Sex Hormone and Gender Difference—Role of Testosterone on Male Predominance in Brugada Syndrome

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Testosterone in Brugada Syndrome. ***Introduction:*** **The clinical phenotype is 8 to 10 times more prevalent in males than in females in patients with Brugada syndrome. Brugada syndrome has been reported to be thinner than asymptomatic normal controls. We tested the hypothesis that higher testosterone level associated with lower visceral fat may relate to Brugada phenotype and male predominance.**

Methods and Results: **We measured body-mass index (BMI), body fat percentage (BF%), and several hormonal levels, including testosterone, in 48 Brugada males and compared with those in 96 age-matched control males. Brugada males had significantly higher testosterone (631 ± 176 vs 537 ± 158 ng/dL; P = 0.002), serum sodium, potassium, and chloride levels than those in control males by univariate analysis, and even after adjusting for age, exercise, stress, smoking, and medication of hypertension, diabetes, and hyperlipidemia, whereas there were no significant differences in other sex and thyroid hormonal levels. Brugada males had significantly lower BMI (22.1 ± 2.9 vs 24.6 ± 2.6 kg/m^2; P < 0.001) and BF% (19.6 ± 4.9 vs 23.1 ± 4.7%; P < 0.001) than control males. Testosterone level was inversely correlated with BMI and BF% in both groups, even after adjusting for the confounding variables. Conditional logistic regression models analysis showed significant positive and inverse association between Brugada syndrome and hypertestosteronemia (OR:3.11, 95%CI:1.22–7.93, P = 0.017) and BMI (OR:0.72, 95%CI:0.61–0.85, P < 0.001), respectively.**

Conclusions: **Higher testosterone level associated with lower visceral fat may have a significant role in the Brugada phenotype and male predominance in Brugada syndrome.** *(J Cardiovasc Electrophysiol, Vol. 18, pp. 415-421, April 2007)*

Brugada syndrome, gender, sex hormones, testosterone, body mass index

Introduction

Brugada syndrome is characterized by coved-type ST-segment elevation in the right precordial electrocardiographic (ECG) leads (V1–V3) and an episode of ventricular fibrillation (VF) in the absence of structural heart disease.[1-5] The prevalence of the disease is estimated to be up to 5 per 10,000 inhabitants and is one of the important causes of sudden cardiac death of middle-aged males, particularly in Asian countries including Japan.[4]

More than eight dozen distinct mutations in *SCN5A*, the gene encoding the α subunit of the sodium channel, have been so far identified in patients with Brugada syndrome and all mutations display an autosomal-dominant mode of transmission.[6,7] Therefore, males and females are expected to inherit the defective gene equally. However, more than 80% of patients in Western countries and more than 90% of patients in Asian countries affected with Brugada syndrome are males.[8] Recent experimental studies have unveiled the cellular mechanism of Brugada phenotype. The male predominance in the Brugada syndrome is suggested to be due, at least in part, to intrinsic differences in ventricular action potential (AP) between males and females.[9]

A male hormone, testosterone is reported to increase net outward currents[10-12] and is expected to accentuate Brugada phenotype, such as ST-segment elevation and subsequent episodes of VF in patients with Brugada syndrome. Testosterone is also known to decrease visceral fat.[13-15] Since patients with Brugada syndrome have been reported to be

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thinner than asymptomatic normal controls by Matsuo et al.,[16] we speculated that higher testosterone level associated with lower visceral fat may modulate Brugada phenotype and may relate to male predominance in patients with Brugada syndrome.

Methods

Patient Population and Data Collection

The study population consisted of 48 males with Brugada syndrome who agreed to participate in this study and showed Type 1 "coved" ST-segment elevation in V1–V3 leads[17] ranging in age from 30 to 69 years with a mean age of 50 ± 11 years (mean ± SD). Brugada males who were less than 30 years old and more than 70 years old were excluded from this study to minimize the influence of age on the basal sex hormonal levels including testosterone. Forty of the forty-eight Brugada males have been included in our previous clinical studies.[18-20] In all patients, physical examination, chest roentgenogram, laboratory values, echocardiography with wall motion analysis, and Doppler screening excluded structural heart diseases. The clinical, electrocardiographic, and electrophysiologic characteristics of the 48 Brugada males are shown in Table 1. Average age of the 48 Brugada males at diagnosis was 47 ± 12 years old. Aborted cardiac arrest or VF was documented in 21 males (44%), syncope alone in 11 males (23%), and 16 males (33%) were asymptomatic. Family history of sudden cardiac death (SCD) was observed in eight males (17%). An *SCN5A* coding region mutation was identified in seven (17%) of 42 males in whom genetic screening was conducted. Implantable cardioverter defibrillator (ICD) was implanted in all 32 symptomatic males with documented VF and/or syncope. ICD was also implanted in nine of 16 asymptomatic males due to induction of VF during the electrophysiologic study. Type 1 ST-segment elevation was recorded spontaneously in 43 males (90%) and was induced by sodium channel blockers in five males (10%). Complete right bundle branch block was observed in three males (6%). Late potential was recorded by a signal-average ECG system in 27 (59%) of 46 males. During the electrophysiologic study, VF requiring direct cardioversion for termination was induced in 32 (73%) of 44 males. Average HV interval was 46 ± 11 msec.

TABLE 1
Clinical, Electrocardiographic, and Electrophysiologic Characteristics in the 48 Brugada Males

Clinical characteristics	
Age at diagnosis (years)	47 ± 12
Aborted cardiac arrest or VF (%)	21/48 (44%)
Syncope alone (%)	11/48 (23%)
Asymptomatic (%)	16/48 (33%)
Family history of SCD	8/48 (17%)
SCN5A mutation	7/42 (17%)
ICD implantation	41/48 (85%)
Follow-up period (month)	41 ± 2
Arrhythmic event (%)	9/48 (19%)
Electrocardiographic characteristics	
Spontaneous coved-type ST elevation	43/48 (90%)
CRBBB (%)	3/48 (6%)
RR (msec)	939 ± 113
PQ interval (II) (msec)	186 ± 34
QRS duration (V2) (msec)	104 ± 18
Corrected QT interval (V5) (msec)	394 ± 27
ST amplitude at J point (V2) (mV)	0.32 ± 0.16
Late potential (%)	27/46 (59%)
Electrophysiologic characteristics	
Induction of VF	32/44 (73%)
Mode (Triple/Double/Single)	16/15/1
HV interval (msec)	46 ± 11

CRBBB = complete right bundle branch block; ICD = implantable cardioverter defibrillator; SCD = sudden cardiac death; VF = ventricular fibrillation.

We first obtained data, such as the hormonal levels, visceral fat parameters, and ECG parameters in the 48 Brugada males prospectively between January and July in 2003, mainly at regular outpatient clinics for checking ICD. Only a Brugada male refused to participate during the recruitment of the case.

Thereafter, age-matched control males were randomly selected from the municipal population registry in Suita City. The hormonal and visceral fat data were collected sequentially between August and December in 2003. The municipal population registry in Suita City included 5,846 control subjects, among whom 1,052 males were age-matched to the 48 Brugada males. The 96 control males with a mean age of 50 ± 11 years were sequentially recruited from the age-matched 1,052 males. None of the recruited 96 control males refused to participate in this study. There were no significant differences in the clinical characteristics between the 96 control males and the remaining 956 age-matched males. Therefore, we had no way of knowing the body weight of the individuals who were selected to serve as controls from a very large database. Although K. Matsuo is a co-author of this study, none of the Brugada males and control males who appeared in the article by Matsuo[16] are included in the present study population.

All protocols were approved by the Ethical Review Committee in the National Cardiovascular Center. Written informed consent was obtained from all subjects.

Sex and Thyroid Hormonal Levels and Serum Electrolytes

Blood samples for analysis of basal hormone levels and serum electrolytes were obtained between 8:00 and 9:00 AM after an overnight fast. Plasma sex hormonal levels including testosterone, estradiol, DHEA-S, LH, and FSH were measured using commercially prepared immunoassay kits (testosterone, LH, and FSH: Chemiluminescent immunoassay [Bayer HealthCare, New York, NY, USA]; estradiol: Electrochemiluminescent immunoassay [Roche Diagnostics GmbH, Mannheim, Germany]; DHEA-S: Radioimmunoassay [Diagnostic Products Corporation, Los Angeles, CA, USA]). Thyroid hormonal levels including free T3, T4, and TSH, and serum electrolyte levels including sodium, potassium, and chloride were also measured.

Body Mass Index and Body Fat Percentage

Body weight (BW) was measured to the nearest 0.1 kg and height to the nearest cm. Body-mass index (BMI) was calculated as weight/height^2 (kg/m^2) as a parameter of visceral fat. We also measured body-fat percentage (BF%) by using body composition analyzer (Biospace Co., Ltd. Tokyo, Japan). These visceral fat parameters were measured just after blood sampling. In the 32 symptomatic Brugada males who had had documented VF and/or syncope, the BW and BMI were also measured within 48 hours after their clinical events during admission in our hospital or other emergent hospitals.

ECG Parameters

In the 48 males with Brugada syndrome, 12-lead ECG was recorded just before blood sampling, and ECG parameters were assessed by an investigator (WS) blinded to clinical information. The ECG parameters included RR interval, PQ interval measured in lead II, QRS interval measured in lead V2, QT interval, corrected QT (QTc) interval measured in leads V5, and ST amplitude at J point measured in lead V2.

Statistical Analysis

We first conducted univariate analysis by using unpaired *t*-test to compare each data between the Brugada males and the control males. Since several confounding variables, such as age, exercise (none, sometimes, regularly), stress (none, sometimes, regularly), current smoking (no, yes), and medication (no, yes) of hypertension, diabetes, and hyperlipidemia may affect the hormonal levels including testosterone level and the visceral fat parameters, analysis of covariance (ANCOVA) was used to compare least square mean values between the Brugada males and the control males adjusting for these confounding variables. Pearson's correlation coefficients were calculated between the testosterone level and the visceral fat parameters. Partial correlation coefficients were calculated between the testosterone level and the visceral fat parameters after adjusting for age, exercise, stress, current smoking, and medication. Moreover, conditional logistic regression models were used to calculate odds ratios and 95% confidence intervals adjusting for age, BMI, exercise, stress, current smoking, hypertension, diabetes, and hyperlipidemia. Hypertestosteronemia was defined as serum testosterone levels $\geq$700 ng/dL, which is 75 percentiles of testosterone levels among case and control combined groups. In the 32 Brugada males with documented VF and/or syncope, a paired *t*-test was used to compare the visceral fat parameters at the clinical cardiac events and at the measurement of hormonal and visceral fat data. A two-sided P value below 0.05 was considered to indicate significance. All statistical analyses were performed by using SAS software, Ver 8.2.

Results

Hormonal Levels, Serum Electrolytes, and Visceral Fat

Table 2 illustrates univariate analysis for comparing sex and thyroid hormonal levels, serum electrolytes, and visceral fat parameters between the two groups. Testosterone level was significantly higher in the Brugada males than in the control males, whereas there were no significant differences in other sex hormonal levels; estradiol, DHEA-S, LH, FSH, and thyroid hormonal levels; T3, T4, and TSH. Serum sodium, potassium, and chloride levels were all significantly higher in the Brugada males than in the control males. BMI, BF%, and BW were all significantly lower in the Brugada males than in the control males. All variables followed normal distribution, both in the 48 Brugada and 96 control males.

The comparison of the confounding variables that may affect the hormonal levels and the visceral fat parameters between the 48 Brugada males and the 96 control males was shown in Table 3. Even after adjusting for age, exercise, stress, current smoking, and medication (hypertension, diabetes, and hyperlipidemia), the testosterone level, serum sodium, potassium, and chloride levels were all significantly higher, and the visceral fat parameters were significantly lower in the 48 Brugada males than in the 96 control males (Table 4). There were also significant differences in these parameters between the 24 definite Brugada males with documented VF and/or *SCN5A* mutations and the 96 control males after adjusting for the confounding variables (Table 4).

Correlation between Testosterone, Visceral Fat, and Serum Electrolytes

Testosterone level was inversely correlated with all visceral fat parameters, BMI, BF%, or BW in both the Brugada males and the control males, even after adjusting for age,

TABLE 2

Sex and Thyroid Hormonal Levels, Serum Electrolytes, and Visceral Fat Parameters in the 48 Brugada Males and the 96 Age-Matched Control Males

	Brugada Males (n = 48)	Control Males (n = 96)	P Value
Sex hormones			
Testosterone (ng/dL)	631 ± 176	537 ± 158	0.002
Estradiol (pg/mL)	28.9 ± 7.6	31.1 ± 12.6	0.263
DHEA-S (ng/mL)	1,901 ± 850	1,966 ± 861	0.668
LH (mIU/mL)	4.6 ± 2.6	3.9 ± 2.0	0.073
FSH (mIU/mL)	6.2 ± 4.9	5.0 ± 2.9	0.066
Thyroid hormones			
Free T3 (pg/mL)	3.3 ± 0.4	3.4 ± 0.3	0.360
Free T4 (ng/dL)	1.3 ± 0.1	1.3 ± 0.2	0.089
TSH (μIU/mL)	1.9 ± 1.4	1.7 ± 1.4	0.619
Serum electrolytes			
Sodium (mEq/L)	143.7 ± 2.0	142.6 ± 2.0	0.003
Potassium (mEq/L)	4.6 ± 0.3	4.3 ± 0.3	<0.001
Chloride (mEq/L)	105.1 ± 2.1	103.6 ± 2.1	<0.001
Viceral fat			
BMI (kg/m^2)	22.1 ± 2.9	24.6 ± 2.6	<0.001
BF% (%)	19.6 ± 4.9	23.1 ± 4.7	<0.001
BW (kg)	62.9 ± 9.7	70.0 ± 8.6	<0.001

Values are mean ± SD where indicated.
BMI = body-mass index; BF% = body-fat percentage; BW = body weight.

TABLE 3

Comparison of the Confounding Variables Between the 48 Brugada Males and the 96 Age-Matched Control Males

	Brugada Males (n = 48)	Control Males (n = 96)	P Value
Exercise			
None (%)	39.6	44.8	
Sometimes (%)	41.6	43.8	
Regularly (%)	18.8	11.5	0.482
Stress			
None (%)	27.1	21.9	
Sometimes (%)	54.2	54.2	
Regularly (%)	18.8	24.0	0.684
Current smoking (%)	25.0	27.1	0.789
Medication			
Hypertension (%)	20.8	19.8	0.883
Diabetes (%)	2.1	13.5	0.028
Hyperlipidemia (%)	10.4	5.2	0.246

TABLE 4

Testosterone, Serum Electrolytes, and Visceral Fat Parameters in the Brugada Males and the 96 Age-Matched Control Males after Adjusting for Confounding Variables

	Brugada Males	Control Males (n = 96)	P Value
ALL Case (n = 48)			
Testosterone (ng/dL)	631 ± 44	538 ± 40	0.003
Sodium (mEq/L)	144.2 ± 0.5	143.2 ± 0.5	0.007
Potassium (mEq/L)	4.6 ± 0.1	4.3 ± 0.1	<0.001
Chloride (mEq/L)	105.5 ± 0.5	103.9 ± 0.5	<0.001
BMI (kg/m^2)	22.3 ± 0.7	24.9 ± 0.7	<0.001
BF% (%)	20.0 ± 1.3	23.9 ± 1.1	<0.001
BW (kg)	63.4 ± 2.4	70.1 ± 2.1	0.001
Definite Brugada case with VF and/or *SCN5A* (n = 24)			
Testosterone (ng/dL)	656 ± 59	550 ± 48	0.009
Sodium (mEq/L)	143.9 ± 0.7	142.9 ± 0.6	0.042
Potassium (mEq/L)	4.7 ± 0.1	4.4 ± 0.1	<0.001
Chloride (mEq/L)	105.2 ± 0.7	103.9 ± 0.6	0.006
BMI (kg/m^2)	21.5 ± 1.0	24.5 ± 0.8	<0.001
BF% (%)	19.9 ± 1.7	24.1 ± 1.4	<0.001
BW (kg)	60.5 ± 3.1	69.2 ± 2.5	0.001

Values are mean ± SE adjusted for age, exercise, stress, current smoking, and medication of hypertension, diabetes and hyperlipidemia. BMI = body-mass index; BF% = body-fat percentage; BW = body weight; VF = ventricular fibrillation.

exercise, stress, current smoking, and medication (Brugada: BMI, r = –0.394, P = 0.011; BF%, r = –0.390, P = 0.012; BW, r = –0.335, P = 0.032; Control: BMI, r = –0.333, p = 0.002; BF%, r = –0.333, P = 0.001; BW, r = –0.305, P = 0.004), suggesting that Brugada males had higher testosterone level associated with lower visceral fat compared with control males (Fig. 1). No significant correlations were observed between other serum electrolytes and testosterone level or visceral fat parameters. Testosterone level was not correlated with age, even after adjusting for exercise, stress, current smoking, and medication (r = 0.007, P = 0.947).

Conditional Logistic Regression Models Analysis

Conditional logistic regression models analysis showed significant positive and inverse association between Brugada syndrome, hypertestosteronemia (Odd Ratio (OR): 3.11, 95%CI: 1.22–7.93, P = 0.017), and BMI (OR: 0.72, 95%CI: 0.61–0.85, P < 0.001), respectively (Table 5). Other variables did not significantly increase or decrease risks of Brugada syndrome (Table 5).

Visceral Fat at Clinical Cardiac Events in Brugada Males

In the 32 symptomatic Brugada males with documented VF and/or syncope, the time-span between the clinical cardiac events and the measurement of hormonal and the visceral fat data was 42 ± 32 months (mean ± SD, 1–99 months). The BMI and BW at the clinical cardiac events (VF or syncope) were significantly lower than those at the measurement of hormonal and visceral fat data (BMI, 21.0 ± 2.6 vs 22.1 ± 2.9 kg/m^2; BW, 60.0 ± 8.9 vs 62.9 ± 9.7 kg: P < 0.001, respectively).

Testosterone versus ECG Parameters, Symptoms or SCN5A Mutation in Brugada Males

Baseline electrocardiographic data of the 48 Brugada males are shown in Table 1. No significant correlations were observed between testosterone level and ECG parameters, including ST amplitude (r = –0.123, P = 0.406) and QTc interval (r = –0206, P = 0.160), in the 48 Brugada males. There was no significant difference in testosterone level between 32 symptomatic and 16 asymptomatic Brugada males (649 ± 185 vs 593 ± 157 ng/dL: P = 0.298). No significant difference was observed in testosterone level between 43 Brugada males with spontaneous Type 1 ST-segment elevation and five Brugada males with sodium channel blocker-induced Type 1 ST-segment elevation (624 ± 171 vs 688 ± 230 ng/dL: P = 0.448). Testosterone level was also no different between seven Brugada males with *SCN5A* mutation and 41 Brugada males without *SCN5A* mutation (700 ± 198 vs 619 ± 172 ng/dL: P = 0.261).

Follow-Up

Arrhythmic events occurred in nine (19%) of 48 Brugada males during average follow-up periods of 41 ± 2 months after blood sampling for the present study (Table 1). In more detail, arrhythmic events appeared in eight (38%) of 21 Brugada males with a history of aborted cardiac arrest or VF, in one (9%) of 11 Brugada males with syncope alone, but did not appear in any (0%) of 16 asymptomatic Brugada males.

Discussion

The major findings of the present study were: (1) Brugada males had significantly higher testosterone level, serum sodium, potassium, and chloride level, and significantly lower BMI, BF%, and BW than those in control males by univariate analysis, even after adjusting for age, exercise, stress, current smoking, and medications related to hypertension, diabetes and hyperlipidemia. (2) Testosterone level was inversely correlated with the BMI, BF%, and BW in both Brugada males and control males, even after adjusting for the confounding variables. (3) Conditional logistic regression models analysis showed strong positive association between Brugada syndrome and higher testosterone level (hypertestosteronemia) and strong inverse association between Brugada syndrome and BMI.

Testosterone in Brugada Phenotype and Male Predominance

For the past decade, numerous clinical, experimental, and molecular genetic studies have elucidated Brugada syndrome as a distinct clinical entity.[1-5,17] However, several problems remain unresolved, such as genetic heterogeneity, ethnic difference, and gender difference.[7] Di Diego and Antzelevitch recently suggested the cellular basis for male predominance in Brugada syndrome by using arterially perfused canine right ventricular wedge preparations.[9] Transient outward current (I_{to})-mediated phase 1 AP notch was larger in male dogs than in female dogs in the right ventricular epicardium, but not in the left ventricular epicardium, responsible for the male predominance in the Brugada phenotype. Recent clinical studies suggested that male hormone testosterone might be attributable to gender difference of the prevalence in this

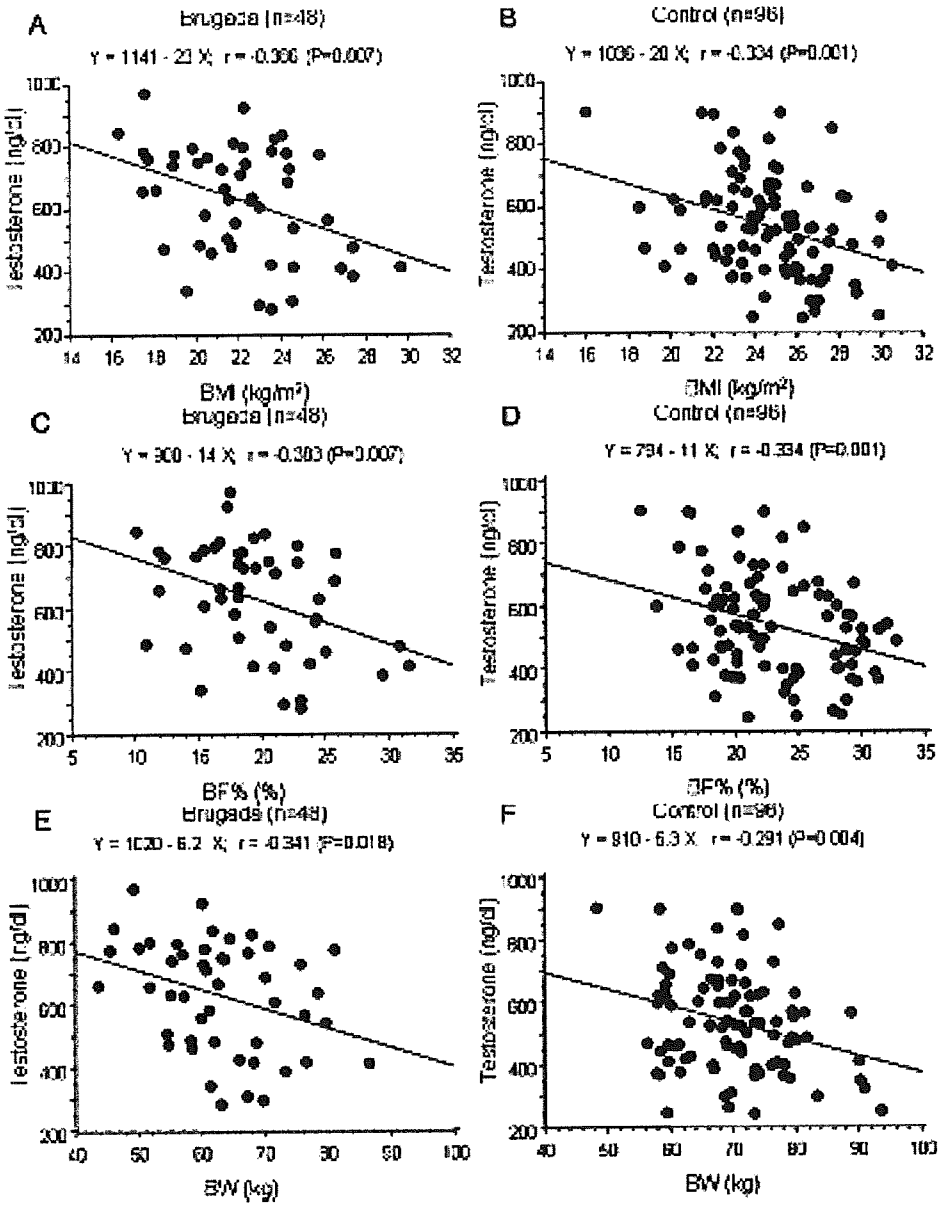

Figure 1. *Correlation between testosterone level and visceral fat parameters; body mass index (BMI) (A and B), body fat percentage (BF%) (C and D), and body weight (BW) (E and F) in the 48 Brugada males and the 96 age-matched control males. Testosterone level was inversely correlated with the BMI, BF%, or BW in both Brugada males and control males.*

syndrome. Matsuo et al. reported two cases of asymptomatic Brugada syndrome in whom typical coved ST-segment elevation disappeared following orchiectomy as therapy for prostate cancer,[21] indicating that testosterone may contribute to the Brugada phenotype in these two cases. Several experimental studies reported that testosterone increased outward potassium currents, such as the rapidly activating component (I_{Kr})[10,11] and the slowly activating component (I_{Ks})[12] of the delayed rectifier potassium current, and the inward rectifier potassium current (I_{K1}),[11] or decreased inward L-type calcium current (I_{Ca-L}).[12] Since the maintenance of the AP dome is determined by the fine balance of currents active at the end of phase 1 of the AP (principally I_{to} and I_{Ca-L}),[22,23] any agents that increase outward currents or decrease inward currents can increase the magnitude of the AP notch, leading to loss of the AP dome (all-or-none repolarization) in the epicardium, but not in the endocardium, contributing to a significant voltage gradient across the ventricular wall during ventricular activation, thus augmenting ST-segment elevation, the Brugada phenotype.[24] Therefore, testosterone would be expected to accentuate the Brugada phenotype. In the present study, males with Brugada syndrome had significantly higher testosterone level than age-matched control males, even after adjusting for age, exercise, stress, current smoking, and medication (hypertension, diabetes, and hyperlipidemia), which may affect the testosterone level. Moreover, conditional logistic regression models analysis showed strong positive association between Brugada syndrome and higher testosterone level (OR: 3.11). Our data suggest a significant role of testosterone, male hormone, in the Brugada phenotype. The

TABLE 5
Odds Ratios of Presence of Hypertestosteronemia and Confounding Risk Factors for Brugada Syndrome in Males

Variable	Odd Ratio	95% Confidence Interval	P Value
Hypertestosteronemia	3.11	1.22–7.93	0.017
Age	0.99	0.95–1.03	0.637
BMI	0.72	0.61–0.85	<0.001
Exercise	1.57	0.87–2.83	0.135
Stress	0.69	0.35–1.35	0.277
Current smoking	0.71	0.26–1.90	0.493
Hypertension	3.12	0.85–11.45	0.087
Diabetes	0.13	0.01–1.27	0.079
Hyperlipidemia	2.14	0.44–10.49	0.348

Hypertestosteronemia was defined as serum testosterone levels ≥ 700 ng/dL.

data also indicate that the male predominance in the Brugada phenotype is at least in part due to testosterone, which is present only in males.

Lower Visceral Fat May Be a Predictor for Brugada Phenotype

Matsuo et al. recently reported in their epidemiologic study that cases with the Brugada-type ECG had significantly lower BMI than that in control subjects.[16] Similarly, in the present study, males with Brugada syndrome had significantly lower visceral fat parameters, BMI, BF%, and BW than those in age-matched control males, even after adjusting for several confounding variables. Moreover, conditional logistic regression models analysis showed strong inverse association between Brugada syndrome and BMI (OR: 0.72). All of the visceral fat parameters were inversely correlated with testosterone level in both Brugada and control males, even after adjusting for the confounding variables. It has been well demonstrated that testosterone level in obese males is decreased compared to normal males of similar age.[13] Tsai et al. reported that lower baseline total testosterone level independently predicted an increase in visceral fat in the Japanese-American male cohort for 7.5 years.[15] Reversely, Marin et al. reported that testosterone treatment of middle-aged abdominally obese males was followed by a decrease of visceral fat mass measured by computerized tomography.[14] These data suggest that primarily higher level of testosterone in Brugada males compared to that in control males may result in lower visceral fat in Brugada males, which would be an "innocent bystander" sign of Brugada phenotype. In reverse, if primary lower visceral fat (body weight loss) would result in higher testosterone level, the weight loss could be a trigger for Brugada phenotype, just like fever is.[25] It is noteworthy that the visceral fat parameters at the clinical cardiac events (VF or syncope) in the 32 symptomatic Brugada males were significantly lower than those at the time of blood sampling for this study. This indicates that testosterone level is expected to be additively higher at the clinical cardiac events, which may contribute to spontaneous episodes of VF or syncope.

Other Hormonal Levels and Serum Electrolytes

Estradiol, female hormone, is reported to reduce the expression of Kv4.3 channels, which are important molecular components of I_{to} currents.[26] However, in contrast to testosterone, other sex hormonal levels including estradiol were not different between the Brugada males and the control males in the present study. Although thyroid hormones are also demonstrated to alter membrane currents, such as I_{to} and I_{Ca-L},[27,28] no significant differences were observed in the thyroid hormonal levels between the two groups in the present study.

On the other hand, serum sodium, potassium, and chloride levels were all significantly higher in the Brugada males than in the control males, even after adjusting for several confounding variables. Recently, many agents and conditions that cause an outward shift in current activity at the end of phase 1 AP have been known to unmask ST-segment elevation, as found in the Brugada syndrome, leading to the acquired form of this disorder.[4,29] Electrolyte abnormalities, such as hyperkalemia, are reported to amplify ST-segment elevation like that in Brugada syndrome.[30] The lower visceral fat found in the Brugada males is expected to decrease serum level of insulin, leptine, a novel adipocyte-derived hormone, or ghrelin, a novel growth hormone-releasing peptide, suppressing β-adrenergic receptor or plasma norepinephrine level, resulting in an increase of serum potassium level.[31,32] Further studies including measurement of levels of insulin, leptine, and ghrelin will be required to elucidate the precise mechanism.

Study Limitations

Although the testosterone level was significantly higher in the Brugada males than in the control males, no statistically significant correlations were observed between the testosterone level and the ST amplitude in the Brugada males. The degree of the ST-segment elevation is variable between Brugada patients because it is influenced by several factors other than sex hormonal levels or electrolytes levels, such as basal autonomic tone, presence of *SCN5A* mutation, or probably intrinsic current density of I_{to}, etc., in the right ventricular epicardial cells. The threshold of ST-segment elevation for spontaneous induction of VF also varies between Brugada patients. Therefore, the Brugada phenotype, such as ST-segment elevation or spontaneous induction of VF, may correlate with the testosterone level day to day individually (intra-personally) in each Brugada male, but may not correlate among the pooled data obtained from many Brugada males, probably due to inter-person difference of the ST-segment elevation.

There were no significant differences in testosterone level between symptomatic and asymptomatic Brugada males, between Brugada males with spontaneous ST elevation and those with sodium channel blocker-induced ST elevation, or between Brugada males with and without *SCN5A* mutation, all of which are probably due to a relatively small number of Brugada males in the present study. Further evaluation with increasing number of Brugada males will be required.

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Prehypertension Increases the Risk for Renal Arteriosclerosis in Autopsies: The Hisayama Study

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ABSTRACT

Information regarding the association between prehypertension BP level and renal arteriosclerosis is limited. In 652 consecutive population-based autopsy samples without hypertension treatment before death, the relationship between the severity of renal arteriosclerosis and BP levels classified according to the criteria of the Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure was examined. The age- and gender-adjusted frequencies of renal arteriosclerosis linearly increased with elevating BP levels; both hypertensive and prehypertensive subjects had significantly higher frequencies of renal arteriosclerosis than subjects with normal BP (normal 11.9%; prehypertension 28.5%; stage 1 hypertension 32.9%; stage 2 hypertension 58.2%; all $P < 0.01$ *versus* normal). In a logistic regression model, prehypertension was significantly associated with renal arteriosclerosis after adjustment for other cardiovascular risk factors (prehypertension multivariate-adjusted odds ratio [mOR] 5.99 [95% confidence interval (CI) 2.20 to 15.97]; stage 1 hypertension mOR 6.99 [95% CI 2.61 to 18.72]; stage 2 hypertension mOR 22.21 [95% CI 8.35 to 59.08]). This significant association was observed for all renal arterial sizes. The similar association was also observed for arteriolar hyalinosis. When the subjects were divided into those with and those without target organ damage, the impact of prehypertension on renal arteriosclerosis was similar for both groups (subjects without target organ damage mOR 5.04 [95% CI 1.36 to 18.62]; subjects with target organ damage mOR 6.42 [95% CI 1.29 to 32.04]). These findings suggest that both hypertension and prehypertension are associated significantly with the severity of renal arteriosclerosis, regardless of the presence or absence of target organ damage.

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Hypertension has been recognized as one of the major risk factors for the development of ESRD.[1,2] Nephrosclerosis is characterized pathologically by focal or global glomerular sclerosis and renal arteriosclerosis and is frequently found in individuals with hypertension.[3–5] Meanwhile, several prospective studies have indicated that the impressive increase in the risk for cardiovascular disease or in the risk for progression to hypertension started at a BP level of ≥120/80 mmHg.[6–8] On the basis of these findings, the Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure (JNC-7) introduced a "prehypertension" category in which BP is 120 to 139/80 to 89 mmHg and for which health-promoting lifestyle modifications are recommended to prevent cardiovascular disease.[9]

A prospective population-based study of cardiovascular disease has been carried out since 1961 in

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the town of Hisayama on Kyushu Island in southern Japan. The most characteristic feature of the Hisayama Study is that the cause of death has been verified by autopsy for approximately 80% of deceased subjects in the study population.[10–13] Our previous autopsy reports of Hisayama residents showed that systolic BP (SBP) was closely related to the progression of glomerular sclerosis, renal arteriolar hyalinosis, and renal arteriosclerosis.[12,13] To our knowledge, the Honolulu Heart Program is the only other population-based study that has examined this issue, although the autopsy rate was not high (20.6%).[14,15] Their findings also suggested that diastolic BP (DBP) was an independent predictor of glomerular sclerosis, renal arteriolar hyalinosis, and renal arteriosclerosis.[14,15] However, the association between categorized BP levels and renal vascular changes was not assessed in these studies or in ours. In this study, we examined the relationship between BP levels and renal arteriosclerosis, focusing on prehypertension, in population-based autopsy samples of the Hisayama Study, taking into account other cardiovascular risk factors as well as renal artery size.

RESULTS

The baseline characteristics of the 652 autopsy subjects are represented according to the BP levels in Table 1. The subjects with stage 1 or stage 2 hypertension were older than those with normal BP. The proportions of women gradually increased with elevating BP levels. The mean GFR values decreased significantly in stage 2 hypertension relative to normal BP level, whereas serum creatinine levels did not change across BP levels. The frequencies of proteinuria and history of cardiovascular disease were significantly higher in subjects with stage 2 hypertension, and that of electrocardiogram (ECG) abnormalities increased linearly with elevating BP levels. The mean values of total cholesterol were significantly higher in subjects with hypertensive or prehypertensive BP levels, whereas the frequency of glucose intolerance and the mean value of body mass index (BMI) did not change across BP levels. The frequency of current smoking decreased gradually with elevating BP levels, but no such tendency was observed for the frequency of alcohol intake.

Figure 1 presents the age- and gender-adjusted frequencies of renal arteriosclerosis, arteriolar hyalinosis, and glomerular sclerosis according to BP classification. The frequencies of renal arteriosclerosis and arteriolar hyalinosis linearly increased with elevating BP levels; not only hypertensive subjects but also prehypertensive subjects had a significantly higher frequency of renal arteriosclerosis and arteriolar hyalinosis compared with subjects with normal BP. Likewise, the age- and gender-adjusted mean values of the wall-lumen ratio of renal arteries decreased linearly (normal 5.10; prehypertension 4.16; stage 1 hypertension 3.96; stage 2 hypertension 3.47; all $P < 0.01$ *versus* normal), and those of the arteriolar hyalinosis index increased gradually with elevating BP levels (normal 1.21; prehypertension 1.29 [$P < 0.05$ *versus* normal]; stage 1 hypertension 1.29 [$P < 0.05$]; stage 2 hypertension 1.38 [$P < 0.01$]). The severity of glomerular sclerosis increased significantly in only stage 2 hypertension. The age- and gender-adjusted odds ratios (OR) of renal arteriosclerosis and arteriolar hyalinosis were significantly higher in prehypertension subjects and in hypertension subjects than in normal ones (Table 2). This association remained substantially unchanged even after adjustment for age at death, gender, total cholesterol, glucose intolerance, BMI, smoking habits, and alcohol intake. Furthermore, we di-

Table 1. Mean values or frequencies of potential risk factors and laboratory variables according to BP classification for 652 autopsy subjects[a]

Variables	BP Classification			
	Normal ($n = 106$)	Prehypertension ($n = 172$)	Stage 1 HT ($n = 176$)	Stage 2 HT ($n = 198$)
Age at death (yr)	70 ± 12	72 ± 13	75 ± 12[b]	79 ± 11[b]
Women (%)	37.7	40.7	43.2	52.5[c]
SBP (mmHg)	109 ± 8	129 ± 6[b]	148 ± 7[b]	179 ± 18[b]
DBP (mmHg)	66 ± 7	73 ± 10[b]	80 ± 11[b]	91 ± 13[b]
GFR (ml/min per 1.73 m^2)	77.1 ± 15.8	75.5 ± 18.8	76.3 ± 20.6	70.2 ± 19.5[c]
Serum creatinine (μmol/L)	83.5 (57.3 to 121.8)	85.4 (54.7 to 133.4)	84.0 (51.5 to 137.2)	88.2 (49.0 to 158.7)
Proteinuria (%)	9.2	6.8	10.8	23.4[b]
History of cardiovascular disease (%)	4.7	11.6	9.1	12.1[c]
Electrocardiogram abnormalities (%)	6.8	15.4[c]	25.7[b]	40.8[b]
Total cholesterol (mmol/L)	4.25 ± 1.07	4.67 ± 1.07[b]	4.60 ± 1.22[c]	4.63 ± 1.10[c]
Glucose intolerance (%)	13.2	21.5	18.2	22.2
BMI (kg/m^2)	20.2 ± 2.8	20.7 ± 3.2	20.7 ± 3.3	20.7 ± 3.1
Smoking habits (%)	47.6	41.3	39.7	35.4[c]
Alcohol intake (%)	24.0	32.6	31.2	30.6

[a]Data are means ± SD or percentage. GFR determined by Modification of Diet in Renal Disease Study Group formula. Glomerular filtration rate and serum creatinine were measured in 442 subjects who died after 1977. Geometric mean values and 95% confidence intervals (CI) of serum creatinine are shown because of the skewed distribution. BMI, body mass index; DBP, diastolic BP; HT, hypertension; SBP, systolic BP.
[b]$P < 0.01$, [c]$P < 0.05$ *versus* normal.

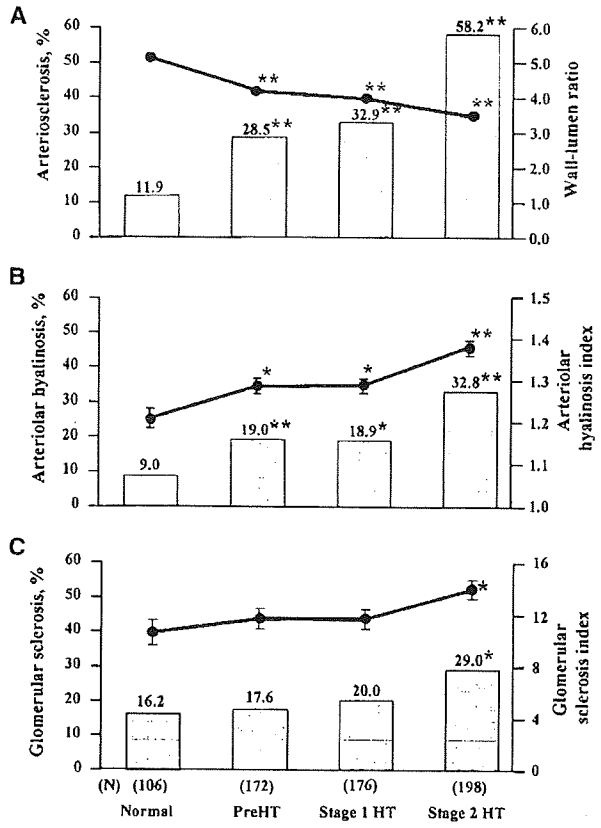

Figure 1. Age- and gender-adjusted frequencies of renal arteriosclerosis (A), arteriolar hyalinosis (B), and glomerular sclerosis (C) according to BP classification among 652 autopsy subjects. Solid lines indicate age- and gender-adjusted mean values of wall-lumen ratio, arteriolar hyalinosis index, and glomerular sclerosis, respectively. PreHT, prehypertension; HT, hypertension. *$P < 0.05$, **$P < 0.01$ *versus* normal.

vided prehypertension into two subcategories—BP 120 to 129/80 to 84 mmHg and BP 130 to 139/85 to 89 mmHg—and examined the association between BP level and renal arteriosclerosis. As a result, the risk for having renal arteriosclerosis was significantly increased in both BP subcategories even after adjustment for the previously mentioned cardiovascular risk factors (BP 120 to 129/80 to 84 mmHg OR 5.93 [95% confidence interval (CI) 2.08 to 16.91; $P < 0.01$]; BP 130 to 139/85 to 89 mmHg OR 5.92 [95% CI 2.02 to 17.29; $P < 0.01$]).

We examined whether the association between BP and renal arteriosclerosis differs by the presence or absence of target organ damage. As shown in Figure 2A, the age- and gender-adjusted frequencies of renal arteriosclerosis were higher in the group with target organ damage than in the group without it, regardless of BP level. In both groups, however, the frequencies of renal arteriosclerosis increased significantly with elevating BP levels; the difference was significant between the normal and both the prehypertension and hypertension categories. Likewise, the mean values of the wall-lumen ratio of renal arteries were significantly lower in BP levels of prehypertension and hypertension than in normal BP level for both the target-organ-damaged and target-organ-undamaged groups (Figure 2B). As shown in Table 3, the impact of prehypertension on renal arteriosclerosis was similar for both the damaged and undamaged groups after adjustment for the previously mentioned cardiovascular risk factors (without target organ damage OR 5.04 [95% CI 1.36 to 18.62; $P < 0.05$]; with target organ damage OR 6.42 [95% CI 1.29 to 32.04; $P < 0.05$]).

Finally, we examined the associations between BP levels and renal arteriosclerosis by the size of renal arteries using logistic regression analysis (Table 4). After adjustment for the previously mentioned cardiovascular risk factors, both prehypertension and hypertension significantly increased the risk for having renal arteriosclerosis in all arterial sizes. For smaller arteries (<300 μm), the risk for arteriosclerosis significantly and linearly increased with elevating BP levels, whereas this linear association was diminished for larger arteries (≥300 μm).

DISCUSSION

In this population-based autopsy survey, we histopathologically examined the relationship between categorized BP levels classified according to the JNC-7 criteria and renal arteriosclerosis. The results showed that both hypertension and prehypertension were associated significantly with renal arteriosclerosis, without regard for the presence or absence of target organ damage or for the size of intrarenal arteries. The relationships between BP and renal histopathologic changes also have been reported in the several biopsy-based studies of living subjects. Lhotta *et al.*[16] showed in patients who underwent biopsy that SBP was associated significantly with the frequencies of glomerular sclerosis and arteriolar hyalinosis. According to the study for patients with biopsy-proven IgA nephropathy, patients with hypertension, defined as BP ≥140/90 mmHg, had more severe glomerular sclerosis, interstitial fibrosis/tubular atrophy, interstitial infiltration, and atherosclerosis compared with those without hypertension.[17] In a similar biopsy study for IgA nephropathy, prehypertension (BP 120 to 139/80 to 89 mmHg) was associated significantly with the severity of mesangial proliferation and arteriolar changes, including intimal thickening, intimal duplication or hyalinosis, but not glomerular sclerosis.[18] These findings are in accordance with those of our study.

Several recent reports have shown that the risk for the development of cardiovascular disease or the risk for the progression to hypertension initiates an increase in BP levels of ≥120/80 mmHg. A meta-analysis of individual data for 1 million adults in 61 prospective studies indicated that the mortality from both ischemic heart disease and stroke increased progressively and linearly from BP levels as low as SBP of 115 mmHg and DBP of 75 mmHg in middle and old age.[6] In addi-

Table 2. Age- and gender-adjusted or multivariate-adjusted OR for renal arteriosclerosis, arteriolar hyalinosis, and glomerular sclerosis according to BP classification among 652 autopsy subjects

Parameter	BP Classification			
	Normal	Prehypertension	Stage 1 HT	Stage 2 HT
Arteriosclerosis				
age and gender adjusted				
OR[b]	1.00	4.21[d]	4.97[d]	16.57[d]
CI	Reference	1.85 to 9.60	2.20 to 11.21	7.41 to 37.05
multivariate adjusted				
OR[c]	1.00	5.99[d]	6.99[d]	22.21[d]
CI	Reference	2.20 to 15.97	2.61 to 18.72	8.35 to 59.08
Arteriolar hyalinosis				
age and gender adjusted				
OR[b]	1.00	2.70[e]	2.59[e]	5.84[d]
CI	Reference	1.19 to 6.13	1.14 to 5.89	2.65 to 12.88
multivariate adjusted				
OR[c]	1.00	2.36[e]	2.19	5.42[d]
CI	Reference	1.01 to 5.50	0.93 to 5.16	2.37 to 12.38
Glomerular sclerosis				
age and gender adjusted				
OR[b]	1.00	1.03	1.24	2.06[e]
CI	Reference	0.50 to 2.12	0.62 to 2.49	1.06 to 4.02
multivariate adjusted				
OR[c]	1.00	1.01	1.21	2.21[e]
CI	Reference	0.46 to 2.21	0.56 to 2.61	1.06 to 4.64

[a]OR odds ratio.
[b]Adjusted for age at death and gender.
[c]Adjusted for age at death, gender, total cholesterol, glucose intolerance, BMI, smoking habit, and alcohol intake.
[d]$P < 0.01$, [e]$P < 0.05$ *versus* normal.

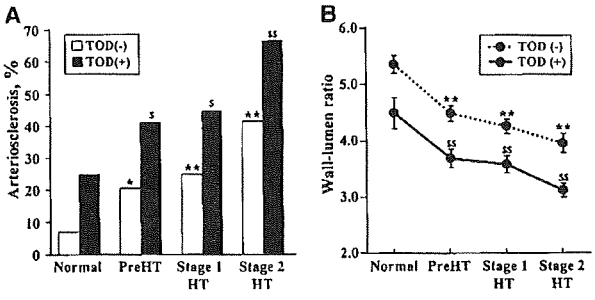

Figure 2. Age- and gender-adjusted frequencies of renal arteriosclerosis (A) and mean values of wall-lumen ratio of the renal arteries (B) according to BP classification by the presence or absence of target organ damage among 652 autopsy subjects. TOD, target organ damage; $^{*}P < 0.05$, $^{**}P < 0.01$ *versus* normal in target organ damage (−); $^{\$}P < 0.05$, $^{\$\$}P < 0.01$ *versus* normal in target organ damage (+).

tion, longitudinal data that were obtained from the Framingham Heart Study indicated that high-normal BP and normal BP, defined by JNC-6, were associated with the occurrence of cardiovascular disease.[7] Moreover, according to the randomized, controlled trial conducted in the Modification of Diet in Renal Disease (MDRD) study, low target BP (mean BP <92 mmHg, equivalent to a BP <125/75 mmHg) reduced the risk for developing kidney failure by approximately 30% compared with the usual target BP (mean BP <107 mmHg, equivalent to a BP <140/90 mmHg).[19] Our findings also showed that prehypertension levels were significantly associated with renal arteriosclerosis and arteriolar hyalinosis. It may be reasonable to suppose that prehypertension promotes systemic arteriosclerosis including renal vascular changes and causes cardiovascular disease and renal dysfunction.

It is possible that prehypertension is not the cause of renal arteriosclerosis but the result of renal vascular changes or organ damages by other cardiovascular risk factors. In this study, however, prehypertension was clearly associated with renal arteriosclerosis, regardless of the presence or absence of target organ damage, and this association was significant even after adjustment for other cardiovascular risk factors. This suggests that a slight increase in BP to prehypertension levels was associated independently with the severity of renal arteriosclerosis. Therefore, it is possible that antihypertensive treatment with BP-lowering <120/80 mmHg prevents the progression of renal arteriosclerosis, regardless of the presence or absence of target organ damage.

In our study, the relationship between BP levels and renal arteriosclerosis differed somewhat according to the size of renal arteries; the risk for renal arteriosclerosis increased significantly and linearly with elevating BP levels in smaller arteries (<300 μm), including arterioles, whereas this phenomenon was diminished in larger arteries (≥300 μm). Instead, the impact of total cholesterol levels was reinforced with elevating renal arterial size in our subjects (data not shown). In autopsy

Table 3. Multivariate-adjusted OR for renal arteriosclerosis according to BP classification by the presence or absence of target organ damage.

Parameter	BP Classification			
	Normal	Prehypertension	Stage 1 HT	Stage 2 HT
Target organ damage (−)[a]				
population at risk	83	109	103	70
OR[b]	1.00	5.04[c]	6.05[d]	18.81[d]
95% CI	Reference	1.36 to 18.62	1.65 to 22.20	4.98 to 71.01
Target organ damage (+)[a]				
population at risk	23	63	73	128
OR[b]	1.00	6.42[c]	7.21[c]	18.02[d]
95% CI	Reference	1.29 to 32.04	1.46 to 35.65	3.75 to 86.47

[a]Target organ damage was defined as the presence of preexisting cardiovascular disease, electrocardiogram abnormalities, proteinuria, or GFR <60 ml/min per 1.73 m^2.
[b]Adjusted for age at death, gender, total cholesterol, glucose intolerance, BMI, smoking habits, and alcohol intake.
[c]$P < 0.05$, [d]$P < 0.01$ *versus* normal.

Table 4. Multivariate-adjusted OR for renal arteriosclerosis according to BP classification by the size of renal arteries among 652 autopsy subjects

Size of Renal Arteries (μm)	BP Classification			
	Normal	Prehypertension	Stage 1 HT	Stage 2 HT
60 to 149				
OR[a]	1.00	4.01[b]	4.13[b]	12.00[b]
95% CI	Reference	1.59 to 10.10	1.64 to 10.39	4.84 to 29.68
150 to 299				
OR[a]	1.00	2.39[c]	4.27[b]	9.94[b]
95% CI	Reference	1.13 to 5.07	2.05 to 8.89	4.77 to 20.72
300 to 499				
OR[a]	1.00	4.21[b]	2.73[c]	6.21[b]
95% CI	Reference	1.68 to 10.60	1.06 to 6.99	2.49 to 15.47
≥500				
OR[a]	1.00	3.80[c]	2.59	3.08
95% CI	Reference	1.09 to 13.30	0.72 to 9.34	0.86 to 11.04

[a]Adjusted for age at death, gender, total cholesterol, glucose intolerance, BMI, smoking habits, and alcohol intake.
[b]$P < 0.01$, [c]$P < 0.05$ *versus* normal.

findings from the Honolulu Heart Program, BP was associated strongly with the intimal thickness of renal arteries with an outer diameter of 80 to 300 μm, but there were no correlations between the intimal thickness of these renal arteries and other cardiovascular risk factors, such as total cholesterol, triglycerides, blood glucose, and smoking.[14] It is feasible to speculate that the degree of the atherogenic effects of risk factors varies according to artery size and that hypertension affects small arteries notably.

Several limitations of our study should be discussed. First, our findings might be biased by the exclusion of 187 subjects who were taking antihypertensive medications. The mean values of SBP, DBP, serum creatinine, and total cholesterol and the frequencies of proteinuria, ECG abnormalities, glucose intolerance, and history of cardiovascular disease were significantly higher and the mean values of wall-lumen ratio were significantly lower in subjects who were taking antihypertensive medications than in the 652 subjects without antihypertensive medications in the present study. This bias has the potential to underestimate the impact of hypertension or other cardiovascular risk factors on renal arteriosclerosis. However, it is unlikely that this bias affects the association between prehypertension and renal arteriosclerosis, because prehypertensive subjects did not use antihypertensive medication. Second, only a single BP measurement was obtained at the baseline examination in the recumbent position. This imperfect measurement of BP might have resulted in a misclassification of our study subjects into different BP categories and a consequent dilution of our estimates of BP's impact on renal arteriosclerosis. Third, this is a cross-sectional study. Therefore, it is difficult to infer causality between prehypertension and risk for progression of renal arteriosclerosis, because it may be presumed that BP increased as a result of renal ischemia by preexisting renal arteriosclerosis, acting mainly through the renin-angiotensin system. In any case, our findings suggest that subjects with prehypertension should be considered as those with more progressive renal arteriosclerosis. Fourth, several variables used in this study were less accurate. We used the MDRD equation to estimate GFR; this formula is notoriously inaccurate in patients with normal kidney function. Proteinuria was established as 1+ or more on the dipstick; this would have missed all subjects with microalbuminuria. In addition,

the definition of glucose tolerance varied depending on when the examination was done. These facts might lead to the misclassification of a normal subgroup without any risk factor and affect the cutoff value of each histologic parameter. However, it seems to be unlikely that this limitation distorted the associations between BP levels and severity of renal arteriosclerosis, because BP levels showed the dosage-dependent association with the continuous values of wall-lumen ratio. Finally, this study is based on autopsy and the proportion of aged people is extremely high. Therefore, its findings cannot be applied to the overall living population. However, we believe that our findings provide useful information toward a better understanding of the pathogenesis of renal arteriosclerosis.

CONCLUSION

Prehypertension level classified by JNC-7 was associated significantly with the severity of renal arteriosclerosis. Therefore, prehypertensive individuals should be considered a high-risk population, regardless of the presence or absence of target organ damage. Our findings emphasize the need to determine whether the lowering of goal BP in hypertension management can prevent the progression of renal and systemic arteriosclerosis.

CONCISE METHODS

Study Population

The population of the town of Hisayama is approximately 7500, and data from the national census show it to be representative of Japan as a whole.[10,11] The study design and characteristics of the subject population have been described in detail elsewhere.[12,13] Briefly, from January 1962 to December 1994, a total of 1742 Hisayama residents of all age groups died, 1394 (80.0%) of whom underwent autopsy. The autopsy rate was not different between men (78.7%) and women (81.6%). Among these consecutive autopsy subjects, 1168 participated in at least one of the six health examinations conducted in 1961, 1967, 1974, 1978, 1983, and 1988. For every examination, the participation rate exceeded >80% of all Hisayama residents 40 yr or older. After exclusion of 98 subjects who lacked preserved renal tissues, 33 with degenerated or small renal tissues, 80 who underwent autopsy at other hospitals, 118 who had no health examination data within 7 yr before death, and 187 who had been treated with antihypertensive medications, 652 subjects (362 men and 290 women) were included in this study. The mean period from the most recent health examination to death was 3.6 ± 1.8 yr.

Morphologic Examination of Renal Tissue

The methods of morphologic examination of renal tissue have been described in detail elsewhere.[13] Briefly, for light microscopic study, paraffin-embedded renal tissues that were obtained by standard autopsy methods were cut at a 2-μm thickness and stained with periodic acid-Schiff reagent. The wall-lumen ratio was evaluated as the severity of arteriosclerosis by the method of Kernohan *et al.*[20] For each specimen, all arteries with an outer diameter >60 μm were examined using an eyepiece micrometer. The outer diameter and the lumen diameter of the least axis of the elliptic profile were directly measured. The wall-lumen ratio was calculated in each artery as lumen diameter/(outer diameter − lumen diameter)/2, and the mean value for all arteries in all subjects was used as the index of arteriosclerosis. We further classified all arteries into four categories according to the outer diameters of the renal arteries—60 to 149, 150 to 299, 300 to 499, and ≥500 μm—and calculated the mean values of the wall-lumen ratio by the previously mentioned categories.

The severity of arteriolar hyalinosis was assessed semiquantitatively by the method of Barder *et al.*[21] For each tissue specimen, 50 arterioles were examined and the severity of the lesion in each arteriole was graded from 1+ to 4+ according to the extent of arteriolar hyalinosis. The arteriolar hyalinosis index was calculated by the following formula: Arteriolar hyalinosis index = $(n_1 \times 1 + n_2 \times 2 + n_3 \times 3 + n_4 \times 4)/50$. Here, n_1, n_2, n_3, and n_4 indicate the number of arterioles showing hyalinosis scores of 1+ to 4+, respectively.

The semiquantitative score was used to evaluate the severity of glomerular sclerosis by the method of Raij *et al.*[22] For each tissue specimen, 100 glomeruli from the superficial to deep cortex were examined uniformly, and the severity of the lesion in each glomerulus was graded from 0 to 4+ according to the percentage of glomerular sclerosis. The glomerular sclerosis index was calculated by the following formula: Glomerular sclerosis index = $(n_0 \times 0 + n_1 \times 1 + n_2 \times 2 + n_3 \times 3 + n_4 \times 4)/4$. Here, n_0, n_1, n_2, n_3, and n_4 indicate the number of glomeruli showing sclerotic lesion scores of 0 to 4+, respectively.

Definition of Renal Arteriosclerosis, Arteriolar Hyalinosis, and Glomerular Sclerosis

To differentiate the effects of cardiovascular risk factors from age-related changes, we selected 103 subjects who had none of the following characteristics: Proteinuria, kidney failure, hypertension, glucose intolerance, or primary renal disease at autopsy. Using this subgroup, the cutoff limits were defined as below the 10th percentile or above the 90th percentile of each histologic parameter distribution; that is, renal arteriosclerosis, arteriolar hyalinosis, and glomerular sclerosis were defined as a wall-lumen ratio <3.37, an arteriolar hyalinosis index >1.44, and a glomerular sclerosis index >17.0, respectively. In the analysis by the size of renal arteries, furthermore, renal arteriosclerosis was defined as below the lower 10th percentile for mean values of the wall-lumen ratio by size (60 to 149 μm: wall-lumen ratio <3.56; 150 to 299 μm: wall-lumen ratio <2.65; 300 to 499 μm: wall-lumen ratio <2.64; ≥500 μm: wall-lumen ratio <2.44).

Risk Factors

BP was measured three times after a single rest period of at least 5 min using a standard mercury sphygmomanometer with the subject in the recumbent position. The mean of the three measurements was used for the analysis. BP levels were categorized according to the criteria recommended by JNC-7[9] (normal: SBP <120 mmHg and DBP <80 mmHg; prehypertension: SBP 120 to 139 mmHg or DBP 80 to 89 mmHg; stage 1 hypertension: SBP 140 to 159 mmHg or DBP 90 to 99

mmHg; stage 2 hypertension: SBP ≥160 mmHg or DBP ≥100 mmHg).

Glucose intolerance was defined by an oral glucose tolerance test in the subjects with glycosuria in 1961 and 1967; by fasting and postprandial glucose concentrations in 1974, 1978, and 1983; and by a 75-g oral glucose tolerance test in 1988, in addition to medical history of diabetes. ECG abnormalities were defined as Minnesota codes 3-1 and/or 4-1, -2, -3. Serum total cholesterol levels were measured by the Zak-Henly method with a modification by Yoshikawa in 1961 and 1967, by the Zurkowski method in 1974, and by the enzymatic method after 1978. Serum creatinine concentration was measured by the Jaffe method after 1974, and GFR was calculated by the MDRD Study Group formula.[23] Freshly voided urine samples were tested by the sulfosalicylic acid method in 1961 and 1967 and by the dipstick method after 1974. Proteinuria was defined as 1+ or more. Body height and weight were measured in light clothing without shoes, and the BMI (kg/m^2) was calculated. Information on antihypertensive medication, alcohol intake, and smoking habits was obtained through a standard questionnaire and classified as current habitual use or a lack thereof. All available information about potential cardiovascular diseases, including stroke, myocardial infarction, and coronary intervention, was gathered and reviewed by a panel of physician members of the Hisayama Study to determine the occurrence of cardiovascular disease under the standard criteria. A history of cardiovascular disease was determined on the basis of this information. Target organ damage was defined as the presence of ECG abnormalities, proteinuria, GFR <60 ml/min per 1.73 m^2, or a history of cardiovascular disease.

Statistical Analyses

SAS software (SAS Institute, Cary, NC) was used to perform all statistical analyses. The crude or age- and gender-adjusted mean values and frequencies of variables were compared among BP levels using Dunnett *t* test or logistic regression analysis as appropriate. The age- and gender-adjusted or multivariate-adjusted OR and 95% CI were calculated by a logistic regression analysis. $P < 0.05$ was considered statistically significant in all analyses.

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DISCLOSURES

None.

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Impact of Metabolic Syndrome on the Development of Cardiovascular Disease in a General Japanese Population

The Hisayama Study

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Background and Purpose—The metabolic syndrome (MetS) is associated with an increased risk of cardiovascular disease (CVD) events in general populations. However, well-designed prospective studies in Asian populations are very limited.

Methods—We prospectively evaluated a total of 2452 community-dwelling Japanese individuals aged 40 years or older from 1988 to 2002 and examined the effects of MetS defined by the modified National Cholesterol Education Program Adult Treatment Panel III criteria on incident CVD.

Results—The prevalence of the MetS was 21% in men and 30% in women at baseline. During the follow up, 307 CVD events occurred. Compared with those without MetS, the age-adjusted incidence of CVD (per 1000 person-years) was significantly higher in subjects with the MetS in both men (21.8 versus 11.6, $P<0.01$) and women (12.9 versus 6.5, $P<0.01$). The risk of CVD events was significantly higher even after adjusting for the following confounding factors: age, proteinuria, electrocardiographic abnormalities, serum total cholesterol, smoking habits, alcohol intake, and regular exercise (hazard ratio, 1.86; 95% CI, 1.32 to 2.62 in men and hazard ratio, 1.70; 95% CI, 1.22 to 2.36 in women). The risk of incident CVD was found to increase with the number of components of MetS and became significantly predictive when the number of components reached 3. Similar associations were also observed when CVD was divided into coronary heart disease and stroke.

Conclusions—Our findings suggest that MetS is a significant risk factor for the development of CVD in the Japanese middle-aged population. (***Stroke*. 2007;38:2063-2069.)**

Key Words: cardiovascular disease ■ epidemiology ■ metabolic syndrome ■ myocardial infarction ■ stroke

Metabolic syndrome (MetS), also known as syndrome X,[1] the insulin resistance syndrome,[2] and deadly quartet,[3] is a constellation of dyslipidemia, central obesity, elevated blood pressure, and impaired glucose tolerance. It is associated with high risk for the development of type 2 diabetes mellitus and cardiovascular disease (CVD).[4–7] In the past several years, a great deal of attention has been directed to it attributable to increases in its prevalence worldwide[6] and its association with CVD morbidity and mortality. Although each of the components of MetS has been shown to increase CVD risk,[8–12] the presence of MetS has been reported to identify additional risk.[7] Different prospective studies[7,13–25] based on the definitions from the National Cholesterol Education Program's (NCEP) Third Adult Treatment Panel Report III[5] and World Health Organization[26] showed that subjects with MetS are at increased risk of incident CVD, CVD mortality, and all-cause mortality in the general population with or without diabetes mellitus. However, most of these studies were based on Western populations, and well-designed prospective studies in Asian populations are very limited.[27–29] Thus, there is a dearth of literature regarding the relationship of MetS with incident CVD based on general population cohorts with a reasonable length of follow-up time in ethnic groups other than whites. In this study, we examined the impact of MetS on CVD events in a general Japanese population cohort based on 14-year prospective follow-up data.

Materials and Methods

Study Population

The Hisayama Study, an epidemiological study of cerebro- and cardiovascular diseases, was established in 1961 in Hisayama Town, a suburban community adjacent to Fukuoka City, a metropolitan area of Kyushu Island in southern Japan. The population of the town is

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approximately 7500, and full community surveys of the residents have been repeated since 1961.[30] In 1988, a screening survey for the present study was performed in the town. A detailed description of this survey was published previously.[31] Briefly, a total of 2736 residents aged 40 years or over (80.7% of the total population of this age group) consented to participate in the examination and underwent a comprehensive assessment. After excluding 102 subjects with a history of coronary heart disease or stroke, as determined by a questionnaire and medical records, one subject for whom no blood sample was obtained, 120 subjects with postprandial blood sample, and 61 subjects without the measurements of their waist circumferences, the remaining 2452 subjects (1050 men and 1402 women) were enrolled in this study.

Follow-Up Survey

The subjects were followed prospectively from December 1988 to November 2002 by repeated health examinations. Health status was checked yearly by mail or telephone for any subjects who did not undergo a regular examination or who had moved out of town. We also established a daily monitoring system among the study team and local physicians or members of the town's health and welfare office. When a subject died, an autopsy was performed at the Department of Pathology of Kyushu University. During the follow-up period, only one subject was lost to follow up and 479 subjects died, of whom 362 (75.6%) underwent autopsy.

Definition of Cardiovascular Events

CVD was defined as first-ever development of coronary heart disease (CHD) or stroke. The criteria for a diagnosis of CHD included first-ever acute myocardial infarction, silent myocardial infarction, sudden cardiac death within 1 hour after the onset of acute illness, or coronary artery disease followed by coronary artery bypass surgery or angioplasty.[32] Acute myocardial infarction was diagnosed when a subject met at least 2 of the following criteria: (1) typical symptoms, including prolonged severe anterior chest pain; (2) abnormal cardiac enzymes more than twice the upper limit of the normal range; (3) evolving diagnostic electrocardiographic changes; and (4) morphological changes, including local asynergy of cardiac wall motion on electrocardiography, persistent perfusion defect on cardiac scintigraphy, or myocardial necrosis or scars >1 cm long accompanied by coronary atherosclerosis at autopsy. Silent myocardial infarction was defined as myocardial scarring without any historical indication of clinical symptoms or abnormal cardiac enzyme changes. Stroke was defined as a sudden onset of nonconvulsive and focal neurological deficit persisting for >24 hours. The diagnosis of stroke and the determination of its pathological type were based on the clinical history, neurological examination, and all available clinical data, including brain CT/MRI and autopsy findings. Stroke was classified as either ischemic or hemorrhagic.[32]

Risk Factor Measurement

At the baseline examination, each participant completed a self-administered questionnaire covering medical history, exercise, treatment for hypertension or diabetes, smoking habits, and alcohol intake. The questionnaire was checked by trained interviewers at the screening. The subjects engaging in sports or other forms of exertion ≥3 times a week during their leisure time made up a regular exercise group. Smoking habits and alcohol intake were classified into currently habitual or not.

Blood pressure was measured 3 times using a standard mercury sphygmomanometer in the sitting position after rest for at least 5 minutes. The mean of the 3 measurements was used for the analysis. Hypertension was defined as blood pressure ≥140/90 mm Hg and/or current use of antihypertensive agents. The waist circumference was measured at the umbilical level in a standing position by a trained staff member. Body height and weight were measured in light clothing without shoes and the body mass index (kg/m^2) was calculated. Electrocardiographic abnormalities were defined as left ventricular hypertrophy (Minnesota code, 3 to 1) and/or ST depression (Minnesota code, 4 to 1, 2, or 3).

Blood samples were collected from an antecubital vein after an overnight fast for the determination of lipids and blood glucose levels. Serum total cholesterol, triglycerides, and high-density lipoprotein cholesterol concentrations were determined enzymatically. Fasting blood glucose levels were measured by the glucose oxidase method. Diabetes was defined as fasting blood glucose ≥126 mg/dL (7.0 mmol/L) and/or current use of insulin or oral medication for diabetes. Fresh voided urine samples were collected at the examination and proteinuria was defined as 1+ or more using a reagent strip.

Definition of Metabolic Syndrome

MetS was defined by using criteria recommended in the NCEP Adult Treatment Panel III guideline[5] with a modification. Specifically, abdominal obesity was defined as a waist circumference >90 cm in men and >80 cm in women according to International Obesity Task Force central obesity criteria for Asia.[33] Elevated blood pressure was defined as average systolic/diastolic blood pressures of ≥130/85 mm Hg and/or current use of antihypertensive medicine. Hypertriglyceridemia was defined as serum triglycerides of ≥1.69 mmol/L. Low high-density lipoprotein cholesterol was defined as serum high-density lipoprotein cholesterol levels of <1.03 mmol/L in men and of <1.29 mmol/L in women. Elevated blood glucose level was defined as fasting blood glucose of ≥6.10 mmol/L and/or current use of insulin or oral medication for diabetes. MetS was defined as the presence of 3 or more of these components.[5]

Statistical Analysis

The SAS software package (SAS Institute, Inc, Cary, NC) was used to perform all statistical analyses. Serum triglycerides were transformed into logarithms to improve the skewed distribution. The statistical significance of differences in mean values of continuous variables and frequencies of categorical variables was examined using the Student t test and χ^2 test as appropriate. The incidences were calculated by the person-year method. Differences in incidences between MetS status were tested by the Cox proportional hazards regression analysis after adjustment for age. The age- or multivariate-adjusted hazard ratios (HRs) and 95% CIs were also estimated with the use of the Cox proportional hazards model. $P<0.05$ was considered statistically significant in all analyses.

Results

The overall prevalence of MetS at baseline was 25.9%. The baseline characteristics on the basis of sex and MetS are shown in Table 1. Men with MetS had significantly higher mean values of blood pressures, waist circumference, body mass index, fasting blood glucose, and serum triglycerides and lower mean values of serum high-density lipoprotein cholesterol compared with those without MetS. Moreover, the frequencies of antihypertensive medication, hypertension, proteinuria, diabetes, and alcohol intake were higher in men with MetS than in those without MetS. A similar distribution was observed in women with MetS in terms of the previously mentioned variables except for alcohol intake. In addition, women with Mets were significantly older and had higher serum total cholesterol compared with those without MetS.

During the 14-year follow up, 307 first-ever CVD events (158 men and 149 women) occurred. Of these, there were 125 CHD (78 men and 47 women) and 209 stroke events (94 men and 115 women). The age-adjusted incidences of CVD were significantly higher in subjects with MetS compared with those without MetS for both sexes (men: 21.8 versus 11.6 per 1000 person-years, $P<0.01$; women: 12.9 versus 6.5, $P<0.01$) (Table 2). The same was true for CHD incidence in both sexes (men: 9.2 versus 5.7, $P<0.01$; women: 5.1 versus 1.5, $P<0.01$) and for stroke in men (14.1 versus 6.4, $P<0.01$). When we divided strokes into ischemic and hemorrhagic type, the age-adjusted incidences of ischemic stroke were

TABLE 1. Clinical Characteristics of Study Population in 1988

Variables	Men		Women	
	MetS (−) (N=834)	MetS (+) (N=216)	MetS (−) (N=983)	MetS (+) (N=419)
Age, years	58±11	58±11	57±11	62±10†
Systolic blood pressure, mm Hg	132±20	145±18†	126±19	145±19†
Diastolic blood pressure, mm Hg	79±11	87±10†	74±10	81±11†
Antihypertensive medication, %	11.8	21.3†	9.0	29.6†
Hypertension, %	37.4	70.4†	24.0	67.5†
Proteinuria, %	7.0	11.6*	3.0	6.9†
Electrocardiogram abnormalities, %	18.7	19.7	12.5	14.6
Waist circumference, cm	80.3±7.6	88.7±6.9†	78.1±9.2	88.2±8.4*
Body mass index, kg/m^2	22.3±2.8	25.0±2.6†	22.1±2.9	24.9±3.1†
Fasting blood glucose, mmol/L	5.7±1.1	6.7±1.7†	5.5±1.0	6.3±1.7†
Diabetes, %	6.7	29.2†	3.0	17.7†
Serum total cholesterol, mmol/L	5.09±1.06	5.19±1.14	5.47±1.05	5.78±1.09†
Serum triglycerides, mmol/L	1.13 (0.43–2.95)	2.46 (0.83–7.32)†	0.90 (0.44–1.86)	1.58 (0.61–4.10)†
Serum high-density lipoprotein cholesterol, mmol/L	1.31±0.29	1.06±0.27†	1.42±0.28	1.14±0.22†
Smoking habits, %	51.6	45.8	6.2	7.9
Alcohol intake, %	59.5	69.4*	8.6	9.8
Regular exercise, %	12.2	8.8	9.2	9.3

Values are mean±SD or percentage.
Electrocardiogram abnormalities are defined as left ventricular hypertrophy (Minnesota code, 3–1) and/or ST depression (Minnesota code, 4–1, 2, 3).
Geometric mean values and 95% CIs of serum triglycerides are shown attributable to the skewed distribution.
*P<0.05, †P<0.01 vs MetS (−).
HDL indicates high-density lipoprotein.

significantly higher in subjects with MetS than in those without MetS for both sexes (men: 9.0 versus 4.8, P=0.03; women: 6.2 versus 3.4, P=0.01). The similar tendency was observed for hemorrhagic stroke only in men (5.1 versus 1.6, P=0.01).

Age- and multivariate-adjusted hazard ratios of MetS for the development of CVD were estimated for both sexes (Table 3). The age-adjusted analysis showed that MetS was a significant risk factor for CVD in men and women. These

TABLE 2. Age-Adjusted Incidence Rates of CVD, CHD, and Stroke According to MetS Status in 2452 Subjects During a 14-Year Follow Up by Sex

	Men				Women			
	Person-Years at Risk	No. of Events	Age-Adjusted Incidence Rate	P Value	Person-Years at Risk	No. of Events	Age-Adjusted Incidence Rate	P Value
Cardiovascular disease								
MetS (−)	9958	108	11.6		12 759	78	6.5	
MetS (+)	2416	50	21.8	<0.01	5078	71	12.9	<0.01
Coronary heart disease								
MetS (−)	10 213	53	5.7		13 010	17	1.5	
MetS (+)	2533	25	9.2	<0.01	5279	30	5.1	<0.01
Stroke								
MetS (−)	10 099	63	6.4		12 817	65	5.3	
MetS (+)	2477	31	14.1	<0.01	5122	50	8.8	0.06
Ischemic stroke								
MetS (−)	10 099	46	4.8		12 817	40	3.4	
MetS (+)	2477	20	9.0	0.03	5122	39	6.2	0.01
Hemorrhagic stroke								
MetS (−)	10 099	17	1.6		12 817	25	2.0	
MetS (+)	2477	11	5.1	0.01	5122	11	2.6	0.72

TABLE 3. Age- or Multivariate-Adjusted HRs for Development of CVD, CHD, or Stroke According to MetS Status in 2452 Subjects During a 14-Year Follow Up by Sex

	Men						Women					
	Age-Adjusted			Multivariate-Adjusted*			Age-Adjusted			Multivariate-Adjusted*		
	HR	(95% CI)	*P* Value	HR	(95% CI)	*P* Value	HR	(95% CI)	*P* Value	HR	(95% CI)	*P* Value
Cardiovascular disease												
MetS (−)	1.00	(reference)		1.00	(reference)		1.00	(reference)		1.00	(reference)	
MetS (+)	1.93	(1.38–2.70)	<0.01	1.86	(1.32–2.62)	<0.01	1.68	(1.22–2.33)	<0.01	1.70	(1.22–2.36)	<0.01
Coronary heart disease												
MetS (−)	1.00	(reference)		1.00	(reference)		1.00	(reference)		1.00	(reference)	
MetS (+)	1.95	(1.21–3.13)	<0.01	1.94	(1.19–3.17)	<0.01	3.11	(1.71–5.65)	<0.01	2.86	(1.56–5.24)	<0.01
Stroke												
MetS (−)	1.00	(reference)		1.00	(reference)		1.00	(reference)		1.00	(reference)	
MetS (+)	2.04	(1.33–3.14)	<0.01	1.92	(1.23–2.98)	<0.01	1.43	(0.99–2.08)	0.06	1.50	(1.03–2.19)	0.03
Ischemic stroke												
MetS (−)	1.00	(reference)		1.00	(reference)		1.00	(reference)		1.00	(reference)	
MetS (+)	1.80	(1.07–3.05)	0.03	1.68	(0.98–2.89)	0.06	1.77	(1.14–2.76)	0.01	1.78	(1.13–2.79)	0.01
Hemorrhagic stroke												
MetS (−)	1.00	(reference)		1.00	(reference)		1.00	(reference)		1.00	(reference)	
MetS (+)	2.67	(1.25–5.69)	0.01	2.54	(1.18–5.49)	0.02	0.88	(0.43–1.80)	0.72	0.99	(0.48–2.05)	0.91

*Adjusted for age, proteinuria, electrocardiogram abnormalities, serum total cholesterol, smoking habits, alcohol intake, and regular exercise.

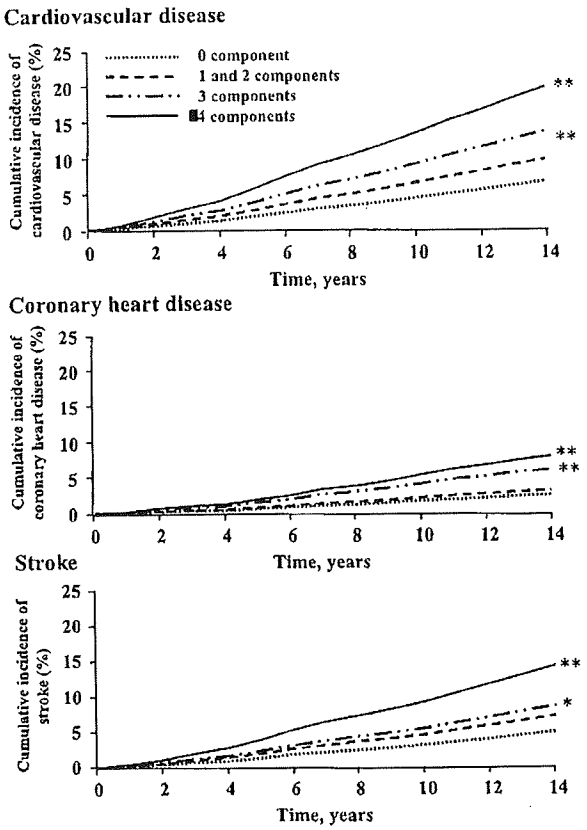
relationships remained substantially unchanged even after adjustment for the following confounding factors: age, proteinuria, electrocardiographic abnormalities, serum total cholesterol, smoking habits, alcohol intake, and regular exercise. Furthermore, MetS was found to be an independent risk factor for the development of CHD and stroke after adjustment for the confounding factors in men and women. When strokes were divided into ischemic and hemorrhagic type, multivariate-adjusted HR of MetS for ischemic stroke was marginally higher in men and significantly higher in women, whereas MetS is an independent risk factor for hemorrhagic stroke only in men.

The age- and sex-adjusted cumulative incidences of CVD, CHD, and stroke according to the number of MetS components are shown in the Figure. Because the cumulative incidence curves for one and 2 components overlapped, we combined these components. The incidences of CVD, CHD, and stroke were significantly higher among the subjects with 3 or more MetS components compared with those without any MetS component. A significant graded relationship between the number of components of MetS and the HR for developing CVD was identified from 3 MetS components and onward (Table 4). Compared with individuals with no MetS component, individuals with one, 2, 3, and 4 or more components had gradually increased HRs, respectively, for developing CVD after adjusting the confounding factors. A similar relationship was found when CVD was divided into CHD and stroke.

Because hypertension and diabetes are strong risk factors for CVD, we examined the combined as well as separate effects of MetS and hypertension or diabetes on the development of CVD. As shown in Table 5, the age- and sex-adjusted HR of CVD was significantly higher in normotensive subjects with MetS, hypertensive subjects without MetS, and hypertensive subjects with MetS compared with those without hypertension and MetS. Furthermore, there was a significant excess risk of CVD in hypertensives with MetS than in those without MetS. Similarly, the age- and sex-adjusted HR of CVD was significantly higher in nondiabetic subjects with MetS and diabetic subjects with MetS compared with those without diabetes and MetS. However, no significant difference was found in the risk of CVD in diabetic subjects without MetS. Among diabetic subjects, the risk of CVD was significantly higher in subjects with MetS than in those without MetS. These relationships remained substantially unchanged even after adjusting for the confounding factors. Furthermore, we examined the association of MetS with CVD by the multivariate analysis using hypertension and diabetes in addition to the previously mentioned risk factors as confounding factors. As a result, MetS remained a significantly independent risk factor for the development of CVD (HR, 1.38; 95% CI, 1.07 to 1.78, $P=0.01$). The risks of other risk factors were as follows: age (HR, 2.00 [per increment of 10 years]; 95% CI, 1.79 to 2.26, $P<0.01$), male sex (1.45; 1.07 to 1.97, $P=0.02$), hypertension (1.64; 1.26 to 2.12, $P<0.01$), diabetes (1.55; 1.14 to 2.13, $P<0.01$), smoking habits (1.69; 1.28 to 2.23, $P<0.01$), regular exercise (0.58; 0.39 to 0.87, $P<0.01$), proteinuria (1.64; 1.13 to 2.38, $P<0.01$), electrocardiographic abnormalities (1.29; 0.98 to 1.69, $P=0.07$), serum total cholesterol (0.99 [per increment of 1 mmol/L]; 0.89 to 1.11, $P=0.92$), and alcohol intake (0.97; 0.73 to 1.30, $P=0.84$).

Discussion

To our knowledge, our study is the first prospective cohort study of a general Japanese population with a long duration of follow up reporting the association of MetS with incident CVD using the modified NCEP definition. The sole study from Japan, which examined a similar association, was based on a diabetic population.[27] We found a clearly increased incidence of CVD during 14 years of follow up in both men



Age- and sex-adjusted cumulative incidences of CVD, CHD, and stroke according to the number of the metabolic syndrome components in 2452 subjects during a 14-year follow up. *$P<0.05$, **$P<0.01$ versus 0 component.

and women with MetS compared with those without MetS. Besides, the risk of MetS for the development of CVD remained significant even after adjustment for hypertension, diabetes, and other potentially confounding factors.

In our study, subjects with MetS had little over 70% increases in CVD risk compared with those without MetS. Similar or higher HRs (1.4- to 5.0-fold) of MetS for CVD/CHD were reported from different European and American studies.[7,13–25] Differences in the study populations, prevalence of individual components of MetS, follow-up length, and MetS definition used seem to be the main causes behind the variation in the HRs. In our study, CHD risk related to MetS is higher in women than in men, which is consistent with the studies from the Western world.[17]

Our study showed that the risk of incident combined CVD, and CHD and stroke separately, was found to increase with the number of components of MetS and increased by 3-fold or more in those with 4 or more MetS components compared with those without any component. It also revealed that the risk of CVD increased in incremental fashion with the number of components of MetS and became predictive of CVD (also CHD and stroke separately) when the number of components reached 3. This phenomenon gives credence to the requirement of ≥3 components in the NCEP definition for establishing the diagnosis of MetS. Thereby, it can be assumed that the modified NCEP definition of MetS is well predictive for CVD in the general Japanese population.

One prospective study based on a Japanese diabetic population mentioned that MetS based on the NCEP definition was predictive for CVD in men and was not in women.[27] The same authors again reported that the new International Diabetes Federation definition[34] was not predictive for CVD in either male or female patients with diabetes.[35] On the other hand, in our study, MetS based on the NCEP definition was consistently predictive of CVD not only in both men and women, but also in subjects with diabetes. We speculate that this discrepancy resulted from the difference in the cutoff point of the waist circumference between the 2 studies. The former used the waist circumference definition for abdominal obesity proposed by the Japan Society for the Study of Obesity (85 cm for men and 90 cm for women),[36] whereas in our study, we used the waist circumference definition for Asian populations (90 cm for men and 80 cm for women), which was recommend by the International Diabetes Federation to use for the Japanese population.[37] Further research is needed to refine the MetS definition, which would be applicable to various populations, including Japanese.

There was a possibility that the increased risk of MetS for CVD resulted from the influences of hypertension or diabetes, which are components of MetS and major risk factors for developing CVD. However, our stratified analysis showed that the MetS was a significant risk factor for CVD in normotensive subjects as well as in nondiabetic individuals and has a similar risk for CVD as hypertension; the risk is even higher than that of diabetes. Moreover, in the multivariate analysis, MetS was found to be a significant risk factor for CVD independent of hypertension, diabetes, and other confounding risk factors. These results imply the significant roles of MetS in the development of CVD and the need for prevention and early management of the MetS components. In addition, diabetes is not predictive of CVD in subjects without MetS in our study. This finding might suggest that good diabetic control is useful. However, because the number of our diabetic subjects without MetS is small, further studies are necessary to elucidate this issue in detail.

The strengths of our study include its longitudinal population-based study design, long duration of follow up, sufficient number of CVD events and almost perfect follow up of subjects, examining the data in men and women separately, and exclusion of patients with CVD at baseline. Moreover, it is the first study to examine prospectively CVD in relation to MetS based on a general Japanese population. One limitation of our study is that the diagnosis of MetS was based on a single measurement of its components at baseline as was the case in other epidemiological studies.[13–25,27–29] During the follow up, risk factor levels could be changed attributable to modification of lifestyle or medication, and misclassification of the MetS is possible. Thus, it would weaken the association found in this study, biasing the results toward the null hypothesis. Therefore, the true association may be stronger than that shown in our findings.

In conclusion, we have shown that the prevalence of MetS is sizeable in Japanese middle-aged men and women and it is predictive of future CVD in both sexes based on a prospective study with 14 years of follow up. Our findings suggest that early identification of MetS and appropriate behavioral and therapeutic intervention may reduce the burden of CVD in the long run.