Heart Rate Variability and Cardiac Autonomic Dysfunction: Prevalence, Risk Factors, and Relationship to Arterial Stiffness in the Treatment Options for Type 2 Diabetes in Adolescents and Youth (TODAY) Study

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OBJECTIVE

To determine whether prior type 2 diabetes (T2D) treatment or glycemic control over time are independently associated with heart rate variability (HRV) and whether the presence of cardiac autonomic dysfunction is associated with arterial stiffness in young adults with youth-onset T2D enrolled in the Treatment Options for Type 2 Diabetes in Adolescents and Youth (TODAY) study.

RESEARCH DESIGN AND METHODS

Heartbeats over 10 min were measured to derive the normal R-Rs (NN intervals). Outcomes included the standard deviation of the NN intervals (SDNN), the root mean square differences of successive NN intervals (RMSSD), percent of NN beats that differ by more than 50 ms (PNN50), and the low-frequency (LF) power domain, high-frequency (HF) power domain, and their ratio (LF:HF). Autonomic dysfunction was defined as ≥3 of 5 abnormal HRV indices compared with obese controls from a separate study.

RESULTS

A total of 397 TODAY participants were evaluated 7 years after randomization. TODAY participants had reduced HRV (SDNN 58.1 ± 29.6 ms vs. controls 67.1 ± 25.4 ms; $P < 0.0001$) with parasympathetic loss (RMSSD 53.2 ± 36.7 ms vs. controls 67.9 ± 35.2 ms; $P < 0.0001$) with sympathetic overdrive (LF:HF ratio 1.4 ± 1.7 vs. controls 1.0 ± 1.1; $P < 0.0001$). Cardiac autonomic dysfunction was present in 8% of TODAY participants, and these participants had greater pulse wave velocity compared with those without dysfunction ($P = 0.0001$). HRV did not differ by randomized treatment, but higher hemoglobin A1c (HbA_{1c}) over time was independently associated with lower SDNN and RMSSD and higher LF:HF ratio after adjustment for age, race-ethnicity, sex, and BMI.

CONCLUSIONS

Young adults with youth-onset T2D show evidence of cardiac autonomic dysfunction with both parasympathetic and sympathetic impairments that are associated with higher HbA_{1c}.

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Cardiovascular autonomic dysfunction results from damage to the nerves and blood vessels innervating the heart and can lead to dysfunctional heart-rate control and abnormal vascular dynamics. Cardiac autonomic dysfunction is a frequent but underdiagnosed complication of type 2 diabetes (T2D) that is associated with arrhythmia, myocardial infarction, and sudden death (1,2). Symptomatic manifestations of autonomic dysfunction include sinus tachycardia, exercise intolerance, orthostatic hypotension, and asymptomatic myocardial ischemia. One of the earliest subclinical manifestations of cardiac autonomic dysfunction is reduction in heart rate variability (HRV) with parasympathetic loss preceding sympathetic dysfunction (3).

Compared with their peers without diabetes, adolescents with T2D have early signs of cardiac and vascular abnormalities including left ventricular hypertrophy and diastolic dysfunction (4–8) as well as vascular stiffness and thickness (9–14). However, cardiac autonomic function and HRV have been evaluated less frequently in young adults with youth-onset T2D (15–17). The SEARCH for Diabetes in Youth (SEARCH) study found cardiac autonomic dysfunction in 17% of their cohort of young adults with youth-onset T2D (compared with a lean control group), a higher prevalence than that seen in young adults with type 1 diabetes (17% vs. 12%) (17). However, SEARCH was not able to identify risk factors associated with the presence of autonomic dysfunction, perhaps because of inadequate statistical power. Thus, given the limited number of studies and lack of data on risk factors for cardiac autonomic function in adolescents with youth-onset T2D, we sought to examine HRV and the presence of cardiac autonomic dysfunction in a large cohort of young adults previously enrolled in the Treatment Options for Type 2 Diabetes in Adolescents and Youth (TODAY) randomized clinical trial.

The goals of this study were *1*) to evaluate HRV in TODAY participants compared with an obese control group and establish the prevalence of cardiac autonomic dysfunction in the TODAY cohort at follow-up (T2P1); *2*) to determine whether prior T2D treatment assignment in TODAY (metformin alone, metformin + rosiglitazone, or metformin + intensive lifestyle intervention) or glycemic control over time are independently associated with impaired HRV; and *3*) to assess of the association of cardiac autonomic function with noninvasive measures of arterial stiffness.

RESEARCH DESIGN AND METHODS

TODAY Participants

The TODAY randomized clinical trial has assembled the largest group of adolescents with T2D to date (18). HRV was assessed in those TODAY participants who participated in T2P1, the observational follow-up study. Details of the TODAY study design have previously been reported (18). Briefly, 699 youth with recent-onset T2D were recruited between July 2004 and February 2009. Inclusion criteria for the study were age 10–17 years, BMI ≥85th percentile for age and sex, diagnosis of T2D for ≤2 years, and negativity for islet cell autoantibodies. Participants were then randomized to one of three treatment arms (metformin alone, metformin + intensive lifestyle intervention, or metformin + rosiglitazone) and followed for 2–6.5 years. The primary end point of the TODAY clinical trial was time to treatment failure, defined as hemoglobin A1c (HbA_{1c}) ≥8% (≥64 mmol/mol) for 6 months or metabolic decompensation requiring insulin therapy (19).

The TODAY follow-up study, T2P1, lasted 3 years and began immediately after the TODAY clinical trial was completed. HRV measurements were collected in the last year of T2P1. One objective of T2P1 was to examine the persistence of effects of the TODAY randomized treatment assignment on long-term glycemic control following discontinuation of randomized treatment.

Of the 699 participants in the TODAY clinical trial, 572 (82%) were subsequently enrolled in the T2P1, the first study follow-up phase. HRV and arterial stiffness were assessed in the last year of T2P1. Data collection was attempted on all participants who attended their last visit in T2P1 (median time from randomization was 7.0 years), except pregnant female participants. A total of 453 participants underwent HRV measurements (65% of the original TODAY cohort and 79% of participants enrolled in the T2P1 phase). HRV data from 53 participants were excluded because of movement artifact or too few heart beats to calculate HRV indices, and 3 were excluded because they reported being on a β blocker, a medication known to decrease heart rate and affect HRV. Comparing participants excluded versus those with valid HRV data, there were no differences in age, sex, race-ethnicity, blood pressure, or heart rate. However, BMI was slightly lower in patients with valid data versus those excluded (BMI average 34.6 kg/m^2 vs. 37.2 kg/m^2 in those excluded; $P = 0.0309$). The 397 included in these analyses did not differ from the rest of the original cohort ($n = 302$) at baseline with respect to sex, race-ethnicity, BMI, HbA_{1c}, and T2D duration. However, the 397 included were slightly younger compared with those excluded (13.7 ± 2.0 vs. 14.3 ± 2.0; $P < 0.0001$). The study was approved by the Institutional Review Board for the Protection of Human Subjects of each participating institution. All participants provided informed consent and minor children confirmed assent according to local guidelines.

Study Variables for TODAY Participants

The demographics, anthropometrics, and laboratory data reported here were obtained within 1 month of the HRV measurements. Study visits were conducted after a 10–14 h overnight fast. Race-ethnicity was self-reported using 2000 U.S. Census–based questions. Participants checked yes or no for Hispanic/Latino ethnicity and then checked as many racial categories as needed. All physical measurements were made by trained staff according to a study-wide protocol (18). Height without shoes was measured using a stadiometer. Weight was measured twice using a Seca scale (model 882; Seca USA, Hanover, MA), with a third measurement taken if the first two differed by >0.2 kg, and measurements were averaged. BMI was calculated as weight in kilograms divided by the square of height in meters. Resting systolic blood pressure and diastolic blood pressure were measured using a CAS 740 monitor with standardized oscillometric cuff sizes while the participant was seated. Information on cigarette smoking and tobacco use in the past month (yes/no) was obtained from the participants.

All laboratory tests, including assessments of HbA_{1c} and fasting lipids, were performed in a central laboratory as previously described (18). HbA_{1c} was

measured with a dedicated high-performance liquid chromatography method (Tosoh Biosciences Inc., South San Francisco, CA) every 2 months during year 1 of TODAY and then quarterly. Urine albumin-to-creatinine ratio (UACR) was measured by spot urine samples after a 10–14 h overnight fast. Elevated urinary albumin excretion (referred to as albuminuria in previous TODAY publications) was defined as an UACR of ≥30 mg/g on two of three urine samples collected over a 3-month minimal period (20). Macroalbuminuria was defined as UACR ≥300 mg/g. Estimated glomerular filtration rate was calculated by the full age spectrum combined serum creatinine and cystatin C equation, which has been newly validated in both pediatric patients and adults and lends itself well to studies examining the transition from the pediatric stage to early adulthood (21). Concentrations of creatinine in serum and urine were determined using the Creatinine Plus enzymatic Roche reagent on a Modular P analyzer (Roche Diagnostics, Inc., Indianapolis, IN). Concentration of cystatin C in serum was determined at baseline and annually by immunochemistry using Siemens reagents (Siemens Healthcare Diagnostics, Inc., Newark, DE) on a Siemens nephelometer autoanalyzer (BN II). Treatments for dyslipidemia, hypertension, and confirmed elevated albumin excretion were initiated as needed per study protocol (18,20).

Control Participants

A control population of adolescents was not part of the T2P1 follow-up study design. However, obese control participants were recruited between 2010 and 2015 in Cincinnati, OH, as part of "Accelerated CV [Cardiovascular] Aging in Youth is related to CV Risk Clusters" (National Institutes of Health grant R01-HL-105591). This study recruited 133 obese (BMI ≥95th percentile for age and sex) control subjects. Obese control subjects had no evidence of T2D/prediabetes by fasting and postprandial blood glucose levels based on oral glucose tolerance testing, defined as a fasting glucose of <100 mg/dL and a 2-h postprandial value of <140 mg/dL. HRV in obese control subjects was obtained using the same protocol and training procedures as those used in TODAY. Lean control subjects were also recruited in CV Aging and their data were used only to define the prevalence of cardiac autonomic dysfunction as the primary comparison was to obese control subjects to study the added effects of T2D (see below). Lean control subjects had no history of chronic disease and had a BMI of <85th percentile for age and sex.

Outcome Measures

Heartbeats over 10 min from an electrocardiogram (ECG) (SphygmoCor device; AtCor Medical, Naperville, IL) were used to determine HRV. All TODAY staff were trained centrally and were certified by a central Vascular Reading Center to perform the HRV measurements. All prescriptions and over-the-counter medications were held until testing was complete. All tracings were reviewed and analyzed to ensure that R waves were adequately identified from artifacts and ectopic beats. The term "NN interval" was used instead of RR interval of the ECG to emphasize that the processed beats are normal sinus rhythm. Both time- and frequency-domain HRV parameters were included. Three time domains were assessed: *1*) the standard deviation (SD) of the NN intervals (SDNN), *2*) the root mean square differences of successive NN intervals (RMSSD), *3*) and the percent of adjacent NN intervals with a difference greater than 50 ms (PNN50). Three frequency domain indices were also assessed: *1*) normalized high-frequency (HF) power, *2*) normalized low-frequency (LF) power, and *3*) the LF:HF ratio. The SphygmoCor device derives the normalized LF and HF power by expressing them as a fraction of the total power (LF n.u. = LF/[total power (TP) − very low frequency (VLF)] × 100 and HF n.u. = HF/[TP − VLF] × 100), where n.u. are normalized units. Using fast Fourier analysis, the device separates the heart rate spectrum into various components and allows the quantification of sympathetic and vagal influences on the heart. SDNN is a measure of overall HRV. RMSSD, PNN50, and HF power represent the parasympathetic component of the HRV (22), thus parasympathetic loss is quantified by the reduction in RMSSD and HF power. The LF power is indicative of the sympathetic control of the cardiac function, and an increased LF:HF ratio denotes the increased sympathovagal balance (23).

Cardiac autonomic dysfunction was defined if three or more (out of five) HRV indices were greater or less than 2.5 SDs compared with control adolescents (17). For example, a HRV measure less than the 5th percentile (for SDNN, RMSSD, and HF) or greater than the 95th percentile (for LF and LF:HF ratio) compared with obese control subjects was considered abnormal, a classification previously used by the SEARCH for Diabetes in Youth study (17). Since PNN50 was not normally distributed, it was not considered in the definition. An obese control group was used as a primary control population. However, lean control subjects were used as a healthy comparison group.

Arterial stiffness measurements were also obtained at the same time as the HRV measurements. Arterial stiffness data in T2P1 participants have already been reported (9), but they are briefly summarized here. Measurements included pulse wave velocity (PWV) and augmentation index (AIx) using the SphygmoCorCPVH (AtCor Medical, Lisle, IL) and brachial distensibility (BrachD) using the DynaPulse 2000 (PulseMetric, San Diego, CA). All measurements were conducted after the participant rested for >10 min. PWV calculates the speed of pressure waves generated by cardiac ejection to reach the periphery, with a higher PWV indicating a higher arterial stiffness. PWV was assessed at three locations: between the carotid and femoral artery (PWV carotid femoral), the carotid and radial artery (PWV carotid radial), and the femoral and dorsalis pedis artery (PWV femoral foot). AIx is a measure of wave reflections and systemic arterial stiffness (24). A higher AIx suggests higher arterial stiffness. BrachD was obtained with the DynaPulse Pathway instrument that uses pulse dynamic analysis of arterial pressure signals obtained from a standard cuff sphygmomanometer to measure stiffness (25). A lower BrachD indicates greater stiffness.

Statistics

Statistical analyses were performed using SAS for Windows (version 9.4; SAS Institute Inc., Cary, NC). Statistical significance was defined as P value of <0.05. Data are mean and SD or percent. Because of a varying time at which measures of HbA_{1c} were collected during the study (values collected every 2 months

during the 1st year and quarterly thereafter), a time-weighted HbA_{1c} average was computed that included all values from the start of TODAY until the time of the vascular assessment.

HF power and LF power were normally distributed. SDNN and RMSSD were skewed and required log-transformation prior to testing. PNN50 was not normally distributed and transformation did not help normalize the variable so we used nonparametric testing to analyze PNN50. To compare HRV in TODAY youth to obese control subjects, we used general linear models (nonparametric rank-based test for PNN50) before and after adjustment for age, sex, race-ethnicity, smoking, and BMI. Unadjusted linear regression models were used to compare the mean of the arterial stiffness measures by presence of cardiac autonomic dysfunction.

Univariate models were used to assess the relationship between glycemic control (time-weighted HbA_{1c} or failure to maintain glycemic control during TODAY or T2P1) or randomized treatment arm and the dependent variables (HRV indices). Pearson correlations (Spearman for PNN50) were computed. Subsequently, multivariate general linear regression models (nonparametric rank-based test for PNN50) were constructed. Covariates included in the full model included time-weighted HbA_{1c}, age, sex (referent group = female), race-ethnicity (referent group = non-Hispanic white), BMI at the time of the HRV assessment, and smoking in the last month (referent group = No). Randomized treatment group and duration of diabetes were considered as additional potential covariates. Model diagnostics were used to assess multicollinearity. R^2 and P values of the models were evaluated to assess the robustness of the models.

RESULTS

Participant Characteristics

Characteristics of the 397 TODAY participants who had HRV measurements performed are shown in Table 1. HRV was measured at a median time of 7.0 years from randomization when average duration of T2D was 7.7 ± 1.5 years. At that time, TODAY participants were a mean age of 20.7 ± 2.5 years. A total of 31.7% were Hispanic; 41.6% were non-Hispanic black; 19.4% were non-Hispanic white; 7.3% were classified as other (mainly American Indian or Asian); and 64.7% were female. Also, 43% were taking an ACE inhibitor, and 14% reported taking lipid-lowering medication. At baseline for these 397 TODAY participants, the randomized treatment group was as follows: metformin (32.7%), metformin + lifestyle (33.5%), and metformin + rosiglitazone (33.8%). These participants had a mean of 27 HbA_{1c} values collected (median [minimum–maximum] was 27 [7–39]) at the time of the HRV measurements, and the mean time-weighted HbA_{1c} for the group was 7.8 ± 2.0%.

HRV and Cardiac Autonomic Function

Compared with obese control subjects, TODAY subjects were younger (mean age 20.7 ± 2.5 vs. 22.6 ± 3.7 years) and had lower LDL cholesterol (98 vs. 107 mg/dL). However, they were not different by sex, BMI, or blood pressure (35% male, 80% non-Caucasian, and BMI 36.6 ± 8.1 kg/m^2) (Supplementary Table 1). Table 2 shows differences between TODAY participants and obese control subjects for HRV measurements unadjusted and adjusted for age, sex, race-ethnicity, smoking, and BMI. After adjustment, TODAY participants had significantly reduced overall HRV (lower SDNN), including a pattern of parasympathetic loss (lower RMSSD, PNN50, and HF power) with sympathetic overdrive (higher LF) and sympathovagal imbalance (higher LF:HF ratio).

Cardiac autonomic dysfunction was defined as having three or more HRV indices at the extremes of the distribution, compared with an obese or lean control group. Cardiac autonomic dysfunction was thus present in 32/397 (8.1%) of the T2D cohort when compared with obese control subjects and in 12.6% of the T2D cohort when compared with lean control subjects. Participants with cardiac autonomic dysfunction had higher femoral PWV compared with those without autonomic dysfunction ($P = 0.0001$). Additionally, participants with cardiac autonomic dysfunction were more likely to have longer diabetes duration, albuminuria, and hypertension (see Supplementary Table 2). The presence of cardiac autonomic dysfunction was evaluated in a linear regression model of PWV femoral and was found to be independently associated with PWV femoral (parameter estimate 0.96; $P = 0.0018$). The full model (model $R^2 = 0.17$; $P < 0001$) was adjusted for age (parameter estimate 0.15; $P < 0.001$), sex (P = NS), race-ethnicity (P = NS), mean arterial pressure (parameter estimate 0.04; $P < 0.001$), and BMI (P = NS), which are risk factors we previously found to be independently associated with PWV carotid femoral in the TODAY cohort (9). PWV carotid radial (arterial stiffness in the arm), PWV femoral foot (arterial stiffness in the leg), AIx (mixed central and peripheral arterial stiffness), or BrachD (stiffness in a medium muscular artery) were not different between those with and without autonomic dysfunction.

Risk Factors and HRV

Univariate association between randomized treatment group, loss of glycemic control (defined as treatment failure or two consecutive HbA_{1c} measurements of ≥8% [≥64 mmol/mol]), time-weighted HbA_{1c}, and duration of diabetes with each of the HRV indices were evaluated (Supplementary Table 3). No association was seen between randomized treatment group and any of the HRV measurements. However, higher HbA_{1c} over time (time-weighted HbA_{1c}), loss of glycemic control, and duration of T2D were each associated with lower SDNN, RMSSD, and PNN50 (all $P < 0.05$). Adjusting for time in the study did not affect any of these associations.

Multivariate Models for HRV

Linear regression models were constructed to determine the independent effects of higher HbA_{1c} over time on HRV indices (Table 3). In models adjusted for age, race-ethnicity, sex, current smoking status, and BMI, we found HbA_{1c} over time (time-weighted HbA_{1c}) was independently associated with lower SDNN, RMSSD, and PNN50 and higher LF:HF ratio. When time-weighted mean HbA_{1c} was replaced in the model by loss of glycemic control (treatment failure or sustained elevated HbA_{1c} during TODAY or T2P1), the results were similar except for the LF:HF ratio. The relation between loss of glycemic control with LF:HF ratio was no longer significant in the multivariate model (P = 0.0932). Adding randomized diabetes treatment to the models did not change any of the associations. Older age and higher BMI were also associated with overall SDNN,

Table 1—Characteristics of the TODAY participants at the time of HRV measurements in TODAY

Variable	Value, $n = 397$
T2D duration (years)	7.7 ± 1.5
Age (years)	20.7 ± 2.5
Sex, female	64.7
Race-ethnicity	
Non-Hispanic black	31.7
Hispanic	41.6
Non-Hispanic white	19.4
Other	7.3
Cigarette smoking in the past month	20.2
BMI (kg/m^2)	36.6 ± 8.1
Systolic blood pressure (mmHg)	118.6 ± 12.3
Diastolic blood pressure (mmHg)	73.2 ± 9.8
Heart rate (beats per minute)	78.2 ± 11.3
On antihypertensive medications	43.2
Total cholesterol (mg/dL)	167.2 ± 37.0
LDL cholesterol (mg/dL)	98.1 ± 30.7
HDL cholesterol (mg/dL)	42.9 ± 12.5
Triglycerides (mg/dL)	137.3 ± 126.4
HbA_{1c} (%)	8.8 ± 2.9
Time-weighed HbA_{1c} (%)	7.8 ± 2.0

Data are mean ± SD or percent.

RMSSD, PNN50, and higher LF:HF ratio. Smoking in the last month was associated with higher LF:HF ratio only. Male sex was associated with a higher LF:HF ratio and non-Hispanic black race-ethnicity (compared with non-Hispanic white) was associated with improved RMSSD, PNN50, and LF:HF ratio at a trend level for SDNN ($P = 0.0578$).

CONCLUSIONS

In the largest cohort of adolescents and young adults with youth-onset T2D reported to date, we found evidence of cardiac autonomic dysfunction and worse HRV indices in participants with T2D compared with obese controls. HRV was independently associated with prior glycemic control over time, and cardiac autonomic dysfunction was associated with higher PWV carotid femoral.

Cardiac autonomic dysfunction is a complication of T2D and, in adults, carries an approximately fivefold increased risk of mortality (3). Damage to the autonomic innervations of the heart and blood vessels can lead to lethal arrhythmias and sudden cardiac death (26). Hyperglycemia is thought to be associated with abnormal signaling of autonomic neurons via accumulation of advanced glycation end products, activation of polyol pathway, and ischemia-induced atrophy of the autonomic nerve fibers innervating the cardiac and vascular tissue (27). Both divisions of the autonomic nervous system are typically affected, with parasympathetic impairment preceding the sympathetic dysfunction (3). Loss of HRV is one of the earliest manifestations of this process. In the Framingham Heart Study, HRV was found to be inversely associated with the risk of mortality (28). Similarly, the Atherosclerosis Risk In Communities (ARIC) study found decreased HRV was independently associated with the risk of developing coronary heart disease (29).

Compared with obese control subjects without diabetes, we found young adults with youth-onset T2D had significantly reduced overall HRV (lower SDNN), including a pattern of parasympathetic loss (lower RMSSD and PNN50) with sympathetic overdrive (higher LF:HF ratio). These differences persisted even after adjustment for differences in age, sex, race-ethnicity, smoking, and BMI. Although both groups had a mean SDNN of <70 ms, TODAY participants had a much lower mean SDNN (58.1 ± 29.6 ms) compared with obese control participants (67.1 ± 25.4 ms). To put these data in perspective, a meta-analysis of 21 studies that included nearly 3,500 adults postmyocardial infarction found an SDNN of <70 ms was associated with a four times higher risk of death compared with an SDNN >70 ms (30). Thus, the overall low mean SDNN <70 ms in this young adult cohort suggests their HRV impairment may be clinically important. Additionally, since a low SDNN is also observed in young adults with obesity, it may reflect the etiology for increased cardiovascular risk in youth-onset T2D.

We found evidence of cardiac autonomic dysfunction in 8.1% of the T2P1 cohort compared with obese control subjects. As expected, the prevalence was higher at 12.1% when compared with lean control subjects. However, this was less than that reported in the SEARCH study, where 17% of their youth with T2D were noted to have cardiac autonomic dysfunction. Differences in study design and cohort characteristics may also explain differences in the percent of adolescents with cardiac autonomic

Table 2—HRV indices in TODAY participants versus obese control subjects

Variable	TODAY, $n = 397$	Obese control subjects, $n = 133$	P value	
			Unadjusted	Adjusted
SDNN (ms)*	58.1 ± 29.6	67.1 ± 25.4	<0.0001	<0.0001
RMSSD (ms)*	53.2 ± 36.7	67.9 ± 35.2	<0.0001	<0.0001
PNN50 (%)*	26.3 ± 23.7	39.7 ± 23.0	<0.0001	<0.0001
LF Power (n.u.)†	47.3 ± 20.0	39.5 ± 19.7	0.0001	<0.0001
HF Power (n.u.)*	52.7 ± 20.0	60.5 ± 19.7	0.0001	<0.0001
LF:HF ratio†	1.4 ± 1.7	1.0 ± 1.1	<0.0001	<0.0001

Unadjusted means ± SD are shown in the table. Total power for TODAY participants was 2,576 ± 2,919. P value from general linear model comparing mean of the obese control subjects to the TODAY participants. SDNN, RMSSD, and LF:HF ratio were log transformed prior to testing because of skewed distribution. A nonparametric rank-based test was used to compare the PNN50 values. Unadjusted and adjusted P values for age, sex, race-ethnicity, smoking, and BMI are given for the cardiac autonomic function measures. n.u., normalized units. *Lower = worse. †Higher = worse.

Table 3—Determinants of HRV, $n = 397$

	SDNN (ms)*		RMSSD (ms)*		PNN50 (%)*		LF:HF Ratio†	
Variable	Estimate	*P* value	Estimate	*P* value	Estimate	*P* value	Estimate	*P* value
Intercept	5.56	<0.0001	5.82	<0.0001	546.2	<0.0001	−1.90	0.0001
Time-weighted HbA_{1c}	−0.08	<0.0001	−0.12	<0.0001	−19.4	<0.0001	0.06	0.0166
Age (years)	−0.03	0.0131	−0.04	0.0100	−7.1	0.0024	0.03	0.0920
Smoking in the past month	0.02	0.7675	−0.06	0.5028	−6.0	0.6671	0.29	0.0162
Male	0.07	0.1878	0.02	0.8384	−2.3	0.8450	0.21	0.0420
Race-ethnicity								
Non-Hispanic black	0.14	0.0578	0.37	0.0005	55.1	0.0007	−0.45	0.0012
Hispanic	−0.02	0.7243	0.05	0.5839	10.2	0.5016	−0.11	0.3907
Other	0.12	0.3013	0.19	0.2491	28.4	0.2661	−0.04	0.8568
BMI (kg/m^2)	−0.01	0.0006	−0.01	0.0026	−1.95	0.0048	0.02	0.0013
Model R^2	0.14	<0.0001	0.16	<0.0001	0.17	<0.0001	0.11	<0.0001

The full model includes a measure of glycemic control (time-weighted HbA_{1c}), sex (referent group = Female), race-ethnicity (referent group = non-Hispanic white), smoking in the past month (referent group = No), and age and BMI at the time of the HRV assessment. Parameter estimates for each covariate included in the model and the full model R^2 are given. *Lower = worse. †Higher = worse.

dysfunction. The TODAY cohort participated in a randomized controlled trial aimed at lowering HbA_{1c} and treating diabetes-related comorbidities, whereas SEARCH is an observational study. Furthermore, compared with TODAY youth, SEARCH participants were older with a mean age of 22 years and had higher HbA_{1c} levels of 9.3%, factors which were associated with cardiac autonomic dysfunction in the current analysis.

Risk factors associated with HRV measurements have not previously been reported in young adults with youth-onset T2D. Here, we show that independent effects of HbA_{1c} over time are associated with overall lower HRV, parasympathetic loss, and sympathetic overdrive. These results concur with data in adults where a recent meta-analysis showed poorer glycemic control is associated with reduced HRV (31). Although we found no differences in any of the HRV indices by prior treatment modalities used in the TODAY clinical trial (metformin alone, metformin and lifestyle, and metformin and rosiglitazone), we cannot exclude a treatment-modifying effect on our HRV measures independent of glycemia. It is also possible that longer follow-up of the cohort is needed to see the effects of diabetes treatments (metabolic memory) or that medication effects do not persist after their discontinuation.

We identified other risk factors associated with HRV in T2P1 participants. Older age and BMI were associated with overall lower HRV and individual measures of parasympathetic loss and sympathetic overdrive. Non-Hispanic black race-ethnicity was associated with less parasympathetic and sympathetic dysfunction and a trend toward improved HRV similar to findings in a recent meta-analysis in adults (32). Smoking and male sex were associated with higher LF:HF ratio or sympathetic overdrive. These data also suggest additional modifiable risk factors including BMI and smoking may be related to HRV.

Finally, we observed that cardiac autonomic function is independently associated with PWV carotid femoral after adjustment for age, race-ethnicity, sex, blood pressure, and BMI. While both are potential complications of diabetes, we postulated that since the autonomic nervous system is responsible for regulating heart rate and vascular tone, it may be related to arterial stiffness. Prior work in youth (33) and adults with type 1 diabetes, including work from the Pittsburgh Epidemiology of Diabetes Complications (34) study, have shown a direct association with cardiac autonomic function and follow-up measures of arterial stiffness independent of traditional cardiovascular disease risk factors. Furthermore, studies in adults with T2D have shown an inverse relationship between HRV and higher PWV (35,36). This study suggests that a relationship exists between cardiac autonomic function and PWV carotid femoral, a noninvasive measure of arterial stiffness in young adults with diabetes. Multiple arterial stiffness measurements were used in this study in order to comprehensively assess the vasculature because atherosclerosis develops in a nonuniform fashion (37). However, it is unclear from our work why no association between cardiac autonomic dysfunction and BrachD or AIx was observed, but the low overall percent of youth with cardiac autonomic dysfunction could be contributing.

Limitations of the current study should be noted. First, a single assessment of HRV was used, so we cannot comment on change in HRV over time or how glycemic control, prior diabetes treatment, or arterial stiffness may have influenced the progression of HRV. However, follow-up HRV measurements and arterial stiffness are planned for this cohort. In addition, although the current analysis used a relatively short length of recording (10 min) and was not able to account for control of breathing, which can influence HRV, the methods used here are considered standard practice for clinical and research purposes and are advocated by the Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology (38), as opposed to the HRV measures derived from the 24-h Holter recordings where fewer data on long-term stability are available. Second, although data related to physical activity and diet were collected in the parent TODAY study, these data were not obtained during the T2P1 follow-up phase. Alcohol consumption was obtained for participants of legal age only. For those reasons, we were unable to account for the effect of physical activity, diet, and alcohol in the analyses, but we recognize that lifestyle choices are important variables

influencing HRV (39). Thirdly, control subjects were recruited from a different study. Although HRV were obtained using the same protocol and data were adjusted for age, race-ethnicity, sex, and BMI, there could have been additional characteristics that differed between the cohorts that were not accounted for. Furthermore, in all study participants, we lacked absolute values of LF and HF power and reported only normalized values (calculated as the absolute value divided by the total variance ∗ 100), which we have found is useful to stabilize the variance of the measures in a 10-min ECG (40,41). Finally, these are cross sectional data and do not demonstrate causality. However, the major strengths of our study are the relatively large and diverse sample of contemporary youth with T2D, which allowed us to quantify cardiac autonomic dysfunction, assess HRV compared with obese control subjects, identify risk factors associated with HRV, and evaluate the relationship between cardiac autonomic function and arterial stiffness.

In conclusion, young adults with youth-onset T2D in TODAY demonstrate evidence of overall reduced HRV with parasympathetic loss and sympathetic overdrive. This appears to be related to modifiable risk factors including glycemic control, BMI, and smoking.

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References

1. Vinik AI, Maser RE, Mitchell BD, Freeman R. Diabetic autonomic neuropathy. Diabetes Care 2003;26:1553–1579
2. Maser RE, Mitchell BD, Vinik AI, Freeman R. The association between cardiovascular autonomic neuropathy and mortality in individuals with diabetes: a meta-analysis. Diabetes Care 2003;26:1895–1901
3. Vinik AI, Ziegler D. Diabetic cardiovascular autonomic neuropathy. Circulation 2007;115:387–397
4. Levitt Katz L, Gidding SS, Bacha F, et al.; TODAY Study Group. Alterations in left ventricular, left atrial, and right ventricular structure and function to cardiovascular risk factors in adolescents with type 2 diabetes participating in the TODAY clinical trial. Pediatr Diabetes 2015;16:39–47
5. Nadeau KJ, Zeitler PS, Bauer TA, et al. Insulin resistance in adolescents with type 2 diabetes is associated with impaired exercise capacity. J Clin Endocrinol Metab 2009;94:3687–3695
6. Shah RV, Abbasi SA, Neilan TG, et al. Myocardial tissue remodeling in adolescent obesity. J Am Heart Assoc 2013;2:e000279
7. Whalley GA, Gusso S, Hofman P, et al. Structural and functional cardiac abnormalities in adolescent girls with poorly controlled type 2 diabetes. Diabetes Care 2009;32:883–888
8. Shah AS, Khoury PR, Dolan LM, et al. The effects of obesity and type 2 diabetes mellitus on cardiac structure and function in adolescents and young adults. Diabetologia 2011;54:722–730
9. Shah AS, El Ghormli L, Gidding SS, et al. Prevalence of arterial stiffness in adolescents with type 2 diabetes in the TODAY cohort: relationships to glycemic control and other risk factors. J Diabetes Complications 2018;32:740–745
10. Shah AS, Urbina EM. Vascular and endothelial function in youth with type 2 diabetes mellitus. Curr Diab Rep 2017;17:36
11. Urbina EM, Dolan LM, McCoy CE, Khoury PR, Daniels SR, Kimball TR. Relationship between elevated arterial stiffness and increased left ventricular mass in adolescents and young adults. J Pediatr 2011;158:715–721
12. Urbina EM, Kimball TR, Khoury PR, Daniels SR, Dolan LM. Increased arterial stiffness is found in adolescents with obesity or obesity-related type 2 diabetes mellitus. J Hypertens 2010;28:1692–1698
13. Urbina EM, Kimball TR, McCoy CE, Khoury PR, Daniels SR, Dolan LM. Youth with obesity and obesity-related type 2 diabetes mellitus demonstrate abnormalities in carotid structure and function. Circulation 2009;119:2913–2919
14. Dabelea D, Stafford JM, Mayer-Davis EJ, et al.; SEARCH for Diabetes in Youth Research Group. Association of type 1 diabetes vs type 2 diabetes diagnosed during childhood and adolescence with complications during teenage years and young adulthood. JAMA 2017;317:825–835
15. Faulkner MS, Quinn L, Fritschi C. Microalbuminuria and heart rate variability in adolescents with diabetes. J Pediatr Health Care 2010;24:34–41
16. Faulkner MS, Quinn L, Rimmer JH, Rich BH. Cardiovascular endurance and heart rate variability in adolescents with type 1 or type 2 diabetes. Biol Res Nurs 2005;7:16–29
17. Jaiswal M, Divers J, Urbina EM, et al.; SEARCH for Diabetes in Youth Study Group. Cardiovascular autonomic neuropathy in adolescents and young adults with type 1 and type 2 diabetes: the SEARCH for Diabetes in Youth Cohort Study. Pediatr Diabetes 2018;19:680–689
18. Zeitler P, Epstein L, Grey M, et al.; TODAY Study Group. Treatment options for type 2 diabetes in adolescents and youth: a study of the comparative efficacy of metformin alone or in combination with rosiglitazone or lifestyle intervention in adolescents with type 2 diabetes. Pediatr Diabetes 2007;8:74–87
19. Zeitler P, Hirst K, Pyle L, et al.; TODAY Study Group. A clinical trial to maintain glycemic control in youth with type 2 diabetes. N Engl J Med 2012;366:2247–2256
20. Bacha F, Pyle L, Nadeau K, et al.; TODAY Study Group. Determinants of glycemic control in youth with type 2 diabetes at randomization in the TODAY study. Pediatr Diabetes 2012;13:376–383
21. Pottel H, Hoste L, Dubourg L, et al. An estimated glomerular filtration rate equation for the full age spectrum. Nephrol Dial Transplant 2016;31:798–806
22. Fagard RH, Pardaens K, Staessen JA, Thijs L. Power spectral analysis of heart rate variability by autoregressive modelling and fast Fourier transform: a comparative study. Acta Cardiol 1998;53:211–218

23. Pagani M, Lombardi F, Malliani A. Heart rate variability: disagreement on the markers of sympathetic and parasympathetic activities. J Am Coll Cardiol 1993;22:951–953
24. Laurent S, Cockcroft J, Van Bortel L, et al.; European Network for Non-Invasive Investigation of Large Arteries. Expert consensus document on arterial stiffness: methodological issues and clinical applications. Eur Heart J 2006;27:2588–2605
25. Urbina EM, Brinton TJ, Elkasabany A, Berenson GS. Brachial artery distensibility and relation to cardiovascular risk factors in healthy young adults (The Bogalusa Heart Study). Am J Cardiol 2002;89:946–951
26. Ewing DJ, Boland O, Neilson JM, Cho CG, Clarke BF. Autonomic neuropathy, QT interval lengthening, and unexpected deaths in male diabetic patients. Diabetologia 1991;34:182–185
27. Verrotti A, Loiacono G, Mohn A, Chiarelli F. New insights in diabetic autonomic neuropathy in children and adolescents. Eur J Endocrinol 2009;161:811–818
28. Tsuji H, Larson MG, Venditti FJ Jr., et al. Impact of reduced heart rate variability on risk for cardiac events. The Framingham Heart Study. Circulation 1996;94:2850–2855
29. Liao D, Cai J, Rosamond WD, et al. Cardiac autonomic function and incident coronary heart disease: a population-based case-cohort study. The ARIC Study. Am J Epidemiol 1997;145:696–706
30. Buccelletti E, Gilardi E, Scaini E, et al. Heart rate variability and myocardial infarction: systematic literature review and metanalysis. Eur Rev Med Pharmacol Sci 2009;13:299–307
31. Benichou T, Pereira B, Mermillod M, et al. Heart rate variability in type 2 diabetes mellitus: a systematic review and meta-analysis. PLoS One 2018;13:e0195166
32. Hill LK, Hu DD, Koenig J, et al. Ethnic differences in resting heart rate variability: a systematic review and meta-analysis. Psychosom Med 2015;77:16–25
33. Jaiswal M, Urbina EM, Wadwa RP, et al. Reduced heart rate variability is associated with increased arterial stiffness in youth with type 1 diabetes: the SEARCH CVD study. Diabetes Care 2013;36:2351–2358
34. Prince CT, Secrest AM, Mackey RH, Arena VC, Kingsley LA, Orchard TJ. Cardiovascular autonomic neuropathy, HDL cholesterol, and smoking correlate with arterial stiffness markers determined 18 years later in type 1 diabetes. Diabetes Care 2010;33:652–657
35. Chorepsima S, Eleftheriadou I, Tentolouris A, et al. Pulse wave velocity and cardiac autonomic function in type 2 diabetes mellitus. BMC Endocr Disord 2017;17:27
36. Bagherzadeh A, Nejati-Afkham A, Tajallizade-Khoob Y, et al. Association of cardiac autonomic neuropathy with arterial stiffness in type 2 diabetes mellitus patients. J Diabetes Metab Disord 2013;12:55
37. Solberg LA, Eggen DA. Localization and sequence of development of atherosclerotic lesions in the carotid and vertebral arteries. Circulation 1971;43:711–724
38. Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology. Heart rate variability: standards of measurement, physiological interpretation and clinical use. Circulation 1996;93:1043–1065
39. Carnethon MR, Prineas RJ, Temprosa M, Zhang ZM, Uwaifo G, Molitch ME; Diabetes Prevention Program Research Group. The association among autonomic nervous system function, incident diabetes, and intervention arm in the Diabetes Prevention Program. Diabetes Care 2006;29:914–919
40. Malliani A, Pagani M, Lombardi F, Cerutti S. Cardiovascular neural regulation explored in the frequency domain. Circulation 1991;84:482–492
41. Pagani M, Lombardi F, Guzzetti S, et al. Power spectral analysis of heart rate and arterial pressure variabilities as a marker of sympathovagal interaction in man and conscious dog. Circ Res 1986;59:178–193