in middle-aged women between Japan and the United States around 1990. Data from the Suita Study, the only population-based study in urban area in Japan where >70% of Japanese live, show that in 1990–1994 mean levels of LDL-C and HDL-C (mg/dL) were 126 and 57 in women aged 40–49 years, respectively and 146 and 57 in women aged 50–59 years, respectively. NHANES III (1988–1994) reported that corresponding numbers were 131 and 56 in women aged 45–54 years, respectively and 144 and 56 in women aged 55–64 years, respectively.[59]

We observed that levels of BP in currently aged 50–69 years have been higher in Japan than in the United States at least for the past two decades. The difference in clinical guidelines for hypertension in the past accounts to some extent for the differences in levels of BP. For example, the 1990 National Survey of Circulatory Disorders in Japan used the criteria of hypertension as systolic BP ≥ 160 mmHg, or diastolic BP ≥ 95 mmHg, or on hypertension medication, based on the WHO criteria,[60] whereas the Fifth Report of the Joint National Committee on Detection, Education, and Treatment of High Blood Pressure in the United States in 1993 defined hypertension as systolic BP ≥ 140 mmHg or diastolic BP ≥ 90 mmHg.[61]

Rates of cigarette smoking have been lower in women in Japan than in the United States, which could partly contribute to lower CHD rates in women in Japan. Meanwhile rates of cigarette smoking in men in Japan have been much higher than in the United States for the past 3 decades (e.g., 70 to 50% in Japan[6,20] versus 40 to 30% in the United States).[15] Passive smoking is an independent risk factor for CHD both in Japan[62] and the US,[63] about 30% higher risk for CHD in passive smokers compared to never smokers. Thus, it is unlikely that the difference in rates of cigarette smoking largely accounts for the difference in CHD rates between women in Japan and the United States.

Our results showed that prevalence of diabetes in women currently aged 60–69 years has been similar between Japan and the United States for the past two decades, whereas that in women currently aged 50–59 years in the United States was almost twice as high as in Japan. Because CHD rates in women aged 50–59 years is lower than that in women aged 60–69 years, the difference in prevalence of diabetes in women aged 50–59 years is unlikely to largely contribute to the difference in CHD rates between Japan and the United States. It is speculated that the difference in the prevalence of diabetes in younger generation is due to the difference in recent trend in BMI between Japan and the United States.[64] Between 1980 and 2010, BMI in women in Japan remained similar whereas that in women in the United States much increased.

It is possible that the difference in BMI contributes to the difference in CHD in women between Japan and the United States. Major plausible intermediary factors linking obesity and CHD are BP, total cholesterol, and type 2 diabetes.[65,66] The current study has shown that it is very unlikely that the difference in these factors contributes to the difference in CHD in women between Japan and the United States. Although we have investigated other possible intermediary factors including C-reactive protein,[12] fibrinogen,[12] adiponectin,[67] D-dimer,[68] von Willebrand factor,[68] and lipoprotein-associated phospholipase A_2[69] in our study in men, none of these factors significantly contributed to the difference in atherosclerosis. These observations need to be confirmed in the future study in women.

Early menopause is significantly associated with increased risk of CHD both in Japan[52] and the United States.[70] Meanwhile, associations of age at menarche with CHD are equivocal.[52,71,72] Available data show that the age at natural menopause in women in Japan is earlier whereas the age at menarche is similar between the two countries: 49.3 in Japan versus 51.3 in the United States for menopause and 12.5 in Japan and 12.8 in the United States for menarche.[73] Thus, the difference in age at menopause is unlikely to contribute to the lower rate of CHD in women in Japan. Although a rate of oral contraceptive use in women in Japan is very low as compared with other developed countries[74] because it was introduced in 1999,[75] recent epidemiological studies show that use of oral contraceptives including past use is not associated with increased risk of CHD.[76]

Lower prevalence of hysterectomy in Japan than in the United States[77] is unlikely to be a significant factor responsible for lower CHD rates in Japan. This is because hysterectomy is not the major determinant of CHD over traditional risk factors. We have recently reported from the Women's Health Initiative Observational Study[78] that the hazard ratio of hysterectomy for incident cardiovascular disease was 1.26 (95% CI: 1.16–1.36, $p < 0.001$), which was attenuated after adjusting traditional CHD risk and other factors to 1.10 (95% CI: 1.00–1.21, $p = 0.042$). The results suggest that a more adverse profile of CHD risk factors in women who had undergone hysterectomy compared with those who had not, rather than hysterectomy itself, is associated with CHD. Moreover, given the hazard ratio of 1.10 after adjusting for CHD and other risk factors, population-attributable risk[79] of hysterectomy for CHD is very low.

Evidence from migrant studies of Japanese and multiethnic studies in the United States does not support the hypothesis that the Japanese are genetically protected against CHD. Migrant studies of Japanese to the United States clearly show an increase in CHD and atherosclerotic burden in Japanese Americans as compared to Japanese in Japan. The NIHON-SAN study, a cross-sectional study of cardiovascular disease and its risk factors in middle-aged Japanese men living in Japan, Hawaii, and California in the 1960s, has shown that CHD mortality is significantly higher in Japanese Americans than in Japan.[80] We have reported from a population-based study of Japanese and Japanese and white American men aged 40–49 years that levels of atherosclerosis assessed as coronary artery calcification and carotid IMT in Japanese Americans are higher or similar compared with U.S. whites.[12] Although CHD mortality in Japanese Americans is reported to be lower compared with white Americans,[81–83] a more recent study has shown that CHD mortality is similar between young Japanese and white American women. Using U.S. Census data and California mortality data in 1990 and 2000, Palaniappan et al. reported age-, sex-, and ethnic-specific CHD mortality for six ethnic groups in California.[84] Although standardized mortality ratios (SMR) of CHD in women aged 45–64 years and 65–84 years are much lower in Japanese Americans than in white Americans, SMR of CHD in women aged 24–44 years is very similar between the two groups.

Given that the lower rates of CHD and atherosclerosis in Japan than in the United States are unlikely to be primarily due to differences in traditional risk or genetic factors, the most likely hypothesis is that there are common source exposures in the diet among Japanese in Japan, which accounts

for their low CHD rates and atherosclerosis. The international collaborative study of macro- and micro-nutrients and blood pressure, which provides the most comprehensive dietary data in the United States and Japan[85] and other dietary studies[86] show that Japanese have markedly high intake of marine n-3 fatty acids (1,000 mg/day in Japan vs. 100 mg/day in the U.S.)[87] and isoflavones (25～50 mg/day in Japan vs. <2 mg/day in the U.S.).[86]

A large prospective cohort study in Japan supports the hypothesis that dietary intake of marine n-3 fatty acids and isoflavones are protective against CHD in Japan. The Japan Public Health Center-Based Study, a population-based cohort of individuals aged 40–59 years in Japan, following more than 40,000 individuals for about 10 years, reported that dietary intake of marine n-3 fatty acids had a significant inverse association with myocardial infarction after adjusting for history of hypertension and diabetes, medication for hyperlipidemia and other potential confounders.[88] This study also reported that that dietary intake of soy isoflavones had a significant inverse association with incidence of myocardial infarction in women even after adjusting for the above-mentioned factors.[89] Additionally, we have recently reported that high serum percentage of marine n-3 fatty acids significantly contributed to the difference in levels of atherosclerosis assessed as coronary artery calcification and carotid IMT between Japanese and whites.[12] It is unlikely that marine n-3 fatty acids or soy isoflavones exert their potential protective effects against CHD through total cholesterol, HDL-C, BP, or glucose homeostasis because recent systematic reviews show that effects of marine n-3 fatty acids or soy isoflavones on these factors are clinically insignificant.[90–92] A large-scale randomized clinical trial (RCT) of marine n-3 fatty acids recently conducted in Japan demonstrated their clear benefit on CHD events,[93] although several recent RCTs failed to show their benefits.[94–96] The discrepancy is at least due to the difference in dosage of marine n-3 fatty acids in RCT and background dietary intake of marine n-3 fatty acids as described above. No RCT of isoflavones on CHD has been reported, although many RCTs of isoflavones on CHD risk factors have been reported.[90,97 98]

Limitations of this study warrant discussion. Mortality statistics are subject to misclassification without validation study. However, the fact that validated AMI incidence is much lower in Japan than in all the registries in other countries strongly indicate that low CHD mortality in Japan is not due to misclassification. Although we compared the trend in risk factors between Japan and the United States using primarily national survey data, these data may not be directly compared, because measurements are not strictly standardized between the countries. Factors other than those described in this paper that may be independently associated with CHD risk in each population, such as physical inactivity, psychosocial factors, and medical practice[55,99,100] could contribute to the difference in CHD rates in women between Japan and the Unites States.

In conclusion, we have shown that CHD mortality in women in Japan is much lower than in women in the United States. Differences in risk factors and their trends are unlikely to explain the difference in CHD rates in women in Japan and the United States. The results from migrant studies of the Japanese to the United States and international studies do not support the notion that Japanese are genetically protected against atherosclerosis and CHD. Investigating factors responsible for low CHD rates in women in Japan is important and may lead to new strategy for CHD prevention.

Disclosure Statement

No competing financial interests exist.

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Address correspondence to:
Akira Sekikawa, MD, PhD
Department of Epidemiology
Graduate School of Public Health
University of Pittsburgh
130 North Bellefield Avenue, Suite 546
Pittsburgh, PA 15213

E-mail: akira@pitt.edu

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Original Article

Effect of Age on the Association Between Waist-to-Height Ratio and Incidence of Cardiovascular Disease: The Suita Study

Yukako Tatsumi[1,2], Makoto Watanabe[1], Yoshihiro Kokubo[1], Kunihiro Nishimura[1], Aya Higashiyama[3], Tomonori Okamura[4], Akira Okayama[5], and Yoshihiro Miyamoto[1]

[1]Department of Preventive Cardiology, National Cerebral and Cardiovascular Center, Suita, Osaka, Japan
[2]Department of Mathematical Health Science, Graduate School of Medicine, Osaka University, Suita, Osaka, Japan
[3]Department of Environmental and Preventive Medicine, Hyogo College of Medicine, Nishinomiya, Hyogo, Japan
[4]Department of Preventive Medicine and Public Health, Keio University, Tokyo, Japan
[5]The First Institute for Health Promotion and Health Care, Japan Anti-tuberculosis Association, Tokyo, Japan

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ABSTRACT

Background: Waist-to-height ratio (WHtR) has been shown to be a useful screening tool for metabolic syndrome and cardiovascular disease (CVD). We investigated the association of WHtR with CVD incidence by age group.
Methods: We conducted a 13.0-year cohort study of Japanese adults (2600 men and 2888 women) with no history of CVD. WHtR was calculated as waist circumference (cm) (WC) divided by height (cm). We stratified participants by sex and age group (30–49, 50–69, ≥70 years). Using the Cox proportional hazards model, we calculated hazard ratios (HRs) and 95% CIs for CVD in relation to WHtR quartile for participants aged 50 to 69 years and 70 years or older.
Results: Men aged 50 to 69 years in the highest quartile had significantly increased risks of CVD and coronary heart disease as compared with the lowest quartile; the HRs (95% CI) were 1.82 (1.13–2.92) and 2.42 (1.15–5.12), respectively. Women aged 50 to 69 years in the highest quartile had a significantly increased risk of stroke (HR, 2.43; 95% CI, 1.01–5.85). No significant results were observed in men or women aged 70 years or older. The likelihood ratio test showed that the predictive value of WHtR was greater than that of WC among men aged 50 to 69 years.
Conclusions: The association between WHtR and CVD risk differed among age groups. WHtR was useful in identifying middle-aged Japanese at higher risk of CVD and was a better predictor than WC of CVD, especially in men.

Key words: waist-to-height ratio; age difference; cardiovascular disease

INTRODUCTION

Obesity and central obesity are closely tied to metabolic risks.[1,2] Waist circumference (WC) is an index of central obesity[3] and is an important component in the diagnostic criteria for metabolic syndrome.[4] Several meta-analyses have reported an association of WC with cardiovascular disease (CVD) and mortality.[5,6] Recently, waist-to-height ratio (WHtR) was shown to be a useful global clinical screening tool for cardiometabolic risk and CVD.[7,8]

WHtR is easy to measure, and the cut-off point for WHtR is subject to less ethnic variation.[7,8] However, WHtR could differ among age groups because whole-body fat distribution and WC change considerably with age[9,10] and because height differs among generations.[11] It is thus important to consider age in assessing the association between WHtR and CVD risk, but few previous studies have done so.[12,13] Therefore, in this long-term prospective cohort study of a Japanese urban population, we investigated the effect of WHtR on CVD risk among participants classified by age group.

METHODS

Study population

The Suita Study is a prospective population-based cohort study of an urban area of Japan and was established in 1989. The details of this study have been described elsewhere.[14–16] Briefly, 6407 men and women aged 30 to 83 years underwent

Address for correspondence. Yukako Tatsumi, Department of Preventive Cardiology, National Cerebral and Cardiovascular Center, 5-7-1 Fujishiro-dai, Suita, Osaka 565-8565, Japan (e-mail: y-tatumi@sahs.med.osaka-u.ac.jp).

a baseline survey at the National Cerebral and Cardiovascular Center between September 1989 and March 1994. Among them, a total of 919 were excluded due to past history of CVD ($n = 208$), loss to follow-up ($n = 535$), and missing data ($n = 176$). The remaining 5488 participants (2600 men and 2888 women) were included in the analysis. This cohort study was approved by the Institutional Review Board of the National Cerebral and Cardiovascular Center.

Baseline examination

Blood samples were centrifuged immediately after collection, and a routine blood examination was performed, including measurement of serum levels of total cholesterol and glucose. About 96% of participants had fasted for at least 8 hours before the blood test. Well-trained physicians used a standard mercury sphygmomanometer to measure blood pressure in triplicate on the right arm after 5 minutes of rest. Hypertension was defined as systolic blood pressure of at least 140 mm Hg, diastolic blood pressure of at least 90 mm Hg, or use of antihypertensive agents. Diabetes was defined as a fasting plasma glucose level of at least 7.0 mmol/L (126 mg/dL), a non-fasting plasma glucose level of at least 11.1 mmol/L (200 mg/dL), or use of antidiabetic agents. Hypercholesterolemia was defined as a total cholesterol level of at least 5.7 mmol/L (220 mg/dL) or use of antihyperlipidemic agents. Participants were wearing light clothing during height and weight measurement. WC was measured at the umbilical level, with the participant in a standing position. WHtR was defined as WC (cm) divided by height (cm). Body mass index (BMI) was defined as weight (kg) divided by the height (m) squared. Public-health nurses obtained information on participants' smoking, drinking, and medical histories.

Endpoint determination

The endpoint determination has been previously reported.[14–16] The endpoints of the present study were (1) date of first coronary heart disease (CHD) or stroke event; (2) date of death; (3) date of departure from Suita city; or (4) December 31, 2007. The first step in the survey of CHD and stroke was checking the health status of all participants by means of clinical visits every 2 years and a yearly questionnaire (by mail or telephone). For the second step, in-hospital medical records of participants suspected of having CHD or stroke were reviewed by registered hospital physicians, who were blinded to the baseline information. In addition, to complete the survey, we also conducted a systematic search of death certificates to identify cases of fatal CHD and stroke. In Japan, all death certificates are forwarded to the Ministry of Health, Welfare, and Labour and coded for the National Vital Statistics. The criteria for myocardial infarction were based on the World Health Organization Monitoring of Trends and Determinants in Cardiovascular Disease projects.[17] In addition to myocardial infarction, we also evaluated coronary angioplasty, coronary artery bypass grafting, and sudden cardiac death, all of which were included in the definition of CHD. Stroke was defined according to criteria from the US National Survey of Stroke and was confirmed by computed tomography.[18] Classification of stroke was based on examination of computed tomography scans, magnetic resonance images, and autopsy findings.

Statistical analysis

To assess the association between age and WHtR, we analyzed mean WC, height, and WHtR according to age in men and women. Pearson product-moment correlation coefficients between height and waist were calculated by sex and age group (30–49, 50–69, ≥70 years). Participants were categorized based on quartiles of WHtR by sex and age group. To compare baseline characteristics among WHtR quartiles, analysis of variance was used for continuous variables and the χ^2 test was used for dichotomous and categorical variables.

The Cox proportional hazards model was used to investigate the association between WHtR and CVD risk only among participants aged 50 to 69 years and 70 years or older, because there were too few CVD cases (men: 17, women: 11) for statistical analysis among those aged 30 to 49 years. Interaction terms were added to the models to assess the interaction between age and WHtR quartile for the risk of CVD. Hazard ratios (HRs) and 95% CIs were computed, and the lowest quartile of WHtR was defined as the reference group. To adjust for confounding factors, we included age, smoking status (current, quit, or never), and drinking status (current, quit, or never) in the model. Cardiometabolic risk factors such as hypertension, diabetes, and hypercholesterolemia were not included in the model because central obesity is upstream in the "metabolic domino".[19] However, in sensitivity analysis, we adjusted for hypertension, diabetes, and hypercholesterolemia to confirm that WHtR was an independent risk factor. The same analysis was performed for WC. In addition, to further assess cut-off points for WHtR, the highest quartile was dichotomized by median WHtR (ie, upper Q4 and lower Q4), and HRs and 95% CIs were estimated. The likelihood ratio test was used to compare the predictive values of WHtR with WC, as follows. First, we calculated the −2 logarithm likelihood for the model including the confounding factors, age, smoking, and drinking status ($-2\,ln[L_c]$). Second, we calculated the −2 logarithm likelihood for the model including the confounding factors plus WHtR ($-2\,ln[L_{c+WHtR}]$). The difference, ie, ($-2\,ln[L_c] - (-2\,ln[L_{c+WHtR}])$), had an approximate χ^2 distribution with 1 degree-of-freedom. The same analysis was performed for WC.

All P values were 2-tailed, and a P value less than 0.05 was considered statistically significant. All statistical analyses were performed with SPSS (Version 20.0J; Japan IBM, Tokyo, Japan).

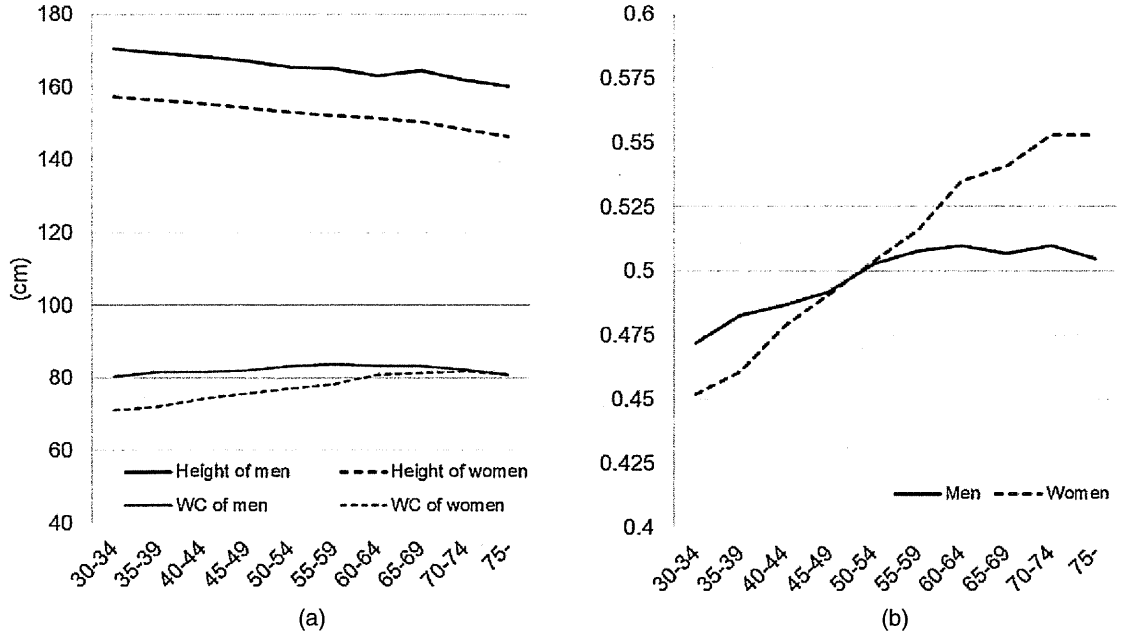

Figure. (a) Average WC (waist circumference), height, and (b) waist-to-height ratio according to age (The Suita Study, Japan)

RESULTS

During the follow-up period (mean, 13.0 years), 428 CVD events (184 CHD and 244 strokes) were observed. The Figure shows average WC, height, and WHtR by sex and age. WC in men increased up to age 50 years, remained almost unchanged from age 50 to 69 years, and decreased at age 70 years or older. WC in women younger than 75 years increased with advancing age and decreased in women aged 75 years or older, as compared with women aged 70 to 74 years. Height decreased with advancing age in both sexes. WHtR in men increased until approximately age 60 years. WHtR in women younger than 75 years increased with advancing age. The Pearson product-moment correlation coefficients (95% CI) between height and WC were 0.16 (0.09–0.22), 0.24 (0.19–0.30), and 0.13 (0.04–0.22) among men aged 30 to 49, 50 to 69, and 70 years or older, respectively, and 0.07 (0.01–0.13), 0.07 (0.02–0.13), 0.09 (−0.003–0.19) among women in the respective age groups.

Tables 1 and 2 summarize the baseline characteristics according to WHtR quartile (results among men and women aged 30–49 years are shown in eTable 1.) The prevalence of hypertension significantly differed by WHtR quartile, except among men aged 70 years or older. The prevalence of hypercholesterolemia and diabetes significantly differed by WHtR quartile among men and women aged 50 to 69 years.

Table 3 shows multivariable-adjusted HRs and 95% CIs for CVD and its subtypes according to WHtR quartile. A significant interaction was observed between age and WHtR for CVD among men (P for interaction = 0.02). Men aged 50 to 69 years in the highest quartile had significantly higher risks of CVD and CHD as compared with men in the lowest quartile; the HRs (95% CI) were 1.82 (1.13–2.92) and 2.42 (1.15–5.12), respectively. There were significant linear increases in the HRs for CVD, CHD, and ischemic stroke in men aged 50 to 69 years. After further adjustment for hypertension, diabetes, and hypercholesterolemia, the HRs (95% CI) were 1.46 (0.90–2.36) and 1.89 (0.89–4.03), respectively (eTable 3). Women aged 50 to 69 years in the highest quartile had a significantly higher risk of stroke than did those in the lowest quartile; the HR (95% CI) was 2.43 (1.01–5.85). There were significant linear increases in the HRs of CVD and stroke in women aged 50 to 69 years. After further adjustment for hypertension, diabetes, and hypercholesterolemia, the HR (95% CIs) was 2.06 (0.84–5.04) (eTable 3).

When men aged 50 to 69 years in the highest quartile were dichotomized by median WHtR (0.56), the HR (95% CI) for CVD was 1.37 (0.76–2.46) for those in the lower WHtR group and 2.34 (1.38–3.97) for those in the upper WHtR group (eTable 2). When women aged 70 years or older in the highest quartile were dichotomized by median WHtR (0.65), the HR for CVD was 1.42 (0.63–3.18) for those in the lower WHtR group and 2.33 (1.10–4.94) for those in the upper WHtR group. After adjustment for hypertension, diabetes, and hypercholesterolemia, the HRs in the upper WHtR decreased but remained significant, ie, 1.78 (1.04–3.05) among men aged 50 to 69 years and 2.16 (1.02–4.61) among women aged 70 years or older.

Table 4 shows the HRs and 95% CIs for CVD in relation to WC quartile. Among men aged 50 to 69 years in the highest quartile, the HR for CVD was 1.63 (1.03–2.59), although the HRs of CVD did not show a significant linear increase in this group. Among women aged 50 to 69 years, a significant linear

Table 1. Baseline characteristics of men, according to age group and quartile of waist-to-height ratio: The Suita Study, Japan

	Q1 (low)	Q2	Q3	Q4 (high)	*P*-value
Age 50–69 years					
No. of subjects	308	304	304	308	
Waist-to-height ratio	0.374–0.475	0.476–0.508	0.509–0.536	0.537–0.761	
Waist, cm	74.0 ± 4.3	81.2 ± 2.9	85.7 ± 3.1	92.8 ± 5.5	<0.01
Height, cm	165.0 ± 5.3	164.9 ± 5.6	164.4 ± 5.4	163.7 ± 5.3	0.01
Age, years	59.0 ± 5.3	59.1 ± 5.2	59.1 ± 5.5	59.4 ± 5.3	0.77
Body mass index, kg/m^2	20.1 ± 1.7	22.1 ± 1.5	23.7 ± 1.5	25.9 ± 2.3	<0.01
Hypertension, %	31	35	45	51	<0.01
Diabetes, %	6	7	9	11	0.045
Hypercholesterolemia, %	23	28	40	35	<0.01
Smoking status (current/quit/never), %	58/25/17	50/31/19	46/35/19	44/38/19	0.01
Drinking status (current/quit/never), %	79/2/19	74/4/22	79/4/17	76/4/21	0.58
Age ≥70 years					
No. of subjects	120	120	124	119	
Waist-to-height ratio	0.352–0.472	0.473–0.508	0.509–0.543	0.544–0.688	
Waist, cm	70.6 ± 5.0	79.8 ± 3.4	84.9 ± 3.3	92.2 ± 5.6	<0.01
Height, cm	162.5 ± 6.0	162.2 ± 5.7	161.3 ± 5.3	159.3 ± 6.0	<0.01
Age, years	74.0 ± 3.0	73.5 ± 2.7	74.1 ± 2.7	73.7 ± 2.9	0.40
Body mass index, kg/m^2	18.5 ± 1.7	21.3 ± 1.7	22.7 ± 1.4	25.6 ± 2.0	<0.01
Hypertension, %	42	44	51	57	0.07
Diabetes, %	4	7	7	8	0.70
Hypercholesterolemia, %	23	29	26	31	0.46
Smoking status (current/quit/never), %	37/48/16	42/41/18	38/47/15	30/50/19	0.66
Drinking status (current/quit/never), %	58/8/33	62/11/28	62/6/32	65/8/28	0.73

Continuous data with a normal distribution were analyzed with analysis of variance: mean ± SD.
Dichotomous and categorical data were analyzed with the χ^2 test.
Q, quartile; hypertension was defined as systolic blood pressure/diastolic blood pressure ≥ 140/90 mm Hg or current use of antihypertensive medications; diabetes was defined as a fasting plasma glucose level ≥ 7.0 mmol/L, a non-fasting plasma glucose level ≥ 11.1 mmol/L, or current use of antidiabetic medications; hypercholesterolemia was defined as a total serum cholesterol level ≥ 5.7 mmol/L or current use of antihyperlipidemic medications.

increase was observed in the HRs for CVD (P for trend = 0.04). However, after further adjustment for hypertension, diabetes, and hypercholesterolemia, these associations were no longer significant among men or women.

The χ^2 values for the likelihood ratio test were 6.49 ($P = 0.01$) for WHtR and 3.63 ($P = 0.06$) for WC among men aged 50 to 69 years, and 4.45 ($P = 0.03$) for WHtR and 4.54 ($P = 0.03$) for WC among women aged 50 to 69 years.

DISCUSSION

Our main findings were that WHtR was significantly positively associated with CVD and CHD risk among men aged 50 to 69 years and with stroke risk among women aged 50 to 69 years. Among men, there was a significant interaction between age and WHtR for CVD incidence. Among women aged 50 to 69 years, there was a borderline association between a WHtR in the highest quartile and increased CVD risk. In addition, among women aged 70 years or older, a WHtR in the upper level of the highest quartile was associated with significantly elevated CVD risk. These findings suggest that the association between WHtR and CVD incidence differs according to age and sex.

Two previous studies, in the United States and China, reported that the association between WHtR and CVD risk was stronger among younger adults as compared with elderly adults.[12,13] We too observed a significantly stronger association between WHtR and CVD risk among relatively young adults (age 50–69 years) as compared with elderly adults (age ≥70 years), which supports the results of previous studies. Consequently, these findings suggest that age stratification is important in estimating the association between WHtR and CVD risk.

In this population, physical frame, eg, WC and height, differed by age group. It has been reported that WC and the ratio of abdominal fat to whole-body fat differ by age.[9,10] In addition, the National Health and Nutrition Examination Survey in Japan noted that height clearly differed by generation.[11] This generational difference in physical frame, as well as aging, could lead to age differences in the association between WHtR and CVD risk.

A recent meta-analysis reported an optimal cut-off point of 0.50 for WHtR in both sexes.[7] However, the present findings suggest that, regardless of age or sex, a cut-off of 0.50 is somewhat low for identifying individuals at higher risk for CVD. The association with CVD risk was of at least

Table 2. Baseline characteristics of women, according to age group and quartile of waist-to-height ratio: The Suita Study, Japan

	Q1 (low)	Q2	Q3	Q4 (high)	*P*-value
Age 50–69 years					
No. of subjects	337	340	335	339	
Waist-to-height ratio	0.348–0.472	0.473–0.520	0.521–0.568	0.569–0.838	
Waist, cm	67.3 ± 4.1	75.4 ± 3.3	82.7 ± 3.4	92.1 ± 6.6	<0.01
Height, cm	153.0 ± 4.7	151.8 ± 4.9	152.1 ± 5.1	150.3 ± 5.2	<0.01
Age, years	57.6 ± 5.3	58.5 ± 5.3	59.5 ± 5.2	60.5 ± 5.4	<0.01
Body mass index, kg/m^2	19.8 ± 2.0	21.7 ± 2.0	23.1 ± 2.3	25.9 ± 3.3	<0.01
Hypertension, %	21	32	36	52	<0.01
Diabetes, %	2	3	5	9	<0.01
Hypercholesterolemia, %	49	57	57	62	0.01
Smoking status (current/quit/never), %	11/2/86	11/3/86	9/3/88	12/5/84	0.43
Drinking status (current/quit/never), %	26/2/73	29/2/69	28/2/71	31/1/68	0.75
Postmenopausal, %	90	94	95	94	0.06
Age ≥70 years					
No. of subjects	103	103	103	103	
Waist-to-height ratio	0.379–0.496	0.497–0.554	0.556–0.602	0.603–0.812	
Waist, cm	68.1 ± 4.4	77.3 ± 4.1	85.6 ± 3.6	95.2 ± 6.4	<0.01
Height, cm	148.4 ± 5.5	147.7 ± 6.1	148.1 ± 5.1	145.8 ± 5.1	<0.01
Age, years	73.8 ± 2.9	73.4 ± 2.7	73.8 ± 2.7	74.0 ± 2.6	0.56
Body mass index, kg/m^2	19.1 ± 2.1	21.3 ± 2.3	23.1 ± 2.1	26.2 ± 2.9	<0.01
Hypertension, %	53	44	50	64	0.03
Diabetes, %	2	5	6	4	0.54
Hypercholesterolemia, %	42	51	53	52	0.32
Smoking status (current/quit/never), %	12/6/83	9/4/87	6/5/89	7/5/88	0.78
Drinking status (current/quit/never), %	22/5/73	18/2/81	19/1/80	19/4/77	0.62
Postmenopausal, %	100	100	100	100	1.00

Continuous data with a normal distribution were analyzed with analysis of variance: mean ± SD.
Dichotomous and categorical data were analyzed with the χ^2 test.
Q, quartile; hypertension was defined as systolic blood pressure/diastolic blood pressure ≥ 140/90 mm Hg or current use of antihypertensive medications; diabetes was defined as a fasting plasma glucose level ≥ 7.0 mmol/L, a non-fasting plasma glucose level ≥ 11.1 mmol/L, or current use of antidiabetic medications; hypercholesterolemia was defined as a total serum cholesterol level ≥ 5.7 mmol/L or current use of antihyperlipidemic medications.

borderline significance for a WHtR in the fourth quartile, except among men aged 70 years or older. Additional analyses showed that the risks markedly increased, particularly in the upper level of the fourth WHtR quartile, among men aged 50 to 69 years and women aged 70 years and older. These results suggest the presence of a threshold rather than a dose-response relation for WHtR, although the present sample was too small to confirm this hypothesis. Additionally, we think that cut-offs should be set in relation to age and sex. On the basis of our results, we propose the following cut-offs (which do not include men aged 70 years or older): 0.560 for men aged 50 to 69 years, 0.569 for women aged 50 to 69 years, and 0.647 for women aged 70 years or older.

The risk of CVD among men aged 50 to 69 years, and women aged 70 years, in the upper level of the highest quartile was significantly elevated even after adjustment for hypertension, hyperlipidemia, and diabetes. We believe that there are 2 possible explanations for this finding. First, an extremely high WHtR might actually be an independent risk factor ie, separate from classical cardiometabolic risks. It has been reported that abdominal obesity is related to increased levels of plasminogen activator inhibitor-1, which can lead to blood coagulation.[20] Such background mechanisms might be important. Second, our findings could be due to insufficient adjustment for confounders in the Cox regression model. Irrespective of the reason, men aged 50 to 69 years, and women aged 70 years or older, with extremely high WHtRs have a considerably higher risk for CVD and should be closely monitored.

We previously investigated the association between WC and CVD risk without age stratification[21] and found a significant association between WC and the risks of CVD and stroke among women but no significant association among men. However, the present age-stratified analysis of WC suggests that our previous results were substantially influenced by age. Therefore, we compared WHtR and WC in relation to CVD in analysis stratified by age group and found that the HRs associated with the highest quartile of WHtR were higher than those associated with WC among middle-aged men and that the predictive value of WHtR was greater than that of WC. Several previous studies reported similar results[12,22–24]; therefore our findings are consistent with those

Table 3. Multivariable-adjusted hazard ratios for cardiovascular disease according to sex, age group, and quartile of WHtR: The Suita Study, Japan

	Q1 (low)	Q2	Q3	Q4 (high)	*P* for trend
Men					
Age 50–69 years					
Person-years	4070	3069	3879	3842	
CVD, no. of cases	28	31	32	47	
HRs	1	1.14 (0.68–1.90)	1.23 (0.74–2.05)	1.82 (1.13–2.92)	0.01
CHD, no. of cases	10	16	16	23	
HRs	1	1.57 (0.71–3.47)	1.72 (0.77–3.80)	2.42 (1.15–5.12)	0.02
Stroke, no. of cases	18	15	16	24	
HRs	1	0.91 (0.46–1.81)	0.95 (0.48–1.87)	1.56 (0.84–2.89)	0.16
Ischemic stroke, no. of cases	10	9	15	18	
HRs	1	0.99 (0.40–2.43)	1.59 (0.71–3.56)	2.06 (0.94–4.49)	0.04
Age ≥70 years					
Person-years	1055	1128	1193	1155	
CVD, no. of cases	21	29	27	30	
HRs	1	1.36 (0.77–2.39)	1.09 (0.62–1.93)	1.36 (0.78–2.38)	0.45
CHD, no. of cases	13	11	10	15	
HRs	1	0.87 (0.39–1.97)	0.63 (0.28–1.45)	1.09 (0.52–2.30)	0.99
Stroke, no. of cases	8	18	17	15	
HRs	1	2.09 (0.90–4.81)	1.79 (0.77–4.15)	1.84 (0.78–4.35)	0.29
Ischemic stroke, no. of cases	4	12	10	11	
HRs	1	2.84 (0.91–8.83)	2.22 (0.69–7.07)	2.71 (0.86–8.53)	0.18
Women					
Age 50–69 years					
Person-years	4811	4863	4477	4470	
CVD, no. of cases	16	18	21	33	
HRs	1	1.09 (0.56–2.14)	1.32 (0.69–2.54)	1.80 (0.98–3.32)	0.04
CHD, no. of cases	9	4	4	13	
HRs	1	0.47 (0.14–1.51)	0.47 (0.14–1.54)	1.35 (0.56–3.22)	0.43
Stroke, no. of cases	7	14	17	20	
HRs	1	1.85 (0.75–4.60)	2.35 (0.97–5.70)	2.43 (1.01–5.85)	0.04
Ischemic stroke, no. of cases	3	7	9	10	
HRs	1	2.09 (0.54–8.10)	2.78 (0.75–10.33)	2.35 (0.63–8.77)	0.22
Age ≥70 years					
Person-years	1095	1259	1164	1094	
CVD, no. of cases	15	15	13	24	
HRs	1	1.00 (0.48–2.08)	0.91 (0.43–1.93)	1.83 (0.95–3.53)	0.08
CHD, no. of cases	6	7	5	9	
HRs	1	1.23 (0.40–3.77)	0.98 (0.29–3.32)	1.78 (0.62–5.14)	0.34
Stroke, no. of cases	9	8	8	15	
HRs	1	0.85 (0.32–2.23)	0.88 (0.34–2.29)	1.92 (0.83–4.45)	0.11
Ischemic stroke, no. of cases	5	4	4	9	
HRs	1	0.83 (0.22–3.16)	0.77 (0.21–2.91)	1.99 (0.66–6.04)	0.21

Multivariable adjustment was performed for age, smoking, and drinking status. Parentheses indicate 95% CIs for HRs.
Abbreviations: WHtR, waist-to-height ratio; Q, quartile; CVD, cardiovascular disease; CHD, coronary heart disease; HR, hazard ratio.

of previous studies. In contrast, WHtR and WC had similar predictive values for CVD among women in the present study. Many previous studies found that WHtR was similar to WC in predicting CVD risk among women.[12,22,24–26] The effect of dividing WC by height might be limited because the correlation of WC with height is weaker among women than among men. Consequently, we believe that WHtR is a better predictor than WC, particularly among middle-aged men.

The superiority of WHtR might be explained by the fact that WHtR, as measured by computed tomography, was more closely correlated than WC with intra-abdominal fat,[27] and a previous study reported that intra-abdominal fat was positively associated with number of cardiometabolic risk factors.[28] In addition, shorter adults tend to have more cardiometabolic risk factors than do taller individuals with a similar WC.[29] This suggests that WHtR, ie, dividing WC by height, is more strongly related than WC to cardiometabolic risk factors. Thus, we believe that WHtR better reflects the accumulation of cardiometabolic risks and leads to superior prediction of CVD.

BMI, along with indices of central obesity, has been an important obesity index in predicting CVD incidence,[30] although a meta-analysis reported that the predictive power of WHtR for CVD was higher than that of BMI.[7] Another report found a significant association between BMI and CVD after adjustment for WHtR[12] and suggested that WHtR and BMI are independently associated with CVD risk. Therefore, it might be better to use both BMI and WHtR to assess obesity.

Table 4. Multivariable-adjusted hazard ratios for cardiovascular disease according to sex, age group, and quartile of WC: The Suita Study, Japan

	Q1 (low)	Q2	Q3	Q4 (high)	*P* for trend
Men					
Age 50–69 years					
Person-years	4078	4004	3872	3806	
CVD, no. of cases	32	33	29	44	
HRs	1	1.07 (0.66–1.75)	0.97 (0.58–1.61)	1.63 (1.03–2.59)	0.06
CHD, no. of cases	13	17	12	23	
HRs	1	1.28 (0.62–2.63)	0.96 (0.44–2.12)	2.02 (1.02–4.02)	0.07
Stroke, no. of cases	19	16	17	21	
HRs	1	0.97 (0.50–1.88)	0.96 (0.49–1.86)	1.43 (0.76–2.67)	0.31
Ischemic stroke, no. of cases	13	9	13	17	
HRs	1	0.80 (0.34–1.87)	1.07 (0.49–2.31)	1.64 (0.79–3.41)	0.15
Age ≥70 years					
Person-years	999	1208	1200	1124	
CVD, no. of cases	25	28	27	27	
HRs	1	0.94 (0.55–1.62)	0.91 (0.53–1.58)	1.06 (0.61–1.84)	0.87
CHD, no. of cases	14	11	12	12	
HRs	1	0.67 (0.30–1.47)	0.65 (0.30–1.43)	0.82 (0.38–1.78)	0.60
Stroke, no. of cases	11	17	15	15	
HRs	1	1.29 (0.60–2.77)	1.21 (0.55–2.66)	1.36 (0.62–2.99)	0.52
Ischemic stroke, no. of cases	5	10	10	12	
HRs	1	1.70 (0.58–4.98)	1.82 (0.62–5.37)	2.26 (0.79–6.47)	0.14
Women					
Age 50–69 years					
Person-years	4669	4685	5046	4221	
CVD, no. of cases	15	18	25	30	
HRs	1	1.19 (0.60–2.36)	1.43 (0.75–2.71)	1.87 (1.00–3.51)	0.04
CHD, no. of cases	7	5	5	13	
HRs	1	0.74 (0.24–2.34)	0.65 (0.21–2.08)	1.86 (0.73–4.72)	0.18
Stroke, no. of cases	8	13	20	17	
HRs	1	1.56 (0.65–3.77)	2.06 (0.90–4.70)	1.93 (0.82–4.54)	0.11
Ischemic stroke, no. of cases	4	6	9	10	
HRs	1	1.44 (0.41–5.10)	1.70 (0.52–5.54)	2.00 (0.62–6.52)	0.23
Age ≥70 years					
Person-years	1175	1234	1046	1157	
CVD, no. of cases	16	16	15	20	
HRs	1	1.05 (0.52–2.11)	1.11 (0.54–2.25)	1.45 (0.74–2.83)	0.28
CHD, no. of cases	8	6	7	6	
HRs	1	0.85 (0.29–2.49)	1.21 (0.43–3.43)	0.88 (0.30–2.59)	0.98
Stroke, no. of cases	8	10	8	14	
HRs	1	1.24 (0.49–3.14)	1.10 (0.41–2.93)	2.00 (0.83–4.87)	0.15
Ischemic stroke, no. of cases	5	4	4	9	
HRs	1	0.85 (0.23–3.21)	0.93 (0.25–3.47)	1.86 (0.61–5.61)	0.24

Multivariable adjustment was performed for age, smoking, and drinking status. Parentheses indicate 95% CIs for HRs.
Abbreviations: WC, waist circumference; Q, quartile; CVD, cardiovascular disease; CHD, coronary heart disease; HR, hazard ratio.

Our study has several limitations. First, the number of cases of CVD among participants aged 30 to 49 years was insufficient for statistical analysis. Further study is required to confirm an association between WHtR and CVD risk among younger adults. Second, the effect of visceral fat could not be estimated because we did not use computed tomography to measure abdominal fat distribution. Third, changes in WHtR during the follow-up period were not considered in the present study. Finally, because WC was measured once, the estimated risks might have been underestimated because of regression dilution bias.[31]

In conclusion, the present findings suggest that WHtR is useful in identifying middle-aged Japanese at higher risk of CVD and is more predictable than WC in determining CVD risk, especially among men. In addition, the data indicate that WHtR cut-off points should be set according to sex and age. This study enrolled a limited Japanese population, and further studies with larger and more ethnically diverse samples are required to confirm our findings.

ONLINE ONLY MATERIALS

eTable 1. Baseline characteristics and CVD incidence among men and women aged 30–49 years according to quartile of waist-to-height ratio: the Suita Study, Japan.

eTable 2. Multivariable-adjusted hazard ratios for cardiovascular disease in the upper and lower fourth quartile of WHtR according to sex and age group: the Suita Study, Japan.

eTable 3. Multivariable-adjusted hazard ratios for cardiovascular disease according to sex, age group, and quartile of WHtR: the Suita Study, Japan.
Abstract in Japanese.

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Conflicts of interest: None declared.

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Original Article

Renal Insufficiency without Albuminuria is Associated with Peripheral Artery Atherosclerosis and Lipid Metabolism Disorders in Patients with Type 2 Diabetes

Takahiro Yamashita[1], Hisashi Makino[1], Rieko Nakatani[1], Yoko Ohata[1], Yoshihiro Miyamoto[2] and Ichiro Kishimoto[1]

[1]Division of Endocrinology and Metabolism, National Cerebral and Cardiovascular Center, Suita, Osaka, Japan
[2]Division of Preventive Cardiology, National Cerebral and Cardiovascular Center, Suita, Osaka, Japan

Aims: A high prevalence of a low glomerular filtration rate (GFR) has recently been reported in patients with diabetes without albuminuria. We aimed to clarify the clinical characteristics of such patients, including the associations between these characteristics and atherosclerosis.
Methods: We investigated the correlations between the estimated GFR (eGFR) and lipid profiles, the ankle-brachial index (ABI) and the intima-media thickness (IMT) in 450 patients with type 2 diabetes without macroalbuminuria.
Results: The prevalence of renal insufficiency (RI) (GFR < 60 mL/min/1.73 m^2) in the patients without albuminuria was 19.1%. The ABI values of the patients with RI were significantly lower than those of the patients without RI, regardless of the presence of microalbuminuria, while there were no significant differences in IMT between the patients with and without RI. In a multivariate analysis, a low ABI was found to be significantly associated with a low eGFR, independent of age, sex, smoking, history of hypertension and/or dyslipidemia and duration of diabetes ($\beta = 0.134$, $p = 0.013$), whereas no significant associations were observed between the ABI and the urinary albumin excretion rate (UAER). The ApoB/LDL-C ratios and levels of ApoC3 were significantly higher in the patients with RI than those observed in the patients without RI, regardless of the presence of albuminuria.
Conclusions: RI without albuminuria is closely associated with atherosclerosis of the peripheral arteries in diabetic patients. Furthermore, alterations in lipid metabolism may underlie this association.

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Key words: Diabetic nephropathy, Macrovascular complications, Lipids

Introduction

Diabetic nephropathy is a strong risk factor for cardiovascular disease and early mortality[1, 2] as well as end-stage renal disease (ESRD). The first clinical sign of classic diabetic nephropathy is the development of microalbuminuria, which gradually leads to macroalbuminuria, a decreased glomerular filtration rate (GFR) and finally ESRD. Therefore, microalbuminuria is considered to be an early marker for the detection of advanced diabetic nephropathy, another precursor of ESRD. However, several recent studies have found low GFR values in some patients with type 2 diabetes without macroalbuminuria[3-5]. For example, the Japan Diabetes Clinical Data Management Study Group (JDDM), which consists of general practitioners conducting research in the context of standard clinical practice, reported that the percentage of patients with diabetes and normoalbuminuric renal insufficiency (RI) is 12.3%[3]. Therefore, in patients with type 2 diabetes, diabetic nephropathy may have two clinical courses. Diabetic nephropathy can lead to a decreased GFR with or without macroalbuminuria. The clinical

Address for correspondence: Hisashi Makino, Division of Endocrinology and Metabolism, National Cerebral and Cardiovascular Center, 5-7-1 Fujishiro-dai, Suita, Osaka565-8565, Japan
E-mail: makinoh@hsp.ncvc.go.jp
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characteristics of normo- and microalbuminuric patients with a low GFR remain unclear.

Recent epidemiological studies have demonstrated that chronic kidney disease (CKD), defined as an eGFR of < 60 mL/min, is an independent risk factor for atherosclerotic diseases, such as coronary artery disease (CAD) and cerebral vascular disease (CVD)[6, 7]. Furthermore, it has been reported that a high resistance index of the interlobular arteries is associated with a low GFR[8]. These reports indicate that atherosclerosis is more closely associated with the progression of RI than albuminuria. To test this hypothesis, we investigated the correlation between the renal function and atherosclerotic markers (ankle-brachial index [ABI] and intima-media thickness [IMT]) in patients with or without albuminuria.

Dyslipidemia is an important risk factor for atherosclerotic disease, even in patients with CKD[9, 10]. Previous studies have demonstrated that dyslipidemia is also a risk factor for renal disease. The Atherosclerosis Risk Communities study demonstrated that a high level of triglycerides (TGs) and a low level of high-density lipoprotein cholesterol (HDL-C) are associated with an increased risk of developing renal insufficiency[11]. The Framingham Offspring Study also demonstrated that a low HDL-C level is an independent risk factor for incident CKD, defined as a GFR of < 60 mL/min/1.73 m^2 [12]. Therefore, we further investigated the associations between the lipid profiles and the renal function.

Methods

Study Subjects

A total of 513 consecutive patients with type 2 diabetes mellitus were hospitalized for glycemic control at the Department of Endocrinology and Metabolism of the National Cerebral and Cardiovascular Center between January 2005 and November 2009. Type 2 diabetes was diagnosed according to the Japanese Diabetes Society (JDS) criteria, i.e., a fasting blood glucose level of ≥ 126 mg/dL or a casual blood glucose level of ≥ 200 mg/dL, and usually not treated with insulin during the first year after diagnosis. Sixty-three patients with macroalbuminuria, defined as a UAER of ≥ 300 mg/g Cr, or who were receiving maintenance dialysis were excluded from the study. Finally, a cross-sectional study was performed with a population of 450 consecutive patients (304 men, mean age: 67.4 ± 9.5 years) with type 2 diabetes mellitus. This study was conducted with the approval of the National Cerebral and Cardiovascular Center Trust Ethics Committee.

Analytical Methods

The UAER was determined using one 24-hour urine collection during hospitalization. Blood samples were obtained after a 12-hour fast to measure the levels of fasting blood sugar (FBS), HbA1c, total cholesterol (TC), TG, HDL-C, ApoA1, ApoA2, ApoB, ApoC2, ApoC3, ApoE, Lp(a) and creatinine. The level of HbA1c (%) was estimated as the NGSP equivalent value (%), calculated as HbA1c (%) = HbA1c (JDS) (%) + 0.3% if HbA1c (JDS) < 5, + 0.4% if 5 ≤ HbA1c (JDS) < 10, or + 0.5% if 10 ≤ HbA1c (JDS), based on the relationship between the HbA1c (JDS) (%) measured according to the previous Japanese standard substance and measurement methods and the HbA1c (NGSP)[13]. The eGFR was calculated using the following equation established by the Modification of Diet in Renal Disease (MDRD) Study Group and adapted for Japanese individuals, as recently recommended by the Japanese Society of Nephrology:

$$eGFR\ (mL/min/1.73\ m^2) = 194 \times sCr^{-1.094} \times Age^{-0.287} \times 0.739\ (\text{if female})$$ [14].

The ABI was calculated using the higher of two systolic blood pressure (SBP) measurements obtained for each lower extremity divided by the higher of two SBP measurements for each upper extremity. The SBP was measured manually. The lowest ABI obtained for each patient was used in the subsequent statistical analyses.

The presence of CAD was defined as a physician's diagnosis of past myocardial infarction or angina pectoris or a history of coronary intervention. Stroke was defined as a brain infarction with or without symptoms, a transient ischemic attack or brain hemorrhage. The presence of PAD was defined as an ABI of ≤ 0.90 or > 1.30. Overweight was defined as a BMI of ≥ 25.0 kg/m^2. Smoking was categorized as never, past or current. Smokers who had quit smoking within the past five years were considered current smokers. Hypertension was defined as a SBP of > 140 mmHg or a diastolic blood pressure (DBP) of > 90 mmHg or both and/or the use of antihypertensive medications. Dyslipidemia was defined as a serum TC level of > 220 mg/dL, a TG level of > 150 mg/dL, an HDL-C level of < 40 mg/dL or the use of lipid-lowering agents. RI, i.e., a low eGFR, was defined as an eGFR of < 60 mL/min/1.73 m^2. Normoalbuminuria (NA) was defined as a UAER of < 30 mg/g Cr, and microalbuminuria (MA) was defined as a UAER of ≥ 30 mg/g Cr.

The study subjects were divided into four groups: NA (RI −) with a UAER < of 30 mg/g Cr and an eGFR of ≥ 60 mL/min/1.73 m^2; MA (RI −) with 30 ≤ UAER < 300 mg/g Cr and an eGFR of ≥ 60 mL/

Table 1. Baseline characteristics of the patients in each group

	RI (−)		RI (+)	
	NA	MA	NA	MA
n	219	83	86	62
Age (years)	65±10	69±8[d]	70±9	72±8[a, b, c]
Sex (Male/Female)	144/75	62/21	56/33	42/20
BMI (kg/m^2)	25.8±4.7	25.5±4.4	25.2±4.3	25.2±3.8
Duration of diabetes (years)	13±10	16±12[d]	15±10	21±11[a, b, c]
SBP (mmHg)	133±16	142±17[a, c]	129±17	141±20[a, c]
DBP (mmHg)	73±11	70±11	76±10	70±11
HbA1c (%)	9.1±1.8	9.5±1.7[e]	8.9±1.2	9.2±2.1
Fasting blood glucose (mg/dL)	161±67	178±71	165±72	153±65
eGFR (mL/min)	83.1±16.8	80.8±16.0	46.3±10.0[a, c]	43.2±10.7[a, c]
Log UAER	2.20±0.73	4.84±1.17[a, b]	2.24±0.75	4.38±0.65[a, b]
Log urinary β2 microglobulin	4.03±0.74	4.33±1.30[d]	4.04±1.25	5.15±1.80[a, b, c]
Retinopathy (Yes/No)	51/168 (23.3%)	37/46 (44.6%)[a]	30/56 (34.9%)[d]	28/54 (45.2%)[a]
Smoking (number)				
current	63	17	19	10
never	90	26	33	23
past	66	40	34	29
Dyslipidemia (Yes/No)	162/57 (74.0%)	61/22 (73.5%)	76/10 (88.4%)	57/5 (91.9%)
Hypertension (Yes/No)	160/59 (73.1%)	72/11 (86.7%)	71/15 (82.3%)	55/7 (88.8%)
Use of ACEI or ARB (Yes/No)	85/134 (38.8%)	45/38 (54.2%)[d]	49/37 (57.0%)[a]	44/18 (71.0%)[d, e, f]
Use of statin (Yes/No)	111/108 (50.7%)	39/44 (47.0%)	50/36 (58.1%)	36/26 (58.1%)
History of CAD (Yes/No)	85/134 (38.8%)	33/50 (39.8%)	44/42 (51.2%)[d]	30/32 (48.4%)
History of CVD (Yes/No)	41/178 (18.7%)	24/59 (28.9%)[d, f]	12/74 (14.0%)	19/43 (30.6%)[d, f]
History of PAD (Yes/No)	61/158 (27.9%)	27/56 (32.5%)	31/55 (36.0%)	27/35 (42.9%)[d]

The values are expressed as the mean ± SD. [a]$p<0.01$ vs. NA (RI−), [b]$p<0.05$ vs. NA (RI+), [c]$p<0.01$ vs. MA (RI−), [d]$p<0.05$ vs. NA (RI−), [e]$p<0.05$ vs. MA (RI−), and [f]$p<0.05$ vs. NA (RI+)

Abbreviations: renal insufficiency (RI), normoalbuminuria (NA), microalbuminuria (MA), estimated glomerular filtration rate (eGFR), urinary albumin excretion rate (UAER), angiotensin-converting enzyme inhibitor (ACEI), angiotensin receptor blocker (ARB), coronary artery disease (CAD), cerebral vascular disease (CVD), peripheral artery disease (PAD)

min/1.73 m^2; NA (RI+) with a UAER of <30 mg/g Cr and an eGFR of <60 mL/min/1.73 m^2; and MA (RI+) with 30 ≤ UAER <300 mg/g Cr and an eGFR of <60 mL/min/1.73 m^2.

Statistical Methods

The results are presented as the mean ± SD. Differences between groups were compared using the chi-squared test for categorical variables and unpaired Student's *t*-test for continuous variables. Two-tailed $p<0.05$ was considered to be statistically significant. A univariate analysis and multiple logistic regression analysis were performed to investigate the relationship between eGFR and ABI, age, sex, duration of diabetes, smoking, HbA1c, history of hypertension, history of dyslipidemia and angiotensin-converting enzyme inhibitor (ACEI) or angiotensin receptor blocker (ARB) use. Patients with an ABI of >1.30 were excluded from the statistical analysis.

Results

The NA (RI+) patients constituted 19.1% (86/450) of all patients (**Table 1**). The NA (RI+) patients had a higher prevalence of dyslipidemia than both the NA (RI−) and MA (RI−) patients. There were no significant differences in sex, HbA1c, SBP or smoking between these two groups. There were no significant differences in urinary β2 microglobulin (β2MG) excretion between the NA (RI+) and MA (RI−) patients, although the MA (RI+) patients had significantly higher levels of β2MG excretion than the patients in the other three groups (**Table 1**). There were no significant differences in the prevalence of retinopathy between the NA (RI+) and MA (RI−) patients (**Table 1**). The prevalence of CAD in the NA

Table 2. Lipid profiles of the patients in each group

	RI (−)		RI (+)	
	NA	MA	NA	MA
n	219	83	86	62
TC (mg/dL)	182±36[b]	183±35[b]	170±34	182±36[b]
Log triglycerides (mg/dL)	4.83±0.50	4.95±0.56	4.99±0.59[d]	5.07±0.58[a]
LDL-C (mg/dL)	112±31	109±30	99±25[a]	107±31
HDL-C (mg/dL)	45±13	47±14	41±11[a, c]	40±12[a, c]
Log Lp(a) (mg/dL)	3.06±0.58	3.06±0.60	3.18±0.61	3.23±0.60[d]
ApoA1 (mg/dL)	126±24	128±22	121±19[d]	118±26[d, e]
ApoA2 (mg/dL)	24.8±4.9	23.8±3.9	23.3±4.3[d]	23.7±7.8
ApoB (mg/dL)	93±23	88±21	89±21	92±24
ApoC2 (mg/dL)	4.6±2.1	4.3±1.7	4.8±2.1	5.3±3.2[d, e]
ApoC3 (mg/dL)	9.4±4.1	9.0±3.2	11.4±5.5[a, c]	10.8±4.5[d, e]
ApoE (mg/dL)	3.4±1.2	3.5±1.0	3.5±1.5	3.7±1.5
ApoB/LDL-C	0.83±0.13	0.84±0.19	0.91±0.21[a, e]	0.88±0.19

The values are expressed as the mean ± SD. [a]$p<0.01$ vs. NA (RI−), [b]$p<0.05$ vs. NA (RI+), [c]$p<0.01$ vs. MA (RI−), [d]$p<0.05$ vs. NA (RI−), and [e]$p<0.05$ vs. MA (RI−).
Abbreviations: renal insufficiency (RI), normoalbuminuria (NA), microalbuminuria (MA)

(RI+) patients was significantly higher than that observed in the NA (RI−) patients (**Table 1**). On the other hand, the prevalence of CVD in the MA (RI−) and MA (RI+) patients was higher than that observed in the NA (RI−) and NA (RI+) patients (**Table 1**). There were no significant differences in the frequency of ACEI and ARB use between the NA (RI+) and MA (RI−) patients. The MA (RI+) patients had a higher frequency of ACEI and ARB use than the patients in the other three groups.

The levels of TGs in the NA (RI+) and MA (RI+) patients were significantly higher than those observed in the NA (RI−) and MA (RI−) patients (**Table 2**). The levels of HDL-C in the NA (RI+) and MA (RI+) patients were significantly lower than those observed in the NA (RI−) and MA (RI−) patients (**Table 2**). The NA (RI+) and MA (RI+) patients had significantly higher ApoB/LDL-C ratios than the NA (RI−) and MA (RI−) patients (**Table 2**). The levels of Lp(a) in the MA (RI+) patients were significantly higher than those observed in the NA (RI−) patients (**Table 2**). The patients with RI had significantly higher ApoC3 levels than the patients without RI, regardless of the presence of albuminuria (**Table 2**).

The NA (RI+) and MA (RI+) patients had significantly lower ABI values than the NA (RI−) and MA (RI−) patients (**Fig. 1A**). On the other hand, the MA (RI−) and MA (RI+) patients had significantly higher IMT values than the NA (RI−) patients, whereas there were no significant differences in IMT between the NA (RI−) and NA (RI+) patients (**Fig. 1B**).

In the univariate analysis, the ABI was found to be significantly associated with the eGFR, whereas the UAER was not associated with the ABI (**Fig. 2**). A multiple regression analysis showed that the ABI was significantly associated with the eGFR, independent of age, sex, duration of diabetes, smoking, HbA1c, history of hypertension and history of dyslipidemia, whereas the UAER was not associated with the ABI (**Table 3**). Furthermore, there was a significant association between the ABI and the eGFR in a multivariate analysis that included ACEI and ARB use, in which the GFR decreased concomitantly with the UAER (**Table 3**).

Discussion

Evidence has been accumulating that CKD, which presents as renal dysfunction, persistent albuminuria or both, is a strong risk factor for cardiovascular diseases, such as CAD, PAD and CVD. However, it remains unclear how significantly the type of renal dysfunction, that is albuminuria or RI, contributes to atherosclerotic diseases. In the present study of diabetic patients, we demonstrated that RI is significantly associated with ABI but not IMT, whereas albuminuria is significantly associated with IMT but not ABI.

The clinical course of typical diabetic nephropathy includes the development of microalbuminuria

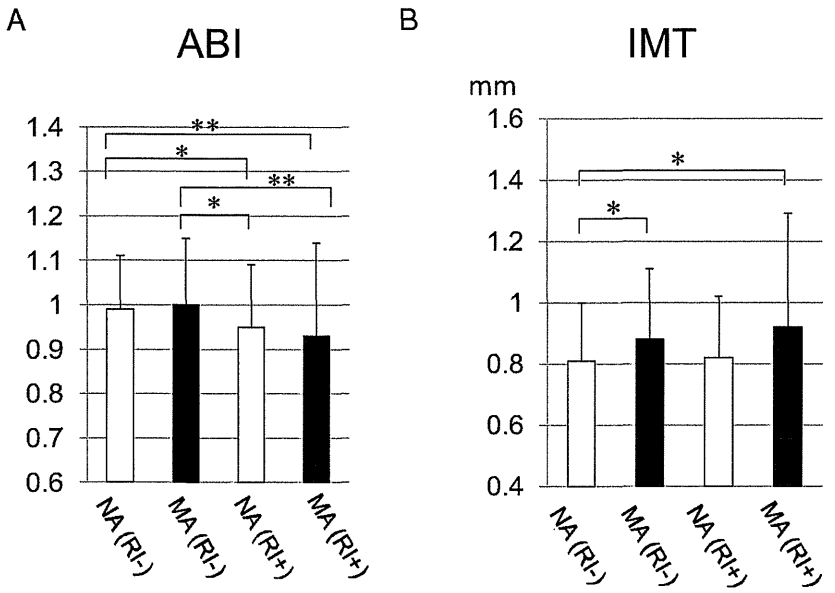

Fig. 1. Ankle-brachial index (ABI) and intima-media thickness (IMT). A: Comparison of ABI among the NA (RI−), MA (RI−), NA (RI+) and MA (RI+) patients. B: Comparison of IMT among the NA (RI−), MA (RI−), NA (RI+) and MA (RI+) patients. NA, normoalbuminuria; MA, microalbuminuria; RI, renal insufficiency. The data are expressed as the mean ± SD. $^{*}p<0.05$, $^{**}p<0.01$

progressing to overt proteinuria and a decline in GFR. However, several reports have demonstrated that RI occurs in some patients with normoalbuminuria[3]. The histological characteristics of diabetic nephropathy include mesangial matrix accumulation, podocyte loss and glomerular basement membrane thickening. These glomerular injuries cause albuminuria. However, the diabetic kidney also experiences tubulointerstitial injury and vascular changes in addition to glomerular injury. Interstitial fibrosis, which is common even during the early stages of diabetic nephropathy with mild glomerular changes[15], leads to a decline in GFR[16]. Furthermore, since the arteries in the diabetic kidney invariably exhibit typical atherosclerotic changes[17], the diabetic kidney is at risk of ischemic damage. Indeed, in a study of 393 renal biopsy samples obtained from patients with type 2 diabetes[18], 15% primarily demonstrated ischemic changes. Therefore, it has also been suggested that interstitial fibrosis and ischemic vascular disease contribute to renal injury in patients with type 2 diabetes, especially those with RI without albuminuria. In this study, a low ABI, reflecting atherosclerosis of the peripheral arteries, was associated with RI. An epidemiological study also demonstrated that the GFR is associated with large artery elasticity[19]. MacIsaac *et al.* reported that a

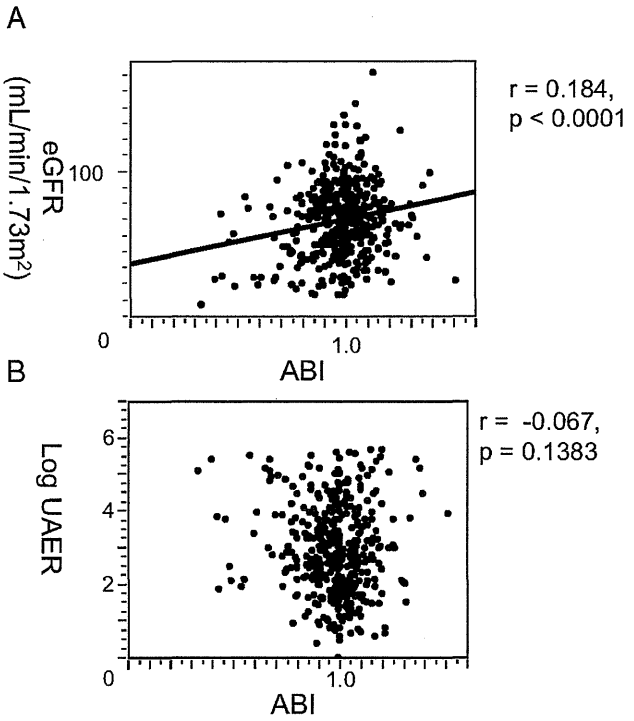

Fig. 2. A: Correlation between ABI and eGFR. B: Correlation between ABI and UAER according to the univariate analysis.

Table 3. Correlations between ABI, eGFR and log UAER in the multivariate analysis

	Model 1		Model 2	
	β	p	β	p
eGFR	0.148	0.0023	0.138	0.0042
Log UAER	−0.027	0.5946	−0.018	0.7154

Model 1 is adjusted for age, sex, duration of diabetes, the smoking status, HbA1c, history of hypertension and history of dyslipidemia. Model 2 is adjusted for the same variables as Model 1 in addition to angiotensin-converting enzyme inhibitor or angiotensin receptor blocker use.
Abbreviations: ankle-brachial index (ABI), estimated glomerular filtration rate (eGFR), urinary albumin excretion rate (UAER)

high resistance index of the interlobular arteries is associated with a low GFR[8], thus suggesting that a decline in eGFR may be caused by atherosclerosis of the intrarenal arteries. These results as well as the present findings indicate the possibility that patients with peripheral artery atherosclerosis may also have intrarenal atherosclerosis that causes RI without albuminuria.

Since the IMT reflects early atherosclerotic changes, an increased IMT is expected to be associated with both albuminuria and low-grade RI. Indeed, in this study, the IMT values of the patients with albuminuria were significantly higher than those of the NA (RI−) patients. However, there were no significant differences in IMT between the NA (RI+) and NA (RI−) patients, even though the NA (RI+) patients had lower ABI values. The Dong-gu study showed that a low eGFR, but not IMT, is associated with carotid plaque[20]. That report also demonstrated that a low eGFR is associated with PAD[20]. Taken together, these findings suggest that the mechanism of atherosclerosis observed in NA (RI+) patients is similar to that of plaque formation and a lower ABI, rather than intima-media thickening.

In this study, the UAER was not significantly associated with the ABI. Previous reports indicate that the UAER is closely associated with early atherosclerotic markers, such as endothelial dysfunction and IMT[20, 21]. On the other hand, decreases in ABI may reflect relatively advanced atherosclerosis since a decreased ABI indicates the presence of peripheral artery stenosis.

Our findings demonstrate albuminuria to be significantly associated with cerebrovascular disease. Ito *et al.* proposed that albuminuria may be an early marker of strain vessel injury. The arterioles in the kidneys are relatively similar to the perforating arteries in the brain[22]. Indeed, it has been reported that albuminuria is associated with decreased small artery elasticity[19].

In this study, RI, but not albuminuria, was found to be significantly associated with lipid metabolism abnormalities. Compared to the patients without RI, those with RI had high ApoB/LDL-C ratios, indicating the existence of small, dense LDL-C particles[23]. This form of LDL-C is strongly atherogenic. Indeed, a clinical study demonstrated that small, dense LDL-C is an independent risk factor for cardiovascular disease[24]. ApoB has also been reported to be a predictor of renal failure progression[25], suggesting that small, dense LDL-C is involved in the progression of renal injury as well as cardiovascular disease. We also demonstrated high Apo C3 levels in the patients with RI compared to those observed in the patients without RI. Since ApoC3 is a well-known endogenous lipoprotein lipase (LPL) inhibitor[26], our data suggest that the LPL activity, which contributes to increases in small, dense LDL-C, may be lower in patients with diabetes and RI. ApoC3 has also been reported to directly activate vascular endothelial cells via the activation of protein kinase C β[27]. Therefore, an elevation of the ApoC3 level is considered to play a role in the progression of atherosclerosis. It has also been suggested that a high level of ApoC3 is involved in renal injury via atherosclerosis of the intrarenal arteries. Moreover, several clinical trials have demonstrated that statin therapy can prevent declines in GFR[25, 28, 29]. These reports indicate that amelioration of dyslipidemia contributes to the prevention of renal function deterioration. Altogether, atherogenic dyslipidemia may be associated with a declining GFR as well as cardiovascular disease progression in patients with diabetes.

Another possible mechanism underlying the association between RI and dyslipidemia is the direct alteration of lipid metabolism by RI. Renal failure impacts lipid metabolism via impaired carbohydrate tolerance as well as a decreased LPL activity due to endogenous LPL inhibition involving pre-β-HDL accumulation and insulin resistance[30]. In this study, however, the contribution of RI to abnormalities in