Microalbuminuria, Cardiovascular Autonomic Dysfunction, and Insulin Resistance in Patients With Type 2 Diabetes Mellitus

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Urinary albumin excretion/microalbuminuria and cardiovascular autonomic dysfunction are associated with high mortality in type 2 diabetic patients. We tested the hypothesis that the presence of microalbuminuria would correlate with cardiovascular autonomic dysfunction and insulin resistance in type 2 diabetic patients. The study group consisted of 15 Japanese patients with type 2 diabetes and microalbuminuria (age: 56 ± 10 years, mean ± SD). The control group consisted of 19 age-matched patients with normalbuminuria (56 ± 7 years). Cardiovascular autonomic function was assessed by baroreflex sensitivity (BRS), heart rate variability, plasma norepinephrine concentration, and cardiac ^{123}I-metaiodobenzylguanidine (MIBG) scintigraphy. BRS was lower in the microalbuminuria group than in the normalbuminuria group ($P < .05$). Early and delayed ^{123}I-MIBG myocardial uptake values were lower ($P < .05$ and $P < .005$, respectively) and the percent washout rate of ^{123}I-MIBG was higher ($P < .0005$) in the microalbuminuria group than in the normalbuminuria group. Fasting plasma glucose ($P < .05$) and insulin concentrations ($P < .05$), and the homeostasis model assessment (HOMA) index ($P < .01$) were higher in the microalbuminuria group than in the normalbuminuria group. Multiple regression analysis showed that urinary albumin excretion was independently predicted by the myocardial uptake of ^{123}I-MIBG at delayed phase, fasting plasma insulin concentration, and the HOMA index. Our results indicate that the presence of microalbuminuria in our Japanese patients with type 2 diabetes is characterized by depressed cardiovascular autonomic function and insulin resistance, and that the myocardial uptake of ^{123}I-MIBG at delayed phase, fasting plasma insulin, and HOMA index are independent predictors of urinary albumin excretion.

THE CARDIOVASCULAR mortality rate is high in insulin-dependent type 1 diabetic patients, and is further increased in association with the presence of proteinuria.[1,2] Similarly, urinary albumin excretion is a predictor of mortality from cardiovascular disease in type 2 diabetic patients.[2,3] Cardiovascular autonomic neuropathy is strongly related to cardiovascular mortality in diabetics,[4] and to increased urinary albumin excretion in type 1[5,6] and type 2 diabetic patients.[7] While the pathophysiology of the association of urinary albumin excretion with cardiovascular autonomic dysfunction remains to be elucidated, age, sex, hypertension, dyslipidemia, poor glycemic control, and endothelial dysfunction may be involved.[8,9] We have recently reported that insulin resistance may play a role in the depressed cardiovascular reflex vagal and sympathetic function in type 2 diabetes patients associated with essential hypertension.[10]

Accordingly, we hypothesized that the presence of microalbuminuria correlates with cardiovascular autonomic dysfunction and insulin resistance in type 2 diabetic patients. In the present study, we compared baroreflex sensitivity (BRS), heart rate variability, plasma norepinephrine concentration, and cardiac ^{123}I-metaiodobenzylguanidine (MIBG) scintigraphic findings in addition to the metabolic profiles in Japanese type 2 diabetic patients with and without microalbuminuria, followed by evaluating the predictors of urinary albumin excretion in these patients.

MATERIALS AND METHODS

Ninety-two consecutive Japanese patients with type 2 diabetes mellitus who were admitted to our department in 2001 were screened. Among them, 64 patients who did not have organic heart disease as determined by physical examination, chest x-ray, 12-lead electrocardiography (ECG), echocardiography, treadmill exercise ECG, and thallium 201 cardiac scintigraphy were enrolled. Albuminuria was measured in two 24-hour urine samples. Patients with macroalbuminuria (>300 mg/d) or abnormal plasma creatine concentrations (≥1.2 mg/dL) were excluded from the study. Patients treated with insulin were also excluded. All female patients were not pregnant and were not treated with postmenopausal hormonal replacement or contraceptives. Fifteen (23%) patients had microalbuminuria (30 to 300 mg/d, microalbuminuria group). We also recruited 19 age-matched patients without microalbuminuria (<30 mg/d, normalbuminuria group), who were selected from the original 64 patients enrolled. The clinical characteristics of patients of both groups are summarized in Table 1. All patients underwent clinical examination to exclude the presence of secondary hypertension. Essential hypertension was defined as diastolic blood pressure ≥ 90 mm Hg, systolic blood pressure ≥ 140 mm Hg, or self-reported use of antihypertensive medication.[11] Twelve of 19 normalbuminuria patients and 10 of 15 microalbuminuria patients met this criterion and all of these patients were being treated with calcium channel antagonists, angiotensin-converting enzyme (ACE) inhibitors, and/or angiotensin II receptor blockers. No patients were treated with diuretics or beta- or alpha-blockers. The study was approved by the ethics review board of our institution and prior informed consent was obtained from all patients.

Echocardiography

M-mode 2-dimensional echocardiography and cardiac Doppler recordings were obtained using a phase-array echo-Doppler system. Echocardiograms were obtained in a standard manner using standard parasternal, short axis, and apical views. The left ventricular mass was calculated according to Devereux et al[12]: left ventricular mass = (1.04

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Table 1. Clinical Characteristics of Studied Patients

	NA Group (n = 19)	MA Group (n = 15)	*P* Value
Age (yr)	56 ± 7	56 ± 10	NS
Gender (male/female)	10/9	9/6	NS
Albuminuria (mg/24 h)	10 ± 6	104 ± 61	<.0001
Duration of diabetes (yr)	8.5 ± 8.4	10.6 ± 5.9	NS
Hypertension (%)	63	67	NS
Smoking history (%)	42	40	NS
Drugs administered (%)			
SU	53	47	NS
Alpha glucosidase inhibitors	42	40	NS
Pioglitazone	5	7	NS
Calcium channel antagonists	32	53	NS
ACE inhibitors	47	40	NS
ARB	16	13	NS
BMI (kg/m^2)	24.9 ± 5.1	25.7 ± 4.4	NS
HR (beats/min)	67 ± 8	70 ± 6	NS
SBP (mm Hg)	127 ± 27	132 ± 18	NS
DBP (mm Hg)	76 ± 15	82 ± 16	NS
FPG (mg/dL)	142 ± 35	176 ± 38	<.05
F-IRI (μU/mL)	4.8 ± 2.4	7.3 ± 3.0	<.05
HOMA index	1.7 ± 0.9	3.1 ± 1.3	<.01
HbA_{1c} (%)	8.0 ± 1.4	8.7 ± 1.8	NS
T-chol (mg/dL)	188 ± 36	208 ± 43	NS
TGL (mg/dL)	115 ± 47	149 ± 70	NS
HDL-c (mg/dL)	46 ± 8	50 ± 11	NS
UA (mg/dL)	5.3 ± 1.6	5.6 ± 1.6	NS
Cr (mg/dL)	0.7 ± 0.2	0.8 ± 0.3	NS
Ccr (mL/min)	111 ± 27	99 ± 25	NS

NOTE. Data are means ± SD.

Abbreviations: NA, normalbuminuria; MA, microalbuminuria; SU, sulfonylurea; ACE, angiotensin-converting enzyme; ARB, angiotensin receptor blocker; BMI, body mass index; HR, heart rate; SBP, systolic blood pressure; DBP, diastolic blood pressure; FPG, fasting plasma glucose; F-IRI, fasting immunoreactive insulin; HOMA, homeostasis model assessment; HbA_{1c}, hemoglobin A_{1c}; T-chol, total cholesterol; TGL, triglyceride; HDL-c, high-density lipoprotein cholesterol; UA, uric acid; Cr, creatinine; Ccr, creatinine clearance; NS, not significant.

[{LVIDd + IVSTd + PWTd}^{3} − LVIDd^3] −14 g), where LVIDd = left ventricular internal dimension at end-diastole; IVSTd = intraventricular septal thickness at end-diastole; and PWTd = posterior wall thickness at end-diastole. The left ventricular mass was divided by body surface area to calculate the left ventricular mass index. Pulsed Doppler recordings were made from standard apical 4-chamber view. Mitral inflow velocity was recorded with the sample volume at the mitral annulus level taking the average of ≥3 cardiac cycles. The peak velocity of early (E) and late ventricular filling (A) was determined and the ratio (E/A) and deceleration time were recorded.

Cardiovascular Autonomic Function Tests

All subjects were studied while in supine position in a quiet room with dimmed lights between 9 and 11 AM.[10,13] A catheter was inserted in the right cubital vein, and arterial blood pressure was recorded noninvasively by tonometry (Jentow-7700; Nihon Colin, Komaki, Japan). The tonometric sensor was attached over the left radial artery. The accuracy of continuous blood pressure monitoring has been demonstrated previously.[14] Arterial blood pressure and a standard 12-lead ECG were monitored simultaneously; data were stored in a PCM data recorder (RD-200T; TEAC, Tokyo, Japan). Three-lead precordial Holter ECG recordings (model-459; Del Mar Avionics, Irvine, CA) were also obtained throughout the procedure for analysis of heart rate variability.

After an interval of 30 minutes to permit stabilization of the cardiovascular baroreflex mechanism, the patient was asked to breathe at a rate of 15 breaths/min using a metronome, to maximize regularity between respiration and cardiovascular function. Blood samples were obtained from the venous catheter to measure plasma norepinephrine concentration. BRS was assessed by the phenylephrine method as described previously.[10,13] Phenylephrine (2 to 3 μg/kg) was injected over 15 seconds to obtain a 15- to 40-mm Hg systolic blood pressure increase. BRS was calculated as the slope of the linear regression line relating systolic blood pressure changes to RR interval changes. Regression lines with more than 20 data points and a correlation coefficient (r) greater than 0.8 were accepted for analysis. The mean of the 2 slope values was taken as the BRS value.[13]

Heart rate variability was analyzed using a 300-second interval on Holter ECG recordings (MARS 8000; Marquette Electronics, Milwaukee, WI) immediately before phenylephrine injection. The power spectrum of the RR interval was computed by a fast Fourier transform and expressed as the area under the power spectrum.[15] We calculated the power of 2 spectral bands, the low frequency component (LF) at 0.04 to 0.15 Hz and the high frequency component (HF) at 0.15 to 0.40 Hz. Based on their skewed distribution, the measured values of heart rate variability were transformed to natural logarithmic values. The ratio of LF to HF (LF/HF) also was computed.

Planar and single-photon emission computed tomography studies were performed both at 15 minutes (early) and 4 hours (delayed) after the injection of 111 MBq of ^{123}I-MIBG using a rotating gamma camera (ZLC 7500; Siemens, Munich, Germany). Data were analyzed with analysis software (SCINTIPAC; Shimadzu, Kyoto, Japan). The anterior planar images from early and delayed ^{123}I-MIBG studies were analyzed visually. For semiquantitative analysis, regions of interest were drawn over the whole heart and a 10 × 10 mm area over the upper mediastinum on the early and delayed planar images was used to calculate the mean heart-to-mediastinum (H/M) ratio. After correcting for the physical decay of ^{123}I, the percent washout rate (WR) of the tracer from the myocardium was determined over a 4-hour period. Insulin resistance was evaluated by the homeostasis model assessment (HOMA) index = ([fasting plasma insulin {μU/mL} × fasting plasma glucose {mmol/L}]/22.5).[16]

Statistical Analysis

Data are presented as means ±SD. Differences between 2 groups were analyzed by unpaired Student's t test, chi-square test, or Fisher's exact probability test. A value of $P < .05$ was considered statistically significant. Simple (Spearman's rank) correlation coefficients between urinary albumin secretion and various variables were calculated, and a stepwise multiple regression analysis was then used to evaluate the independent association of these variables with urinary albumin secretion. In our multivariate analysis, F values ≥ 4 were considered significant.

RESULTS

As shown in Table 1, the mean age was similar between the microalbuminuria and normalbuminuria groups and there were no significant differences with respect to gender, duration of diabetes, body mass index, smoking history, and administered medications. Fasting plasma glucose and insulin concentrations were significantly higher ($P < .05$ for each) in the microalbuminuria group, resulting in a higher HOMA index ($P < .01$). There was no significant difference in hemoglobin A_{1c} between the 2 groups. The hemodynamic data listed in Table 1 were

Table 2. Echocardiographic Findings

	NA Group (n = 19)	MA Group (n = 15)	*P* Value
EF (%)	72 ± 5	69 ± 6	NS
LVIDd (mm)	48 ± 5	50 ± 5	NS
LVIDs (mm)	30 ± 2	32 ± 3	NS
IVSTd (mm)	9.0 ± 1.1	9.1 ± 1.4	NS
PWTd (mm)	9.2 ± 1.1	9.3 ± 1.6	NS
LVMI (g/m^2)	112 ± 23	118 ± 24	NS
E-peak velocity (cm/s)	64 ± 17	56 ± 11	NS
A-peak velocity (cm/s)	63 ± 18	75 ± 16	<.05
E/A ratio	1.07 ± 0.29	0.78 ± 0.25	<.01
Deceleration time (ms)	227 ± 60	248 ± 50	NS

NOTE. Data are means ± SD.

Abbreviations: EF, ejection fraction; LVIDd, left ventricular internal dimension at end-diastole; LVIDs, left ventricular internal dimension at end-systole; IVSTd, interventricular septal thickness at end-diastole; PWTd, posterior wall thickness at end-diastole; LVMI, left ventricular mass index; NS, not significant.

obtained immediately before BRS assessment. The resting heart rate, systolic and diastolic blood pressures, plasma total cholesterol, triglyceride, and high-density lipoprotein (HDL)-cholesterol, uric acid, creatinine, and creatinine clearance were not significantly different between the 2 groups.

Table 2 presents a summary of echocardiographic findings. The left ventricular dimensions at end-diastole and end-systole, intraventricular septal and posterior wall thickness at end-diastole, ejection fraction, and left ventricular mass index were essentially similar in the 2 groups. With regard to the left ventricular diastolic function, the peak velocity of late ventricular filling (A) was higher and the E/A ratio was lower in the microalbuminuria group compared with the normalbuminuria group ($P < .05$ and $P < .01$, respectively). However, there was no significant difference in the deceleration time.

Figure 1 summarizes the results of the cardiovascular autonomic function tests. BRS was lower in the microalbuminuria group than in the normalbuminuria group (8.2 ±5.0 *v* 12.7 ±5.3 ms/mm Hg, $P < .05$, Fig 1A). Plasma norepinephrine concentrations were similar in both groups (216 ± 121 *v* 229 ± 86 pg/ml, P = not significant [NS], Fig 1B). Analysis of heart rate variability revealed that the HF power and the LF/HF ratio were not significantly different between the 2 groups (3.7 ± 1.3 *v* 4.0 ± 1.1 ln-ms^2, P = NS, 1.4 ± 1.2 *v* 1.2 ± 1.1, P = NS, respectively, Fig 1C). Cardiac ^{123}I-MIBG scintigraphy disclosed that the H/M ratios at early and delayed phases were

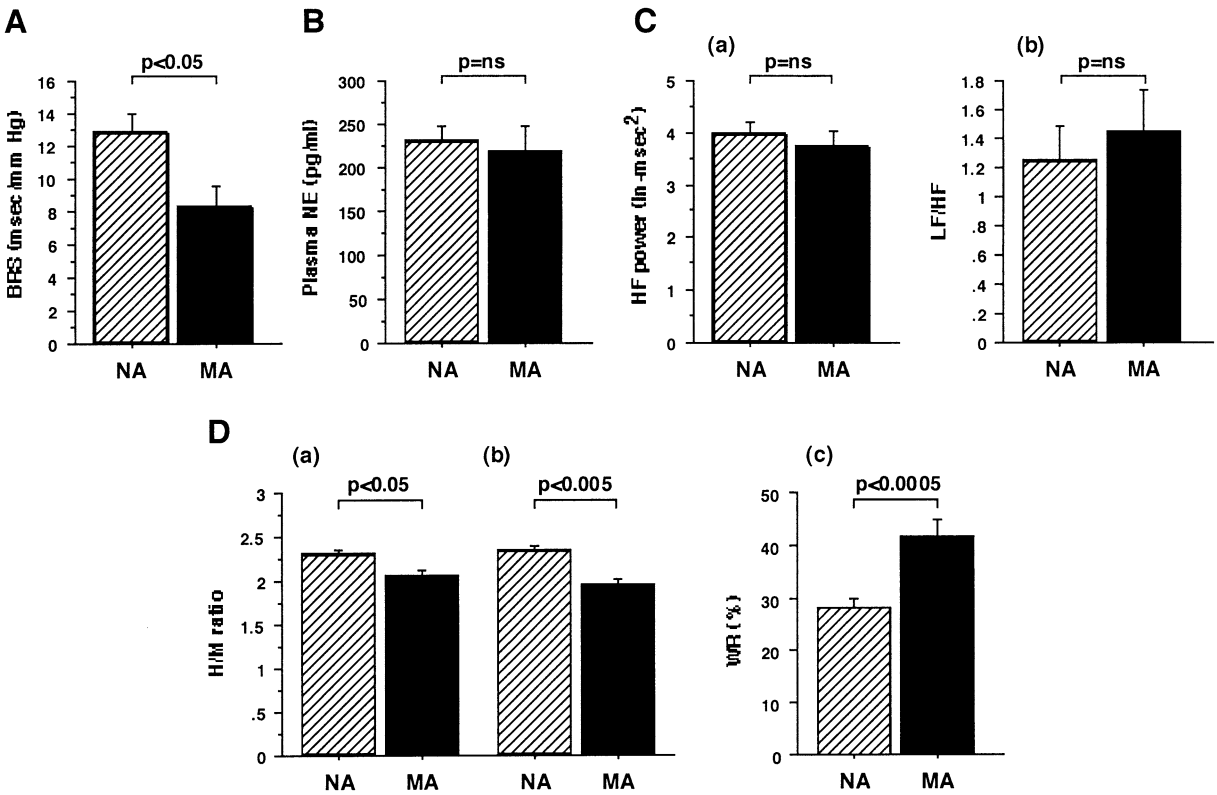

Fig 1. Comparison of autonomic function tests between type 2 diabetic patients with normalbuminuria (NA) and with microalbuminuria (MA). (A) Baroreflex sensitivity (BRS). (B) Plasma norepinephrine (NE) concentration. (C) Heart rate variability (HRV). Power of high frequency component (HF, 0.15 to 0.40 Hz, a) and the ratio of the low frequency power (LF, 0.04 to 0.15 Hz) to HF power (LF/HF, b). The distributions of HRV values were skewed and were thus transformed to natural logarithmic values. (D) Cardiac ^{123}I-metaiodobenzylguanidine (MIBG) scintigraphic findings. Myocardial uptake of ^{123}I-MIBG at early (a) and delayed (b) phases. Myocardial uptake of ^{123}I-MIBG is expressed as the mean heart-to-mediastinum (H/M) ratio. (c) Percent washout rate (WR) of ^{123}I-MIBG. Data are means ± SD. ns = not significant.

Table 3. Correlation of Urinary Albumin Excretion to Measures of Variables

	Univariate	
	r	*P* Value
Age	0.027	.879
Duration of diabetes	0.080	.6521
BMI	0.291	.0954
HR	0.188	.2876
SBP	0.054	.7627
DBP	0.109	.5384
FPG	0.420	.0133
F-IRI	0.379	.0272
HOMA index	0.540	.0010
HbA_{1c}	0.261	.1354
T-chol	0.187	.2898
TGL	0.045	.8020
HDL-c	0.216	.2208
UA	0.089	.6184
Cr	0.070	.6960
Ccr	−0.257	.1415
EF	0.273	.1236
LVMI	0.010	.9641
E/A	−0.318	.0672
BRS	−0.378	.0277
Plasma NE	−0.149	.3997
HF power	−0.028	.8769
LF/HF	0.159	.3693
H/M ratio at early phase	−0.432	.0107
H/M ratio at delayed phase	−0.542	.0009
WR	0.443	.0087

Abbreviations: BRS, baroreflex sensitivity; NE, norepinephrine; HF, high frequency; LH, low frequency; H/M, heart-to-mediastinum ratio; WR, washout rate. See Tables 1 and 2 for other abbreviations.

lower in the microalbuminuria group than in the normalbuminuria group (2.04 ± 0.32 *v* 2.29 ±0.31, $P < .05$ and 1.94 ± 0.35 *v* 2.33 ± 0.31, $P < .005$, respectively, Fig 1D). The percent WR of ^{123}I-MIBG was higher in the microalbuminuria group than in the normoalbuminuria group (41.7% ± 11.5% *v* 28.1% ± 8.3%, $P < .0005$, Fig 1D).

Table 3 illustrates the correlation between urinary albumin excretion and age, body mass index, and various other variables in all patients of both the microalbuminuria and the normoalbuminuria groups. Urinary albumin excretion correlated positively with fasting plasma glucose, fasting plasma insulin, HOMA index, and percent WR, and negatively with H/M ratio at delayed phase and BRS. Multiple regression analysis was performed using the stepwise procedure. Urinary albumin excretion was independently predicted by H/M ratio at delayed phase, fasting plasma insulin, and HOMA index (Table 4).

DISCUSSION

In the present study, diabetic patients with microalbuminuria manifested lower BRS, and lower myocardial uptake and enhanced clearance of ^{123}I-MIBG. Among the metabolic parameters, the fasting plasma concentrations of glucose and insulin and the HOMA index were higher in patients with microalbuminuria than in those with normalbuminuria. Furthermore, multiple regression analysis revealed that urinary albumin excretion was independently predicted by H/M ratio at delayed phase, fasting plasma insulin, and HOMA index in our Japanese patients with type 2 diabetes.

Microalbuminuria is considered as a marker of generalized endothelial damage, which leads to nephropathy in patients with diabetes.[17] Hyperglycemia, common in type 2 diabetes, impairs endothelial function due to impaired generation of nitric oxide (NO) and increased formation of reactive oxygen species.[18,19] However, conflicting results have been reported regarding the effects of hyperinsulinemia on endothelial function. Stout[19] reported that the effect of insulin, acting like a growth factor, is detrimental on the vasculature. However, others[20-22] have shown its beneficial actions on the vasculature by demonstrating that insulin increased endothelial constitutive NO synthase (ecNOS) gene expression and its activity, resulting in enhanced NO bioavailability. Because atherosclerosis affects conduit arteries, the effect of insulin on large arteries has attracted attention to understand the pathogenic link between the insulin resistance syndrome and endothelial function. Recently, Arcaro et al[23] reported that endothelium-dependent vasodilation in large conduit arteries such as brachial and common femoral arteries was abolished by insulin infusion using a euglycemic insulin clamp techniques. They speculated that insulin could lead to endothelial dysfunction by increasing the availability of endothelin-1, resulting in downstream effects on NAD(P)H oxidase and superoxide anion production. Together with our finding that the fasting plasma insulin concentration and HOMA index were independent predictors of urinary albumin excretion, we suggest that insulin resistance is an important underlying factor of microalbuminuria possibly via endothelial dysfunction.

In the present study, patients of the microalbuminuria group had depressed BRS compared to the normalbuminuria group, indicating a decrease in cardiovascular vagal reflex activity. This finding is in agreement with those of others.[24,25] The ^{123}I-MIBG is an analog of guanidine that shares the same neuronal transport and storage mechanisms with norepinephrine. In the heart, it is considered that reduced uptake of ^{123}I-MIBG (H/M ratio) reflects reduced norepinephrine content at presynaptic sites or reduced neural density while an enhanced

Table 4. Stepwise Regression Analysis Between Urinary Albumin Excretion and Various Parameters

Independent Variables	Regression Coefficient	Standard Error	Standard Regression Coefficient	F Value
To urinary albumin excretion intercept	178.303			
H/M ratio at delayed phase	−71.423	22.183	−0.435	10.367
F-IRI	−12.378	6.048	−0.574	4.138
HOMA index	43.671	13.482	0.913	10.493

Abbreviations: F-IRI, fasting immunoreactive insulin; HOMA, homeostasis model assessment.

washout rate of ^{123}I-MIBG reflects enhanced release of norepinephrine from presynaptic sites.[26] In our study, the myocardial uptake of ^{123}I-MIBG was lower and its clearance was higher in the microalbuminuria group than the normalbuminuria group. Furthermore, it is noteworthy that multiple regression analysis revealed the H/M ratio at delayed phase as an independent predictor of urinary albumin excretion. To our knowledge, there are no reports demonstrating the association of urinary albumin excretion/microalbuminuria with cardiac ^{123}I-MIBG scintigraphic findings. Although the precise mechanism for this association remains to be investigated, it has been reported that poor glycemic control worsens and strict glycemic control improves both urinary albumin excretion and cardiac ^{123}I-MIBG scintigraphic findings.[27] On the other hand, in patients with essential hypertension, hyperinsulinemia correlated with higher clearance of ^{123}I-MIBG.[28] Similarly, we demonstrated previously that type 2 diabetic patients with essential hypertension showed insulin resistance, low myocardial uptake of ^{123}I-MIBG and high ^{123}I-MIBG clearance.[10] The clinical usefulness of cardiac ^{123}I-MIBG scintigraphy to predict urinary albumin excretion/microalbuminuria in diabetic patients should be further investigated.

Compared with the normalbuminuria group, patients with microalbuminuria manifested depressed cardiac diastolic dysfunction although no significant difference was noted with respect to the left ventricular mass index. Diastolic dysfunction is associated with depressed cardiovascular autonomic dysfunction and insulin resistance.[10] Interestingly, Liu et al[29] recently reported that albuminuria was independently associated with left ventricular systolic and diastolic dysfunction in patients with type 2 diabetes.

The present study has some limitations. First, 67% and 63% of our patients with microalbuminuria and normalbuminuria, respectively, had been diagnosed earlier with associated essential hypertension. All these patients were being treated with one or more antihypertensive drugs, including ACE inhibitors, angiotensin II receptor blockers and calcium channel antagonists, prior to enrolment. In this regard, all 3 of these drug classes have been reported to improve insulin resistance,[30,31] cardiovascular autonomic function,[32-34] and urinary albumin excretion.[35] Therefore, these medications might have beneficially affected our results. As to antidiabetic medications, a considerable number of patients were being treated with sulfonylurea and/or alpha glucosidase inhibitors while only one patient in each group was with pioglitazone, an insulin-sensitizing drug. Pioglitazone but not glibenclamide or voglibose was reported to reduce urinary albumin excretion in type 2 diabetic patients with microalbuminuria,[36] suggesting that thiazolidinedione derivative rather than sulfonylurea or alpha glucosidase inhibitors may be useful in terms of improving insulin resistance and microalbuminuria in type 2 diabetic patients. Second, ambulatory blood pressure monitoring was not performed in our study. However, because it was demonstrated that urinary albumin excretion is greater in nondipper essential hypertensive patients than in dippers[37] and insulin resistance is closely related to nondipper essential hypertension,[38] the diminished nocturnal blood pressure fall might be involved in the development of microalbuminuria. Third, it has been recognized that there is gender difference in various aspects of cardiovascular autonomic function and metabolism. In the present study, there was no significant difference in these measures between male and female (data not shown). A large-scale study is needed to clarify the gender difference. Finally, although we considered the predominant involvement of endothelial dysfunction in the development of microalbuminuria and depressed cardiovascular autonomic dysfunction, we did not assess in vivo endothelial function. Other mechanisms, apart from endothelial dysfunction, may be operative. Further clinical investigation is needed to determine the role of endothelial function in these patients.

In conclusion, the present results indicate that in our Japanese type 2 diabetic patients the presence of microalbuminuria is associated with depressed cardiovascular autonomic function and insulin resistance, and that myocardial uptake of ^{123}I-MIBG at delayed phase, fasting plasma insulin, and HOMA index are independent predictors of urinary albumin excretion in these patients.

REFERENCES

1. Borch-Johnsen K, Kreiner S: Proteinuria: Value as predictor of cardiovascular mortality in insulin dependent diabetes mellitus. Br Med J 294:1651-1654, 1987

2. Mogensen CE: Microalbuminuria and hypertension with focus on type 1 and type 2 diabetes. J Intern Med 254:45-66, 2003

3. Savage S, Estacio RO, Jeffers B, et al: Urinary albumin excretion as a predictor of diabetic retinopathy, neuropathy, and cardiovascular disease in NIDDM. Diabetes Care 19:1243-1248, 1996

4. Ewing DJ: Diabetic autonomic neuropathy and the heart. Diabetes Res Clin Pract 30:S31-S36, 1996 (suppl)

5. Clarke CF, Eason M, Reilly A, et al: Autonomic nerve function in adolescents with type 1 diabetes mellitus: Relationship to microalbuminuria. Diabet Med 16:550-554, 1999

6. Lafferty AR, Werther GA, Clarke CF: Ambulatory blood pressure, microalbuminuria, and autonomic neuropathy in adolescents with type 1 diabetes. Diabetes Care 23:533-538, 2000

7. Rutter MK, McComb JM, Brady S, et al: Autonomic neuropathy in asymptomatic subjects with non-insulin-dependent diabetes mellitus and microalbuminuria. Clin Auton Res 8:251-257, 1998

8. Neil HA, Thompson AV, John S, et al: Diabetic autonomic neuropathy: the prevalence of impaired heart rate variability in a geographically defined population. Diabet Med 6:20-24, 1989

9. Smulders YM, Jager A, Gerritsen J, et al: Cardiovascular autonomic function is associated with (micro-)albuminuria in elderly Caucasian subjects with impaired glucose tolerance or type 2 diabetes: The HOORN Study. Diabetes Care 23:1369-1374, 2000

10. Takahashi N, Nakagawa M, Saikawa T, et al: Effect of essential hypertension on cardiac autonomic function in type 2 diabetic patients. J Am Coll Cardiol 38:232-237, 2001

11. Liao D, Sloan RP, Cascio WE, et al: Multiple metabolic syndrome is associated with lower heart rate variability. The Atherosclerosis Risk in Communities Study. Diabetes Care 21:2116-2122, 1998

12. Devereux RB, Alonso DR, Lutas EM, et al: Echocardiographic assessment of left ventricular hypertrophy: Comparison to necropsy findings. Am J Cardiol 67:450-458, 1986

13. Takahashi N, Nakagawa M, Saikawa T, et al: Noninvasive assessment of the cardiac baroreflex: Response to downward tilting and

comparison with the phenylephrine method. J Am Coll Cardiol 34:211-215, 1999

14. Sato T, Nishinaga M, Kawamoto A, et al: Accuracy of a continuous blood pressure monitor based on arterial tonometry. Hypertension 21:866-874, 1993

15. Nakagawa M, Iwao T, Ishida S, et al: Circadian rhythm of the signal averaged electrocardiogram and its relation to heart rate variability in healthy subjects. Heart 79:493-496, 1998

16. Matthews DR, Hosker JP, Rudenski AS, et al: Homeostasis model assessment: Insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia 28:412-419, 1985

17. Goligorsky MS, Chen J, Brodsky S: Endothelial cell dysfunction leading to diabetic nephropathy: Focus on nitric oxide. Hypertension 37:744-748, 2001

18. Gutterman DD: Vascular dysfunction in hyperglycemia: Is protein kinase C the culprit? Circ Res 90:5-7, 2002

19. Stout RW: Insulin and atheroma: 20-yr perspective. Diabetes Care 13:631-654, 1990

20. Zeng G, Quon MJ: Insulin-stimulated production of nitric oxide is inhibited by wortmannin: Direct measurement in vascular endothelial cells. J Clin Invest 98:894-898, 1996

21. Kuboki K, Jiang ZY, Takahara N, et al: Regulation of endothelial constitutive nitric oxide synthase gene expression in endothelial cells and in vivo: A specific vascular action of insulin. Circulation 101:676-681, 2000

22. Steinberg HO, Paradisi G, Hook JG, et al: Free fatty acid elevation impairs insulin-mediated vasodilation and nitric oxide production. Diabetes 49:1231-1238, 2000

23. Arcaro G, Cretti A, Balzano S, et al: Insulin causes endothelial dysfunction in humans: Sites and mechanisms. Circulation 105:576-582, 2002

24. Gottsater A, Ahmed M, Fernlund P, et al: Autonomic neuropathy in type 2 diabetic patients is associated with hyperinsulinaemia and hypertriglyceridaemia. Diabet Med 16:49-54, 1999

25. Szelag B, Wroblewski M, Castenfors J, et al: Obesity, microalbuminuria, hyperinsulinemia, and increased plasminogen activator inhibitor 1 activity associated with parasympathetic neuropathy in type 2 diabetes. Diabetes Care 22:1907-1908, 1999

26. Sakata K, Shirotani M, Yoshida H, et al: Cardiac sympathetic nervous system in early essential hypertension assessed by ^{123}I-MIBG. J Nucl Med 40:6-11, 1999

27. Muhr-Becker D, Weiss M, Tatsch K, et al: Scintigraphically assessed cardiac sympathetic dysinnervation in poorly controlled type 1 diabetes mellitus: One-year follow-up with improved metabolic control. Exp Clin Endocrinol Diabetes 107:306-312, 1999

28. Watanabe K, Sekiya M, Tsuruoka T, et al: Relationship between insulin resistance and cardiac sympathetic nervous function in essential hypertension. J Hypertens 17:1161-1168, 1999

29. Liu JE, Robbins DC, Palmieri V, et al: Association of albuminuria with systolic and diastolic left ventricular dysfunction in type 2 diabetes: The Strong Heart Study. J Am Coll Cardiol 41:2022-2028, 2003

30. Gavras HP: Issues in hypertension: drug tolerability and special populations. Am J Hypertens 14:231S-236S, 2001 (suppl)

31. Lender D, Arauz-Pacheco C, Breen L, et al: A double blind comparison of the effects of amlodipine and enalapril on insulin sensitivity in hypertensive patients. Am J Hypertens 12:298-303, 1999

32. Kontopoulos AG, Athyros VG, Didangelos TP, et al: Effect of chronic quinapril administration on heart rate variability in patients with diabetic autonomic neuropathy. Diabetes Care 20:355-361, 1997

33. Rodgers JE, Patterson JH: Angiotensin II-receptor blockers: Clinical relevance and therapeutic role. Am J Health Syst Pharm 58:671-683, 2001

34. Lefrandt JD, Heitmann J, Sevre K, et al: The effects of dihydropyridine and phenylalkylamine calcium antagonist classes on autonomic function in hypertension: The VAMPHYRE study. Am J Hypertens 14:1083-1089, 2001

35. Lasaridis AN, Sarafidis PA: Diabetic nephropathy and antihypertensive treatment: What are the lessons from clinical trials? Am J Hypertens 16:689-697, 2003

36. Nakamura T, Ushiyama C, Shimada N, et al: Comparative effects of pioglitazone, glibenclamide, and voglibose on urinary endothelin-1 and albumin excretion in diabetes patients. J Diabetes Complications 14:250-254, 2000

37. Bianchi S, Bigazzi R, Baldari G, et al: Diurnal variations of blood pressure and microalbuminuria in essential hypertension. Am J Hypertens 7:23-29, 1994

38. Anan F, Takahashi N, Ooie T, et al: Role of insulin resistance in nondipper essential hypertensive patients. Hypertens Res 26:669-676, 2003