Baroreflex Sensitivity Predicts Cardiovascular Events in Patients With Type 2 Diabetes Mellitus Without Structural Heart Disease

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**Background:** Cardiovascular autonomic neuropathy is a major complication in patients with diabetes mellitus (DM). However, the relationship between cardiovascular autonomic neuropathy and the incidence of cardiovascular events has been poorly investigated in type 2 DM. The present study aimed to assess the long-term cardiovascular predictive value of baroreflex sensitivity (BRS) in Japanese patients with type 2 DM without structural heart disease.

**Methods and Results:** BRS was evaluated using the phenylephrine method in 210 patients with type 2 DM who did not have structural heart disease or other severe complications. BRS was considered depressed if <6 ms/mmHg. Accurate follow-up information for 3–10 years (mean 4.7 years) was obtained in 184 patients (90 females, 94 males; mean age 58±12 years). The initial onset of a major adverse cardiovascular event (MACE) was investigated. During follow-up, 19 patients presented with a MACE (4 cardiovascular deaths, 3 nonfatal myocardial infarctions, 4 coronary revascularizations, 5 strokes, 2 congestive heart failures). Cox proportional hazards regression analysis revealed that depressed BRS was independently associated with the incidence of MACE (hazard ratio 1.93, 95% confidence interval 1.09–3.82, P=0.0236).

**Conclusions:** Depressed BRS at baseline has long-term cardiovascular predictive value in Japanese patients with type 2 DM without structural heart disease. (Circ J 2010; 74: 1379–1383)

**Key Words:** Autonomic nervous system; Baroreflex sensitivity; Cardiovascular event; Diabetes mellitus

Evaluation of baroreflex sensitivity (BRS), obtained by measuring changes in heart rate in response to changes in blood pressure (BP) induced by vasoactive drugs, is an established tool for assessing autonomic control of the cardiovascular system. Early experimental studies using dogs demonstrated that myocardial infarction (MI) attenuates baroreflex control of heart rate and that depressed BRS following MI identifies subgroups of dogs at higher risk for ventricular fibrillation and sudden death. Those experimental studies led to the ATRAMI (Autonomic Tone and Reflexes After Myocardial Infarction) study, which clinically demonstrated that, independent of left ventricular ejection fraction, depressed BRS using the phenylephrine method was an independent predictor of cardiac mortality in patients with previous MI. Diabetes mellitus (DM) is known to impair cardiovascular autonomic function, termed “cardiovascular autonomic neuropathy”. In our series of clinical studies investigating BRS using the phenylephrine method in patients with type 2 DM without structural heart disease, the presence of essential hypertension, microalbuminuria, high levels of either high-sensitivity C-reactive protein or brain natriuretic peptide, or hypoadiponectinemia was associated with low BRS. However, the relationship between cardiovascular autonomic neuropathy and the incidence of cardiovascular events has been poorly investigated in patients with type 2 DM. In fact, there have been no Japanese reports on the relationship between cardiovascular autonomic neuropathy and the incidence of cardiovascular events. We tested the hypothesis that depressed BRS could be a long-term cardiovascular predictor in Japanese patients with type 2 DM and no structural heart disease.

**Methods**

**Patient Selection**
A total of 210 patients with type 2 DM who were admitted to...
Table 1. Baseline Patient Characteristics

<table>
<thead>
<tr>
<th></th>
<th>Preserved BRS (n=99)</th>
<th>Depressed BRS (n=85)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>55.6±10.5</td>
<td>60.6±12.0</td>
<td>0.0031**</td>
</tr>
<tr>
<td>Sex (F/M)</td>
<td>36/63</td>
<td>54/31</td>
<td>0.0002**</td>
</tr>
<tr>
<td>Body mass index (kg/m²)</td>
<td>25.2±5.2</td>
<td>25.8±5.8</td>
<td>0.5352</td>
</tr>
<tr>
<td>Current smoker (%)</td>
<td>43.5</td>
<td>57.6</td>
<td>0.0570</td>
</tr>
<tr>
<td>Duration of diabetes (years)</td>
<td>9.1±7.9</td>
<td>10.7±8.1</td>
<td>0.1796</td>
</tr>
<tr>
<td>Fasting plasma glucose (mg/dl)</td>
<td>147±45</td>
<td>159±53</td>
<td>0.1087</td>
</tr>
<tr>
<td>Hemoglobin A₁c (%)</td>
<td>8.1±1.8</td>
<td>8.3±1.9</td>
<td>0.3528</td>
</tr>
<tr>
<td>Hypertension (%)</td>
<td>47.5</td>
<td>64.7</td>
<td>0.0186</td>
</tr>
<tr>
<td>Systolic BP (mmHg)</td>
<td>121±18</td>
<td>133±20</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Diastolic BP (mmHg)</td>
<td>67±13</td>
<td>70±12</td>
<td>0.0441</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>68±8.0</td>
<td>70±9.0</td>
<td>0.0766</td>
</tr>
<tr>
<td>Dyslipidemia (%)</td>
<td>71.7</td>
<td>74.1</td>
<td>0.7149</td>
</tr>
<tr>
<td>Hyperuricemia (%)</td>
<td>23.2</td>
<td>21.2</td>
<td>0.7381</td>
</tr>
<tr>
<td>Chronic kidney disease (%)</td>
<td>21.2</td>
<td>31.7</td>
<td>0.1044</td>
</tr>
<tr>
<td>BRS (ms/mmHg)</td>
<td>11.5±5.1</td>
<td>3.11±1.8</td>
<td>&lt;0.0001**</td>
</tr>
</tbody>
</table>

Data are mean±SD. **P<0.01.
BRS, baroreflex sensitivity; BP, blood pressure.

Table 2. Number of Patients Who Developed Study Endpoints

<table>
<thead>
<tr>
<th></th>
<th>Preserved BRS (n=99)</th>
<th>Depressed BRS (n=85)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cardiovascular death</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>Nonfatal MI</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>Coronary revascularization</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>Stroke</td>
<td>1</td>
<td>4</td>
</tr>
<tr>
<td>Congestive heart failure</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Total</td>
<td>4</td>
<td>15</td>
</tr>
</tbody>
</table>

MI, myocardial infarction. Other abbreviation see in Table 1.

Oita University Hospital from 1998 to 2004 for blood glucose control were recruited. Patients were excluded if they were more than 80 years old. Type 2 DM was defined as a fasting plasma glucose (FPG) concentration ≥126 mg/dl, a 2-h plasma glucose concentration following a 75-g oral glucose load ≥200 mg/dl, or the self-reported use of antidiabetic medication. None of the patients had organic heart disease as determined by physical examination, chest X-ray, 12-lead ECG, echocardiography, and thallium cardiac scintigraphy. Myocardial ischemia was excluded by treadmill exercise ECG testing. Essential hypertension was defined as diastolic BP ≥90 mmHg, systolic BP ≥140 mmHg or self-reported use of antihypertensive medication. Patients treated with α- or β-adrenergic blocking agents, antiplatelet agents or with macroalbuminuria (≥500 mg/day) or abnormal plasma creatinine concentrations (≥1.2 mg/dl) were also excluded from the study. Dyslipidemia was defined as fasting triglycerides >200 mg/dl, or high-density lipoprotein cholesterol ≤45 mg/dl in women and ≤35 mg/dl in men. Hyperuricemia was defined as uric acid >6.0 mg/dl in women or >7.0 mg/dl in men. Chronic kidney disease (CKD) was defined as an estimated glomerular filtration rate (eGFR) <60 ml·min⁻¹·1.73 m⁻². Patients with an eGFR <30 ml·min⁻¹·1.73 m⁻² were excluded.

This investigation was conducted according to the principles expressed in the Declaration of Helsinki. Prior informed consent in terms of BRS measurement was obtained from all patients, and the study protocol was approved by the Institutional Review Board of Oita University.

BRS Measurement
For BRS assessment, all subjects were studied while lying supine in a quiet room between 9 and 11 am. A catheter was inserted into the right cubital vein, and arterial BP was recorded noninvasively using tonometry (Jentow-7700; Nihon Colin, Komaki, Japan), while 12-lead ECG was simultaneously monitored. Data were stored in a PCM data recorder (RD-200T; TEAC, Tokyo, Japan). After a 30-min interval to allow stabilization, the patient was asked to breathe at a rate of 15 breaths/min using a metronome. BRS was assessed using the phenylephrine method. Phenylephrine (2–3 μg/kg) was injected over 15 s to increase systolic BP by 15–40 mmHg. BRS was calculated as the slope of the linear regression line relating the systolic BP changes to the RR interval changes. Regression lines with more than 20 data points and a correlation coefficient (r) >0.8 were accepted for analysis. The mean of the 2 slope values was taken as the BRS value. BRS was considered depressed if it was <6 ms/mmHg.

Follow-up
Most of the follow-up of patients occurred at Oita University Hospital. Information was obtained for those patients whose follow-up was performed by a general practitioner and for those hospitalized in other departments. For the patients who died, the cause of death was documented with the help of the patients’ family and general practitioner.

The study endpoint was the occurrence of a major adverse cardiovascular event (MACE), which included cardiovascular mortality, nonfatal MI, coronary revascularization by angioplasty or bypass, stroke and congestive heart failure requiring admission. Using this criterion, only the first event was taken into account in the statistical analysis.

Statistical Analysis
Data are presented as mean±SD. The chi-square test was used for categorical variables, and the analysis of variance test was used for continuous variables. Kaplan-Meier MACE-free analysis was used to compare MACE-free times between the preserved and depressed BRS groups. To test for differences between the resulting curves, the log-rank test was used. Univariate and multivariate stepwise Cox proportional hazard
regression analyses were performed to identify independent predictors (risk factors) of MACE. Risk factors entered into the risk model included age, sex, body mass index (BMI), smoking status, duration of DM, FPG, hemoglobin A\textsubscript{1c} (HbA\textsubscript{1c}), hypertension, dyslipidemia, hyperuricemia, CKD, and BRS. Results are given as hazard ratios (HR) with 95% confidence intervals (CI). P<0.05 was considered significant. All computations were performed with SPSS software version 12.0.1 (SPSS Inc, Chicago, IL, USA), running under Windows 2003 XP (Microsoft, Redmond, WA, USA).

### Results

#### Patient Characteristics

Of the 210 patients enrolled, we obtained accurate follow-up information for 184 patients (90 females, 94 males; mean age 58±12 years (31–79 years)). Baseline patient characteristics of 2 BRS groups are presented in Table 1. The mean age was higher in the depressed BRS group than in the preserved BRS group (P<0.01). The number of current smokers tended to be higher in the depressed BRS group than in the preserved BRS group, but did not reach statistical significance. Females and patients with hypertension were largely included in the depressed BRS group (P<0.01 and P<0.05, respectively), while no significant difference was observed between the 2 groups in the number of patients with dyslipidemia. Regarding DM, no significant difference was observed in terms of duration, FPG or HbA\textsubscript{1c}. The number of patients with hyperuricemia or CKD did not differ between the 2 groups.

#### Patient Outcomes

During the follow-up, 8 patients died (4.3%): 4 were cardiovascular deaths, and 4 were attributed to non-cardiovascular causes. The all-cause mortality rate was not significantly different between groups. Total cardiovascular mortality (1.0% (1/99) in the preserved BRS group) was not significantly different from the 3.5% (3/85) in the depressed BRS group.

#### Univariate and Multivariate Predictors of MACE

During the follow-up, 19 patients presented with a MACE (10.3%): 4 cardiovascular deaths, 3 nonfatal MIs, 4 coronary revascularizations, 5 strokes, and 3 congestive heart failures.
MACE occurred more frequently in the depressed BRS group (17.6%) than in the preserved BRS group (4.0%, P<0.01). Results of univariate and multivariate Cox proportional hazards regression analysis of MACE are presented in Table 3. Univariate analysis revealed that female sex, BMI, the prevalence of hypertension, dyslipidemia, CKD, and depressed BRS were associated with MACE. Based on multivariate analysis, among risk factors including age, sex, BMI, smoking status, duration of DM, FPG, HbA1c, hypertension, dyslipidemia, hyperuricemia, CKD, and depressed BRS, only depressed BRS independently predicted the incidence of MACE (HR 1.93, 95%CI 1.09–3.82, P=0.0236).

Kaplan-Meier MACE-Free Estimation

The MACE-free ratio as evaluated by Kaplan-Meier analysis was significantly higher in the preserved BRS group than in the depressed BRS group (log-rank 9.63, P=0.002) (Figure).

Discussion

In the present study, 184 Japanese patients with type 2 DM were followed for a mean period of 4.7 years, and MACE occurred in 19 patients. The most important finding is that among the 12 risk factors studied only depressed BRS was independently associated with the development of MACE. To the best of our knowledge, this report is the first to demonstrate the long-term cardiovascular predictive value of BRS measured using the phenylephrine method in Japanese type 2 DM patients without structural heart disease or severe complications. The cut-off value of <6 ms/mmHg was used to define depressed BRS, because the ATRAMI study categorized post-MI patients into 3 groups: BRS <3, BRS 3.0–6.1, and BRS >6.1. By using a cut-off index of 6 ms/mmHg, 85 of 184 total patients (46%) were classified into the depressed BRS group, and 15 of the 19 patients (79%) who developed MACE were included in the depressed BRS group. These observations appear to support the validity of our definition of depressed BRS (<6 ms/mmHg). Cardiovascular autonomic function tests in diabetic patients have been generally performed using the Ewing method. A recently standardized method that includes 3 tests for heart rate variability, including the expiration-to-inspiration ratio, and responses to the Valsalva maneuver and to postural changes from lying to standing has been recommended by the American Diabetes Association. We did not intend to demonstrate the superiority of BRS over other noninvasive tests; however, the need for intravenous cannulation and use of a vasoactive drug appear to be impracticable in the daily clinical setting. In fact, analysis of “spontaneous” oscillations in BP and RR intervals has been developed as a noninvasive method of evaluating BRS.

Total and cardiovascular mortality of the 184 patients for a mean of 4.7 years was 4.3% and 2.7%, respectively. These low levels are consistent with our strict inclusion criteria. Approximately 30 years ago, Ewing et al reported a 5-year mortality rate of 53% in diabetic patients with abnormal autonomic function tests compared with a mortality rate of 15% in those with normal autonomic function test results. Most patients with abnormal autonomic function tests also suffered from postural hypotension, intermittent diarrhea, hypoglycemic unawareness, sweating abnormalities and gastric fullness, and half of the deaths were from renal failure, suggesting that they recruited patients with severe complications. Similar to our study, Rathmann et al assessed the risk of mortality in diabetic patients without clinical manifestations, including proteinuria, proliferative retinopathy, coronary artery disease or stroke, and reported that diabetic autonomic neuropathy increased the mortality rate from 2.9% to 14% after 4 years, which is still higher than the mortality rate in the present study.

The relationship between cardiovascular autonomic neuropathy and MACE has been poorly investigated. Valensi et al recruited 108 diabetic patients, in whom cardiovascular autonomic neuropathy was assessed by the Ewing method. Major cardiac events (cardiac death, nonfatal MI, congestive heart failure, resuscitation from ventricular tachycardia/fibrillation and need for coronary revascularization) occurred in 11 (10.2%) during a mean follow-up period of 4.5 years. Of the 11 events, 8 (24.2%) occurred in patients with neuropathy while 3 (7.1%) occurred in patients without neuropathy, which appears to be similar to our observations.

A worsened outcome for diabetic patients with cardiovascular autonomic neuropathy who have had an acute MI has been established. Sudden cardiac death and ventricular tachycardia/fibrillation are major events in this population. In contrast, in our study most cases of MACE were MI, stroke, or coronary revascularization. Sudden cardiac death or ventricular tachycardia/fibrillation did not occur in any of the present patients. Taken together, the findings suggest that type 2 DM patients without structural heart disease do not have the arrhythmic background for developing life-threatening ventricular tachyarrhythmias. Rather, an association of diabetic cardiovascular autonomic neuropathy with an increased incidence of stroke has been reported.

The American Diabetes Association recommends low-dose aspirin as a primary prevention strategy for diabetic patients at high risk for cardiovascular events on the basis of following risk factors, including family history of cardiovascular disease, cigarette smoking, obesity, macro- and microalbuminuria, and dyslipidemia. Based on our observations, we suggest that patients with cardiovascular autonomic neuropathy should be recognized as at high risk for cardiovascular events. Early aspirin may be recommended in this population, although our patients did not take any antiplatelet agents at the time of enrollment. Interestingly, a recent study in experimentally induced hypercholesterolemic rats demonstrated that aspirin improves the baroreflex response and prevents a rise in BP and heart rate, possibly by reducing sympathetic activity because of its antioxidant effect. The effects of aspirin and other drugs that have been suggested to improve baroreflex response, including angiotensin-converting enzyme inhibitors and β-blockers, should be evaluated to determine whether they could prevent MACE in diabetic patients with cardiovascular autonomic neuropathy.

Study Limitations

First, because we enrolled patients without severe complications, the incidence of MACE was low, which might make it difficult to analyze the data. Conversely, most of the patients with preserved BRS were free from MACE. Thus, compared with the positive predictive value of depressed BRS (17.6%), the negative predictive value of preserved BRS (96.0%) may be more clinically relevant. Second, the clinical manifestations of diabetic autonomic neuropathy is multifaceted, including the cardiovascular, gastrointestinal, genitourinary, metabolic, sudomotor, and papillary systems. Because we did not quantitatively evaluate other autonomic neuropathies, the association between them and BRS remains unclear. Third, we excluded patients with eGFR <30 ml/min/1.73 m², mac-
Conclusion

Our observations suggest that baseline assessment of BRS using the phenylephrine method may be a long-term cardiovascular predictor in type 2 diabetic patients without structural heart disease or severe complications. Intensive management and/or therapy may be needed in this population.

References


