The Association of Heart-Rate Variability With Cardiovascular Risk Factors and Coronary Artery Calcification

A study in type 1 diabetic patients and the general population

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OBJECTIVE — To examine the association of heart-rate variability with cardiovascular risk factors and coronary calcification in type 1 diabetic and nondiabetic subjects without a history of cardiovascular disease. Reduced heart-rate variability is associated with increased risk of coronary events. Whether it is associated with coronary atherosclerosis is unknown.

RESEARCH DESIGN AND METHODS — Power spectral analysis was used to define heart-rate variability in a cross-sectional study of 160 type 1 diabetic patients and 163 randomly selected nondiabetic adults from the general population aged 30–55 years. Coronary artery calcification was measured using electron beam–computed tomography.

RESULTS — Reduced heart-rate variability was associated with similar risk factors in the diabetic and nondiabetic subjects, namely higher HbA_{1c}, triglycerides, systolic blood pressure, BMI, and albumin excretion rate. Reduced heart-rate variability was significantly associated with coronary artery calcification in all subjects (odds ratio per tertile lower total power = 1.5, P = 0.01). This association was not independent of blood pressure or BMI (odds ratio on adjustment = 1.3, P = 0.1).

CONCLUSIONS — Reduced heart-rate variability clusters with other cardiovascular disease risk factors, especially those that are more common in the insulin resistance syndrome, and is associated with increased coronary calcification in asymptomatic young adults. Whether reduced heart-rate variability leads to other risk factor disturbances or mediates the effects of other risk factors on atherosclerosis deserves further study.

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Heart-rate variability is a measure of cardiac autonomic function. Broadly speaking, high-frequency oscillations in heart rate reflect parasympathetic input to variability, whereas low-frequency oscillations reflect sympathetic input. Reduced heart-rate variability is indicative of reduced autonomic modulation of heart rate, although it does not necessarily indicate reduced nerve firing or reduced autonomic tone (1,2).

In the nondiabetic aging population, reduced heart-rate variability is associated with an increased incidence of cardiac events (3,4). Other measures of cardiac autonomic dysfunction are associated with an increased incidence of cardiac events in type 1 diabetic patients (5). The suggested mechanisms by which cardiac autonomic dysfunction leads to increased cardiac events include arrhythmia, cardiac failure, and impaired myocardial blood flow. Whether autonomic dysfunction is involved in the pathogenesis of atherosclerosis itself has received little attention but would have important implications for our understanding of the pathogenesis of coronary atherosclerosis in diabetic patients. A direct effect of autonomic dysfunction on atherosclerosis is certainly plausible. Sympathetic denervation may cause dedifferentiation of vascular smooth muscle cells and alteration to a synthetic phenotype (6,7). This phenotype is associated with extracellular matrix production and migration to the intima, changes that have been observed in atherosclerosis (8). Other possible mechanisms include vascular stiffening, which may be an independent risk factor for atherosclerosis (9,10).

The purpose of this study was to examine the association between heart-rate variability and coronary heart disease risk factors and coronary artery calcification (CAC). CAC is highly correlated (r = 0.9) with the degree of atherosclerosis in the coronary arteries and is accurately measured by electron beam–computed tomography (EBCT) (11,12). The study was performed in young type 1 diabetic and nondiabetic adults without ischemic heart disease, which allowed us to examine whether any association was a specific feature of diabetes. Because type 1 diabetes is associated with a much greater elevation of coronary risk in women than in men (13), a particular focus of the study was to establish whether there is a more adverse effect of diabetes on heart-rate variability in women than in men.

RESEARCH DESIGN AND METHODS

— A random sample of type 1 diabetic men and women aged 30–55 years was taken from the diabetes registers of five hospitals in London. Type

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Abbreviations: CAC, coronary artery calcification; EBCT, electron beam–computed tomography; ECG, electrocardiography.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

1 diabetes was defined by age of onset ≤25 years and need for insulin treatment within 1 year of diagnosis. A random sample of the nondiabetic general population, stratified for age and sex to be similar to the distribution of diabetic patients, was drawn from the lists of two general medical practices in London. Subjects were included regardless of any history of heart disease; only one subject had a history of angina and no subjects had a history of infarction. A total of 199 type 1 diabetic patients (95 women) and 201 nondiabetic subjects (107 women) were examined. A higher proportion of the nondiabetic than diabetic subjects were in the manual social class (25 vs. 15%, $P = 0.02$). The detailed methods and comparison of calcification data in the diabetic and nondiabetic subjects have been published elsewhere (14). Of the 400 subjects who underwent EBCT scanning, electrocardiography (ECG) was performed for analysis of heart-rate variability in 323. ECG data were not collected in 55 subjects because the ECG recording equipment was not available for part of the study, and in the remaining 22 patients, recordings were unusable because of excessive movement during the recording. The 323 subjects for whom heart-rate variability data were available did not differ from the 77 subjects in whom these data were unavailable in terms of prevalence of calcification (44 vs. 39%, $P = 0.4$), sex (% female = 52 vs. 46%, $P = 0.3$), or diabetes (50 vs. 51%, $P = 0.9$). Approval of the ethics committee was obtained, and all subjects gave written consent after receiving details of study procedures and risks in written and oral form.

History and examination

Respondents completed a standardized questionnaire regarding medical history, use of medications, and lifestyle. The average weekly consumption of alcohol units was calculated, and smoking exposure was quantified as pack-years. The duration and intensity of weekly walking, cycling, sporting, and occupational activity was used to define low or high levels of physical activity (<10 vs. ≥10 MJ energy expenditure per week) (15). Blood pressure was recorded three times after 5 min of rest using an Omron 705c oscillometric device (Omron, Tokyo, Japan); the mean of the second and third readings was used. A fasting blood sample was obtained. Obesity was defined as a BMI ≥30 kg/m^2. Waist circumference was measured midway between the iliac crest and the lower rib. Hip circumference was measured at the level of the greater trochanters.

Heart-rate variability

Approximately 30 min after eating a light breakfast, subjects rested for 5–10 min in a supine position in a quiet room. Subjects were asked to relax but not fall asleep. A 5-min ECG recording was obtained using a lead ensuring a prominent R wave. The ECG signal was digitized at 1,000 Hz using a computer with an analog-to-digital card (AT-MIO-16E2). The R-R intervals were sampled at 5 Hz, with premature beats identified and corrected by linear interpolation with the previous and following beats. Autoregressive power spectral analysis was used to determine the spectral power in the following bands: high frequency (0.15–0.45 Hz), low frequency (0.04–0.15 Hz), and very low frequency (0.01–0.04 Hz) (16). It has been suggested that the ratio of high- to low-frequency band power may confer additional information on parasympathetic to sympathetic imbalance; therefore, this ratio was also calculated.

EBCT scan

An ultrafast CT scanner (Imatron C-150XL; Imatron, San Francisco, CA) was used to measure coronary calcification. Two sets of 20 transverse tomograms of 3-mm thickness were obtained from the lower margin of the bifurcation of the right branch of the pulmonary artery to the apex of the heart during two breaths. The radiation exposure was <1 mSv. A radiologist, blinded to the sex and diabetes status of the subject, placed a region of interest around each potentially calcific lesion (peak density >130 Hounsfield units) within right coronary, circumflex, left anterior descending, and left main coronary arteries. The area and peak density in Hounsfield units of each lesion were measured. A density score of 1–4 was defined, based on the peak density of the lesion, and the Agatston calcification score was then calculated as the product of the area of the lesion and its density score, as described previously (17). To be included in the calcification score, the area of the lesion had to be at least 0.51 mm^2 (i.e., two contiguous pixels) and the peak density had to be at least 130 Hounsfield units. A total score for each artery and for the entire heart was calculated by summing the lesion scores.

Laboratory methods

Fasting total cholesterol, HDL cholesterol, and triglyceride levels were measured using enzymatic colorimetric methods (18,19). HDL cholesterol was measured directly after stabilization of other lipoproteins (20). LDL cholesterol was calculated as described by Friedewald et al. (21). Three diabetic subjects and five nondiabetic subjects had triglyceride levels >4.5 mmol/l, invalidating the calculated LDL cholesterol levels. The LDL cholesterol data for these subjects have been excluded from Table 1. HbA_{1c} was measured using a latex-enhanced immunoassay. Respondents completed two timed overnight urine collections. Urinary albumin was measured using an immunoturbidimetric method.

Statistical analysis

All analyses were performed using Stata 5 Software (Stato, College Station, TX). Heart-rate variability was compared across the four diabetes/sex groups. Because these data had non-normal distributions, the medians and interquartile ranges were presented. Variables with skewed distributions were normalized using log-transformation. Differences in heart-rate variability between diabetic and nondiabetic respondents were then tested using multiple linear regression, adjusting for age. We tested whether the effect of diabetes on heart-rate variability differed by sex by including a diabetes-by-sex interaction term in these regression models. This is equivalent to testing whether the sex difference in heart-rate variability differs by diabetes status.

Calcification scores (for the total heart) were positively skewed with a high frequency of zero values. Because data transformation would not have normalized this distribution, we used logistic regression to examine the odds of having any calcification (a score >0) associated with the total spectral power. We then adjusted for other risk factors in this model. The association between the high-frequency:low-frequency band power ratio and calcification was examined in the same way.

RESULTS

— The distribution of risk factors by diabetes and sex is shown in Table 1. Diabetic men and women had

Table 1—*Risk factors, spectral power, and other characteristics by diabetes and sex*

	Men		Women	
	Nondiabetic	Diabetic	Nondiabetic	Diabetic
n	74	82	89	78
Mean (SEM)				
Age (yr)	37.7 (0.5)	38.4 (0.5)	37.8 (0.4)	37.3 (0.5)
Diabetes duration (yr)		23.8 (0.9)		23.9 (0.8)
HbA_{1c}% (g)	5.3 (0.05)	8.3 (0.14)‡	5.3 (0.04)	8.9 (0.20)‡
BMI (kg/m^2)	25.2 (0.4)	25.6 (0.3)	25.5 (0.6)	25.6 (0.4)
Waist-to-hip ratio	0.92 (0.01)	0.91 (0.01)	0.81 (0.01)	0.82 (0.01)
HDL (mmol/l)	1.57 (0.05)	1.70 (0.04)†	1.83 (0.04)	1.94 (0.06)*
LDL (mmol/l)	3.3 (0.11)	3.0 (0.1)*	2.96 (0.08)	2.84 (0.1)
Total cholesterol/HDL ratio	3.8 (0.14)	3.2 (0.11)‡	3.1 (0.1)	2.9 (0.1)
Triglyceride [mmol/l (g)]	1.4 (0.09)	1.2 (0.06)*	1.0 (0.05)	0.92 (0.04)
Systolic blood pressure (mmHg)	124 (1.5)	128 (1.3)*	112 (1.3)	120 (1.6)‡
Diastolic blood pressure (mmHg)	76 (1.1)	76 (0.9)	69 (0.9)	72 (1.0)*
% (SEM)				
Obese	8 (3)	7 (3)	21 (4)	10 (3)†
AER ≥ 20 μg/min	4 (2)	22 (5)†	3 (2)	8 (3)
Ever smoked	59 (6)	51 (6)	53 (5)	45 (6)
Exercise score <10	75 (5)	78 (4)	86 (4)	85 (4)
Median (interquartile range)				
High-frequency power (ms^2)	253 (495)	149 (276)†	389 (484)	127 (190)‡
Low-frequency power (ms^2)	505 (583)	297 (573)†	457 (589)	171 (323)‡
Very-low-frequency power (ms^2)	420 (507)	266 (422)†	412 (609)	162 (268)†
Total power (ms^2)	1,349 (955)	697 (883)†	1,365 (1,056)	491 (567)‡
Heart rate (beats/min)	64 (11)	67 (18)†	64 (13)	72 (13)‡
High frequency/low frequency ratio	0.53 (0.7)	0.45 (0.5)	0.94 (0.8)	0.71 (0.98)

Data are means (SE). *$P < 0.05$, †$P < 0.01$, ‡$P < 0.001$ for the difference between subjects of the same sex with and without diabetes adjusted for age using regression analysis. Interquartile range defines the magnitude of the difference between the 25th and 75th centiles.

significantly higher blood pressures than nondiabetic subjects. However, the lipid profile of diabetic subjects, particularly diabetic men, was better than nondiabetic subjects.

Heart-rate variability in diabetic and nondiabetic subjects

In both sexes, diabetes was associated with a significant reduction in total heart-rate variability and variability in both high- and low-frequency spectral bands (Table 1). The difference between diabetic and nondiabetic subjects was slightly, although not significantly, greater for women than men ($P > 0.09$ for all variability indexes). High-frequency power and low-frequency power were strongly correlated with total spectral power; therefore, the remainder of the analysis focused on total spectral power. The associations reported for total spectral power were similar to those found for high- and low-frequency power.

Factors associated with reduced total spectral power

Total spectral power declined with age in both diabetic and nondiabetic subjects ($P = 0.02$). Longer duration of diabetes was associated with reduced spectral power, although not independent of age. The partial correlation coefficients for the association of total spectral power with other factors adjusted for age are shown in Table 2. Although the coefficients vary slightly, the same pattern of associations

Table 2—*Partial correlation coefficient (r) for the association between total spectral power and coronary risk factors by diabetes, adjusted for sex and age*

	Nondiabetic subjects	Diabetic subjects	All subjects adjusted for diabetes
Age	−0.18†	−0.09	−0.13†
Systolic blood pressure (mmHg)	−0.13	−0.25‡	−0.21§
BMI (kg/m^2)	−0.12	−0.14	−0.13†
Waist-to-hip ratio	−0.15	−0.08	−0.11†
HbA_{1c} (%)	−0.20‡	−0.28‡	−0.26§
LDL cholesterol (mmol/l)	−0.06	−0.09	−0.08
HDL cholesterol (mmol/l)	0.11	0.01	0.05
Triglycerides (mmol/l)*	−0.21‡	−0.10	−0.15§
Exercise score*	0.13	0.21†	0.18‡
Albumin excretion rate (μg/min)*	−0.24‡	−0.17	−0.18‡

*Variables were log-transformed; all correlations other than the correlation with age itself were adjusted for age. †$P < 0.05$, ‡$P < 0.01$, §$P < 0.001$.

Table 3—*Odds ratio for CAC per tertile of total spectral power*

	All subjects			Non-diabetic			Diabetic		
	Odds ratio	95% CI	*P*	Odds ratio	95% CI	*P*	Odds ratio	95% CI	*P*
Unadjusted	1.6	(1.2–2.2)	0.003	1.6	1.01–2.6	0.04	1.6	1.03–2.3	0.03
Adjusted for:									
Age	1.5	1.1–2.05	0.01	1.5	0.93–2.4	0.09	1.5	0.99–2.24	0.06
Age, heart rate	1.5	1.03–2.2	0.03						
Age, systolic blood pressure	1.4	0.99–1.9	0.06						
Age, BMI	1.3	0.98–1.9	0.08						
Age, triglycerides	1.4	1.02–1.9	0.04						
All of the above	1.2	0.87–1.7	0.3						

was seen in the diabetic and nondiabetic subjects, and there were no significant differences between diabetic and nondiabetic subjects in the strength of these associations. Subjects with lower spectral power had higher blood pressure, BMI, waist-to-hip ratio, triglycerides, and HbA_{1c} and performed less exercise. In multiple regression analysis adjusting for these factors simultaneously, spectral power was independently associated with HbA_{1c} and systolic blood pressure in diabetic patients and HbA_{1c} and triglycerides in nondiabetic patients. Lower spectral power was associated with higher albumin excretion rate in the subset of subjects ($n = 271$) for whom these data were available (Table 2). In the nondiabetic group, the association of spectral power with albumin excretion rate was independent of BMI, systolic blood pressure, triglycerides, and HbA_{1c} ($P = 0.01$ on adjustment). When the 21 diabetic patients and 3 nondiabetic subjects who were taking blood pressure–lowering drugs were excluded from these analyses, the same associations were found. There was no association between social class and spectral power.

The lower spectral power in diabetic patients compared to nondiabetic subjects remained unchanged on adjustment for systolic blood pressure, BMI, and lipids (data not shown). Adjusting for social class did not alter the difference in spectral power between diabetic and nondiabetic subjects in either sex. Adjusting for HbA_{1c} abolished the difference in heart-rate variability between diabetic and nondiabetic subjects.

The prevalence of calcification

As described previously, the prevalence of CAC was 54% in nondiabetic men and 52% in diabetic men ($P = 0.7$) (14). The prevalence of calcification was 22% in nondiabetic women and 47% in diabetic women. Therefore, the effect of diabetes on calcification prevalence was fivefold greater in women than in men ($P < 0.001$ for the diabetes-by-sex interaction term). Diabetes was, however, associated with a greater severity of calcification in both sexes.

Total spectral power and CAC

Age was associated with both spectral power (Table 2) and calcification (odds ratio for calcification per year of age = 1.07, $P = 0.01$); therefore, the association of spectral power and CAC was adjusted for age (Table 3). Both diabetic and nondiabetic subjects with lower total spectral power had a higher odds of CAC; however, when adjusted for age, this did not reach significance within either group (Table 3). This association was significant for all subjects combined with an odds ratio of 1.5 per tertile of spectral power adjusted for age (Table 3). There was a stepwise increase in the odds of CAC with every decreasing tertile of spectral power, such that in all subjects combined, those in the lowest tertile for spectral power had a 2.3-fold odds of CAC compared with those in the top tertile adjusted for age ($P = 0.01$). The association of spectral power with CAC was independent of triglyceride level but not systolic blood pressure or BMI (Table 3). The association was not altered by adjustment for HbA_{1c}, albumin excretion rate, or physical activity level in those for whom these data were available. Exclusion of data regarding subjects who were taking drugs affecting blood pressure did not alter the association between spectral power and calcification. There was no significant difference between men and women in the strength of the association ($P = 0.1$ for the interaction between sex and spectral power on calcification).

High-frequency:low-frequency band power ratio

Diabetes was not associated with a significant difference in the ratio of high-frequency to low-frequency band power in either sex, and there was no association between this ratio and CAC (data not shown). As previously reported in other studies, this ratio was much higher in women than in men (Table 1).

CONCLUSIONS — This study demonstrates that reduced heart-rate variability is associated with increased CAC. As judged by the odds ratio, the association was of similar magnitude in the diabetic and nondiabetic subjects, although it did not reach significance within each group. The study also demonstrates that reduced heart-rate variability has similar risk factors in the general population as in diabetic patients, including risk factors that cluster in the insulin-resistance syndrome and HbA_{1c}.

The association between heart-rate variability and CAC is not secondary to ischemia

The cross-sectional observation that subjects with cardiac autonomic dysfunction have more coronary atherosclerosis may reflect the common antecedents of these two conditions or that one may cause the other. Therefore, it is possible that ischemia arising from atherosclerosis might lead to reduced heart-rate variability. However, we studied young adults, only one of whom had a clinical history of angina. The levels of CAC we observed were mostly consistent with mild atherosclerosis (90% of those with CAC had EBCT scores <100). Even the two subjects with

the highest scores (>400), who had been referred for thallium stress testing, had no evidence of ischemia. Our data are, therefore, more consistent with reduced heart-rate variability being associated with atherosclerosis itself rather than being a secondary effect of ischemia.

Previous studies in the general population have shown an association between heart-rate variability and severity of atherosclerosis on angiography. However, these latter studies were of patients with angina; therefore, a reduction in heart-rate variability secondary to ischemic cardiac failure or ischemic damage to cardiac nerves could not be excluded (22). This explanation also could not be excluded in prospective cohort studies in which reduced heart-rate variability was associated with an increased incidence of coronary heart disease. The short duration of follow-up in these cohorts meant that ischemia may already have been present at baseline in those subjects in whom coronary heart disease subsequently developed (3,4).

Is reduced heart-rate variability a risk factor for atherosclerosis?

Does the association between heart-rate variability and CAC simply reflect the fact that these conditions share common risk factors? The association between heart-rate variability and CAC was independent of some shared risk factors (e.g., triglycerides) but was not independent of systolic blood pressure and BMI. Therefore, the association may represent confounding by systolic blood pressure and BMI, but it is equally possible that reduced heart-rate variability is part of the pathway through which obesity and hypertension lead to calcification. Reduced heart-rate variability may even be an antecedent of hypertension. Consistent with this, in the Atherosclerosis Risk in the Community Study, reduced high-frequency spectral power was independently associated with incident hypertension (23). The observed association might also be attributable to other shared factors not measured in this study. For example, although HbA_{1c} was not associated with calcification, it is possible that measures of long-term glycemic control or advanced glycation end products might have been associated with both heart-rate variability and calcification. This study cannot resolve this, but our data are important in demonstrating that the potential role of autonomic dysfunction in determining atherosclerotic risk factors or in mediating their effects deserves more detailed investigation.

If cardiac autonomic dysfunction is involved in the pathogenesis of atherosclerosis, then we need to revise our understanding of the pathogenic consequences of diabetic cardiac autonomic neuropathy to include atherosclerosis in addition to increased risk of heart failure or arrhythmia. Even in these young, relatively well-controlled type 1 patients, we found a substantial reduction in heart-rate variability, as in other studies (24, 25). These differences were not explained by coronary risk factors, but adjusting for HbA_{1c} abolished the difference in heart-rate variability between diabetic and nondiabetic subjects. However, the interpretation of this is uncertain because there is little overlap between diabetic and nondiabetic subjects in HbA_{1c}; therefore, in any model, HbA_{1c} will be a very good proxy marker of diabetes and would be expected to explain differences between diabetic and nondiabetic subjects for that reason alone.

Heart-rate variability does not explain why diabetes abolishes the sex difference in calcification

It is well known that diabetes has a more adverse effect on coronary heart disease risk in women than in men, abolishing the sex difference in coronary heart disease (26). Consistent with this, we found that the effect of diabetes on calcification was greater in women than in men (14). We examined to what extent this might be related to a more adverse effect of diabetes on heart-rate variability in diabetic women than in men or a stronger relationship between heart-rate variability and calcification in diabetic women than in men. To our knowledge, there are no previous reports on this question. However, we found that heart-rate variability was only slightly more adversely affected by diabetes in women than in men and there was no sex difference in the strength of association with calcification. Therefore, heart-rate variability is unlikely to be the basis of the more adverse effect of diabetes on calcification in women than in men.

Reduced heart-rate variability clusters with risk factors that are associated with insulin resistance

We observed that low heart-rate variability was associated with higher triglycerides, blood pressure, and BMI in both diabetic and nondiabetic subjects, which is consistent with previous studies (27). Heart-rate variability also showed a strong independent relationship with HbA_{1c}, particularly in nondiabetic subjects. An association with HbA_{1c} was reported recently in older subjects in the Hoorn Study, although not independently of use of antihypertensive drug (28). Our data are consistent with an increasing body of evidence that cardiac autonomic dysfunction may be a very early feature of either glycemia in the nondiabetic range or some aspect of the insulin-resistance syndrome, possibly hyperinsulinemia itself (29–33).

Heart-rate variability is inversely correlated with albumin excretion rate in the general population

An interesting finding in this study is that a higher albumin excretion rate was associated with lower spectral power in nondiabetic subjects. This was despite the fact that almost all nondiabetic subjects (96%) had albumin excretion rates in the normoalbuminuric range ($<20\ \mu g/min^{-1}$). This is of interest because, in the general population, a higher albumin excretion rate is believed to be a marker of atherosclerotic risk (34). Possible explanations of our data include that reduced heart-rate variability might be a marker of abnormal renal sympathetic activity. Renal sympathetic dysfunction could lead to glomerular hyperfiltration, an antecedent of albuminuria (35). Our data are in contrast to the Hoorn Study, in which there was no association between cardiac autonomic function and albuminuria in glucose-tolerant subjects after adjustment for age (36).

Methodological considerations

The associations detected in this study were based on a single 5-min recording of heart rate and are likely to be attenuated by within-individual variation. To determine the true strength of the associations would require more extended measurements, such as 24-h heart-rate variability recordings. An obvious limitation of our study was that, because it was cross-sectional, it was not possible to differentiate

whether reduced heart-rate variability precedes elevated blood pressure or is a consequence of it. Resolving this will require a prospective study in very young subjects.

Considering that blood glucose and insulin levels are associated with sympathetic activity and possibly sympathetic modulation of heart rate, a potential limitation is that we were unable to control for blood glucose and insulin at the time of measurement. This might affect our estimates of the differences between the diabetic and nondiabetic subjects. However, within each of the diabetic and nondiabetic groups, the effect of variation in glucose and insulin levels between participants would be to reduce our ability to detect an association (due to regression dilution bias) between heart-rate variability and calcification.

It should be noted that in this study we used CAC as a measure of atherosclerosis volume itself, with which it is highly correlated, and not as a measure of luminal stenosis or plaque vulnerability, with which it is less well correlated (11,37). It is important to consider whether CAC is as good a marker of atherosclerosis in diabetic subjects as in nondiabetic subjects. EBCT-detected CAC is strongly associated with coronary artery disease in type 1 diabetes (38), and at autopsy, plaques in type 1 diabetic subjects were found to have a similar calcium content for a given amount of plaque as in nondiabetic subjects (39). Nonetheless, it remains possible that calcification may be a less close correlate of atherosclerosis volume in type 1 diabetic subjects than in nondiabetic subjects. However, even if this were the case, it would still be of interest to understand why reduced heart-rate variability is associated with calcification itself.

An important consideration in the interpretation of these data is that surgical sympathectomy and diabetic autonomic neuropathy are associated with calcification in the media of peripheral vessels (40,41). This raises the question of whether the calcification we have detected in diabetic patients could be medial rather than atherosclerotic intimal calcification. However, autopsy studies have demonstrated that CAC is intimal and atherosclerosis-related (42), except in the presence of renal failure and severe peripheral vascular disease. None of the subjects we studied had renal failure or a history of peripheral vascular disease. Furthermore, our detection of an association between calcification and heart-rate variability in nondiabetic subjects argues against medial calcification as an explanation for our findings.

In summary, reduced heart-rate variability is associated with similar cardiovascular risk factors in diabetic and nondiabetic subjects and is associated with subclinical atherosclerosis. This association is not independent of blood pressure and BMI. Therefore, the extent to which reduced heart-rate variability leads to atherosclerotic risk factor disturbances or mediates the effects of other risk factors deserves further study. An important issue is whether prevention of autonomic neuropathy might confer the added benefit of reduced coronary artery disease in diabetic patients. New therapies for the prevention of autonomic neuropathy are clearly needed. Meanwhile, the strong association between HbA_{1c} and heart-rate variability in this study emphasizes the importance of good glycemic control for the prevention of autonomic neuropathy, as demonstrated in the Diabetes Control and Complications Trial (43), in addition to control of blood pressure and management of lipids.

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