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to be correlated with γ-GTP levels. However, the analysis restricted to never-drinkers remained unaltered suggesting that the confounding effect might be weak. Finally, since changes in serum γ-GTP levels could occur over time, a single measurement of γ-GTP levels at baseline may have led to nondifferential misclassification of γ-GTP categories among participants. Thus, real associations may have been stronger.

Conclusion

In conclusion, serum γ-GTP levels were positively associated with CVD and stroke mortality independently of alcohol drinking status in both men and women and with CHD mortality in women in the present individual participant data meta-analysis of Japanese cohorts. Whether measurement of serum γ-GTP levels would help predict future mortality from CVD or understanding pathophysiological mechanisms for the association requires further investigations.

Conflicts of Interest

The authors declare no conflicts of interest.

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Appendix

The Evidence for Cardiovascular Prevention from Observational Cohorts in Japan (EPOCH–JAPAN) Research Group is composed of the following investigators. Chairperson: Hirotsugu Ueshima (Shiga University of Medical Science); Co–Chairperson: Tomonori Okamura (Keio University); Executive committee: Hirotsugu Ueshima (Shiga University of Medical Science), Yutaka Imai (Tohoku University Graduate School of Pharmaceutical Sciences), Takayoshi Ohkubo (Teikyo University School of Medicine), Fujiko Irie (Ibaraki Prefecture), Hiroyasu Iso, Akihiko Kitamura (Osaka University Graduate School of Medicine), Yutaka Kiyohara (Kyushu University Graduate School of Medicine), Katsuyuki Miura (Shiga University of Medical Science), Yoshitaka Murakami (Toho University), Hideaki Nakagawa (Kanazawa Medical University), Takeo Nakayama (Kyoto University School of Public Health), Akira Okayama (Research Institute of Strategy for Prevention), Toshimi Sairenchi (Dokkyo Medical University), Shigeyuki Saitoh (Sapporo Medical University), Kiyomi Sakata (Iwate Medical University), Akiko Tamakoshi (Hokkaido University Graduate School of Medicine), Ichiro Tsuji (Tohoku University Graduate School of Medicine), Michiko Yamada (Radiation Effects Research Foundation), Masahiko Kiyama (Osaka Center for Cancer and Cardiovascular Disease Prevention), Yoshihiro Miyamoto (National Cerebral and Cardiovascular Center), Shizukiyo Ishikawa (Jichi Medical University), Hiroshi Yatsuya (Fujita Health University) and Tomonori Okamura (Keio University School of Medicine)

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Original Article

Serum Uric Acid and Mortality Form Cardiovascular Disease: EPOCH-JAPAN Study

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Aim: To investigate the relationship between serum uric acid levels and cardiovascular disease in Asians.
Methods: We examined the above relationship using the data of Evidence for Cardiovascular Prevention from Observational Cohorts in Japan (EPOCH-JAPAN Study). The data of 36,313 subjects (15,628 men and 20,685 women aged 35−89 years without histories of stroke, coronary heart disease, or cancer at baseline) were used for the analyses.
Sex-specific hazard ratios (HRs) of mortality from cardiovascular disease were estimated according to the quintiles of serum uric acid using Cox hazard models stratified by cohorts.
Results: During 441,771 person-years of follow-up, we documented 1,288 cardiovascular deaths. A J- or U-shaped relationship between serum uric acid level and cardiovascular disease mortality was observed. Compared with the lowest quintile of serum uric acid levels, the highest quintile was associated with an increased cardiovascular disease mortality in men [HR: 1.28; 95% confidence interval (CI): 1.01−1.63] and women (HR: 1.51; 95% CI: 1.14−1.99). However, there was no significant association with mortality from stroke, coronary heart disease or heart failure in both men and women.
Conclusion: This large pooled analysis in Japan suggested a J- or U-shaped relationship between serum uric acid levels and cardiovascular mortality. The highest quintile of serum uric acid levels was associated with increased cardiovascular disease mortality in both Japanese men and women.

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Key words: Serum uric acid, Stroke, Cardiovascular disease, Mortality

Introduction

Although high serum uric acid level has been associated with an increased risk of cardiovascular disease[1], there are some findings suggesting its protective effect on oxidative stress. The loss of urate oxidase activity leading to high serum uric acid levels has been hypothesized to protect the body from oxidative damage and the prolonged lifespan of hominoids[2, 3]. In the last decades, a number of epidemiologic studies showed conflicting results[4-15].

The Framingham Study was the first to show an independent association between serum uric acid levels and risk of cardiovascular outcomes in the general population under the careful measurement of known cardiovascular risk factors in a 23-year follow-up period[4]. However, most of the participants in that study were whites, and we do not know whether the results might apply to non-white populations. A 14-year follow-up study of 8,172 Japanese men and women showed that uric acid levels were not associ-

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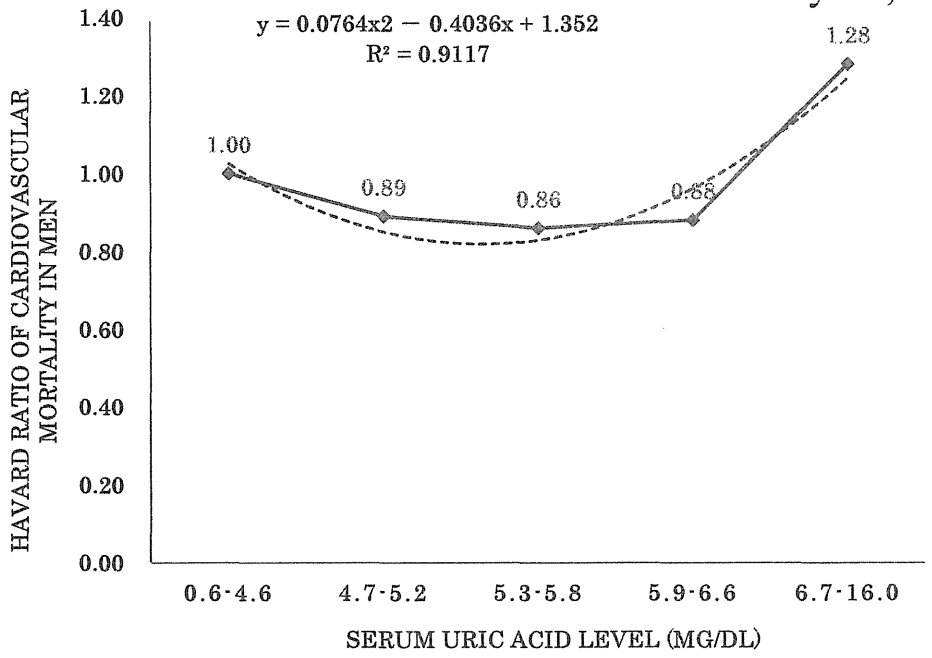

Fig. 1. Fitting curve of association between serum uric acid and cardiovascular mortality in men.

ated with mortality from cardiovascular disease or stroke after adjustment for known cardiovascular risk[10]. Another 8-year follow-up study of 90,393 Taiwanese men and women indicated that hyperuricemia was an independent risk factor of mortality from cardiovascular disease[11]. Two recent cross-sectional surveys showed that serum uric acid level was significantly associated with various metabolic indicators and elevated carotid intima-media thickness in the Asian Mongolian as well as middle-aged and elderly Chinese subjects, respectively[12, 13]. Serum uric acid levels predict the incidence of coronary heart disease but not stroke among atomic bomb survivors in Nagasaki or is a risk factor for ultrasonographically determined carotid arterial intima-media thickness in Japanese elderly persons (≥74 years)[14, 15].

Accordingly, a large-scale prospective study on the association between serum uric acid levels and risk of mortality from cardiovascular disease in Asian adults was still required. For this purpose, the present study has been conducted.

Study Population

The Evidence for Cardiovascular Prevention from Observation Cohorts in Japan (EPOCH-JAPAN) is the pooling project of a number of well-qualified cohort studies, which investigated the relationship between health examination measures (laboratory measures and lifestyle factors) and mortality in the Japanese population. The EPOCH-JAPAN comprises 13 cohort studies in Japan with an average of 10-year follow-up periods. The year range of baseline survey in the cohort was between 1977 and 1995. The details of these projects have been previously described[16-23]. A total of 90,528, of which the endpoint was death owing to cardiovascular disease were included in the study. Serum uric acid was measured using a colorimetric phosphotungstic acid procedure.

Subjects were excluded if they reported a history of stroke, coronary heart disease, or cancer (n=4,144) at baseline or if they were unable to provide data for serum uric acid levels; in addition, those who were older than 90 years or younger than 35 years (total: n=50,071) were also excluded. Data from the remaining 36,313 subjects (15,628 men and 20,685 women) were used for the analyses.

Endpoints

The primary end points for this analysis were deaths from cardiovascular disease (CVD) (the International Classification of Disease, 9th revision, codes 390−459 and 10th revision, codes I01−I99), which

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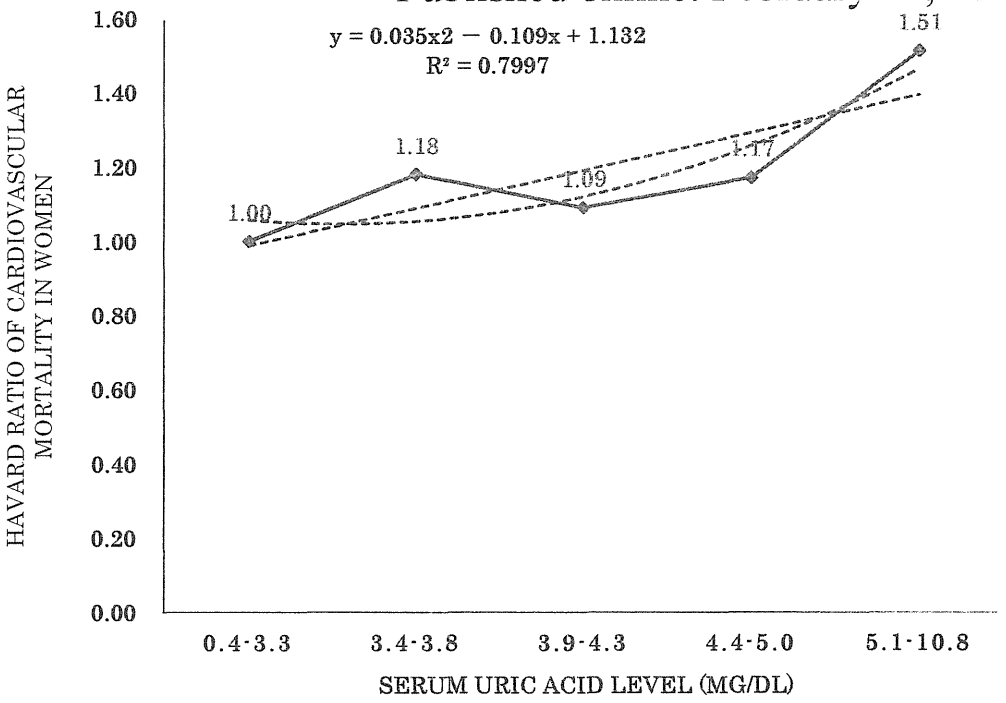

Fig. 2. Fitting curve of association between serum uric acid and cardiovascular mortality in women.

was further divided into hemorrhagic stroke (430–431 and I60–I61) and ischemic stroke (433–434 and I63) as well as coronary heart disease (CHD, 410–414 and I20–I25) and heart failure (428 and I50).

Statistical Methods

The data of serum uric acid were divided into sex-specified quartiles. The median level of serum uric acid of each quintile was 4.0 mg/dl, 4.9 mg/dl, 5.5 mg/dl, 6.2 mg/dl, and 7.3 mg/dl for men and 3.0 mg/dl, 3.6 mg/dl, 4.1 mg/dl, 4.7 mg/dl, and 5.7 mg/dl for women. Hazard ratios (HRs) for mortalities were estimated in both men and women by Cox hazard models, which were stratified by cohorts. The adjustment variables included age (continuous), smoking status (never, past, 1–20/day, or ≥21/day), drinking status (drinkers, ex-drinkers, or never-drinkers), body mass index (quartiles), triglycerides (quartiles), total cholesterol (quartiles), and systolic blood pressure (continuous).

Two fitting curves of association between serum uric acid and cardiovascular mortality in both sexes were plotted to clearly demonstrate the results (**Fig. 1** & **2**).

All statistical analyses for two-tailed tests were conducted using SAS version 9.13 (SAS Institute Inc., Cary). *P* values of <0.05 were regarded as statistically significant.

Results

During 441,771 person-years of follow-up, we documented 1,288 deaths from CVD (649 in men and 639 in women) including 301 total strokes, 131 coronary heart diseases, and 116 heart failures in men as well as 293 total strokes and 136 heart failures in women.

Table 1 shows the age-adjusted mean values and prevalence of cardiovascular risk factors at baseline according to the quintiles of serum uric acid levels. Men with higher uric acid were younger, whereas women with higher uric acid levels were older. Compared with men and women in the lowest quintile of uric acid levels, those in the higher quintiles were likely to be overweight and to have higher levels of total cholesterol, systolic and diastolic blood pressures, and triglycerides. Men with higher uric acid levels drank less but the opposite trend was observed in women. In addition, uric acid levels were inversely associated with high density cholesterol levels for both men and women.

Table 2 shows sex-specified HRs of stroke, coronary heart disease, heart failure, and total CVD

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Table 1. Baseline characteristics according to quintiles of serum uric acid levels.

	Quintile of serum uric acid levels					*P* for trend
	1 (low)	2	3	4	5 (high)	
Men						
No. at risk	3042	3353	2939	3052	3242	
Median uric acid (mg/dl)	4.0	4.9	5.5	6.2	7.3	
Range of uric acid (mg/dl)	0.6-4.6	4.7-5.2	5.3-5.8	5.9-6.6	6.7-16.0	
Mean age (years)	54.6	53.3	52.6	52.0	52.9	<0.001
Mean body mass index (kg/m^2)	21.9	22.2	22.7	23.1	23.8	<0.001
Mean total cholestrerol (mg/dl)	187.8	189.8	192.8	196.4	201.2	<0.001
Mean HDL cholesterol (mg/dl)	53.3	52.7	51.3	50.5	50.0	<0.001
Mean systolic blood pressure (mmHg)	131.0	131.2	132.1	133.3	137.2	<0.001
Mean diastolic blood pressure (mmHg)	79.1	79.7	80.8	81.6	84.4	<0.001
Median triglycerides (mg/dl)	93.0	99.0	108.0	115.0	135.0	<0.001
Current smokers (%)	63.8	68.7	70.6	72.6	74.6	<0.001
Current drinkers (%)	59.2	58.8	56.2	54.8	50.6	<0.001
Women						
No. at risk	4388	3933	4386	3628	4350	
Median uric acid (mg/dl)	3.0	3.6	4.1	4.7	5.7	
Range of Uric acid (mg/dl)	0.4-3.3	3.4-3.8	3.9-4.3	4.4-5.0	5.1-10.8	
Mean age (years)	51.3	52.3	53.3	55.1	58.2	<0.001
Mean body mass index (kg/m^2)	22.1	22.5	22.8	23.3	24.2	<0.001
Mean total cholestrerol (mg/dl)	193.9	199.5	202.5	208.5	214.2	<0.001
Mean HDL cholesterol (mg/dl)	58.2	57.1	56.7	55.9	51.6	<0.001
Mean systolic blood pressure (mmHg)	126.9	128.4	129.5	133.2	138.3	<0.001
Mean diastolic blood pressure (mmHg)	75.8	76.9	77.7	79.5	81.6	<0.001
Median triglycerides (mg/dl)	82.0	87.0	92.0	104.0	120.0	<0.001
Current smokers (%)	5.2	5.4	6.1	7.4	8.2	<0.001
Current drinkers (%)	13.0	15.0	16.0	18.2	16.2	<0.001

according to the quintiles of serum uric acid levels. In multivariable models, we observe a J- or U-shaped relationship between serum uric acid level and total or cause-specific cardiovascular mortality. We did not observe a linear increase in mortality from almost all causes of death associated with the increase in serum uric acid levels. In most causes of death, the 5th quintile of uric acid showed highest mortality in both men and women; however, few of them reached to statistical significance. For both men and women, the highest uric acid levels (=>6.7 mg/dl for men and =>5.1 mg/dl for women) were associated with increased mortality owing to total CVD; we also observed statistical significance for trend test in the association between uric acid quintile and total cardiovascular mortality and in the relationship between uric acid quintile and mortality because of stroke in women.

To check whether there was any reverse causal bias, a sub-analysis was conducted to estimate the associations censoring the first 3 years. The results were shown in **Supplementary Table** and **Supplementary Figs**. In multivariable models, a J- or U-shaped relationship between serum uric acid level and total or cause-specific cardiovascular mortality was also observed. The whole results were quite similar to the ones shown in **Table 2**. Therefore, we considered that there was no or few reverse causal bias.

Discussion

In this large pooled analysis of Japanese cohort studies with a median follow-up of 10 years, we found a J- or U-shaped relationship between serum uric acid levels and cardiovascular mortality. Compared with the lowest quintile of serum uric acid levels, the highest quintile was associated with an increased cardiovascular mortality in both men and women; however, none of the cause-specific death was significantly associated with serum uric acid levels.

Elevated serum uric acid has been recognized as

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Table 2. Sex-specific hazard ratios (95% CI) of mortality from stroke, coronary heart disease, heart failure and total cardiovascular diseases according to quintiles of serum uric acid levels

	Quintiles of serum uric acid levels					*P* for trend
	1 (low)	2	3	4	5 (high)	
Men						
No. at risk	3042	3353	2939	3052	3242	
Person Year	34762	40456	34398	38145	40323	
Total stroke						
No. of death	63	59	46	48	85	
Age-adjusted HR	1.00	0.81 (0.56-1.15)	0.76 (0.52-1.12)	0.75 (0.52-1.10)	1.14 (0.82-1.59)	0.291
Multivariable HR†	1.00	0.83 (0.58-1.18)	0.77 (0.52-1.13)	0.77 (0.52-1.13)	1.19 (0.84-1.68)	0.258
Ischemic stroke						
No. of death	35	34	26	31	47	
Age-adjusted HR	1.00	0.83 (0.52-1.34)	0.76 (0.45-1.26)	0.90 (0.55-1.47)	1.19 (0.76-1.85)	0.286
Multivariable HR†	1.00	0.87 (0.54-1.40)	0.75 (0.45-1.26)	0.91 (0.55-1.50)	1.19 (0.75-1.90)	0.350
Hemorrhagic stroke						
No. of death	17	18	16	15	29	
Age-adjusted HR	1.00	0.89 (0.46-1.73)	1.04 (0.52-2.06)	0.81 (0.41-1.64)	1.37 (0.75-2.51)	0.248
Multivariable HR†	1.00	0.90 (0.46-1.77)	1.07 (0.54-2.14)	0.83 (0.41-1.68)	1.41 (0.75-2.65)	0.252
Coronary heart disease						
No. of death	24	27	18	27	35	
Age-adjusted HR	1.00	0.99 (0.57-1.72)	0.83 (0.45-1.53)	1.15 (0.66-2.00)	1.29 (0.76-2.18)	0.235
Multivariable HR†	1.00	0.98 (0.57-1.71)	0.75 (0.40-1.39)	1.02 (0.58-1.79)	1.12 (0.65-1.93)	0.600
Heart failure						
No. of death	19	23	24	18	32	
Age-adjusted HR	1.00	1.06 (0.57-1.95)	1.38 (0.76-2.54)	0.94 (0.49-1.80)	1.45 (0.82-2.58)	0.229
Multivariable HR†	1.00	1.09 (0.59-2.03)	1.46 (0.79-2.69)	1.05 (0.54-2.03)	1.76 (0.97-3.18)	0.066
Total cardiovascular disease						
No. of death	126	127	102	111	183	
Age-adjusted HR	1.00	0.87 (0.68-1.19)	0.86 (0.66-1.20)	0.87 (0.67-1.12)	1.24 (0.98-1.55)	0.028
Multivariable HR†	1.00	0.89 (0.70-1.14)	0.86 (0.66-1.12)	0.88 (0.68-1.14)	1.28 (1.01-1.63)	0.022

an independent risk factor for heart failure[13, 24, 25]. In the Framingham Offspring Study, a longitudinal observational study of 4,912 children from the original Framingham cohort participants and their spouses showed that the serum uric acid levels were associated with the incidence of heart failure. The hazard ratio (95% CI) for the highest versus lowest quintiles of serum uric acid (>6.3 mg/dl and <3.4 mg/dl, respectively) was 2.10 (1.04–4.22) after adjusting for sex, age, smoking, body mass index, renal dysfunction, diuretics, systolic blood pressure, valvular heart disease, diabetes, alcohol, and use of antihypertension medications[24]. Although the risk ratio did not reach statistical significance level, the magnitude of hazard ratio for both men and women in the highest uric acid quintile in the present study was almost similar to that of the Framingham Offspring Study. The MJ Health Screening Cohort conducted in Taiwan[13] failed to show significant positive associations between serum uric acid levels and mortality from heart failure in men and women but showed a weak positive association for both sexes combined; the multivariable hazard ratio (95% CI) for serum uric acid level of >7.0 mg/dl compared with that of ≤7.0 mg/dl was 1.13 (1.07–1.19); *p* value for trend was <0.01[13].

The associations between hyperuricemia and risk of stroke have been investigated in several studies, but the results also have been controversial[8-14]. The NIPPON DATA 80 performed in 1980 at baseline was the first cohort study to examine the association between hyperuricemia and deaths because of CVDs in a representative sample of Japanese adults[10]. In that study, uric acid levels were not significantly associated with mortality because of CVD or stroke in either age-adjusted or multivariable models, probably because of the limited number of deaths[10]. The present large-

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(Cont Table 2)

	Quintiles of serum uric acid levels					*P* for trend
	1 (low)	2	3	4	5 (high)	
Women						
No. at risk	4388	3933	4386	3628	4350	
Person Year	51098	46069	55526	44685	56309	
Total stroke						
No. of death	33	42	45	51	122	
Age-adjusted HR	1.00	1.22 (0.77-1.93)	0.93 (0.59-1.46)	1.07 (0.69-1.67)	1.45 (0.98-2.15)	0.024
Multivariable HR†	1.00	1.27 (0.90-2.01)	0.98 (0.62-1.54)	1.05 (0.67-1.64)	1.46 (0.98-2.19)	0.036
Ischemic stroke						
No. of death	15	23	18	30	57	
Age-adjusted HR	1.00	1.38 (0.71-2.63)	0.77 (0.39-1.54)	1.22 (0.65-2.29)	1.33 (0.75-2.37)	0.285
Multivariable HR†	1.00	1.42 (0.74-2.74)	0.80 (0.40-1.61)	1.22 (0.65-2.30)	1.35 (0.75-2.44)	0.314
Hemorrhagic stroke						
No. of death	11	14	20	14	38	
Age-adjusted HR	1.00	1.41 (0.64-3.11)	1.32 (0.63-2.76)	1.12 (0.51-2.50)	1.55 (0.78-3.07)	0.269
Multivariable HR†	1.00	1.41 (0.64-3.13)	1.33 (0.63-2.80)	1.09 (0.48-2.43)	1.54 (0.76-3.10)	0.301
Coronary heart disease						
No. of death	10	13	18	23	50	
Age-adjusted HR	1.00	1.28 (0.56-2.93)	1.15 (0.53-2.50)	1.60 (0.75-3.38)	1.83 (0.92-3.64)	0.032
Multivariable HR†	1.00	1.29 (0.56-2.96)	1.20 (0.55-2.61)	1.49 (0.70-3.18)	1.75 (0.87-3.54)	0.067
Heart failure						
No. of death	18	13	23	26	56	
Age-adjusted HR	1.00	0.64 (0.31-1.31)	0.78 (0.42-1.46)	0.86 (0.47-1.57)	1.09 (0.63-1.88)	0.200
Multivariable HR†	1.00	0.69 (0.34-1.41)	0.84 (0.45-1.56)	0.96 (0.52-1.77)	1.29 (0.74-2.25)	0.071
Total cardiovascular diseases						
No. of death	68	82	106	119	264	
Age-adjusted HR	1.00	1.15 (0.83-1.58)	1.04 (0.76-1.41)	1.18 (0.87-1.59)	1.46 (1.11-1.91)	<0.001
Multivariable HR†	1.00	1.18 (0.86-1.64)	1.09 (0.80-1.48)	1.17 (0.86-1.58)	1.51 (1.14-1.99)	<0.001

† Adjusted further for body mass index, smoking status, ethanol intake, systolic blood pressure and total cholesterol.

cohort study showed no significant association between serum uric acid levels and stroke mortality, although increased risk with borderline statistical significance was observed in the highest quintile of serum uric acid levels in women. MJ Health Screening Cohort reported increased mortality from ischemic stroke with higher serum uric acid levels in women only[13]. A meta-analysis comprising 16 cohort studies and 238,449 adults showed that persons with hyperuricemia had higher age-adjusted risk for stroke: stroke incidence (6 studies; risk ratio, 1.41; 95% CI: 1.05−1.76) and mortality (6 studies; risk ratio 1.36, 95% CI: 1.03−1.69)[26]. The subgroup analysis of this study adjusting for known risk of factors including age, hypertension, diabetes mellitus, and cholesterol still showed an increased risk of stroke in persons with hyperuricemia. However, the abovementioned study was a traditional meta-analysis based on bibliographic information, which is not a pooled analysis based on individual data.

The present study showed an excess risk with borderline statistical significance for mortality because of coronary heart disease in women, but not in men. The NIPPON DATA 80[10] and the MJ Health Screening Cohort[13] showed no significant association between serum uric acid levels and mortality from coronary heart disease in either sex. A recent meta-analysis including 26 studies and 402,997 adults showed that hyperuricemia was associated with increased risk of coronary heart disease: an adjusted risk ratio for incidence (95% CI) of 1.09 (1.01−1.16) and an adjusted risk ratio for mortality (95% CI) of 1.16 (1.01−1.30)[27]. However, the sex-specific analysis also showed a significant association of hyperuricemia with the incidence of or mortality from coronary heart disease in women, but not in men. Furthermore,

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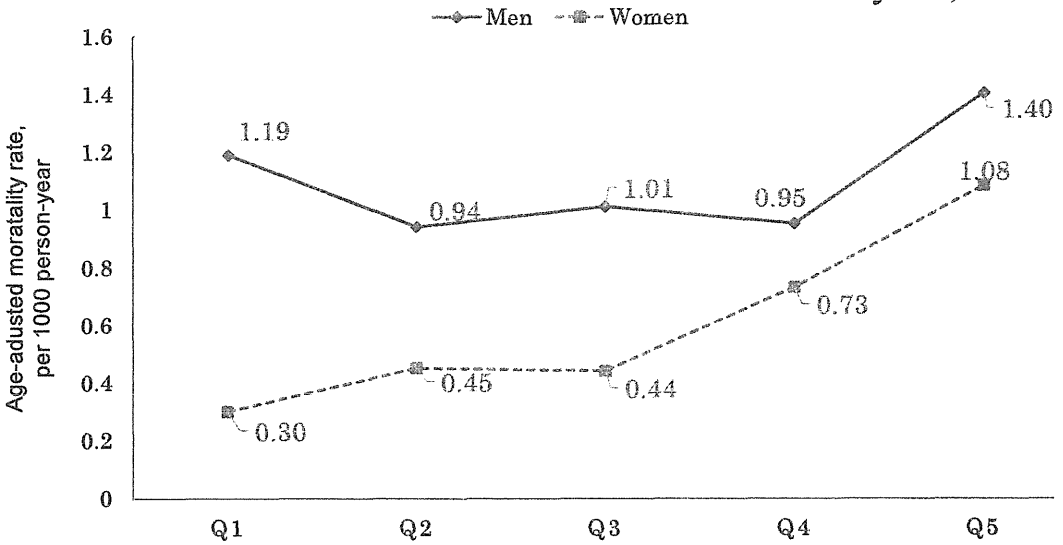

Fig. 3. Sex-specific age –adjusted cardiovascular disease (CVD) mortality rates according to quintiles of serum uric acid levels.

their study is not a pooled analysis based on individual data.

In the present study, the overall mortality rate from CVD in women was lower than that in men, particularly at low serum uric acid levels (**Fig. 3**). Therefore, the hazard ratio for the highest versus lowest quintiles of serum uric acid was higher in women and was more statistically significant than that in men. Several previous studies also showed that elevated serum uric acid levels in women were associated with a higher cardiovascular hazard ratio than that in men[9, 13, 28]. This may be because of estrogen that probably plays a cardioprotective role in women[8, 13, 29], whereas hyperuricemia in women could possibly be a hallmark of escape from estrogen protection[13]. In addition, another possible reason could be that both the mean age level and the mean glucose level were higher in the higher uric acid level (data not shown) groups of women than of men. Uric acid is also associated with diabetes or glucose intolerance and is a risk factor that confers greater relative risk for CVD in women[8, 30, 31].

Hyperuricemia induces endothelial dysfunction[32], which may stimulate glucose assimilation in oxidative process in adipocytes[33]. In a population-based study of 783 men, hyperuricemia was associated with increased renal tubular sodium re-absorption, which may provide a link with hyperinsulinemia and hypertension[34]. In a recent randomized, placebo-controlled, crossover trial of hyperuricemia involving individuals with newly diagnosed hypertension, compared with placebo ($n=15$), casual and 24-hour ambulatory blood pressure levels was reduced to a greater extent with allopurinol treatment ($n=15$)[35]. On the other hand, uric acid has been reported to be an antioxidant that may prevent stress-induced cell transformation and oxidant-induced cardiac and renal toxicity[36]. An *in-vivo* study showed that uric acid protected cultured rat hippocampal neurons against cell death induced by glutamate and NaCN insults, which are relevant to the pathogenesis of cerebral ischemia[37]. A clinical study also supported a potential neuroprotective role of the exogenous uric acid administration in stoke patients treated by thrombolysis[38]. These studies suggested that the effect of hyperuricemia on CVDs was strongly associated with other cardiovascular risk factors such as hypertension and insulin resistance. These findings may explain J- or U-shaped relationship between serum uric acid levels and cardiovascular mortality in the present study. In other words, a certain level of serum uric acid may provide benefit on cardiovascular disease to some extent; however, among the subjects with other cardiovascular risk factors, such as high glucose level or high blood pressure, higher serum uric acid levels would be associated with elevated risk of cardiovascular mortality.

The strengths of the present study include its large population-based individual data from all over Japan as well as its prospective design. To our knowledge, EPOCH-JAPAN is the largest-scale pooled data to examine the associations between uric acid level and risk of CVD in Japan. However, there were several limitations in the present study. First, the pooled

Advance Publication
Journal of Atherosclerosis and Thrombosis
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data for most of the cohorts were from participants in municipal health examinations, but not the representative samples in communities. Second, we did not adjust for medication use for hypertension or diabetes as well as for glucose or creatinine level because the information of more than 30% participants was missing. However, among the participants who had the information of antihypertensive medication use, we essentially obtained the same estimation for HRs of mortality from CVD according to the quintiles of serum uric acid, adjusting further for medication use for hypertension (data not shown).

In conclusion, the results of our large pooled analysis indicated a J- or U-shaped relationship between serum uric acid level and cardiovascular mortality and also showed that the highest quintile of serum uric acid levels compared with the lowest quintiles were associated with an increased CVD mortality in both Japanese men and women.

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Appendix

The Evidence for Cardiovascular Prevention from Observational Cohorts in Japan (EPOCH-JAPAN) Research Group is composed of the following investigators. Chairperson: Hirotsugu Ueshima (Shiga University of Medical Science); Co-Chairperson: Tomonori Okamura (Keio University);

Executive committee: Hirotsugu Ueshima (Shiga University of Medical Science), Yutaka Imai (Tohoku University Graduate School of Pharmaceutical Sciences), Takayoshi Ohkubo (Teikyo University School of Medicine), Fujiko Irie (Ibaraki Prefecture), Hiroyasu Iso, Akihiko Kitamura (Osaka University Graduate School of Medicine), Yutaka Kiyohara (Kyushu University Graduate School of Medicine), Katsuyuki Miura (Shiga University of Medical Science), Yoshitaka Murakami (Toho University), Hideaki Nakagawa (Kanazawa Medical University), Takeo Nakayama (Kyoto University School of Public Health), Tomonori Okamura (Keio University), Akira Okayama (Research Institute of Strategy for Prevention), Toshimi Sairenchi (Dokkyo Medical University), Shigeyuki Saitoh (Sapporo Medical University), Kiyomi Sakata (Iwate Medical University), Akiko Tamakoshi (Hokkaido University Graduate School of Medicine), Ichiro Tsuji (Tohoku University Graduate School of Medicine), Michiko Yamada (Radiation Effects Research Foundation), Masahiko Kiyama (Osaka Center for Cancer and Cardiovascular Disease Prevention), Yoshihiro Miyamoto (National Cerebral and Cardiovascular Center), Shizukiyo Ishikawa (Jichi Medical University) and Hiroshi Yatsuya (Fujita Health University).

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Disclosures

None.

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Supplementary Table

Sex-specific hazard ratios (95% CI) of mortality from stroke, coronary heart disease, heart failure and total cardiovascular diseases according to quintiles of serum uric acid levels censoring the first 3 years

	Quintiles of serum uric acid levels					*P* for trend
	1 (low)	2	3	4	5 (high)	
Men						
No. at risk	2944	3270	2860	2984	3149	
Person Year	34600	40315	34266	38029	40172	
Total stroke						
No. of death	56	53	41	44	78	
Age-adjusted HR	1.00	0.81 (0.56-1.18)	0.77 (0.51-1.16)	0.77 (0.52-1.15)	1.19 (0.84-1.68)	0.211
Multivariable HR†	1.00	0.82 (0.51-1.20)	0.77 (0.51-1.16)	0.78 (0.52-1.17)	1.21 (0.84-1.74)	0.220
Ischemic stroke						
No. of death	29	31	23	29	46	
Age-adjusted HR	1.00	0.91 (0.55-1.51)	0.82 (0.46-1.40)	1.01 (0.60-1.70)	1.40 (0.87-2.24)	0.082
Multivariable HR†	1.00	0.94 (0.56-1.58)	0.79 (0.45-1.37)	1.01 (0.59-1.71)	1.37 (0.83-2.24)	0.142
Hemorrhagic stroke						
No. of death	16	16	16	14	24	
Age-adjusted HR	1.00	0.91 (0.52-1.60)	0.82 (0.44-1.51)	1.06 (0.60-1.86)	1.14 (0.66-1.94)	0.484
Multivariable HR†	1.00	0.85 (0.42-1.71)	1.19 (0.59-2.41)	0.83 (0.40-1.73)	1.29 (0.66-2.51)	0.406
Coronary heart disease						
No. of death	24	25	18	25	31	
Age-adjusted HR	1.00	0.99 (0.57-1.72)	0.83 (0.45-1.53)	1.15 (0.66-2.00)	1.29 (0.76-2.18)	0.485
Multivariable HR†	1.00	0.90 (0.51-1.59)	0.74 (0.40-1.37)	0.92 (0.52-1.64)	0.97 (0.55-1.70)	0.992
Heart failure						
No. of death	15	20	19	16	28	
Age-adjusted HR	1.00	1.13 (0.58-2.21)	1.35 (0.68-2.66)	1.01 (0.50-2.06)	1.55 (0.83-2.92)	0.193
Multivariable HR†	1.00	1.17 (0.60-2.30)	1.39 (0.70-2.77)	1.09 (0.53-2.23)	1.86 (0.97-3.57)	0.068
Total cardiovascular disease						
No. of death	110	114	90	103	164	
Age-adjusted HR	1.00	0.89 (0.68-1.15)	0.87 (0.66-1.15)	0.91 (0.70-1.19)	1.26 (0.99-1.61)	0.023
Multivariable HR†	1.00	0.90 (0.69-1.17)	0.86 (0.65-1.14)	0.91 (0.69-1.19)	1.28 (0.99-1.65)	0.029
Women						
No. at risk	4320	3867	4329	3555	4247	
Person Year	50989	45970	55440	44575	56142	
Total stroke						
No. of death	30	35	44	47	103	
Age-adjusted HR	1.00	1.12 (0.69-1.83)	0.99 (0.62-1.58)	1.08 (0.68-1.72)	1.35 (0.89-2.04)	0.079
Multivariable HR†	1.00	1.16 (0.71-1.89)	1.05 (0.65-1.67)	1.06 (0.67-1.69)	1.35 (0.88-2.06)	0.121
Ischemic stroke						
No. of death	15	21	18	29	49	
Age-adjusted HR	1.00	1.25 (0.64-2.41)	0.77 (0.38-1.53)	1.18 (0.63-2.23)	1.17 (0.65-2.10)	0.562
Multivariable HR†	1.00	1.31 (0.67-2.55)	0.80 (0.40-1.60)	1.19 (0.63-2.25)	1.19 (0.65-2.18)	0.588
Hemorrhagic stroke						
No. of death	10	10	20	11	34	
Age-adjusted HR	1.00	1.11 (0.46-2.68)	1.44 (0.67-3.10)	0.97 (0.41-2.31)	1.53 (0.74-3.14)	0.252
Multivariable HR†	1.00	1.09 (0.45-2.64)	1.45 (0.67-3.12)	0.90 (0.37-2.15)	1.44 (0.69-3.00)	0.360
Coronary heart disease						
No. of death	9	13	17	21	43	
Age-adjusted HR	1.00	1.41 (0.60-3.30)	1.16 (0.51-2.61)	1.58 (0.72-3.48)	1.68 (0.81-3.48)	0.122
Multivariable HR†	1.00	1.41 (0.60-3.33)	1.20 (0.53-2.72)	1.49 (0.67-3.30)	1.61 (0.76-3.39)	0.206
Heart failure						
No. of death	17	11	22	24	47	
Age-adjusted HR	1.00	0.56 (0.26-1.19)	0.76 (0.40-1.43)	0.80 (0.43-1.50)	0.92 (0.52-1.62)	0.548
Multivariable HR†	1.00	0.61 (0.28-1.31)	0.81 (0.43-1.55)	0.91 (0.48-1.73)	1.11 (0.62-1.99)	0.236
Total cardiovascular diseases						
No. of death	62	72	102	108	226	
Age-adjusted HR	1.00	1.10 (0.78-1.54)	1.07 (0.78-1.47)	1.16 (0.84-1.59)	1.35 (1.01-1.80)	0.012
Multivariable HR†	1.00	1.13 (0.81-1.59)	1.12 (0.82-1.55)	1.16 (0.84-1.59)	1.40 (1.04-1.88)	0.010

† Adjusted further for body mass index, smoking status, ethanol intake, systolic blood pressure and total cholesterol.

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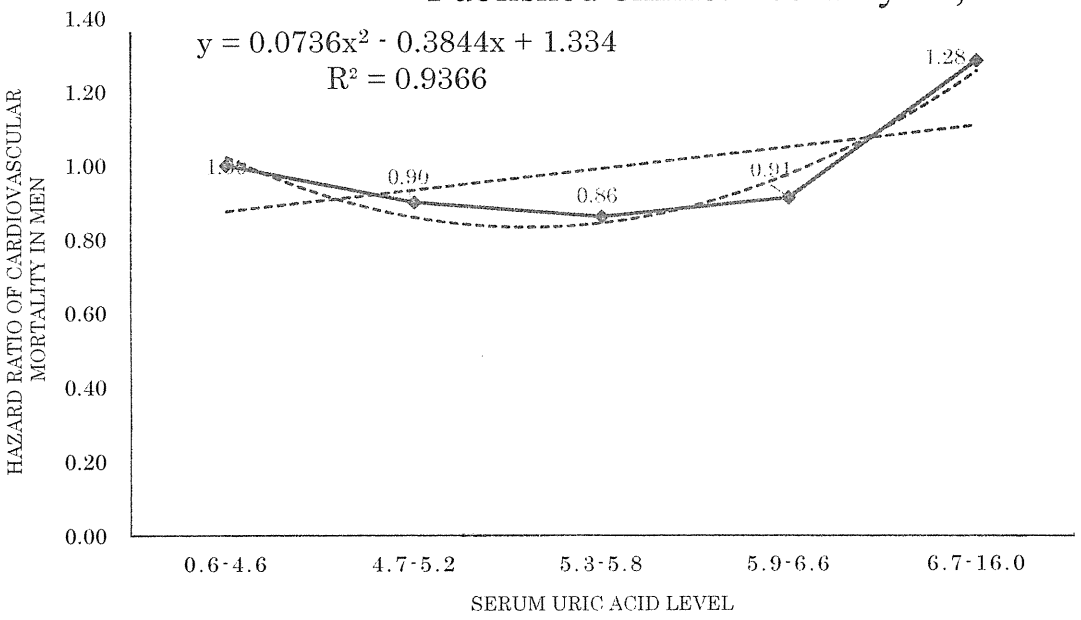
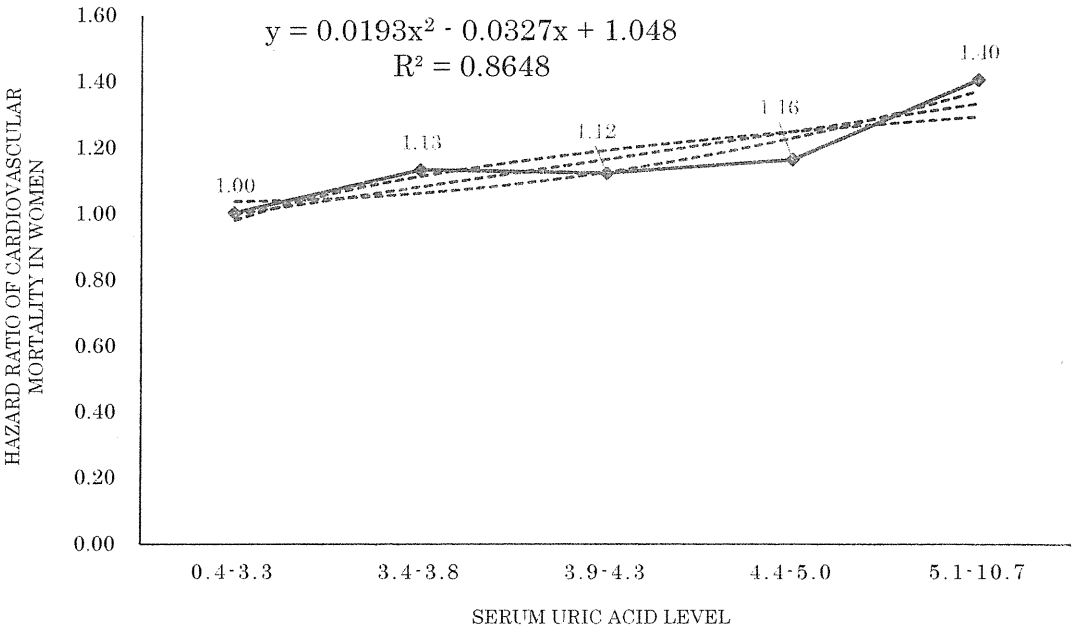


Supplementary Fig. 1.
Fitting curve of association between serum uric acid and cardiovascular mortality in men censoring the first three years



Supplementary Fig. 2.
Fitting curve of association between serum uric acid and cardiovascular mortality in women censoring the first three years

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Original Article

HOMA-IR Values are Associated With Glycemic Control in Japanese Subjects Without Diabetes or Obesity: The KOBE Study

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ABSTRACT

Background: Several studies have reported that insulin resistance was a major risk factor for the onset of type 2 diabetes mellitus in individuals without diabetes or obesity. We aimed to clarify the association between insulin resistance and glycemic control in Japanese subjects without diabetes or obesity.

Methods: We conducted a community-based cross-sectional study including 1083 healthy subjects (323 men and 760 women) in an urban area. We performed multivariate regression analyses to estimate the association between the homeostasis model assessment of insulin resistance (HOMA-IR) values and markers of glycemic control, including glycated haemoglobin (HbA1c), 1,5-anhydroglucitol (1,5-AG), and fasting plasma glucose (FPG) levels, after adjustment for potential confounders.

Results: Compared with the lowest tertile of HOMA-IR values, the highest tertile was significantly associated with HbA1c and FPG levels after adjustment for potential confounders, both in men (HbA1c: $\beta = 1.83$, $P = 0.001$; FPG: $\beta = 0.49$, $P < 0.001$) and women (HbA1c: $\beta = 0.82$, $P = 0.008$; FPG: $\beta = 0.39$, $P < 0.001$). The highest tertile of HOMA-IR values was inversely associated with 1,5-AG levels compared with the lowest tertile ($\beta = -18.42$, $P = 0.009$) only in men.

Conclusions: HOMA-IR values were associated with markers of glycemic control in Japanese subjects without diabetes or obesity. Insulin resistance may influence glycemic control even in a lean, non-diabetic Asian population.

Key words: homeostasis model assessment of insulin resistance; glycemic control; epidemiology

INTRODUCTION

Insulin resistance is a clinical condition characterized by a decreased sensitivity to insulin in peripheral tissues and is strongly associated with metabolic diseases, such as type 2 diabetes mellitus and obesity.[1–3] Prospective cohort studies in subjects without diabetes have also revealed that increased insulin resistance worsened glycemic control and contributed to the development of type 2 diabetes mellitus.[4–6] However, in all previous reports, the average body mass index (BMI) of the subjects was high (28–33 kg/m^2), and >50% of the subjects were obese.[4–6] Thus, it is unclear whether insulin resistance affects glycemic control in subjects without obesity or diabetes.

For assessing glycemic control, temporal variations in the indicative parameters are more important than values obtained at a single point in time. Glycated hemoglobin (HbA1c) and 1,5-anhydroglucitol (1,5-AG) levels are generally used to

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evaluate glycemic control in clinical practice. HbA1c levels are the gold standard marker of glycemic control in patients with diabetes, and they reflect average plasma glucose levels during the past 2–3 months.[7] In contrast, 1,5-AG levels are used as an index that reflects glycemic control during the past few days or weeks and glycemic control fluctuations.[8,9] Thus, it is necessary to use several indices in various time periods to evaluate glucose metabolism. However, to the best of our knowledge, no earlier studies have investigated the association between insulin resistance and glucose metabolism using multiple markers of glycemic control.

Therefore, in the present study, we aimed to investigate the impact of insulin resistance on glucose metabolism of Japanese subjects without diabetes or obesity. We used three markers that are commonly used to evaluate glucose metabolism in Japanese populations: HbA1c, 1,5-AG, and fasting plasma glucose (FPG) levels.

METHODS

Subjects

We used data from the baseline survey in the Kobe Orthopedic and Biomedical Epidemiological (KOBE) study. The KOBE study is a population-based prospective cohort study of risk factors for cardiovascular disease or worsening of quality of life in Kobe City, a major urban area in Japan, that has been ongoing since 2010. The KOBE study has been described in detail elsewhere.[10] The present study was approved by the Ethics Committee of the Institute of Biomedical Research and Innovation (Committee approval number: 11-12). Written informed consent was obtained from all participants.

A total of 1118 subjects (342 men and 776 women) participated in the baseline survey from July 2010 to December 2011. None of the participants had past history of cardiovascular disease or cancer, and none were under therapy with medications for hypertension, dyslipidemia, or diabetes at the time of the survey. We excluded 34 participants who were diagnosed with diabetes or obesity on the basis of FPG level of $\geq$7.0 mmol/L ($n = 8$) and/or HbA1c level of $\geq$6.5% ($n = 22$) or BMI of $\geq$30 kg/m^2 ($n = 4$) at baseline. A participant with missing data ($n = 1$) was also excluded. We ultimately analysed data of 1083 subjects (323 men and 760 women) without diabetes or obesity in this study.

Measurements

Each subject completed a self-reported questionnaire to assess past medical history and lifestyle factors, such as smoking status, alcohol consumption, and regular exercise habits, and trained researchers directly confirmed the responses to the questionnaire. Waist circumference was measured at the level of the umbilicus in a standing position. Height and body weight were measured with patients wearing socks and light clothing, and BMI was calculated by dividing weight in kilograms by the squared height in meters.

Fasting blood samples were drawn from all participants after they had fasted for at least 10 hours. Blood samples were transported to a single commissioned clinical laboratory centre (SRL Inc., Tokyo, Japan) for measurements. Plasma glucose levels (mmol/L) were determined using the glucose oxidase method. 1,5-AG levels were measured using an enzymatic method. HbA1c levels were measured using high-performance liquid chromatography and were expressed as National Glycohemoglobin Standardization Program units and International Federation of Clinical Chemistry and Laboratory Medicine values for the current analysis.[11] Serum immunoreactive insulin (IRI) levels (pmol/L) were determined using the chemiluminescence enzyme immunoassay (CLEIA) method, and homeostasis model assessment-insulin resistance (HOMA-IR) values were calculated using the following formula: HOMA-IR = IRI × glucose/22.5.[12] Estimated glomerular filtration rate (eGFR) was calculated using the following formula: eGFR (mL/min per 1.73 m^2) = 194 × creatinine$^{-1.094}$ × age$^{-0.287}$ (× 0.739 if female),[13] and chronic kidney disease (CKD) defined as eGFR of <60 mL/min per 1.73 m^2. High-molecular-weight adiponectin (HMW-adiponectin) levels were measured using the CLEIA method.

Statistical analysis

Gender-specific analyses were performed in light of observed gender differences in HOMA-IR distribution. HOMA-IR values were divided into tertiles to compare the characteristics. Data were presented as means (standard deviations [SDs]) or medians (interquartile ranges) for continuous variables, or numbers (percentages) for categorical variables. We used one-way analysis of variance for continuous variables and the chi-square test or Fisher's exact test for categorical variables to compare the characteristics among the groups. Multiple adjustments were performed with linear regression models to estimate the association between HOMA-IR values and markers of glycemic control, such as FPG, 1,5-AG, and HbA1c levels. We also performed multivariate logistic regression analysis to clarify the association between HOMA-IR values and any of the higher percentiles (80th or 90th percentile) of HbA1c levels, lower percentiles (10th or 20th percentile) of 1,5-AG levels or higher percentiles (80th or 90th percentile) of FPG levels. Multivariable analyses were adjusted for potential confounders in the following steps: (1) age; (2) BMI, regular exercise habits, current smoking, current alcohol drinking, CKD, and HMW-adiponectin levels, in addition to the variables in step 1; and (3) waist circumference substituted for BMI in step 2. The adjusted coefficient of determination (adjusted R^2) was also calculated. Two-tailed P values of <0.05 were considered statistically significant. All analyses were performed using STATA SE 11 data analysis and statistical software (Stata Corp LP, College Station, TX, USA).

Table 1. Characteristics of the participants according to HOMA-IR values by gender

	HOMA-IR tertile			*P* value
	1st (low)	2nd	3rd (high)	
Men (*n* = 323)				
Number of participants	109	107	107	
HOMA-IR	<3.397	3.397–5.596	≥5.596	
HbA1c (NGSP; %), mean (SD)	5.47 (0.30)	5.43 (0.25)	5.63 (0.36)	<0.001
HbA1c (IFCC; mmol/mol), mean (SD)	36 (3)	36 (3)	38 (4)	<0.001
1,5-AG (μmol/L), mean (SD)	145.9 (46.9)	139.4 (45.0)	123.4 (40.9)	<0.001
FPG (mmol/L), mean (SD)	4.91 (0.34)	5.00 (0.35)	5.38 (0.50)	<0.001
IRI (pmol/L), mean (SD)	11.2 (3.1)	19.8 (3.0)	36.6 (12.4)	<0.001
Age (years), mean (SD)	61.1 (8.6)	60.0 (9.5)	61.3 (8.9)	0.495
Body mass index (kg/m^2), mean (SD)	21.2 (2.2)	22.8 (2.1)	24.5 (2.3)	<0.001
Waist circumference (cm), mean (SD)	78.1 (6.6)	82.7 (6.0)	87.9 (7.7)	<0.001
Regular exercise, *n* (%)	70 (64.2%)	69 (64.5%)	66 (61.7%)	0.916
Current smoker, *n* (%)	15 (13.8%)	15 (14.0%)	5 (4.7%)	0.036
Current alcohol drinker, *n* (%)	86 (78.9%)	82 (76.6%)	82 (76.6%)	0.903
Chronic kidney disease, *n* (%)	9 (8.3%)	12 (11.2%)	15 (14.0%)	0.407
HMW-Adiponectin (μg/mL), median (IQR)	3.6 (2.6–5.2)	3.2 (2.0–4.7)	2.5 (1.6–3.7)	<0.001
Women (*n* = 760)				
Number of participants	254	255	251	
HOMA-IR	<3.126	3.126–4.819	≥4.819	
HbA1c (NGSP; %), mean (SD)	5.53 (0.31)	5.55 (0.27)	5.63 (0.29)	<0.001
HbA1c (IFCC; mmol/mol), mean (SD)	37 (3)	37 (3)	38 (3)	<0.001
1,5-AG (μmol/L), mean (SD)	105.6 (31.8)	107.1 (32.3)	109.5 (38.0)	0.444
FPG (mmol/L), mean (SD)	4.68 (0.34)	4.86 (0.33)	5.08 (0.40)	<0.001
IRI (pmol/L), mean (SD)	11.1 (2.7)	18.3 (2.4)	30.7 (10.1)	<0.001
Age (years), mean (SD)	57.0 (8.9)	58.4 (8.7)	58.5 (8.4)	0.094
Body mass index (kg/m^2), mean (SD)	19.5 (2.1)	21.0 (2.3)	22.1 (2.6)	<0.001
Waist circumference (cm), mean (SD)	73.9 (7.3)	78.5 (7.3)	81.9 (7.6)	<0.001
Regular exercise, *n* (%)	141 (55.5%)	139 (54.5%)	124 (49.4%)	0.341
Current smoker, *n* (%)	7 (2.8%)	2 (0.8%)	6 (2.4%)	0.200
Current alcohol drinker, *n* (%)	107 (42.1%)	79 (31.0%)	89 (35.5%)	0.031
Chronic kidney disease, *n* (%)	18 (7.1%)	18 (7.1%)	20 (8.0%)	0.924
HMW-Adiponectin (μg/mL), median (IQR)	6.6 (4.6–8.8)	5.5 (3.9–7.8)	4.4 (3.1–6.1)	<0.001

1,5-AG, 1,5-anhydroglucitol; FPG, fasting plasma glucose; HMW-Adiponectin, high-molecular-weight adiponectin; HOMA-IR, homeostasis model assessment of insulin resistance; IFCC, International Federation of Clinical Chemistry and Laboratory Medicine; IQR, interquartile range; IRI, immunoreactive insulin; NGSP, National Glycohemoglobin Standardization Program; SD, standard deviation.

RESULTS

Baseline characteristics of participants

Table 1 shows the characteristics of the participants according to HOMA-IR category by gender. The mean (SD) age was 60.8 (9.0) and 58.0 (8.7) years in men and women, respectively. Participants in higher HOMA-IR categories had higher HbA1c and FPG levels, both in men and women, and only men had lower 1,5-AG levels. Participants in higher HOMA-IR categories also had higher BMI and waist circumference, as well as lower HMW-adiponectin levels, both in men and women.

Association between HOMA-IR values and markers of glycemic control

The association between HOMA-IR values and markers of glycemic control, such as HbA1c, 1,5-AG, and FPG levels, in the multivariate linear regression analysis are shown according to gender in Table 2 (men) and Table 3 (women). HbA1c and FPG levels were significantly higher in the highest tertile group of HOMA-IR values than in the lowest tertile group, both in men and women. 1,5-AG levels were significantly lower in the highest tertile group of HOMA-IR values than in the lowest tertile group in men but not in women. The association between HOMA-IR values and markers of glycemic control was unchanged after adjusting for potential confounders, including BMI, waist circumference, and HMW-adiponectin levels. We performed multiple linear regression analysis to estimate the association between HOMA-IR values and markers of glycemic control according to BMI and gender. The results showed that the absolute values of coefficient were larger in the group with high BMI than in the group with low BMI in men (eTable 1), which suggested a strong association between HOMA-IR and these glycemic control parameters; however, these findings were not clearly observed in women (eTable 2). We also performed multivariate logistic regression analysis to clarify the association between HOMA-IR values and any of the higher percentiles of HbA1c levels, lower percentiles of 1,5-AG levels, or higher percentiles of FPG levels, both in men (eTable 3) and women (eTable 4). Compared with the lowest tertile group of HOMA-IR values, the highest tertile group

Table 2. Associations between HOMA-IR values and markers of glycemic control in men (n = 323)

Dependent variables		Independent variables: HbA1c (mmol/mol)				Independent variables: 1,5-AG (μmol/L)				Independent variables: FPG (mmol/L)			
		Coefficient	95% CI	Standardized Coefficient	*P* value	Coefficient	95% CI	Standardized Coefficient	*P* value	Coefficient	95% CI	Standardized Coefficient	*P* value
Model 1													
HOMA-IR	1st (<3.397)	Reference				Reference				Reference			
	2nd (3.397–5.596)	−0.22	(−1.09, 0.65)	−0.03	0.621	−7.36	(−19.11, 4.39)	−0.08	0.219	0.10	(−0.01, 0.21)	0.10	0.064
	3rd (≥5.596)	1.77	(0.90, 2.64)	0.24	<0.001	−22.27	(−34.00, −10.53)	−0.23	<0.001	0.47	(0.36, 0.57)	0.49	<0.001
Age (10 years)		0.95	(0.55, 1.35)	0.25	<0.001	−7.86	(−13.23, −2.49)	−0.16	0.004	0.10	(0.05, 0.15)	0.20	<0.001
		Adjusted coefficient of determination (R^2) = 0.12				Adjusted coefficient of determination (R^2) = 0.06				Adjusted coefficient of determination (R^2) = 0.23			
Model 2													
HOMA-IR	1st (<3.397)	Reference				Reference				Reference			
	2nd (3.397–5.596)	−0.33	(−1.24, 0.58)	−0.05	0.470	−7.04	(−19.16, 5.09)	−0.07	0.254	0.12	(0.01, 0.23)	0.12	0.034
	3rd (≥5.596)	1.65	(0.60, 2.70)	0.22	0.002	−18.62	(−32.60, −4.63)	−0.19	0.009	0.49	(0.36, 0.61)	0.51	<0.001
Age (10 years)		1.03	(0.59, 1.46)	0.26	<0.001	−6.42	(−12.25, −0.60)	−0.13	0.031	0.09	(0.04, 0.14)	0.18	0.001
Body mass index (kg/m^2)		0.09	(−0.07, 0.26)	0.07	0.258	0.30	(−1.89, 2.50)	0.02	0.786	−0.01	(−0.03, 0.01)	−0.04	0.486
Regular exercise (yes)		−0.12	(−0.92, 0.67)	−0.02	0.758	4.20	(−6.36, 14.75)	0.04	0.434	−0.01	(−0.10, 0.09)	−0.01	0.912
Current smoking (yes)		1.02	(−0.18, 2.21)	0.09	0.094	30.94	(15.05, 46.82)	0.21	<0.001	−0.21	(−0.35, −0.06)	−0.14	0.005
Current alcohol drinking (yes)		−0.84	(−1.70, 0.02)	−0.10	0.056	−1.74	(−13.19, 9.72)	−0.02	0.766	0.07	(−0.03, 0.18)	0.07	0.161
Chronic kidney disease (yes)		0.08	(−1.08, 1.24)	0.01	0.895	−4.52	(−20.00, 10.96)	−0.03	0.566	−0.08	(−0.22, 0.06)	−0.06	0.240
HMW-Adiponectin (μg/mL)		0.32	(−0.31, 0.94)	0.06	0.319	3.57	(−4.74, 11.88)	0.05	0.399	0.02	(−0.06, 0.09)	0.03	0.613
		Adjusted coefficient of determination (R^2) = 0.13				Adjusted coefficient of determination (R^2) = 0.09				Adjusted coefficient of determination (R^2) = 0.25			
Model 3													
HOMA-IR	1st (<3.397)	Reference				Reference				Reference			
	2nd (3.397–5.596)	−0.25	(−1.15, 0.66)	−0.03	0.595	−6.93	(−18.97, 5.11)	−0.07	0.258	0.12	(0.01, 0.23)	0.12	0.033
	3rd (≥5.596)	1.83	(0.79, 2.86)	0.25	0.001	−18.42	(−32.20, −4.64)	−0.19	0.009	0.49	(0.36, 0.61)	0.51	<0.001
Age (10 years)		1.00	(0.57, 1.44)	0.26	<0.001	−6.50	(−12.32, −0.69)	−0.13	0.028	0.09	(0.04, 0.14)	0.18	0.001
Waist circumference (10 cm)		0.13	(−0.41, 0.67)	0.03	0.635	0.83	(−6.33, 8.00)	0.01	0.819	−0.03	(−0.09, 0.04)	−0.04	0.441
Regular exercise (yes)		−0.11	(−0.90, 0.69)	−0.01	0.790	4.30	(−6.29, 14.89)	0.05	0.425	−0.01	(−0.10, 0.09)	−0.01	0.861
Current smoking (yes)		1.03	(−0.16, 2.23)	0.09	0.091	30.91	(15.00, 46.82)	0.21	<0.001	−0.21	(−0.35, −0.06)	−0.14	0.005
Current alcohol drinking (yes)		−0.84	(−1.70, 0.02)	−0.10	0.056	−1.76	(−13.22, 9.70)	−0.02	0.763	0.07	(−0.03, 0.18)	0.07	0.158
Chronic kidney disease (yes)		0.12	(−1.04, 1.29)	0.01	0.834	−4.38	(−19.82, 11.07)	−0.03	0.578	−0.09	(−0.23, 0.05)	−0.06	0.221
HMW-Adiponectin (μg/mL)		0.29	(−0.34, 0.91)	0.05	0.373	3.57	(−4.79, 11.93)	0.05	0.402	0.02	(−0.06, 0.09)	0.02	0.643
		Adjusted coefficient of determination (R^2) = 0.13				Adjusted coefficient of determination (R^2) = 0.09				Adjusted coefficient of determination (R^2) = 0.25			

1,5-AG, 1,5-anhydroglucitol; CI, confidence interval; FPG, fasting plasma glucose; HMW-Adiponectin, high-molecular-weight adiponectin; HOMA-IR, homeostasis model assessment of insulin resistance. Multivariate adjustment; Model 1: adjusted by age; Model 2: adjusted by age, body mass index, regular exercise (yes/no), current smoking (yes/no), current alcohol drinking (yes/no), chronic kidney disease (yes/no) and high-molecular-weight (HMW)-Adiponectin (log-transformed); Model 3: adjusted by age, waist circumference, regular exercise (yes/no), current smoking (yes/no), current alcohol drinking (yes/no), chronic kidney disease (yes/no) and HMW-Adiponectin (log-transformed).

Table 3. Associations between HOMA-IR values and markers of glycemic control in women (n = 760)

Dependent variables		Independent variables: HbA1c (mmol/mol)				Independent variables: 1,5-AG (μmol/L)				Independent variables: FPG (mmol/L)			
		Coefficient	95% CI	Standardized Coefficient	*P* value	Coefficient	95% CI	Standardized Coefficient	*P* value	Coefficient	95% CI	Standardized Coefficient	*P* value
Model 1													
HOMA-IR	1st (<3.126)	Reference				Reference				Reference			
	2nd (3.126–4.819)	0.18	(−0.36, 0.72)	0.03	0.516	1.90	(−4.03, 7.84)	0.03	0.529	0.16	(0.10, 0.22)	0.20	<0.001
	3rd (≥4.819)	1.00	(0.45, 1.54)	0.15	<0.001	4.35	(−1.61, 10.31)	0.06	0.152	0.38	(0.32, 0.44)	0.46	<0.001
Age (10 years)		0.88	(0.62, 1.13)	0.24	<0.001	−3.18	(−6.19, −0.59)	−0.09	0.018	0.11	(0.08, 0.14)	0.25	<0.001
		Adjusted coefficient of determination (R^2) = 0.07				Adjusted coefficient of determination (R^2) = 0.01				Adjusted coefficient of determination (R^2) = 0.23			
Model 2													
HOMA-IR	1st (<3.126)	Reference				Reference				Reference			
	2nd (3.126–4.819)	0.01	(−0.55, 0.57)	0.001	0.974	0.83	(−5.34, 6.99)	0.01	0.792	0.16	(0.10, 0.22)	0.19	<0.001
	3rd (≥4.819)	0.80	(0.19, 1.41)	0.12	0.010	2.28	(−4.45, 9.00)	0.03	0.507	0.37	(0.31, 0.44)	0.45	<0.001
Age (10 years)		0.73	(0.44, 1.02)	0.20	<0.001	−3.06	(−6.28, 0.15)	−0.08	0.062	0.10	(0.07, 0.13)	0.23	<0.001
Body mass index (kg/m^2)		0.07	(−0.02, 0.17)	0.06	0.130	0.96	(−0.11, 2.03)	0.07	0.077	0.01	(−0.004, 0.02)	0.04	0.286
Regular exercise (yes)		0.19	(−0.30, 0.68)	0.03	0.438	−2.19	(−7.58, 3.20)	−0.03	0.425	0.04	(−0.02, 0.09)	0.05	0.160
Current smoking (yes)		−1.12	(−2.73, 0.50)	−0.05	0.174	16.49	(−1.27, 34.25)	0.07	0.069	−0.01	(−0.19, 0.17)	−0.003	0.934
Current alcohol drinking (yes)		−0.62	(−1.08, −0.15)	−0.09	0.009	−2.01	(−7.12, 3.11)	−0.03	0.441	0.04	(−0.01, 0.09)	0.05	0.136
Chronic kidney disease (yes)		0.67	(−0.18, 1.52)	0.05	0.122	0.09	(−9.27, 9.44)	0.001	0.985	0.07	(−0.02, 0.17)	0.05	0.128
HMW-Adiponectin (μg/mL)		0.03	(−0.41, 0.47)	0.01	0.884	1.74	(−3.08, 6.57)	0.03	0.479	−0.001	(−0.05, 0.05)	−0.001	0.982
		Adjusted coefficient of determination (R^2) = 0.08				Adjusted coefficient of determination (R^2) = 0.01				Adjusted coefficient of determination (R^2) = 0.23			
Model 3													
HOMA-IR	1st (<3.126)	Reference				Reference				Reference			
	2nd (3.126–4.819)	0.02	(−0.54, 0.58)	0.002	0.956	0.64	(−5.53, 6.80)	0.01	0.840	0.17	(0.11, 0.23)	0.20	<0.001
	3rd (≥4.819)	0.82	(0.21, 1.43)	0.12	0.008	2.09	(−4.59, 8.77)	0.03	0.539	0.39	(0.32, 0.45)	0.46	<0.001
Age (10 years)		0.70	(0.41, 1.00)	0.19	<0.001	−3.49	(−6.76, −0.22)	−0.09	0.036	0.10	(0.07, 0.14)	0.23	<0.001
Waist circumference (10 cm)		0.22	(−0.09, 0.53)	0.06	0.166	3.52	(0.09, 6.94)	0.08	0.044	0.0004	(−0.03, 0.04)	0.001	0.979
Regular exercise (yes)		0.20	(−0.29, 0.69)	0.03	0.425	−2.17	(−7.55, 3.21)	−0.03	0.428	0.04	(−0.01, 0.09)	0.05	0.144
Current smoking (yes)		−1.07	(−2.69, 0.54)	−0.05	0.192	17.11	(−0.64, 34.85)	0.07	0.059	−0.01	(−0.18, 0.17)	−0.002	0.956
Current alcohol drinking (yes)		−0.65	(−1.11, −0.18)	−0.10	0.007	−2.44	(−7.57, 2.69)	−0.03	0.350	0.04	(−0.01, 0.09)	0.05	0.141
Chronic kidney disease (yes)		0.68	(−0.17, 1.53)	0.05	0.119	0.18	(−9.17, 9.53)	0.001	0.971	0.07	(−0.02, 0.17)	0.05	0.130
HMW-Adiponectin (μg/mL)		0.03	(−0.41, 0.47)	0.01	0.883	1.98	(−2.87, 6.82)	0.03	0.424	−0.01	(−0.05, 0.04)	−0.01	0.811
		Adjusted coefficient of determination (R^2) = 0.08				Adjusted coefficient of determination (R^2) = 0.01				Adjusted coefficient of determination (R^2) = 0.23			

1,5-AG, 1,5-anhydroglucitol; CI, confidence interval; FPG, fasting plasma glucose; HMW-Adiponectin, high-molecular-weight adiponectin; HOMA-IR, homeostasis model assessment of insulin resistance. Multivariate adjustment; Model 1: adjusted by age; Model 2: adjusted by age, body mass index, regular exercise (yes/no), current smoking (yes/no), current alcohol drinking (yes/no), chronic kidney disease (yes/no) and high-molecular-weight (HMW)-Adiponectin (log-transformed); Model 3: adjusted by age, waist circumference, regular exercise (yes/no), current smoking (yes/no), current alcohol drinking (yes/no), chronic kidney disease (yes/no) and HMW-Adiponectin (log-transformed).

had significantly higher odds ratios for any of higher percentiles of HbA1c levels, lower percentiles of 1,5-AG levels, or higher percentiles of FPG levels, both in men and women, after adjusting for potential confounders.

DISCUSSION

This is the first report, to the best of our knowledge, to assess the relationship between HOMA-IR values and several indices of glucose metabolism, obtained at various time points, in Japanese subjects without diabetes or obesity. As a result, we found that HOMA-IR values were significantly associated with all indices of glucose metabolism in men, as well as with HbA1c or FPG levels in women.

HOMA-IR is generally considered an index of insulin resistance in the liver.[12] Insulin suppresses the elevation of plasma glucose levels by promoting glucose uptake into cells and by inhibiting glucose release from the liver. However, when insulin resistance increases, the regulatory mechanism fails and blood glucose levels remain elevated.[14–16] In the present study, our results indicate that increased insulin resistance further deteriorates glucose metabolism in patients with not only type 2 diabetes mellitus and obesity but also in those without diabetes or obesity. The relationship between insulin resistance and several indices of glucose metabolism was maintained after adjusting for confounding factors, such as BMI or waist circumference. Multivariate regression analysis in this study revealed that BMI and waist circumference were not correlated with the indices of glucose metabolism. However, in the multivariate regression model that excluded HOMA-IR as an independent variable, BMI (eTable 5) and waist circumference (eTable 6) maintained significant correlation with HbA1c and FPG levels. Therefore, these results indicated that insulin resistance might regulate glucose metabolism downstream of BMI and waist circumference.

The present study showed that HOMA-IR values were not significantly associated with 1,5-AG levels in women. 1,5-AG is monosaccharide excreted in the urine. Approximately, 99%–100% of the excreted 1,5-AG is reabsorbed in the renal tubules, and a constant level is maintained in subjects with normal glucose tolerance. When blood glucose levels reach the threshold at which urinary glucose appears, 1,5-AG levels decrease remarkably because 1,5-AG reabsorption is inhibited.[8,17,18] In other words, 1,5-AG levels do not change if blood glucose levels do not reach the urinary glucose excretion threshold. Considering these mechanisms, it is suspected that most of the participants, especially women, had normal glucose tolerance, although participants both with normal glucose tolerance and mild glucose intolerance were included in the present study. In male participants, HOMA-IR values were weakly correlated with 1,5-AG levels compared to the correlations of HOMA-IR values with HbA1c and FPG levels. Thus, it is possible that the suspected high prevalence of normal glucose tolerance influenced this result.

A cohort study of the general Japanese population revealed that metabolic syndrome increased the risk of onset of type 2 diabetes mellitus, suggesting that insulin resistance contributed to the onset of type 2 diabetes mellitus in the Japanese population.[19] Another cohort study of the general Japanese population showed that both a decrease in insulin secretion ability and an increase in insulin resistance contributed to the onset of type 2 diabetes mellitus.[20] In the present study, a correlation between an index of insulin resistance and several indices of glucose metabolism was observed in Japanese subjects without obesity and with low insulin resistance. These findings suggest that insulin resistance mainly contributed to the onset of type 2 diabetes mellitus in Japanese subjects. Lifestyle interventions, such as healthy diet and regular exercise, have been shown to be effective for the improvement of insulin resistance but not impaired insulin secretion capacity.[21] Thus, we recommend lifestyle interventions for the prevention of onset of type 2 diabetes mellitus not only in obese subjects but also in non-obese subjects. In the present study, we also found that the association between HOMA-IR values and each marker of glycemic control was stronger in subjects with high BMI than in those with low BMI. Therefore, even in non-obese (BMI $<30\,kg/m^2$) Asians, we suggest that the impact of lifestyle intervention on the onset of type 2 diabetes mellitus is larger among subjects with high BMI than among those with low BMI when impaired insulin secretion capacity is suspected.

This study has several limitations. At first, we used HOMA-IR, which is an indirect index for the evaluation of insulin resistance. Although the glucose clamp technique is necessary for direct evaluation,[22] we were unable to apply this test in our subjects. However, a previous study reported that the use of HOMA-IR was appropriate to assess insulin sensitivity in subjects without diabetes.[23] Second, we used a self-reported questionnaire in the present study; thus, recall bias might have affected the evaluation of physical activity. Finally, we could not evaluate postprandial hyperglycemia accurately in this study because we did not measure the blood glucose afterload. We used HbA1c levels as the diagnostic criteria of diabetes in this study; thus, subjects having marked postprandial hyperglycemia were excluded.

In conclusion, the present study showed that HOMA-IR values were significantly associated with several indices of glucose metabolism in Japanese subjects without diabetes or obesity and that insulin resistance prescribed glucose metabolism downstream of BMI or waist circumference. These findings suggest that insulin resistance may mainly influence glycemic control even in non-diabetic subjects without obesity.

ONLINE ONLY MATERIALS

eTable 1. Associations between HOMA-IR values and markers of glycemic control divided by median BMI in men ($n = 323$).