle changes in sympathetic innervation are reflected as changes in oxidative metabolism of the heart.

We are currently obtaining 3-year follow-up studies on these subjects, and thus will be able to evaluate whether the subtle, early changes that we have reported here develop into more clinically evident changes in LV function, as assessed by CMR imaging, and whether cardiac sympathetic innervation and myocardial oxidative metabolism become manifestly abnormal over time.

Study Limitations include the relatively small sample size, and the cross-sectional nature of these initial analyses, which prevents us from attributing causality to the demonstrated correlations. An additional important limitation is the relatively low number of healthy controls that were included, which restricts our ability to better define the combined effects of diabetes and gender in the development and progression of the described deficits. However, the T1DM were asymptomatic and with relatively early disease state, and the healthy controls were age-matched to the diabetic subjects; as such, with this sample size the study had 80% power to detect a difference of 0.9 SD between the two groups. Furthermore, since our assessment of LV myocardial efficiency was based on noninvasive measurements, we were not able to assess the effects of preload on myocardial contractility and efficiency. We measured only resting myocardial perfusion in this study, since all subjects were clinically free of CAD, with normal exercise treadmill tests. However, the marked gender difference we noted in myocardial blood flow, as well as strong correlation between blood flow and oxidative metabolism, raise the possibility that our findings relate mostly to gender effects, rather than to the effects of diabetes, on LV efficiency.

NEW KNOWLEDGE GAINED

Increases in oxidative metabolism in otherwise healthy type 1 diabetics are linked to altered myocardial sympathetic innervation. Women with T1DM show decreased myocardial efficiency compared with T1DM men in this study.

CONCLUSIONS

Our findings provide insights into the relationship between LV sympathetic innervation, a measure of CAN, and substrate metabolism in type 1 diabetes, suggesting that increased oxidative metabolism may be an early event in the natural history of diabetic cardiomyopathy. Gender differences in myocardial blood flow and oxidative metabolism as well as sympathetic innervation may contribute to the adverse prognosis seen in diabetic women, although the differences that we observed may, in part, simply be due to known differences between men and women unrelated to diabetes.

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Disclosures

The authors report no conflicts of interest.

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