mean values were used for analysis. LQTS-related cardiac events were defined as syncope, aborted cardiac arrest, or unexpected sudden death.

Statistical Analyses

All data are expressed as the mean value±SD. The Student's t-test was used to compare continuous data between mutations located in the pore region and those in the non-pore region. Differences in frequencies were analyzed by the chi-square test. Time to the first cardiac event (syncope, cardiac arrest, or sudden cardiac death) before initiation of β-blocker therapy and before age 50 years was determined by Kaplan-Meier cumulative estimates. Two-sided probability values <0.05 were considered statistically significant. Statistical calculations were performed with SPSS software (version 11.01J, Chicago, IL, USA).

Results

Genetic Characteristics

Table 1 lists the *KCNH2* mutations we identified, classified by location, number of patients with these causative mutations, coding effects (missense, insertion, deletion and frameshift) and functional outcomes. We identified 62 different *KCNH2* mutations among the 69 LQTS families: 42 missense, 16 deletion/insertion, 11 frameshift and 4 nonsense mutations. There were 27 (44%) mutations causing amino acid changes in the pore region and 35 (56%) mutations within the non-pore regions (15 in the N-terminus, 8 in the non-pore transmembrane, and 12 in the C-terminus). In the pore mutations there were 25 (93%) missense mutations and the remaining 2 were protein deletions (K638del and F640del).

In contrast, the non-pore mutations included more significantly complex mutations such as deletion, insertion, frameshift or nonsense mutations that resulted in truncation of channel proteins (15/35, 43%). Thirty-five mutations (56%, 11 in the pore region and 24 in the non-pore regions) were novel and indicated by asterisk in Table 1. Functional effects by cellular electrophysiologic tests have been reported in only 12 of the 62 mutations (19%);[22–29] however, all those previous reports indicated that the *KCNH2* mutations had loss-of-function effects and made the I_{Kr} current reduce or disappear. Four pore mutations had dominant-negative effects, 4 pore mutations and 2 non-pore mutations had trafficking defects, and 2 non-pore mutations reduced the I_{Kr} current.

Clinical Characteristics

Table 2 is a comparison of the clinical characteristics of the 56 patients with pore mutations and the 62 patients with non-pore mutations. There were no significant differences between the 2 groups regarding gender, the percentage of probands and the age at baseline ECG recording. Diagnostic LQTS scores of Schwartz et al[30] were noticeably greater in the pore group. RR and QT_{peak} intervals were comparable; however, corrected QT_{end} and $T_{peak-end}$ intervals were much longer in the pore than in the non-pore group. Although the incidence of TdP and T-wave alternans did not differ between groups, notched T waves were more frequently seen in the pore group (p=0.007 vs non-pore group). The incidence of cardiac events and the introduction of β-blocker therapy were not statistically different between the 2 groups.

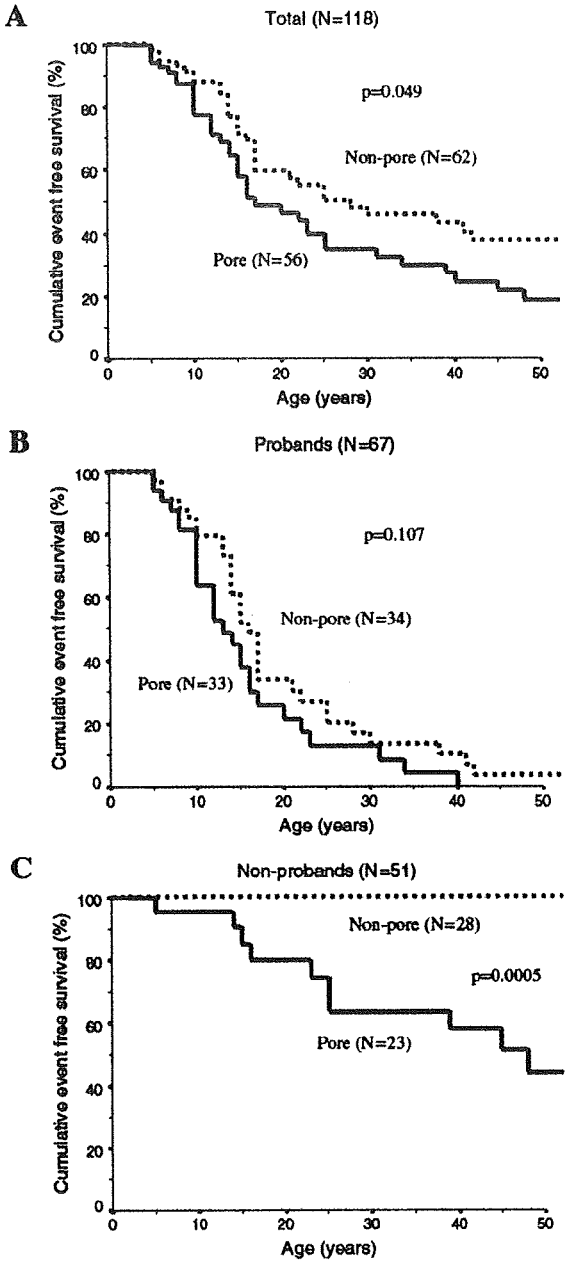

Fig 1. (A) Kaplan-Meier cumulative cardiac event-free survival curves from birth through to age 50 years for the total of 118 patients with *KCNH2* mutations located in the pore (n=56, smooth line) and non-pore (n=62, dotted line) regions. The pore group patients experienced their first cardiac event at a younger age than the non-pore group (log-rank, p=0.049). The difference was caused mainly by the high first-event rate in non-probands. Kaplan-Meier cumulative cardiac event-free survival curves for 67 probands (B) and 51 non-probands (C) with pore mutations (smooth line) and non-pore mutations (dotted line).

Clinical Course by Mutation Location

Fig 1A shows the Kaplan-Meier cumulative cardiac event-free survival curves from birth through to age 50 years for 118 patients (pore group, n=56; non-pore group, n=62). The pore-group patients experienced their first cardiac event at a younger age than the non-pore group (log-rank, p=

Table 3 Clinical Characteristics of Pore and Non-Pore Mutations in Non-Probands

	Pore (n=23)	Non-pore (n=28)	p value
Demographics			
Female gender (%)	*14 (61%)*	*19 (68%)*	*0.769*
Age (years) at baseline ECG (range)	*42±20 (9–74)*	*33±20 (2–71)*	*0.124*
Diagnosis			
Schwartz score	*4.7±1.5*	*3.5±1.7*	*0.008*
Schwartz score ≥4 (%)	*18 (78%)*	*12 (43%)*	*0.021*
ECG measurements			
Heart rate (beats/min)	*65±15*	*70±17*	*0.251*
RR (ms)	*959±179*	*894±179*	*0.201*
QT_{end} (ms)	*480±51*	*441±54*	*0.0011*
QT_{peak} (ms)	*352±47*	*352±53*	*0.974*
$T_{peak-end}$ (ms)	*128±46*	*89±30*	*0.001*
Corrected QT_{end} (ms)	*494±45*	*470±40*	*0.044*
Corrected QT_{peak} (ms)	*364±49*	*374±40*	*0.423*
Corrected $T_{peak-end}$ (ms)	*131±43*	*96±32*	*0.002*
Torsade de pointes (%)	*1 (4%)*	*0*	*0.451*
T-wave alternans (%)	*0*	*0*	–
Notched T wave (%)	*17 (74%)*	*14 (50%)*	*0.095*
Cardiac events			
All cardiac events (%)	*11 (48%)*	*0*	*<0.001*
Syncope (%)	*10 (43%)*	*0*	*<0.001*
Aborted cardiac arrest/SCD (%)	*1 (4%)*	*0*	*0.451*
Therapy			
β-blocker therapy	*6 (26%)*	*0*	*0.006*
Pacemaker (%)	*0*	*0*	–
Sympathectomy (%)	*0*	*0*	–
Defibrillator (%)	*0*	*0*	–

Data are mean value±SD or number (%) of subjects.
Abbreviations see in Table 2.

0.049). We examined the clinical course of the 67 probands and 51 non-probands separately (Figs 1B,C). The clinical courses of the probands were not significantly different according to mutation site (Fig 1B), whereas in the non-pore group 28 non-probands remained asymptomatic and more than half had suffered from cardiac events by the age of 50 (Fig 1C). Therefore, the difference stemmed from markedly distinct prognoses among the non-probands.

Table 3 summarizes the clinical characteristics of the 51 non-probands. The absolute and corrected QT_{end} and $T_{peak-end}$ intervals were all significantly greater in the pore than in the non-pore group. In the non-probands, the incidence of all cardiac events, syncope, and β-blocker therapy were significantly greater in the pore group than in the non-pore group.

Discussion

This study demonstrates that the clinical features of 118 Japanese LQT2 patients who had 62 different *KCNH2* mutations correlated with the mutation sites, but only in non-probands. In probands, there was no significant relationship between mutation site and prognosis. Moss et al[11] reported approximately 179 LQT2 patients based on 44 different *KCNH2* mutations and those patients with pore mutations had significantly ($p<0.0001$) higher frequency of LQTS-related cardiac events and longer QTc intervals than those with non-pore mutations. In contrast to their results, in the present study the mutation-dependent difference in prognosis was relatively small, though significant ($p=0.049$), when analyzed in the total patient cohort (Fig 1A). Indeed, the beneficial outcome of the non-pore patients stemmed from their family members (Fig 1C), and probands showed virtually similar prognosis to that of pore mutation carriers. Although Moss et al did not report separate sub-analysis of probands and family members, the percentage of family members in their non-pore group was significantly larger than that of the pore group (84% vs 57%, $p<0.001$). The very good prognosis of the non-pore mutation group in their study may have reflected that large number of family members.

The character of the mutation per se may be important as another reason for the variance between these 2 studies, as both had a similar number of LQT2 patients. Compared with the study by Moss et al,[11] the type of mutation in the present study was quite different: in our non-pore group, there were significantly more complex mutations, such as nonsense or frameshift, that caused the truncation of channel proteins (15/35, 43%) than in the report of Moss et al (4/30, 13%). For example, nonsense-mediated mRNA decay (NMD) has recently been reported to play an important role in reducing dominant negative suppression effects.[31] Premature termination codon caused by either a deletion or insertion mutation would also cause NMD and thereby attenuate the severity of cardiac phenotypes. This different nature of the mutations may cause the apparently different prognosis of the non-pore mutation groups in each study.

In our pore site mutation group, there were only 2 in-frame deletions, but no frameshift mutations (Table 1). Although it was practically very difficult to conduct every functional assay for each novel *KCNH2* mutation identified here, some cellular electrophysiological effects are available in a small number of *KCNH2* mutations we found (Table 1). Several missense mutations in the pore region (such as A561V and T613M) have been shown to produce dominant negative suppression effects, a greater functional change predisposing to arrhythmic events. In contrast, functional assay of several missense mutations in the non-pore regions has revealed relatively smaller loss-of-function effects (such as with A490T or S818L) Greater functional disruption may also be reflected in the different prognosis

of family members in the pore and non-pore groups (Fig 1).

Previously we reported that LQT1 patients with *KCNQ1* mutations located in the transmembrane regions, including the pore region, are at a higher risk of congenital LQTS-related cardiac events and longer QTc and $T_{peak-end}$ intervals than are patients with C-terminal mutations[13] In LQT2, we have also demonstrated that $T_{peak-end}$, representing transmural dispersion of ventricular repolarization[19] is longer in pore patients than in non-pore patients (Table 2), supporting the finding that family members with pore mutations are more likely to suffer from LQTS-related cardiac events than those with a non-pore mutation.

Study Limitations

Cardiac events are not simply linked to the site of mutation in probands; there are other triggering factors such as modifier genes, including single nucleotide polymorphisms,[32] hypokalemia and bradycardia, which play significant roles in aggravating the symptoms of *KCNH2* mutation carriers. The influence of these factors could be interpreted in the similar occurrence of cardiac events in the probands irrespective of mutation site, because the presence of symptoms usually caused the patient to agree to undergo genetic testing.

Regarding each mutation, the number of study patients was relatively small (at most 5), and the location of the mutations was scattered, even in the same pore region. The coding effect was also so various that we had limited ability to show arrhythmic risk according to a specific mutation site. Our cohort contained 35 novel *KCNH2* mutations, and their functional outcomes were not available. Moreover, our study population included only Japanese, so more subjects per mutation and a greater spectrum of *KCNH2* mutations in a worldwide study are needed to evaluate the arrhythmic risks associated with these mutations.

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Age- and Genotype-Specific Triggers for Life-Threatening Arrhythmia in the Genotyped Long QT Syndrome

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Age and Long QT Syndrome. ***Introduction:*** **Patients with long QT syndrome (LQTS) become symptomatic in adolescence, but some become at age of ≥20 years. Since it remains unknown whether clinical features of symptomatic LQTS patients differ depending on the age of onset, we aimed to examine whether triggers for cardiac events are different depending on the age in genotyped and symptomatic LQTS patients.**

Methods and Results: **We identified 145 symptomatic LQTS patients, divided them into three groups according to the age of first onset of symptoms (young <20, intermediate 20–39, and older ≥40 years), and analyzed triggers of cardiac events (ventricular tachycardia, syncope, or cardiac arrest). The triggers were divided into three categories: (1) adrenergically mediated triggers: exercise, emotional stress, loud noise, and arousal; (2) vagally mediated triggers: rest/sleep; and (3) secondary triggers: drugs, hypokalemia, and atrioventricular (AV) block. In the young group, 78% of the cardiac events were initiated by adrenergically mediated triggers and 22% were vagally mediated, but none by secondary triggers. In contrast, the adrenergically mediated triggers were significantly lower in the intermediate group. The percentage of secondary triggers was significantly larger in the older group than in the other two groups (0% in young vs 23% in intermediate vs 72% in older; $P < 0.0001$). Concerning the subdivision of secondary triggers on the basis of genotype, hypokalemia was only observed in LQT1, drugs mainly in LQT2, and AV block only in LQT2.**

Conclusion: **Arrhythmic triggers in LQTS differ depending on the age of the patients, stressing the importance of age-related therapy for genotyped LQTS patients.** *(J Cardiovasc Electrophysiol, Vol. 19, pp. 794-799, August 2008)*

long QT syndrome, genetic test, age, triggers, drugs, hypokalemia, bradycardia

Introduction

The long QT syndrome (LQTS) is a disease entity characterized by an abnormality in the myocardial repolarization that leads to the prolongation of the QT interval, morphological changes in T waves, and torsade de pointes (TdP) type of ventricular tachycardia on surface electrocardiogram (ECG).[1] The prevalence of LQTS is reported as 1 per 5,000 and it induces syncope and sudden cardiac death usually among young people. Up to date, several different genes have been reported to cause the LQTS.[1-3]

Since the first description on two LQTS-related genes (*KCNH2* and *SCN5A*) in 1995,[1] a number of studies have been performed regarding the relationship between genotype and phenotype. In addition to the genetic background predisposing excessive QT prolongation and TdP, many triggers have been known to modify and aggravate the clinical features of LQTS.[4,5] They are, for example, gender (being female), exercise, emotional stress, loud noise, sudden arousal, drugs, hypokalemia, and bradycardia. Some of them are related to the autonomic nervous tone, and it is well known that LQT1 patients are at a higher risk of TdP during exercise and LQT2 patients, in sudden arousal and auditory stimuli.[5-7]

Although many LQTS patients develop symptoms during adolescence, some of them experience the first cardiac event in their adulthood. In order to study the age-related difference in the LQTS phenotype, we aimed to examine whether the above-mentioned triggers for cardiac events are different depending on the age in genotyped and symptomatic LQTS patients.

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Methods

Study Population

The study population consisted of consecutive 145 symptomatic patients (117 probands and 28 family members) of a known genotype (LQT1, LQT2, and LQT3) from 117 unrelated Japanese families out of 343 genotyped patients (185 probands and 158 family members). They were enrolled from three institutes in Japan – Shiga University of Medical Science, National Cardiovascular Center, and Kyoto University Graduate School of Medicine – between 1996 and 2007. Patients with LQT5, LQT6, LQT7 (Andersen-Tawil syndrome), and compound mutations were excluded from the present study. All of the patients experienced cardiac events, and they were associated with, or triggered by, well-defined conditions. LQTS-related cardiac events were defined as syncope (transient and complete loss of consciousness), documented TdP, aborted cardiac arrest, or unexpected sudden cardiac death without a known cause. We excluded the patients who were genotyped but remained asymptomatic. All subjects or their guardians provided informed consent for the genetic and clinical studies according to each institutional review board's guidelines.

The patients were classified into three groups according to the age of first onset of cardiac events: (1) young group (n = 106): patients who experienced their first cardiac event at age of less than 20 years; (2) intermediate group (n = 20): those who experienced their first cardiac event at age of 20-39 years; and (3) older group (n = 19): those who experienced their first cardiac event after age of 40 years.

Clinical Phenotyping

Routine clinical and electrocardiographic (ECG) parameters were acquired at the time of the first examination for the evaluation of LQTS. Measured parameters on the first recorded ECG included QT and R-R interval in milliseconds, with corrected QT interval (QTc) corrected for heart rate (HR) by Bazett's formula.[8] Measurement for ECG parameters was performed manually on lead V5 (if not available on leads II). A cumulative LQTS diagnostic "Schwartz" score (which is derived in part from the QTc, symptoms, and family history) was assigned.[9] In regard to the family history, we defined positive family history as subjects who have relatives with a Schwartz score of $\geq$4.

Genetic Analysis

Screening for mutations of *KCNQ1*, *KCNH2*, and *SCN5A* was performed using polymerase chain reaction (PCR)/single-strand conformation polymorphism (SSCP) or denatured high-performance liquid chromatography analyses (dHPLC, WAVE system; Transgenomic Inc., Omaha, NE, USA). For aberrant PCR products, DNA sequencing was conducted with a DNA sequencer (ABI 3130 DNA Sequencer; Perkin Elmer, Foster City, CA, USA).

Genetic mutations of amino acid sequence were characterized by a specific location and coding effect (missense, nonsense, splice site, frameshift, insertion, deletion, and intronic variant). The transmembrane regions of *KCNQ1*, *KCNH2*, and *SCN5A* were defined as six membrane segments (S1 to S6, amino acid residues 112 through 354 for *KCNQ1*, 397 through 666 for *KCNH2*, and 127 through 1771 for *SCN5A*, respectively). They, therefore, included cytoplasmic and extracellular linkers, as well as the pore region. As for LQT1 and LQT2, the pore region was defined as the area extending from S5 to the mid portion of S6 involving amino acid residues 262 through 354 for *KCNQ1* and 550 through 650 for *KCNH2*, respectively.[10-14]

Triggering Factors

We divided the triggers into three categories:(1) adrenergically mediated triggers: exercise, emotional stress, loud noise, and arousal; (2) vagally mediated triggers: rest/sleep; and (3) secondary triggers: drugs, hypokalemia, and AV block. There was a small number of undefined conditions associated with cardiac events, and they were classified as other triggers and excluded for analysis in Figures 1–3.

Statistical Analysis

Data are expressed as the mean value ± standard deviation (SD). The clinical characteristics of the study groups were compared with the chi-square test for categorical variables. For continuous variables, we analyzed the normally distributed data with one-way analysis of variance and non-normally distributed data with Kruskal-Wallis tests. For comparisons between two groups, the Student's *t*-test was used for parametric data and the Mann–Whitney's *U*-test for non-parametric data. Differences were accepted as significant for P value of <0.05.

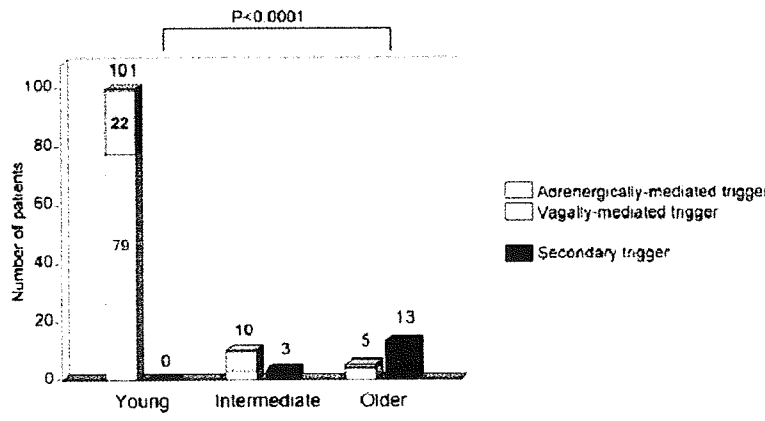

Figure 1. *Triggers for cardiac events in the young, intermediate, and older groups. Incidence of three categorical triggers. Bar graphs show the number of symptomatic patients and their triggers of the first cardiac events: open bars, adrenergically mediated; gray bars, vagally mediated; and black bars, secondary triggers. Other triggers for cardiac events that were undefined were excluded.*

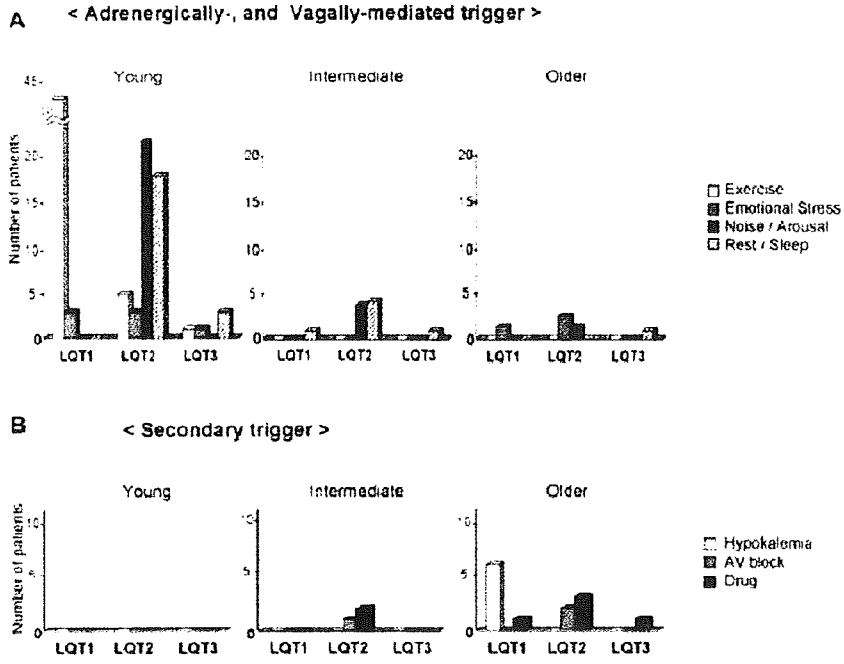

Figure 2. *Genotype-dependent difference of triggers for cardiac events in the young, intermediate, and older groups. A: autonomic triggers. B: secondary triggers: bar graphs indicate the number of patients and their patterns of a specific trigger as summarized in insets. Other triggers for cardiac events that were undefined were excluded.*

Results

Clinical Characteristics

Table 1 summarizes the clinical characteristics of the study subjects. The percentages of females, probands, and patients with positive family history were significantly different among the three groups. In the older group, the percentage of females and probands increased, but that of positive family history decreased. The intermediate group patients showed similar levels of QT prolongation, family history, and Schwartz scores as those of the young group patients. There were no significant differences in basal HR, QTc, and Schwartz scores among the three groups.

Genetic Characteristics

There were 58 LQT1, 75 LQT2, and 12 LQT3 patients (Table 1). In these genotyped patients, we identified 31 *KCNQ1*, 60 *KCNH2*, and 8 *SCN5A* mutations (total 99 different mutations). Among 58 LQT1 patients, most (48/58, 83%) of the first cardiac events occurred at young age. In contrast, first cardiac events occurred less at young age in LQT2 (51/75, 68%) and LQT3 (7/12, 58%) patients compared to the LQT1 patients ($P = 0.019$). The prevalence of transmembrane mutations in LQT1 and LQT3 patients and that of pore site mutations in LQT2 patients was evaluated, but no significant differences were observed among the three groups.

Triggers for Cardiac Events

Figure 1 illustrates the incidence of three categorical triggers in the three age groups. In Figure 1, left-sided bars indicate the number of patients in whom the event was induced by either adrenergically (open bar) or vagally (gray bar) mediated triggers. Right-sided black bars indicate those with secondary triggers. The vertical axis indicates the number of patients. In the young group, all 101 cardiac events were

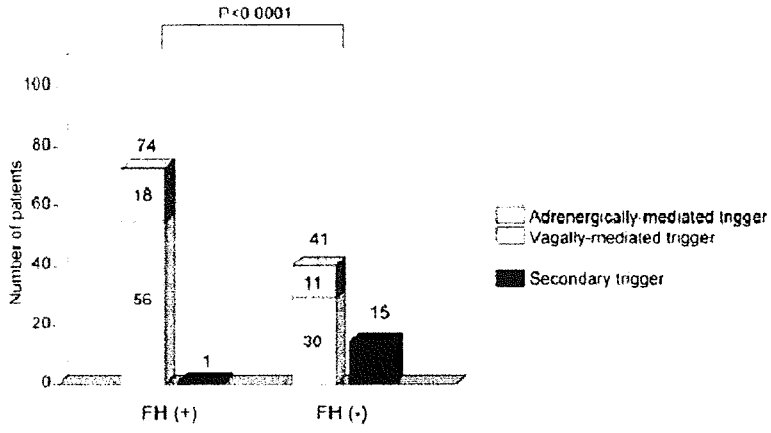

Figure 3. *Triggers for cardiac events in the patients with and without family history. Bar graphs show the number of symptomatic patients and their triggers to induce the cardiac events: open bars, adrenergically mediated; gray bars, vagally mediated; and black bars, secondary triggers. Other triggers for cardiac events that were undefined were excluded.*

TABLE 1

Clinical and Genetic Characteristics in the Three Groups

	Young (n = 106)	Intermediate (n = 20)	Older (n = 19)	P Value
Age at first cardiac event (years)	11.0 ± 0.4	28.0 ± 1.1	59.0 ± 3.5	
Female (%)	63 (59%)	16 (80%)	18 (95%)	0.004
Proband (%)	80 (75%)	18 (90%)	19 (100%)	0.035
Family history	68 (64%)	11 (55%)	3 (16%)	<0.001
HR (bpm)	65 ± 1.2	65 ± 2.8	65 ± 2.8	0.984
QTc (ms)	515 ± 5.7	523 ± 12	485 ± 10	0.084
Schwartz score	6.1 ± 0.2	6.2 ± 0.4	5.3 ± 0.5	0.335
Subtype				
LQT1 (n = 58)	48/58 (83%)	1/58 (2%)	9/58 (15%)	
LQT2 (n = 75)	51/75 (66%)	16/75 (21%)	8/75 (11%)	
LQT3 (n = 12)	7/12 (58%)	3/12 (25%)	2/12 (17%)	
Transmembrane mutation (LQT1)	39/48 (81%)	1/1 (100%)	7/9 (78%)	
Pore site mutation (LQT2)	25/51 (49%)	9/16 (56%)	1/8 (13%)	
Transmembrane mutation (LQT3)	6/7 (86%)	3/3 (100%)	1/2 (50%)	

Data are presented as the mean value ± SD or number (%) of subjects.
HR = heart rate; LQT1 = long QT syndrome caused by the *KCNQ1* potassium channel gene mutations; LQT2 = long QT syndrome caused by the *KCNH2* potassium channel gene mutations; LQT3 = long QT syndrome caused by the *SCN5A* sodium channel gene mutations; QTc = QT interval corrected by Bazett's formula.

associated with autonomic triggers, among which 79 (78%) events were adrenergically mediated and 22 (22%) were vagally mediated. On the other hand, only 5 of 18 cardiac events (28%) were associated with autonomic triggers in the older group, and secondary triggers induced cardiac events in the majority of the older group patients (13/18, 72%). The percentage of secondary triggers was significantly larger in the older group than in the other two groups (0% in young [0/101] vs 23% in intermediate [3/13] vs 72% in older [13/18]; $P < 0.0001$). Among the cardiac events triggered by autonomic factors, the percentage of the adrenergically mediated triggers was significantly lower in the intermediate group patients (79/101 [78%] in young vs 3/10 [30%] in intermediate vs 4/5 [80%] in older group; $P < 0.001$). Thus, triggering factors were significantly different among the three groups.

Figure 2A shows autonomic triggers in the three genotypes in each age group. There were also genotype-dependent differences in triggers for cardiac events in the young group, as previously reported:[5] in young LQT1 patients, 92% (44/48) of the cardiac events occurred during exercise (open bar), but none with noise/arousal (black bar) or rest/sleep (hatched bar). This is in sharp contrast with the pattern in young LQT2 patients: 37% (19/51) of the events occurred during rest/sleep and 43% (22/51) with noise/arousal. Irrespective of onset age, cardiac events triggered by noise/arousal were very specific and observed in only LQT2 patients (35%, 26 of 75 LQT2 patients). In contrast, 44% (3/7) of LQT3 patients experienced cardiac events during rest/sleep. In opposition to the young LQT1 patients, only ~20% of total LQT2 and LQT3 patients experienced cardiac events triggered by exercise or emotional stress.

Figure 2B depicts secondary triggers in the three genotypes in each age group. Hypokalemia (open bar), complete AV block (gray), and drugs (black) were associated with cardiac events in total of 6, 3, and 7 patients, respectively, in the intermediate and older groups. Interestingly, hypokalemia was associated with cardiac episodes in only older LQT1 patients. On the other hand, drugs and AV block triggered cardiac events mainly in LQT2 patients of >20 years. Responsible drugs were amphetamine, aprindine, cisapride (plus pimenol), disopyramide, erythromycin, hydroxyzine, and procainamide.

TABLE 2

Clinical and Genetic Characteristics in Patients With or Without Family History

	FH (+) (n = 82)	FH (−) (n = 63)	P Value
Age at first cardiac event (years)	14.0 ± 1.1	26.0 ± 2.8	<0.001
Female (%)	55 (67%)	42 (67%)	0.960
Proband (%)	54 (66%)	63 (100%)	<0.001
HR (bpm)	65 ± 1.2	64 ± 1.4	0.728
QTc (ms)	512 ± 6.9	513 ± 7.0	0.890
Schwartz score	6.4 ± 0.2	5.6 ± 0.2	0.005
Subtype			
LQT1 (n = 58)	40/58 (69%)	18/58 (31%)	
LQT2 (n = 75)	36/75 (48%)	39/75 (52%)	
LQT3 (n = 12)	6/12 (50%)	6/12 (50%)	
Transmembrane mutation (LQT1)	35/40 (88%)	13/18 (72%)	
Pore site mutation (LQT2)	19/36 (53%)	16/39 (41%)	
Transmembrane mutation (LQT3)	6/6 (100%)	4/6 (67%)	

Data are presented as the mean value ± SD or number (%) of subjects.
FH = family history; HR = heart rate; LQT1 = long QT syndrome caused by the *KCNQ1* potassium channel gene mutations; LQT2 = long QT syndrome caused by the *KCNH2* potassium channel gene mutations; LQT3 = long QT syndrome caused by the *SCN5A* sodium channel gene mutations; QTc = QT interval corrected by Bazett's formula.

Family History

Comparison of clinical and genetic characteristics between patients with and without family history is shown in Table 2. The age at first cardiac event was significantly younger and Schwartz score was significantly higher in the patients with family history than in those without it. LQT1 patients appeared to have more family history compared to those of LQT2 and LQT3 genotypes. Figure 3 illustrates the incidence of three categorical triggers in patients with and without family history. Triggers for cardiac events were also significantly different between the two groups, and secondary trigger was seen in only 1 patient with family history and in 27% (15 of 56) of patients without family history.

Discussion

In the genotyped/symptomatic LQTS patients, the present study demonstrated that factors triggering cardiac events were different depending on the age of their first onset. In general, syncope and sudden death in LQTS are believed to be due to TdP-type of ventricular tachycardia and occur usually in the young.[15,16] However, pathophysiological properties of LQTS-related events were found to be even different among the three groups that were divided by age of less than 20, 20-39, and greater than 40 years. In the young group (<20 years), triggers were closely related to the autonomic nervous tone. In contrast, secondary triggers induced cardiac events in 72% of the older patients (>40 years), suggesting

that "double hit" by secondary trigger(s) appeared to aggravate the clinical phenotype, in addition to genetic variants in ion channel genes, in the older group. The intermediate group patients were at in-between risk in clinical characteristics and the triggers of cardiac events. Interestingly, regarding the triggers of cardiac events, the percentage of the adrenergically mediated trigger was lower in the intermediate group. This may reflect a relatively small number of LQT1 patients in the intermediate group.

Although there was no statistically significant difference in the QTc interval among the three age groups ($P = 0.084$), the older group showed shorter QTc interval compared to that in the other two groups. The QTc in the young group was even shorter than that in the intermediate group. This was probably due to the fact that the QTc in the LQT1 patients was significantly shorter than that in the LQT2 and LQT3 patients (LQT1: 490 ± 6.6, LQT2: 534 ± 8.4, LQT3: 555 ± 26; $P < 0.0001$) and the percentage of LQT1 patients was higher in the young group.

Our results in the young group are consistent with previous reports:[5,7] LQT1 patients experienced the majority of their cardiac events during exercise or emotional stress and only a few occurred during rest/sleep, in opposition to the pattern in LQT2 and LQT3 patients. Cardiac events in LQT2 patients in the young group were mainly associated with noise and sudden arousal and other adrenergic triggers. Cardiac events occurred during rest/sleep in half of the young LQT3 patients.

Among the secondary triggers, hypokalemia was associated with cardiac episodes in only LQT1 patients. Lower extracellular K^+ concentrations are known to reduce outward conductance of both rapid component of delayed rectifier potassium (I_{Kr}) and background inward rectifier potassium (I_{K1}) currents.[17-19] In LQT1, the slow component of delayed rectifier potassium current (I_{Ks}) is impaired, and the function of I_{Kr} and I_{K1} channels remains normal or even upregulated to compensate the total net outward K^+ conductance. Therefore, hypokalemia may unveil the potential repolarization disorder by reducing both "healthy" I_{Kr} and I_{K1}.

On the other hand, AV block and drug intake associated with cardiac events as secondary triggers were seen to be present in most of the intermediate and older LQT2 patients. Tan and colleagues[20] reported that pause-dependence of TdP onset was predominant in LQT2 but absent or rare in LQT1, suggesting that this disparity may reflect different mechanisms. Experimental studies have shown that I_{Ks} blockade (LQT1) causes delayed afterdepolarizations (DAD) but not early afterdepolarizations (EAD);[21] on the contrary, I_{Kr} blockade (LQT2) causes EADs, predominantly at slower HRs.[22] Extreme bradycardia due to AV block may lead to EAD as well as TdP through the postpausal prolongation of action potential plateau. Both a smaller I_{Kr} due to complete deactivation and an enhanced inward Na^+/Ca^{2+} exchanger at low HR may contribute to EAD formation by providing time for recovery and reactivation of L-type Ca^+ channel. In the presence of pathological bradycardia, therefore, I_{Kr} plays a more important role in abbreviating the repolarization and, thereby, keeping the appropriate QT interval because little accumulation of outward I_{Ks} occurs at lower HR.[23]

In this connection, drug-induced TdP has been shown to depend on intervals of preceding pauses.[24] The above-mentioned mechanism on the bradycardia-induced TdP may give an explanation of our result that most of the drug-induced events were observed in LQT2. Because responsible drugs are known to block cardiac I_{Kr} (except hydroxyzine), preexisting repolarization abnormality due to gene mutations may predispose the patients to fatal arrhythmias by further reducing the outward K^+ conductance.[25,26] In preliminary experiments of biophysical assay with heterologous expression systems, we found that these *KCNH2* mutations identified in drug-induced TdP patients produced mild loss-of-function of I_{Kr}.

We evaluated only "already-symptomatic" genotyped patients in this study. The percentage of "still-asymptomatic" patients was 58% of all genotyped patients (198 of 343). The average ages of asymptomatic patients were 19.0 ± 1.9 years (5–67 years) for probands and 34.0 ± 1.8 years (2–68 years) for family members. Asymptomatic probands were still young; therefore, some of them would be symptomatic in the future, being exposed to higher risk of lethal events. The results of our study again emphasize the importance of a careful approach to asymptomatic (preclinical) LQTS patients to decrease their arrhythmic risk, particularly in older patients (≥20 years). Because of lack of apparent phenotypes, most of them were not diagnosed prior to the onset of symptoms. However, one of the most important missions of our genetic testing would be to achieve a preclinical diagnosis of LQTS, particularly in patients with forme-fruste phenotype. Because of low penetrance, inheriting a gene mutation per se does not always mean that the individual mutation carrier will present clinical manifestation,[27] but apparently "healthy" carriers have inherited the risk for developing the clinical phenotype. Once genetic information becomes available, we can introduce the timely beta-blocker therapy and conduct careful follow-up, including ECG recordings, lifestyle modifications (i.e., avoidance of QT-prolonging drugs), avoidance of hyopokalemia, bradycardia, other alarming symptoms, and family education (home automatic electrical defibrillator, etc.).

Study Limitations

Intermediate and older group patients may have a higher possibility to use more drugs. We, therefore, could not exclude such an age-dependent risk accumulation affecting the results of trigger distribution. As for another issue, carriers of milder mutations may induce cardiac events more likely in association with secondary triggers. Our study included only subjects with three major genotypes, although they account for the majority of LQTS patients. Patients with compound mutations of LQT1-3 and 5–7 genotypes were all excluded from analysis. However, we failed to exclude compound mutation carriers with other (LQT4, 8–10) or unknown genotypes, which may result in a minor selection bias.

We evaluated only the Japanese population and there remains a concern about ethnic differences. However, the prevalent mutations found in more than 4 patients were A341V-KCNQ1, A344A/sp-KCNQ1, and A614V-HERG and were all popular in other ethnic cohorts. The genotype-specific triggers were also similar to those observed in previous studies from other countries.

Although syncope may result from diseases other than LQTS-related ventricular arrhythmia, we considered sudden onset/offset nature of loss of consciousness in a genotyped LQTS patient as syncope due to ventricular arrhythmias, if there was no evidence of another explanation, and included as a study subject. In this connection, very short duration of

TdP that did not cause syncope was underestimated if Holter ECG failed to detect it.

Conclusion

Triggers of cardiac events were closely related to the autonomic nervous tone with a higher incidence of family history in younger patients. In contrast, arrhythmic events in older patients were associated with secondary triggers, such as drugs, hypokalemia, and AV block, with genotype specificity. Thus, arrhythmic triggers in LQTS differ depending on the age of the patients, stressing the importance of age- and genotype-related therapy for genotyped LQTS.

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Original Article

Impact of Metabolic Syndrome Components on the Incidence of Cardiovascular Disease in a General Urban Japanese Population: The Suita Study

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Abdominal obesity is a prerequisite for some definitions of metabolic syndrome (MetS). We investigated the impact of MetS defined by two different criteria, which either did or did not require abdominal obesity as a prerequisite, on cardiovascular disease (CVD) incidence in an urban Japanese cohort study. We studied 5,332 Japanese (aged 30–79 years, without CVD at baseline), who completed a baseline survey (September 1989 to March 1994) and were followed up through December 2005. MetS was defined by the NCEP-ATPIII (modified by Asian obesity criteria) and the Japanese criteria. After 61,846 person-years of follow-up, we documented 317 CVD incidences. The MetS frequencies of the Japanese and of the modified NCEP-ATPIII criteria were 17.7% and 25.1% for men and 5.0% and 14.3% for women, respectively. The multivariate hazard ratios (HRs; 95% confidence intervals [CI]) of CVD incidence for MetS by the modified NCEP-ATPIII criteria were 1.75 (1.27–2.41) in men and 1.90 (1.31–2.77) in women, and those for MetS by the Japanese criteria were 1.34 (0.96–1.87) in men and 2.20 (1.31–3.68) in women. The multivariate HRs of CVD incidence for MetS for the Japanese and for the modified NCEP-ATPIII criteria were 2.92 (1.54–5.55) and 1.94 (0.98–3.82) in men under 60 years old, respectively. The CVD incidence risks increased according to the number of MetS components. The risks were similar among participants with the same number of MetS components, regardless of abdominal obesity. In conclusion, the number of MetS components (modified NCEP-ATPIII criteria) may be more strongly associated with CVD incidence than the abdominal obesity essential criteria (the Japanese criteria) in a general urban Japanese population. (*Hypertens Res* 2008; 31: 2027–2035)

Key Words: **metabolic syndrome, cardiovascular risk factor, cohort study, general population**

Introduction

Metabolic syndrome (MetS) is a clustering of impaired glucose metabolism, abdominal fat accumulation, dyslipidemia, and elevated blood pressure (*1*). Previous papers have shown an association between MetS and cardiovascular disease (CVD) (*2*), but most studies conducted thus far have been based on Western populations. There have been several well-designed prospective studies of Asian populations, and those

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studies had various limitations, including the use of body mass index (BMI) (*3*, *4*), non-fasting triglyceride and glucose levels (*3*, *4*), mortality (*4*, *5*), or small sample size (*4–7*). In order to properly define MetS, it is essential to use data on waist circumference and on the levels of both fasting glucose and fasting triglycerides.

MetS has been defined in several ways by several groups, including the World Health Organization (*8*), the European Group for the Study of Insulin Resistance (*9*), the American Association of Clinical Endocrinologists, and the National Cholesterol Education Program Adult Treatment Panel III (NCEP-ATPIII) (*10*). However, these definitions are aimed mainly at Western countries. The International Diabetes Foundation (IDF) (*11*) and the American Heart Association (*12*) have recently introduced alternative definitions that can be applied worldwide (*10*). Stroke incidence is relatively higher in Japan than in Western countries (*13*). It is uncertain whether these criteria can be applied well to Japanese populations. A MetS definition needs to be tailored to the epidemiological background of the area in question.

The Japanese Committee on the Criteria for MetS has recently proposed a definition of Japanese MetS (*14*, *15*). Under both the IDF and the Japanese definitions, the presence of abdominal obesity is necessary for a diagnosis of MetS. However, no prospective study has examined the association between MetS based on the Japanese criteria and CVD, particularly in urban areas, where most Japanese live. Therefore, we undertook this study to examine the impact of MetS under the Japanese and modified NCEP-ATPIII criteria on CVD incidence in a general urban Japanese population.

Methods

Study Population

The Suita study (*16*, *17*), an epidemiological survey of cerebrovascular disease and CVD, was based on a random sampling of 12,200 residents of Suita, a city of approximately 350,000 people in northern Osaka, Japan. As a baseline, in 1989, participants between the ages of 30 and 79 were arbitrarily selected from the municipality population registry and stratified into groups by sex and age in 10-year increments. Of these, 6,406 men and women participated in regular health checkups between September 1989 and March 1994. Since then, these participants have participated in regular health checkups at the National Cardiovascular Center every 2 years and answered health questionnaires every year.

Some cohort members in the study population were excluded from these analyses because they met one or more of the following criteria: past or present CVD illness at baseline (n=208), failure to fast for at least 10 h before venipuncture or missing data (n=170), or failure to follow up after their baseline examination (n=696). After these exclusions, 5,332 individuals remained for analysis.

Baseline Survey

We performed routine blood tests that measured fasting serum total cholesterol, high-density lipoprotein (HDL) cholesterol, triglycerides, and glucose levels. Physicians or nurses administered questionnaires covering the subjects' personal habits and any present illnesses. The subjects were classified as current smokers if they smoked at least one cigarette per day, as non-smokers if they had never smoked, and as past smokers if they had stopped smoking. Blood pressure was measured three times in a sitting position after at least 5 min of rest. Systolic and diastolic blood pressures (SBP and DBP) were taken to be the average of the second and third measurements that were recorded at least 1 min apart by well-trained doctors. Waist circumference was measured in a standing position at the umbilical level to the nearest 1 cm by well-trained technicians. Informed consent was obtained from all participants. This study was approved by the Institutional Review Board of the National Cardiovascular Center.

Definitions of Metabolic Syndrome

MetS was defined using two criteria. First, in accordance with NCEP-ATPIII (*18*) criteria, it was defined as the presence of three or more of the following five components: 1) abdominal obesity modified by the International Obesity Task Force central obesity criteria for Asia (waist circumference ≥90 cm in men and ≥80 cm in women) (*19*), 2) elevated blood pressure (SBP/DBP ≥130/85 mmHg and/or current use of antihypertensive medication), 3) hypertriglyceridemia (serum triglyceride levels ≥1.7 mmol/L [150 mg/dL] and/or current use of cholesterol-lowering medication), 4) low HDL cholesterol (serum HDL levels of ≤1.0 mmol/L [40 mg/dL] in men and of ≤1.3 mmol/L [50 mg/dL] in women), and 5) elevated blood glucose levels (fasting blood glucose ≥6.1 mmol/L [110 mg/dL] and/or current use of insulin or oral medication for diabetes).

Second, we used the definition of MetS recommended by the Japanese Committee on the Criteria for MetS (*14*, *15*). MetS was defined by abdominal obesity (waist circumference ≥85 cm in men and ≥90 cm in women) (*20*) and least two of the following three components: 1) elevated blood pressure (SBP/DBP ≥130/85 mmHg), 2) hyperlipidemia (serum triglyceride levels ≥1.7 mmol/L [150 mg/dL] and/or HDL levels <1.0 mmol/L [40 mg/dL]), and 3) elevated blood glucose levels ≥6.1 mmol/L (110 mg/dL). Subjects taking medication for hypertension, hyperlipidemia, or diabetes were included as having that component.

Endpoint Determination

The endpoint of the follow-up period for each participant was whichever one of the following occurred first: 1) the date of the first myocardial infarction (MI) or stroke event, 2) the date of death, 3) the date the participant moved out of Suita,

Table 1. Baseline Distributions of Cardiovascular Disease Risk Factors According to Metabolic Syndrome under the NCEP-ATPIII Modified by Asian Obesity Definitions

	Men (n=2,492)			Women (n=2,840)		
	MetS(−) (n=2,043)	MetS(+) (n=449)	p*	MetS(−) (n=2,253)	MetS(+) (n=587)	p*
Age at baseline, years	55.4±13.3	58.1±11.5	<0.001	52.2±12.6	61.3±9.8	<0.001
Systolic blood pressure, mmHg	126±20	140±19	<0.001	120±20	141±20	<0.001
Diastolic blood pressure, mmHg	78±12	85±11	<0.001	73±11	83±12	<0.001
Total cholesterol, mg/dL	200±34	210±35	<0.001	210±38	227±38	<0.001
HDL cholesterol, mg/dL	51±13	40±10	<0.001	60±12	45±10	<0.001
Triglyceride, mg/dL#	121±73	241±156	<0.001	90±44	178±113	<0.001
Waist circumference, cm	81.0±7.3	89.7±7.0	<0.001	74.7±8.9	87.4±8.5	<0.001
Elevated blood pressure, %	41.8	85.8	<0.001	30.4	82.1	<0.001
Hypertriglyceridemia, %	21.6	82.9	<0.001	7.2	63.7	<0.001
Lower-HDL cholesterol, %	15.5	64.8	<0.001	18.7	80.1	<0.001
Hyperglycemia, %	8.9	43.9	<0.001	3.6	29.6	<0.001
Current smoker, %	50.5	47.6	0.278	11.9	11.8	0.958
Current drinker, %	75.5	72.6	0.207	34.6	25.4	<0.001

Elevated blood pressure: antihypertensive drug use or >130/85 mmHg; hypertriglyceridemia: antilipidemic drug use or triglyceride >150 mg/dL; lower-HDL cholesterol: HDL cholesterol <40 mg/dL. MetS, metabolic syndrome; HDL, high-density lipoprotein. *ANOVA or χ^2 tests were performed. #Log-transformed triglyceride was performed to statistical analysis.

Table 2. Age-Adjusted Hazard Ratios (Confidence Intervals) for Incidence of Cardiovascular Disease According to Abdominal Obesity at Baseline Examination

	Men				Women			
	Case, n	Person-year	HR (95% CI)	p	Case, n	Person-year	HR (95% CI)	p
Japanese criteria								
<85 cm (men)/<90 cm (women)	111	17,112	1		96	29,960	1	
≥85 cm (men)/≥90 cm (women)	77	11,247	0.97 (0.72–1.30)	0.844	33	3,890	1.64 (1.09–2.46)	0.019
Asian criteria								
<90 cm (men)/<80 cm (women)	145	23,136	1		53	21,139	1	
≥90 cm (men)/≥80 cm (women)	43	5,223	1.18 (0.84–1.67)	0.327	76	12,711	1.44 (1.00–2.07)	0.048
NCEP-ATPIII criteria								
<102 cm (men)/<88 cm (women)	182	27,976	1		91	28,730	1	
≥102 cm (men)/≥88 cm (women)	6	384	2.00 (0.88–4.54)	0.095	38	5,121	1.47 (1.00–2.17)	0.048

HR, hazard ratio; CI, confidence interval.

or 4) December 31, 2005 (censored). As a first-step survey to detect MI and stroke incidence, each participant's health status was checked during a clinical visit at the National Cardiovascular Center every 2 years. Furthermore, every year a health questionnaire was given to each participant *via* mail or telephone.

Confirmation of Strokes and Myocardial Infarctions

In total, five hospitals in this area were capable of performing computed tomographic scans and/or magnetic resonance imaging, and all were major hospitals that admitted acute stroke and MI patients. Medical records were reviewed by registered hospital physicians or research physicians who were blinded to the baseline information. Strokes and MI events were registered if they occurred after the date on which the baseline health examination was held and before January 1, 2006. Strokes were defined according to the National Survey of Stroke criteria (*21*). These criteria require the rapid onset of a constellation of neurological deficits lasting at least 24 h or until death. For each stroke subtype (cerebral infarction [thrombotic or embolic infarction], intracerebral hemorrhage, and subarachnoid hemorrhage), a definite diagnosis was established based on examination of computed tomographic scans, magnetic resonance images, or autopsy. Defi-

Table 3. Age-Adjusted Hazard Ratios (95% Confidence Intervals) for Incidence of Cardiovascular Disease, Myocardial Infarction, and All Strokes According to Metabolic Syndrome under the Japanese and NCEP-ATPIII Definitions

	Men			Women		
	MetS(−)	MetS(+)	*p* value	MetS(−)	MetS(+)	*p* value
Cardiovascular disease						
MetS Japanese definition						
Cases, *n*	140	48		110	19	
Person-year	23,542	4,817		32,325	1,526	
Age-adjusted	1	1.31 (0.94–1.82)	0.109	1	2.16 (1.31–3.54)	0.002
Multivariate-adjusted	1	1.34 (0.96–1.87)	0.080	1	2.20 (1.31–3.68)	0.003
<60 years old						
Cases, *n*	27	15		25	4	
Person-year	14,752	2,366		22,085	529	
Age-adjusted	1	2.76 (1.46–5.23)	0.002	1	5.39 (1.82–15.98)	0.002
Multivariate-adjusted	1	2.92 (1.54–5.55)	0.001	1	6.25 (2.08–18.79)	0.001
≥60 years old						
Cases, *n*	113	33		85	15	
Person-year	8,790	2,451		10,240	997	
Age-adjusted	1	1.04 (0.70–1.53)	0.841	1	1.83 (1.05–3.18)	0.033
Multivariate-adjustcd	1	1.06 (0.71–1.57)	0.764	1	1.80 (1.01–3.20)	0.046
MetS NCEP-ATPIII (Asian) definition						
Cases, *n*	133	55		73	56	
Person-year	23,373	4,986		27,405	6,446	
Age-adjusted	1	1.70 (1.23–2.34)	0.001	1	1.93 (1.35–2.77)	<0.001
Multivariate-adjusted	1	1.75 (1.27–2.41)	<0.001	1	1.90 (1.31–2.77)	<0.001
<60 years old						
Cases, *n*	30	12		19	10	
Person-ycar	14,509	2,606		19,872	2,742	
Age-adjusted	1	1.79 (0.91–3.52)	0.089	1	2.72 (1.23–5.99)	0.013
Multivariate-adjusted	1	1.94 (0.98–3.82)	0.055	1	2.96 (1.34–6.57)	0.007
≥60 years old						
Cases, *n*	103	43		54	46	
Person-year	8,864	2,381		7,533	3.704	
Age-adjusted	1	1.67 (1.16–2.40)	0.005	1	1.78 (1.19–2.66)	0.005
Multivariate-adjusted	1	1.73 (1.20–2.48)	0.003	1	1.70 (1.12–2.59)	0.012
Myocardial infarction						
MetS Japanese definition						
Cases, *n*	56	22		32	7	
Person-year	22,962	4,663		31,697	1,457	
Age-adjustcd	1	1.48 (0.90–2.44)	0.117	1	2.36 (1.02–5.46)	0.043
Multivariate-adjusted	1	1.51 (0.91–2.48)	0.105	1	2.70 (1.15–6.35)	0.023
MetS NCEP-ATPIII (Asian) definition						
Cases, *n*	52	26		18	21	
Person-ycar	22,833	4,795		26,944	6,211	
Age-adjusted	1	2.09 (1.30–3.37)	0.002	1	2.68 (1.41–5.10)	0.003
Multivariate-adjusted	1	2.12 (1.31–3.43)	0.002	1	2.77 (1.44–5.32)	0.002
All strokes						
MetS Japancsc definition						
Cases, *n*	84	26		78	12	
Person-year	23,177	4,659		32,078	1,487	
Age-adjusted	1	1.21 (0.78–1.89)	0.381	1	2.09 (1.12–3.88)	0.019
Multivariate-adjusted	1	1.27 (0.81–1.97)	0.292	1	2.05 (1.07–3.92)	0.031
MetS NCEP-ATPIII (Asian) definition						
Cases, *n*	81	29		55	35	
Person-year	23,010	4,826		27,266	6,299	
Age-adjustcd	1	1.52 (0.99–2.34)	0.053	1	1.70 (1.09–2.64)	0.018
Multivariate-adjusted	1	1.58 (1.02–2.43)	0.037	1	1.62 (1.02–2.58)	0.041

Multivariate adjusted for age, smoking and drinking status. MetS, metabolic syndrome.

nite and probable MI was defined according to the criteria set out by the MONICA (Monitoring Trends and Determinants of Cardiovascular Disease) project (*22*), which requires evidence from ECGs, cardiac enzymes, and/or autopsy. Sudden deaths of unknown origin were deaths that occurred within 24 h from onset and were included in MI. However, there was little difference in hazard ratios between the groups with and without sudden death from CVD, because sudden death constituted a small sample size ($n=6$).

To complete surveillance for fatal stroke and MI, we also systematically searched for death certificates, the purpose of which were permitted to use by the Ministry of Health, Labour and Welfare. We checked for possible stroke and MI using data from 1) the health examination and questionnaire for the stroke and MI registry, without informed consent for the medical records survey and 2) death certificates without registration of CVD incidence, which were defined as probable stroke or MI. CVD was defined as stroke and MI in this study. Informed consent to review in-hospital medical records was obtained from 86.2% of participants who were suspected of having any signs or information suggesting the incidence of stroke or MI. For 13.8% of subjects from whom informed consent was not obtained, final diagnoses of CVD were confirmed by physicians or epidemiologists who had been involved in the diagnostic process throughout the study, in order to avoid the misclassification of diagnoses.

Statistical Analysis

Analyses of variance and χ^2 tests were used to compare mean values and frequencies by sex, respectively, according to MetS based on the modified NCEP-ATPIII criteria. For each subject, the person-years of follow-up were calculated from September 1, 1989, to whichever came first: the first endpoint, MI or stroke event, death, emigration, or December 31, 2005. A Cox proportional hazards regression model was used to detect associations between abdominal obesity for Japanese (≥85 cm in men or ≥90 cm in women), Asian (≥90 cm in men or ≥80 cm in women), and American criteria (≥102 cm in men or ≥88 cm in women) and CVD during the follow-up period. The Cox proportional hazard regressions were fitted to the grouping (positive or negative MetS) after adjusting for age and the other potential confounding factors: baseline age, smoking status (never, ex-smoker, or current smoker), and drinking status (never, ex-drinker, or current drinker). Trend tests were conducted by assigning the number of MetS components to test the significance of these variables. All statistical analyses were conducted using the SAS statistical package (release version 8.2; SAS Institute Inc., Cary, USA).

Results

During the follow-up period (averaging 12.5 years), 200 strokes were documented (160 definite strokes and 40 probable strokes). These strokes comprised 130 cerebral infarctions, 31 intracerebral hemorrhages, 22 subarachnoid hemorrhages, and 17 unclassified strokes. In addition, 117 MIs were documented (61 definite MIs and 56 probable MIs or sudden cardiac deaths).

Table 1 shows the distribution of CVD risk factors at the baseline according to MetS as defined by the modified NCEP-ATPIII criteria. Compared with the non-MetS groups, men and women with MetS were more likely to be older and to have higher frequencies of each MetS component.

Table 2 presents the age-adjusted HRs (95% confidence intervals [CI]) for the incidence of CVD according to waist circumference by the NCEP-ATPIII, Japanese, and Asian obesity criteria. Regardless of the criteria set, abdominal obesity was associated with CVD only in women.

Table 3 shows the association of MetS by the Japanese and the modified NCEP-ATPIII criteria with CVD incidence according to age category and sex. Using the Japanese criteria, MetS was associated only in women with the incidence of CVD, MI, and all strokes (HR [95% CI]: 2.20 [1.31–3.68], 2.70 [1.15–6.35], and 2.05 [1.07–3.92], respectively), whereas in men overall MetS was not associated with the incidence of CVD or its subtypes. However, among men under 60 years old, MetS based on the Japanese criteria was associated with CVD incidence (HR=2.92, 95% CI: 1.54–5.55). Using the modified NCEP-ATPIII definition, MetS was associated with each CVD subtype in both men and women. Multivariate adjusted HRs of CVD incidence for MetS based on the NCEP-ATPIII criteria were 1.94 (0.98–3.82) and 1.73 (1.20–2.48) in men less than or equal to and over 60 years old, respectively.

Figure 1 shows that the frequency of MetS increased with age for men and women based on the NCEP-ATPIII (A) and Japanese (B) criteria, respectively. The frequency based on the NCEP-ATPIII modified by the Asian obesity criteria (25.1% for men and 14.3% for women) was higher than that based on the Japanese criteria (17.7% for men and 5.0% for women), especially in women.

The risk of CVD incidence increased according to the number of components combined in men and women with and without abdominal obesity (Fig. 2). In addition, compared with the non-abdominal obesity and non-component groups, the risks of CVD incidence were similar among participants who had the same numbers of components, regardless of the presence or absence of abdominal obesity in men and women combined.

Figure 3 shows the multivariate HRs for MetS based on the Japanese and NCEP-ATPIII definitions modified by the obesity criteria for waist circumference. When the Japanese definition was adopted and the risk of MetS was monitored through sequential waist circumference changes, the cut-off points for waist circumference, which conferred a risk of CVD in men and women, were 84 cm and 92 cm, respectively. When the definition of MetS-indicative waist circumference was higher than those values, the risk was not statistically significant. When the NCEP-ATPIII definition

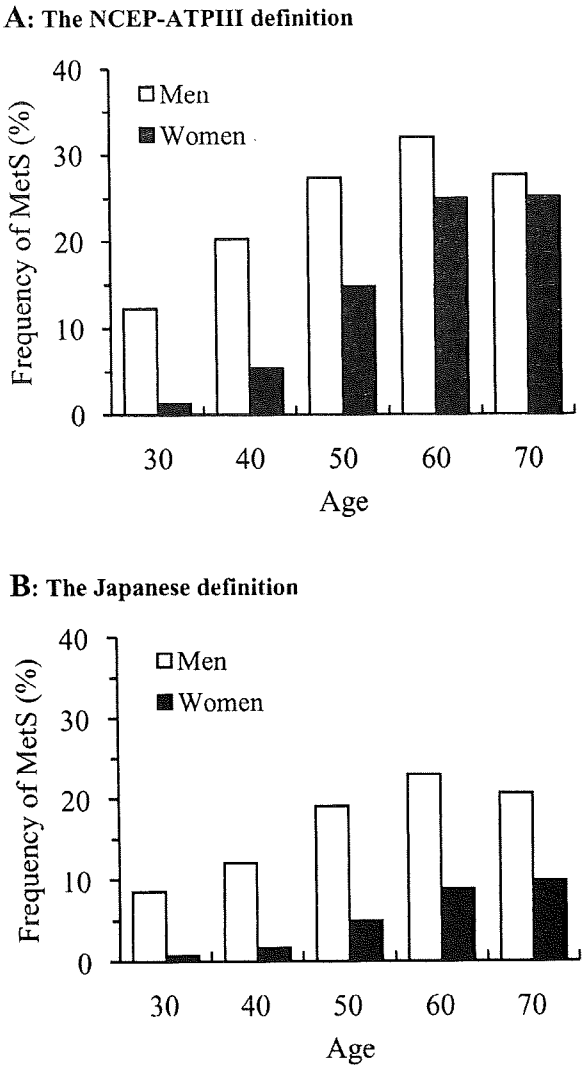

Fig. 1. *Frequencies of MetS components (A: the NCEP-ATPIII definition; and B: the Japanese definition, modified by the Asian waist circumference criteria) by sex. White and solid bars indicate men and women, respectively.*

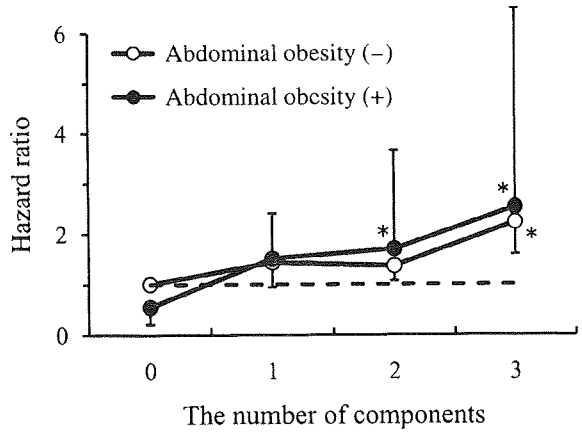

Fig. 2. *Multivariate HRs for the risks of CVD incidence according to the number of components based on the NCEP-ATPIII definition with and without abdominal obesity. White and solid circles indicate non-abdominal and abdominal obesity according to the Asian obesity criteria. *p < 0.05 compared to the reference of non-abdominal obesity and no-components. Bars show 95% CI for the HRs.*

was used, the value of waist circumference did not modify the risk of CVD, implying that the clustering of risk factors may be more important than waist circumference itself for determining CVD risk.

Discussion

In the current cohort study of a general urban Japanese population, the association between MetS and CVD was significant when the NCEP-ATPIII (modified by the Asian criteria) definition was applied. MetS based on the Japanese criteria was associated with CVD incidence in women, whereas in men the association was found only in those under 60 years old. In addition, the risk of CVD incidence was similar among participants who had the same numbers of components regardless of whether they were abdominally obese. To the best of our knowledge, this is the first study of an urban Japanese cohort.

Compared to the previous studies, this study has several methodological strengths. First, previous Japanese cohort studies associating MetS with CVD were based predominantly on BMI (*3*, *4*), non-fasting blood collection (*3*, *4*), and mortality as the endpoint (*4*, *5*). Our baseline subjects were observed in the fasting state, and we used waist circumference and a wide age range. Second, we evaluated a large prospective cohort of people randomly selected from a general Japanese population. A prospective study has little recall bias as well as results from a general population cohort that is more representative than occupational, hospital-based, or volunteer cohorts. Third, our sample size was relatively large for a cohort study and we could therefore perform sub-analysis by age and CVD subtypes. Fourth, our cohort population was selected at random from an urban population, in contrast to most of the other MetS cohort populations, which were selected from rural populations. Our study is the first of its kind in an urban area. Finally, our study examined the risk of CVD incidence, which is a more direct measure of CVD risk than the rate of CVD mortality, because the time to death from CVD is influenced by treatment.

Abdominal obesity induces inflammation in adiposities (*23*), endothelial dysfunction (*24*, *25*), and oxidative stress (*26*), thereby contributing to CVD development (*27*, *28*).

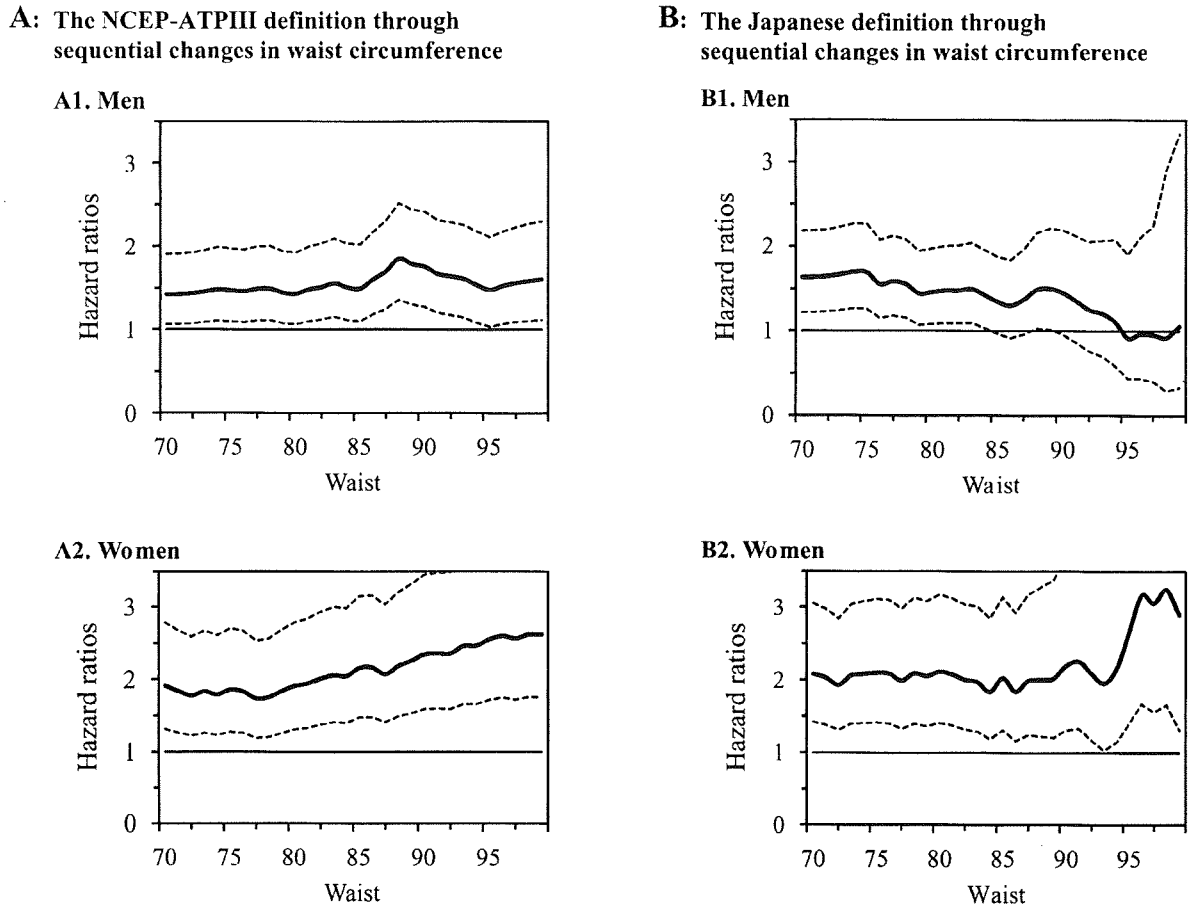

Fig. 3. *Multivariate HRs for MetS based on the NCEP-ATPIII (A) and Japanese (B) definitions through sequential changes in waist circumference by sex. Solid and dotted lines indicate HRs and 95% CI, respectively.*

Accumulating evidence suggests that MetS increases the risk of CVD (*29*). However, there has been a lack of convincing evidence (*29*) that MetS is associated with CVD in Japan. Iso *et al.* reported that MetS was associated with a risk for ischemic CVD in Japan (*3*), although they used BMI as well as non-fasting blood glucose and triglyceride levels to define MetS. Ninomiya *et al.* reported that MetS was a significant risk factor for CVD in a rural Japanese population (*6*). However, that study examined a rural population half the size of that in our study. Takeuchi *et al.* reported that MetS was a risk factor for cardiac disease in a rural cohort (*7*), but their data were based on a small sample that comprised only men. Kadota *et al.* reported that MetS, defined by BMI and non-fasting blood samples, was associated with CVD mortality (*4*).

We have shown that the components of MetS synergistically increase CVD risk. Abdominal obesity did not affect the association between the number of MetS components and the risk of CVD incidence. The risk of CVD was also not related to waist circumference when the NCEP-ATPIII definition was applied (data not shown), suggesting that the combination of risk factors *per se* is more important than abdominal obesity for conferring risk.

The definition of MetS may be reconsidered on the basis of age and sex. According to our results, lifestyle modifications may not be needed for older men who are free of cardiovascular risk factors even if they have abdominal obesity. Therefore, to prevent CVD, it is not adequate for only subjects with MetS to change their lifestyles; subjects with one or two MetS components, even without abdominal obesity, should modify their lifestyles.

When the waist-circumference threshholds were sequentially changed in the Japanese criteria for MetS, our data showed that the clustering of metabolic risk factors was statistically significant for CVD at waist circumferences less than 85 cm for men and 93 cm for women. When the definition of MetS-indicative waist circumferences was higher than those values, the risk clustering was not statistically significant for

CVD in men, and the 95% CI was much wider but still significant in women. Subjects with high risks and non-abdominal obesity with risk clustering aside from abdominal obesity will drop out when the waist-circumference definitions are raised.

Our study has several limitations. First, the annual emigration rate (1.5%) is relatively higher than that in rural areas. Second, about 10% of the subjects who underwent a baseline examination did not respond to our questionnaires afterward. We found no clinical background difference between participants and non-participants, because the main denial reason for participation in this study was not health problems. The frequencies of MetS according to NCEP-ATPIII modified by Asian criteria were 19% and 21% for participants and non-participants, respectively (χ^2 test $p=0.09$). In this study, the main reasons for emigration included job transfer, but not health problems.

In conclusion, the current prospective study for a general urban population showed that MetS, as defined by the Japanese criteria, was associated with CVD in women and middle-aged men; a stronger association was found when the NCEP-ATPIII definition modified by the Asian obesity criteria was applied. The number of MetS components may be more strongly associated with CVD incidence than the essential waist-circumference criteria.

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Advance Publication by J-STAGE

Risk of Smoking and Metabolic Syndrome for Incidence of Cardiovascular Disease

—— Comparison of Relative Contribution in Urban Japanese Population: The Suita Study ——

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Background: Risk factor clustering, the so-called metabolic syndrome (MetS), is an important risk factor for cardiovascular disease (CVD). Smoking is also an important CVD risk factor with still a high prevalence. However, few previous studies have compared the risk for CVD or the population-attributable fraction (PAF) of smoking, MetS, and both.

Methods and Results: The present study was an 11.9-year cohort study of 1,822 men and 2,089 women, aged 40–74 years, selected randomly from an urban general population in Japan. MetS was defined according to the National Cholesterol Education Program on Adult Treatment Panel III (NCEP-ATPIII) guideline modified by the Asian criteria for waist circumference. The prevalence of smoking was 49.5% in men and 11.1% in women, and that of MetS was 19.8% and 23.5%, respectively. In men, the multivariate-adjusted hazard ratio for CVD incidence, compared with non-smoking participants without MetS, was 2.07 (1.26–3.40) in those who smoked, 2.09 (1.08–4.04) in those with MetS, and 3.56 (1.89–6.72) in those with both. In men the PAF for CVD incidence was 21.8% because of smoking, 7.5% because of MetS, and 11.9% because of both.

Conclusions: Although countermeasures for MetS are important, smoking should continue to be considered an important public health problem and antismoking campaigns should be promoted, especially for men, to prevent CVD.

Key Words: Cohort; Hazard ratio; Metabolic syndrome; Smoking

Risk factor clustering, the so-called metabolic syndrome (MetS), is an important risk factor for cardiovascular disease (CVD), and previous studies have shown the risk of MetS for CVD in the Japanese population.[1–4] In addition, health guidance for people aged 40–74 years who fulfill the Japanese MetS criteria[5] began in April 2008 and countermeasures for MetS has become a national project.[6]

Editorial p ????

However, cigarette smoking is a widely accepted risk factor for CVD,[7–9] and the prevalence of smoking is still high in Japan compared with Western developed countries.[10] Accordingly, in Japan, countermeasures for MetS are being applied with a still high prevalence of smoking, which might be different from the situation in Western developed countries with a lower prevalence of smoking.[10] To improve this situation, it is important to examine and show the combined risk of MetS and smoking, and compare the impact of each risk factor and both for CVD from the viewpoint of the impact not only on the individual but also on the population using indicators such as population-attributable fraction (PAF). In addition, such an assessment could be useful for motivating individuals with MetS, smoking, or both because both MetS and smoking are targets of lifestyle modification. However, few studies have compared the risk of smoking, MetS, and both for CVD.

Our a priori hypothesis was that the coexistence of smoking and MetS worsens the CVD risk, and that the PAF of smoking in Japanese men is larger than that of MetS because of their high prevalence of smoking. To examine this hypothesis, we performed a 11.9 year (mean length) cohort study in an urban general Japanese population to compare the effects of smoking, MetS and both on CVD risk.

Methods

Population

The Suita study,[2,11–14] a cohort study of CVD, was established in 1989 in Suita City, Osaka. In that study, 6,485 participants who were randomly selected from the municipal population registry participated in a baseline survey at the National Cardiovascular Center (NCVC) between

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Table 1. Baseline Characteristics of the Participants According to the Combination of Smoking and MetS

	MetS (−)		MetS (+)	
	Non-smoker	Smoker	Non-smoker	Smoker
Men				
n	732	730	189	171
Age (years)	58.9±9.9	56.1±9.4	59.3±8.4	57.4±9.1
Waist (m)	0.82±0.07	0.81±0.07	0.89±0.07	0.89±0.07
BMI (kg/m^2)	22.7±2.7	22.1±2.5	24.9±2.5	24.8±2.5
Total cholesterol (mmol/L)	5.26±0.88	5.08±0.85	5.49±0.88	5.44±0.98
Non-HDL-cholesterol (mmol/L)	3.90±0.88	3.79±0.88	4.41±0.85	4.41±0.98
High blood pressure (%)	48.6	39.3	86.8	84.8
High triglycerides (%)	19.3	22.5	83.1	80.7
Low HDL-cholesterol (%)	13.5	19.6	63.0	69.6
High blood glucose (%)	9.6	10.0	47.1	42.1
Abdominal obesity (%)	13.9	7.5	56.1	59.1
Medication				
For hypertension (%)	32.7	33.7	36.8	39.3
For hypercholesterolemia (%)	1.0	0.5	4.8	4.1
For hypertriglyceridemia (%)	0.5	0.4	2.1	1.2
For diabetes (%)	14.9	12.9	26.9	14.3
Smoking				
Never (%)	37.8	0.0	32.3	0.0
Ex (%)	62.2	0.0	67.7	0.0
Current (%)	0.0	100.0	0.0	100.0
Alcohol drinking				
Never (%)	20.9	19.6	20.6	22.8
Ex (%)	4.2	2.5	5.8	3.5
Current (%)	74.9	77.9	73.5	73.7
Women				
n	1,424	174	433	58
Age (years)	55.3±9.4	52.6±9.1	60.3±8.7	59.3±8.6
Waist (m)	0.77±0.09	0.75±0.09	0.88±0.09	0.87±0.09
BMI (kg/m^2)	21.8±2.8	21.4±3.0	24.8±3.3	24.7±3.2
Total cholesterol (mmol/L)	5.57±0.90	5.39±0.98	5.93±1.00	5.83±0.98
Non-HDL-cholesterol (mmol/L)	4.02±0.90	3.97±1.03	4.75±1.01	4.77±0.95
High blood pressure (%)	35.1	20.1	85.2	70.7
High triglycerides (%)	6.6	6.3	58.0	81.0
Low HDL-cholesterol (%)	18.3	34.5	82.0	87.9
High blood glucose (%)	4.3	1.7	30.5	24.1
Abdominal obesity (%)	30.1	27.6	86.6	79.3
Medication				
For hypertension (%)	33.7	17.4	43.6	44.4
For hypercholesterolemia (%)	1.6	0.0	6.5	3.4
For hypertriglyceridemia (%)	0.1	0.0	1.4	1.7
For diabetes (%)	16.7	0.0	17.5	30.0
Smoking				
Never (%)	97.1	0.0	94.2	0.0
Ex (%)	2.9	0.0	5.8	0.0
Current (%)	0.0	100.0	0.0	100.0
Alcohol drinking				
Never (%)	67.4	50.6	75.5	65.5
Ex (%)	1.0	5.7	1.6	0.0
Current (%)	31.6	43.7	22.9	34.5

Data are value±indicate standard deviation.
MetS=presence of 3 or more of the following: (1) abdominal obesity defined as a waist circumference ≥90 cm in men and ≥80 cm in women; (2) high blood pressure defined as average systolic/diastolic blood pressures of ≥130/85 mmHg and/or current medication for hypertension; (3) high triglycerides defined as serum level ≥1.68 mmol/L; (4) low HDL-cholesterol defined as serum level <1.03 mmol/L in men and <1.29 mmol/L in women; (5) high blood glucose defined as fasting blood glucose ≥6.10 mmol/L and/or current use of insulin or oral medication for diabetes.
MetS, metabolic syndrome; BMI, body mass index; HDL, high-density lipoprotein.

September 1989 and February 1994. Of the 4,285 participants who were aged 40–74 years at baseline, a total of 374 were excluded for the following reasons: past history of CVD (ischemic heart disease and stroke: n=127), non-fasting visit (n=155), and missing information at the time of the baseline survey or lost to follow-up (n=92). The data for the remaining 3,911 participants (1,822 men and 2,089 women) were then analyzed. Informed consent was given by all participants. The present cohort study was approved by the Institutional Review Board of the NCVC.

Baseline Examination

Well-trained nurses obtained information on smoking (never, ex-, or current smoker), alcohol drinking (never, ex-, or current drinker), and the medical history of each participant. If the participant answered yes to "current smoker", information was obtained for how many cigarettes per day were smoked.