Table 3. The Cox regression coefficients for the Suita cohort adjusted for the original FRS variables (TC Suita Score)

3a. TC Suita Score with CKD

	β	HR	*P*-value	95% CI
Age, years	0.0766382	1.08	<0.001	1.06-1.10
Female	−0.5866078	0.56	0.001	0.39-0.78
Smoking	0.4865127	1.63	0.002	1.20-2.21
DM	0.4557071	1.59	0.042	1.02-2.45
Blood Pressure				
Optimal	−0.7183575	0.49	0.003	0.31-0.78
Normal and high normal	Referent	Reference	Reference	Reference
Stage I hypertension	0.3330895	1.40	0.055	0.99-1.96
Stage II hypertension	0.59332684	1.81	0.002	1.25-2.63
TC (mg/dl)				
<160	−1.112393	0.33	0.008	0.14-0.74
160-239	Referent	Reference	Reference	Reference
240-279	0.5110573	1.67	0.003	1.19-2.32
Over 280	0.8511397	2.34	0.002	1.36-4.04
HDL(mg/dl)				
<35	0.6173452	1.85	0.001	1.27-2.71
35-50	Referent	Reference	Reference	Reference
50-59	−0.5096169	0.60	0.008	0.41-0.87
Over 60	−0.4322771	0.65	0.022	0.45-0.94
CKD				
Stage 3	0.3278965	1.39	0.035	1.02-1.88
Stage 4 or 5	1.315004	3.72	0.005	1.48-9.38

3b. TC Suita Score without CKD

	β	HR	*P*-value	95% CI
Age, years	0.0806456	1.08	<.0001	1.07-1.10
Female	−0.7401036	0.48	<.0001	0.35-0.66
Smoking	0.4527364	1.57	0.004	1.16-2.14
DM	0.424255	1.52	0.059	0.98-2.37
Blood Pressure				
Optimal	−0.7017837	0.50	0.004	0.31-0.79
Normal and high normal	Referent	Reference	Reference	Reference
Stage I hypertension	0.3607005	1.43	0.037	1.02-2.01
Stage II hypertension	0.6305927	1.87	0.001	1.30-2.72
TC (mg/dl)				
<160	−1.073273	0.34	0.010	0.16-0.78
160-239	Referent	Reference	Reference	Reference
240-279	0.5408852	1.71	0.001	1.23-2.40
Over 280	0.8678275	2.38	0.002	1.38-4.10
HDL(mg/dl)				
<35	0.6742382	1.96	<.0001	1.35-2.86
35-50	Referent	Reference	Reference	Reference
50-59	−0.5011838	0.61	0.009	0.44-0.93
Over 60	−0.4464421	0.64	0.018	0.15-0.93

CKD, chronic kidney disease; TC, total cholesterol; LDL, low density lipoprotein; 95% CI, 95% confidence interval; CKD, chronic kidney disease; HR, hazard ratio; all other abbreviations are the same as in Table 1. The gender difference was incorporated into the model as a covariate to improve the predictability of the model.

Table 4. The LDL Suita Score with CKD model according to the JAS Guideline 2012 LDL/HDL cut-off (n=5,727) (LDL Suita Score)

4a. The LDL Suita Score with CKD

	β	HR	P-value	95% CI
Age, years	0.0760078	1.08	<0.001	1.06-1.10
Female	−0.6619839	0.52	<0.001	0.36-0.74
Smoking	0.5031949	1.65	0.002	1.20-2.28
DM	0.5533678	1.74	0.031	1.05-2.88
Blood Pressure				
Optimal	−0.6763825	0.51	0.005	0.32-0.82
Normal and high normal	Reference	Reference	Reference	Reference
Stage I hypertension	0.4189501	1.52	0.019	1.07-2.16
Stage II hypertension	0.5935986	1.81	0.001	1.22-2.68
LDL (mg/dl)				
<100	Reference	Reference	Reference	Reference
100-140	0.5319015	1.70	0.039	1.03-2.81
140-160	0.6837867	1.98	0.015	1.14-3.43
160-180	1.021015	2.78	<0.001	1.57-4.91
Over 180	1.128479	3.09	<0.001	1.69-5.66
HDL(mg/dl)				
<40	Reference	Reference	Reference	Reference
40-59	−0.4730423	0.62	0.005	0.45-0.87
≥ 60	−0.5822414	0.56	0.007	0.37-0.85
CKD				
Stage 3	0.2893668	1.34	0.071	0.98-1.83
Stage 4 or 5	1.388216	4.01	0.008	1.43-11.25

4b. The LDL Suita Score without CKD

	β	HR	P-value	95% CI
Age, years	0.0795083	1.08	<0.001	1.06-1.10
Female	−0.804252	0.45	<0.001	0.32-0.62
Smoking	0.4642934	1.59	0.004	1.16-2.19
DM	0.3955565	1.48	0.106	0.91-2.40
Blood Pressure				
Optimal	−0.6602255	0.52	0.006	0.32-0.83
Normal and high normal	Reference	Reference	Reference	Reference
Stage I hypertension	0.4467296	1.56	0.012	1.10-2.21
Stage II hypertension	0.6256262	1.87	0.002	1.27-2.76
LDL (mg/dl)				
<100	Reference	Reference	Reference	Reference
100-140	0.5040579	1.66	0.049	1.00-2.74
140-160	0.664678	1.94	0.017	1.57-4.90
160-180	1.01949	2.77	<0.001	1.73-4.90
Over 180	1.151674	3.16	<0.001	1.73-5.80
HDL (mg/dl)				
<40	Reference	Reference	Reference	Reference
40-59	−0.4798994	0.62	0.004	0.45-0.86
≥ 60	−0.6092216	0.54	0.005	0.36-0.83

JAS Guideline 2012, Japan Atherosclerosis Society(JAS) Guidelines for Prevention of Atherosclerotic Cardiovascular Diseases 2012; CKD, chronic kidney disease; LDL, low density lipoprotein; 95% CI, 95% confidence interval; CKD, chronic kidney disease; HR, hazard ratio; all other abbreviations are the same as in Table 1. Stage 3 and Stage 4 or 5 CKD were defined by estimated GFR levels of 30-60 ml/min/1.73 m^2 and less than 30 ml/min/1.73 m^2, respectively.

Table 5. Prediction score sheets for TC/ LDL Suita score

5a. TC Suita scoore

Risk Factor Variable	Score
Age	
36-45	30
46-55	39
56-65	46
>65	58
Female	−6
Current Smoker	5
DM	5
Blood pressure	
Optimal blood pressure	−7
Normal and high normal	0
Stage 1 hypertension	3
Stage 2 hypertension	6
TC (mg/dl)	
<160	−11
160-239	0
240-279	5
>280	9
HDL (mg/dl)	
<35	6
36-49	0
50-59	−5
>=60	−5
CKD	
eGFR>60	0
Stage 3	3
Stage 4 or 5	15
Total Score	A

Predicted Probability of CHD in 10 years

Total Score	Probability (%)
30<=	<1
31-35	1
36-40	2
41-45	4
46-50	6
51-55	10
56-60	15
>=61	24

5b. LDL Suita Score

Risk Factor Variable	Score
Age	
35-44	30
45-54	38
55-64	45
65-69	51
>=70	53
Female	−7
Current Smoker	5
DM	6
Blood pressure	
Optimal blood pressure	−7
Normal and high normal	0
Stage 1 hypertension	4
Stage 2 hypertension	6
LDL (mg/dl)	
<100	0
100-139	5
140-159	7
160-179	10
>=180	11
HDL (mg/dl)	
<40	0
40-59	−5
>=60	−6
CKD	
eGFR>60	0
Stage 3	3
Stage 4 or 5	14
Total Score	A

Predicted Probability of CHD in 10 years

Total Score	Probability (%)
35<=	<1
36-40	1
41-45	2
46-50	3
51-55	5
56-60	9
61-65	14
66-70	22
71<=	>28

Estimates risk for CHD over a period of 10 years based on Suita Cohort experience at baseline. Summation of risk factor category points yield total score. JAS Guideline 2012, Japan Atherosclerosis Society (JAS) Guidelines for Prevention of Atherosclerotic Cardiovascular Diseases 2012. LDL cholesterol was derived by Fridewald's equation. Those who have triglycerides>400 were omitted for the calculation.

Discussion

In this study, we demonstrated the predictive ability of newly developed coronary prediction algorithms for Japanese subjects developed in the manner of the FRS. Our findings can be summarized as follows: 1) the risk profile for CHD of a Japanese population was considerably different from that of the original Framingham Heart Study cohort; 2) The prediction of CHD obtained with the risk score based on the Suita cohort with CKD variables was superior to that of the FRS or recalibration of the FRS; 3) Clinical reclassification revealed that the FRS overestimates the CHD risk in the Japanese population.

First, the risk profile of the Suita cohort proved to be considerably different from that of a Western population. The crude incidence rate of CHD in the original Framingham Cohort was 8.94 per 1000 person-years, while that of the Suita cohort was only 2.81 per 1000 person-years. The risks of hypertension, low HDL-C for males, and diabetes and smoking for females, in the Suita cohort were weighted higher than the risks in the Framingham cohort. This difference between the Suita and the Framingham cohorts constitutes a major concern for the application of the FRS in Japanese subjects, where the lower CHD incidence

Table 6. Validation of the inclusion of CKD in the TC Suita Score and LDL Suita Score

6a.

Model	Log Likelihood	LR test, *p*-value	C-statistics	BIC
TC Suita Score with CKD	−1610.8	referent	0.835	3233.3
TC Suita Score without CKD	−1618.1	0.013	0.833	3238.5
LDL Suita Score with CKD	−1510.1	referent	0.831	3365.6
LDL Suita Score without CKD	−1513.6	0.032	0.829	3414.5

6b.

Model		TC Suita Score with CKD	LDL Suita Score with CKD	LDL Suita Score with CKD
Reference Model		TC Suita Score without CKD	LDL Suita Score without CKD	TC Suita Score with CKD
Cases	Reclassified Downward (%)	16.7	48.1	48.8
	Reclassified Upward (%)	83.3	51.9	51.2
Non Cases	Reclassified Downward (%)	36.7	26.1	43.8
	Reclassified Upward (%)	63.3	73.9	56.2
Category-free NRI(%)		40.0	43.9	10.1
P-value		<0.001	<0.001	0.256

6a, Comparison of the C-index , LR test and BIC results demonstrating the discrimination for CHD prediction models based on the Suita Score with and without CKD, the FRS and the FRS calibrated for the means of the Suita cohort. Log likelihoods were derived from the multivariate adjusted Cox proportional hazard model.; CKD, chronic kidney disease; FHS, Framingham Heart Study; LR test, likelihood ratio test; BIC, Bayesian information criteria
6b, Comparison of the FRS and Suita Scores and the corresponding reclassification rate for the prediction of CHD events during a 10-year period;. NRI, net reclassification improvement

and different risk factor levels were observed[32-34].

Second, the discriminatory capability of the TC Suita Score with CKD is better than those of the original and recalibrated FRS. Although recalibration with the mean value of the risk factors and baseline survival functions for the study cohort improved the discriminatory capability for various ethnic groups in the U.S., China and the CKD population[6, 12, 16], the recalibration did not improve the discriminatory capability in Japanese subjects. We believe this is probably due to the low incidence of CHD in Japan compared to Western and Chinese populations[35]. The relative risks of various factors were similar between Suita Study cohort and the Framingham cohort. Therefore, the difference between the two prediction tools heavily depends on the difference in the absolute risks between these two cohorts. Accordingly, the clinical reclassification pointed out that the FRS overestimated the risk of CHD in Japanese subjects, especially in the non-CHD group, since the baseline survival function, which was higher than that in the original FRS, affected the estimated risk in an exponential manner and the overestimation was more severe in the high risk groups.

Furthermore, we found that CKD is an independent risk factor for CHD after adjusting for other predictors of the FRS. The cohorts in the Framingham Heart Study and the Offspring study showed no significant association between the presence of kidney disease and the incidence of CVD[36] although some collaborative analyses showed positive associations[17, 37]. Our result is essentially compatible with that of Weiner's study, which reported the HRs of CKD after adjustment of the FRS for whites and blacks[38]. No previous study has dealt with this association for Asian ethnicity as an additional covariate in the prediction tool, although many cohort studies in Japan have demonstrated a significant association between CKD and cardiovascular disease[20, 25, 39].

Finally, we developed a simple prediction sheet for the estimation of CHD based on the TC and LDL Suita Score. For the exact estimation, the beta-coefficient from the TC and LDL Suita Score are preferable. However, the calculation requires computational

Table 7. A comparison of the predicted risks in models based on the Framingham risk score and Suita Score with and without CKD

7a.

Model	Log Likelihood	LR test, *p*-value	C-statistics	BIC
TC Suita Score with CKD	−1610.8	referent	0.835	3233.3
TC Suita Score without CKD	−1618.1	0.013	0.833	3238.5
Original Framingham Score	−1678.5	<0.001	0.768	3365.6
Recalibrated Framingham Score	−1702.9	<0.001	0.740	3414.5

7b.

Model		TC Suita Score with CKD	TC Suita Score with CKD	TC Suita Score without CKD	TC Suita Score without CKD
Reference Model		Original Framingham Score	Recalibrated Framingham Score	Original Framingham Score	Recalibrated Framingham Score
Cases	Reclassified Downward (%)	42.0	50.0	42.8	49.3
	Reclassified Upward (%)	58.0	50.0	57.2	50.7
Non Cases	Reclassified Downward (%)	65.4	63.1	65.4	63.2
	Reclassified Upward (%)	34.6	36.9	34.6	37.0
Category-free NRI(%)		46.8	25.4	45.3	27.5
P-value		<0.001	0.002	<0.001	0.001

7a, Comparison of the C-index, LR test and BIC results demonstrating the discrimination for CHD prediction models based on the Suita Score with and without CKD, the FRS and the FRS calibrated for the means of the Suita cohort. The log likelihoods were derived from the multivariate adjusted Cox proportional hazard model.; CKD, chronic kidney disease; FHS, Framingham Heart Study; LR test, likelihood ratio test; BIC, Bayesian information criteria

7b, A comparison of the FRS and Suita Scores and the corresponding reclassification rate for the prediction of CHD events during a 10-year period;. NRI, net reclassification improvement

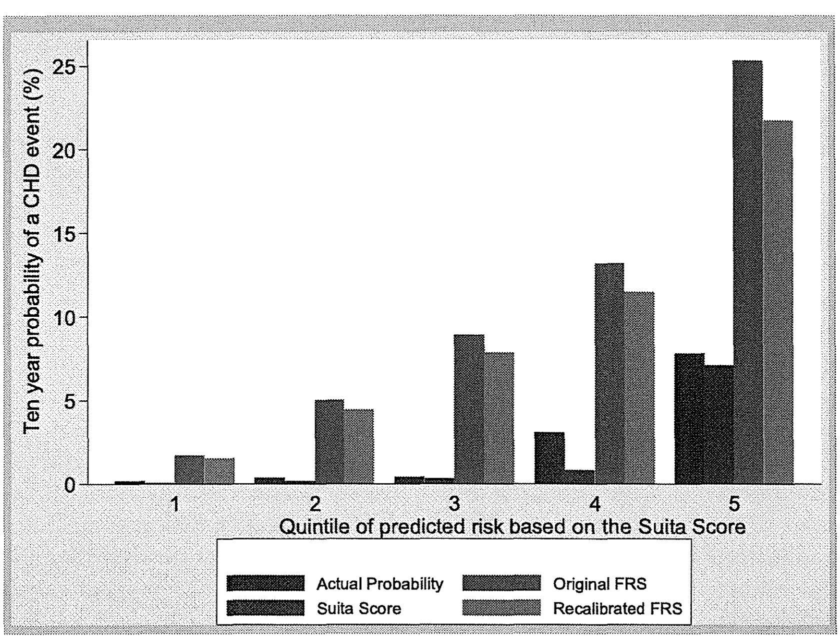

Fig. 2. The ten-year prediction of CHD events in the Suita study using the TC Suita Score with CKD

A graphical representation of the actual 10-year risk of cardiac events in the Suita cohort, along with the predicted risk and the Framingham risk function with and without recalibration for the means of the Suita cohort stratified by the quintile of predicted risk in the Suita cohort. The Suita participants were divided into quintiles of 10-year CHD risk predicted by the Suita score functions with CKD in Table 3. In each quintile, the mean predicted 10-year probabilities and actual probabilities were estimated. The Suita Score, the Suita Score with CKD shown in Table 3. FRS, Framingham risk score. CHD, coronary heart disease.

power, and the more simplified tool is as effective in the clinical setting as the original FRS, since both the models use beta coefficients and a simplified clinical score.

The incorporation of CKD yields limited improvement in the predictive capability in terms of C-statistics. However, the NRI and IDI showed marked improvement by the incorporation of CKD, which is a more clinically relevant index for prediction improvement. These two methods are becoming more popular and widely used in cardiovascular medicine[40, 41] For example, incorporating the homocysteine level into the FRS was evaluated by the NRI[42], since the inclusion of a new biomarker to the existing CHD risk score changed the predictability of events in a very marginal manner (less than 0.01 of the AUC)[43], and an enormously large odds ratio is needed for significant improvement[44, 45]. Clinicians currently do not have a tool for evaluating the CHD risk of patients with CKD but with relatively few other risk factors. These patients might be misjudged as having a very low risk.

Recently, an individualized risk prediction tool including more diverse risk factors increased 43% AMI and Strokes at the same cost[46]. Therefore, we believe that the inclusion of CKD in the prediction score is necessary and effective for populations at high risk for CHD. Currently, there are estimated to be more than 11 million CKD patients in Japan[47], and people have little doubt that CKD has a major impact on the population's health.

Our population had higher risks for developing CHD compared to other Japanese cohorts. The Suita cohort population was selected from an urban population, in contrast to the majority of other cohorts in Japan, which have been selected mainly from rural populations. Because approximately 66% of the Japanese population lives in urban areas according to 2006 Japanese Census[48], this is an important feature of our analysis. Interestingly, the JMS cohort and JALS reported that the crude incidence of AMI was 0.68 and 0.60 per 1000 person-years, respectively[11, 14, 15, 49]. On the contrary, the crude incidence of AMI in the Suita study was 1.40[50]. These findings may suggest that there is a large difference in the incidence of CHD between rural and urban areas in Japan. Thus, our tool is more useful for predicting the risk in urbanized populations with a higher risk of CHD.

Our study is associated with several limitations. First, the single assessment of risk factors at the baseline survey may have led to a regression dilution bias[51]. Second, the response rate of the original cohort was 53.1% (6,825/ 12,200) although the participants were randomly selected from the population of Suita city. In addition, based on the urbanized nature of the study population, it may not be possible to apply this tool in the whole Japanese population. However, since the outcome of the Suita study was the development of CHD, we believe that this tool can be a complement to the NIPPON DATA 80 risk score adopted in the JAS 2012 guidelines[11], in which the outcome was CHD mortality. The external validation of our score must be evaluated in other cohort studies, although a lack of external validation is a common problem with the existing Japanese risk prediction tools, including the NIPPON DATA 80, JALS, JMS cohort and Hisayama study. Considering the increasingly Westernized lifestyle in urban areas[52], these tools should be re-evaluated using a consortium of cohort studies, which include both urban and rural areas, such as the Epoch-Japan study group[53].

Very recently, the new AHA/ACC Guideline on the Treatment of Blood Cholesterol recommended the use of the new Pooled Cohort Equations to estimate the 10-year CHD risk in both white and black males and females, aged 40-75 years, and the FRS is no longer used for risk assessment[54]. However, this guideline is known to inaccurately estimate the CHD risk for Asians, Therefore, the value of the Suita Score for Japanese subjects and other low risk Asian populations is still superior to other systems.

Third, besides CKD, new biomarkers that can predict the CHD risk are emerging[55-57]. However, our study could not access their importance as have other existing prediction tools for Japanese subjects. For, example, the QRISK included rheumatoid arthritis, atrial fibrillation and the BMI. These relatively common, but not classic, cardiac risk factors must also be evaluated in future studies.

In conclusion, for Japanese subjects, the Suita prediction score with the CKD category resulted in better CHD prediction than the original and recalibrated FRS.

Conflict of Interest Disclosures

None.

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ORIGINAL ARTICLE

Blood Pressure, Low-Density Lipoprotein Cholesterol, and Incidences of Coronary Artery Disease and Ischemic Stroke in Japanese: The Suita Study

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BACKGROUND

Blood pressure (BP) and low-density lipoprotein cholesterol (LDL-C) are risk factors for coronary artery disease (CAD) and ischemic stroke. However, the hazards of their coexistence are not fully understood in Asian populations. We investigated whether the relationship between BP and cardiovascular disease (CVD) outcomes are modified by LDL-C level in a Japanese population.

METHODS

Individuals aged 30–79 years (n = 5,151) were classified into 6 groups according to LDL-C levels (<140 and ≥140 mg/dL or lipid medication) and BP levels (optimal BP, prehypertension, and hypertension; reference: low LDL-C and optimal BP). Hazard ratios (HRs) were calculated after adjusting for age, high-density lipoprotein cholesterol, diabetes, smoking status, and alcohol consumption. The effect modification of LDL-C on BP–CVD association was assessed using likelihood ratio tests.

RESULTS

There were 264 CAD and 215 ischemic stroke events during 13 years of follow-up. With low LDL-C, the HRs of prehypertension and hypertension for CAD were 2.01 and 4.71, respectively. Similar trends of HRs were observed with high LDL-C (optimal BP = 2.09, prehypertension = 3.45, hypertension = 5.94). However, the HRs for ischemic stroke did not differ between normal and high LDL-C levels at the same BP level. The apparent effect modification of LDL-C was not observed in the BP–CVD association in either CAD ($P = 0.48$) or ischemic stroke ($P = 0.39$).

CONCLUSIONS

The HRs for CAD in prehypertensive and hypertensive groups were higher than those in the optimal BP group at the same LDL-C levels in a Japanese population; however, there was no statistical effect modification of LDL-C on the BP–CAD association.

Keywords: Asian; blood pressure; cohort study; coronary artery disease; hypertension; incidence; ischemic stroke; low-density lipoprotein cholesterol; Suita Study.

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Cardiovascular disease is a leading cause of mortality and morbidity in Asian countries.[1] Elevated blood pressure (BP)[1–5] and hypercholesterolemia[1,6–10] are well-established independent cardiovascular risk factors. Moreover, the combination of these risk factors is a better predictor of the risk of cardiovascular disease in Western populations.[11,12] In Japan, the Japan Lipid Intervention Trial (J-LIT) study showed that Japanese hypercholesterolemia patients with high systolic BP (SBP; ≥130 mm Hg) and high total cholesterol levels (≥220 mg/dl) treated with low-dose simvastatin had an increased risk of cardiovascular disease events.[13] In Asia, the Asia Pacific Cohort Studies Collaborations (APCSC) demonstrated that the combination of high SBP (≥130 mm Hg) and high total cholesterol (≥212 mg/dl) increased the risks of fatal and nonfatal cardiovascular disease among both Western and Asian populations.[14] However, the J-LIT and APCSC studies have some drawbacks, including relatively short follow-up periods (mean follow-up period

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of approximately 6 years) and lipid profiles based on total cholesterol and not low-density lipoprotein cholesterol (LDL-C). Furthermore, the J-LIT study was a patient-based clinical trial,[13] and the APCSC study[14] did not exclusively involve Asian populations, which have a higher incidence of stroke and lower incidence of coronary artery disease (CAD) than Western populations.[1]

The purpose of our study was to examine whether the relationship between BP and CVD outcomes (CAD and ischemic stroke) is modified by LDL-C levels in a community-based cohort study in a Japanese population.

METHODS

Population

The Suita Study, a cohort study evaluating cardiovascular disease risk in an urban Japanese population, was established in 1989. This cohort study has been extensively used to evaluate risk factors associated with the incidences of CAD and stroke.[4,15–18] The details of this study have been described previously.[4,15–18] Briefly, 6,483 men and women aged 30–79 years underwent a baseline survey at the National Cerebral and Cardiovascular Centre (Japan) between September 1989 and March 1994. Subjects older than 80 years were excluded because it remains unconfirmed whether LDL-C is a risk factor for cardiovascular disease in the elderly population (aged ≥80 years).[19] A total of 1,332 participants were excluded for the following reasons: history of CAD or stroke (n = 208); loss to follow-up (n = 535); lack of participation in the baseline survey (n = 78); nonfasting visit (n = 239); triglyceride level >400 mg/dl (n = 86); LDL-C ≤0 (n = 1); missing total cholesterol, high-density lipoprotein cholesterol (HDL-C), or triglyceride data (n = 30); aged ≥80 years (n = 12); and other missing data (n = 145). Therefore, data from the remaining 5,151 participants (men: n = 2,399; women: n = 2,752) were included in our analysis.

This study was approved by the institutional review board of the National Cerebral and Cardiovascular Centre. Informed consent was obtained from all participants by health professions at the baseline examination. The collected data were anonymized.

Baseline examination

Blood samples were collected at the National Cerebral and Cardiovascular Centre after the participants had fasted for at least 10 hours. The samples were immediately centrifuged, and a routine blood examination that included serum total cholesterol, HDL-C, triglyceride, and glucose levels was performed. LDL-C was estimated for both men and women using the Friedewald formula.[20] Participants with triglyceride levels >400 mg/dl were excluded because LDL-C estimates are inaccurate among such persons.[19,21,22]

BP was measured by well-trained physicians using a standard mercury sphygmomanometer. After the participant had been in the seated position for 5 minutes, BP was measured 3 times on the right arm, and the average of the second and third measurements was used in the analyses to avoid bias due to white coat hypertension. Because HbA1c data from before 1995 were unavailable, diabetes was defined according to the American Diabetes Association 2013 guidelines as a fasting serum glucose level ≥126 mg/dl, the use of diabetes medication, or both.[23,24] Height and weight were measured while the subjects wore socks and light clothing. Public health nurses obtained information about smoking status, alcohol consumption, and medical history of the participants.

The information about smoking and alcohol consumption have been reported previously.[4,15–18] Well-trained nurses obtained information on smoking and alcohol consumption. Smoking status was classified as never, ex-smoker, or current smoker. If a participant responded yes to "current smoker," the number of cigarettes smoked per day was ascertained. Alcohol consumption was categorized as never drinker, ex-drinker, or current drinker (i.e., >1 time per week).

Endpoint determination

The endpoint determination in the Suita Study has been described previously.[4,15–18] The Suita Study is an ongoing cohort study, and the latest endpoint determination was performed on 31December 2007. The endpoints in our follow-up study were as follows: (i) the date of the first CAD or stroke event, (ii) the date of death, (iii) the date of leaving Suita City, and (iv) 31 December 2007.

The first step in the CAD and stroke survey involved checking the health status of all participants at biennial clinical visits every 2 years and through annual questionnaires sent by mail or administered by telephone. The second step involved the review of the in-hospital medical records of participants suspected of having had CAD or stroke; the reviews were performed by registered hospital physicians or research physicians blinded to the baseline information. The diagnosis of stroke was based on the US National Survey of Stroke criteria.[25] Stroke subtypes, including ischemic stroke, intracerebral hemorrhage, and subarachnoid hemorrhage, were diagnosed on the basis of computed tomography, magnetic resonance imaging, or autopsy results. Definite and probable acute myocardial infarction were defined according to the criteria of the Monitoring Trends and Determinants of Cardiovascular Disease (MONICA) Project.[26] In addition to acute myocardial infarction, the criteria for CAD diagnosis included sudden cardiac death within 24 hours from symptom onset or CAD followed by coronary artery bypass or angioplasty. Furthermore, myocardial infarction and stroke fatalities were recorded by searching for systematic death certificates.

Statistical analysis

BP was categorized into 3 groups; optimal BP (SBP <120 mm Hg and diastolic blood pressure (DBP) <80 mm Hg), prehypertension (SBP = 120–139 mm Hg, DBP = 80–89 mm Hg, or both), and hypertension (SBP ≥140 mm Hg, or DBP ≥90 mm Hg, or the use of antihypertensive agents). LDL-C was categorized into 2 groups; normal (LDL-C <140 mg/dl) or high (LDL-C ≥140 mg/dl or lipid medication) according to the diagnostic criteria for screening of the Japan Atherosclerosis Society Guidelines for Prevention of Atherosclerotic Cardiovascular Diseases.[19]

This guideline has set the cutoff point of LDL-C to 140 mg/dl as diagnostic criteria of dyslipidemia. Using the abovementioned BP and LDL-C categories, combinations of BP and LDL-C (3 × 2 groups) were made to estimate hazard ratios (HRs) with optimal BP and normal LDL-C as the reference group. Analysis of variance and the Bonferroni test were used to compare continuous variables, and the χ^2 test was used to compare dichotomous variables.

Sex-stratified Cox proportional hazard models, accounting for different baseline hazards in men and women, were used to estimate the HRs of the combination of BP and LDL-C on cardiovascular disease outcomes. Age, HDL-C, smoking status, alcohol consumption, and diabetes were included in the models as confounders. For the primary analysis, HRs and 95% confidence intervals (CIs) were estimated for CAD and stroke events by analyzing BP as a categorical variable within each LDL-C group. Moreover, the association between BP and LDL-C was examined by comparing the HRs for CAD and stroke events across the 6 groups, adjusting for age, HDL-C, diabetes, smoking status, and alcohol consumption. The interaction between BP and LDL-C on cardiovascular outcomes was assessed using likelihood ratio tests.[27] The level of significance was set at $P < 0.05$. All statistical analyses were performed using STATA release 12 (Stata Corp LP, College Station, TX).

RESULTS

Overall, 5,151 individuals (men: n = 2,399; women: n = 2,752) were analyzed. Table 1 shows the baseline characteristics of groups with BP and LDL-C combination in men. In both LDL-C groups, men with hypertension had the highest mean age, body mass index, and fasting blood glucose, as well as the most cases of diabetes and medications. There were fewer current drinkers in the high LDL-C group than the normal LDL-C group ($P < 0.001$). Moreover, there were more current smokers in the normal LDL-C group than the high LDL-C group ($P < 0.001$).

Table 2 shows the means and prevalence of baseline characteristics with respect to BP and LDL-C groups in women. As with the men, in both LDL-C groups, women with hypertension had the highest mean age, body mass index, and fasting blood glucose, as well as the most cases of diabetes and medications. There were fewer current drinkers in the high LDL-C group than the normal LDL-C group ($P < 0.001$). Moreover, there were more current smokers in the normal LDL-C group than in high LDL-C group ($P < 0.001$).

The associations of alcohol consumption and smoking status with LDL-C levels were similar in both sexes. In addition, the rates of current smokers and drinkers were obviously higher in men than women (Tables 1 and 2).

During the 13-year follow-up (total = 67,287 person-years), 164 CAD cases (men: n = 110; women: n = 54) and 215 stroke cases (ischemic: n = 126; hemorrhagic: n = 48; subarachnoid hemorrhage: n = 22; and unclassified stroke: n = 19) were documented. The adjusted HRs for CAD in hypertension were highest in both the normal and the high LDL-C groups. The high HRs were observed as both BP and LDL-C upgraded in CAD (Figure 1). In the high LDL-C group, the HRs of CAD with optimal BP, prehypertension, and hypertension were 2.09 (95% CI = 0.88–4.98), 3.45 (95% CI = 1.59–7.51), and 5.94 (95% CI = 2.88–12.27), respectively. In the normal LDL-C group, the HR of CAD with prehypertension (2.01; 95% CI = 0.92–4.42) was almost the same as those with optimal BP and high LDL-C, whereas the HR with hypertension (2.95; 95% CI = 1.45–5.9) was significantly higher (Figure 1). The HR for ischemic stroke was 2.70 (95% CI = 1.37–5.35) in the hypertension and normal LDL-C group and 2.95 (95% CI = 1.47–5.90) in the hypertension and high LDL-C group. No apparent interaction between BP and LDL-C was detected with either CAD ($P = 0.48$) or ischemic stroke ($P = 0.39$).

These results have almost no differences when medication use (lipid-lowering medicine and BP-lowering medicine) is not considered the BP and LDL classification.

DISCUSSION

This study showed that the HRs for CAD in prehypertensive and hypertensive groups were higher than those in the optimal BP group at the same LDL-C levels in the Japanese population; however, 95% CIs for these groups almost overlapped, and no apparent modification by LDL-C was observed in the BP–CVD relationship. Furthermore, the HRs for ischemic stroke were not different between normal and higher LDL-C levels at the same BP levels.

Our results support the results from the APCSC, which demonstrated that the combination of elevated BP and elevated total cholesterol increases the risks of fatal and nonfatal cardiovascular disease in Asian, Australian, and New Zealand populations.[14] The APCSC showed that cardiovascular disease events are particularly increased in individuals with SBP ≥130 mm Hg and the highest total cholesterol levels (≥212 mg/dl). Furthermore, the relative risk of cardiovascular disease events in individuals with an SBP of 130–144 mm Hg and total cholesterol levels of 212–241 mg/dl is similar to that of individuals with an SBP of 145–159 mm Hg and total cholesterol levels <212 mg/dl. The J-LIT study, which was an observational study among Japanese patients that investigated the relationship between total cholesterol and BP on cardiovascular disease, also found that the relative risk of cardiovascular disease events was significantly higher in patients with poorly controlled hypercholesterolemia patients (total cholesterol >220 mg/dl), prehypertension (SBP = 130–139 mm Hg; DBP = 80–89 mm Hg), and hypertension (SBP >140 mm Hg; DBP >90 mm Hg) compared with the reference group (SBP <130 mm Hg; DBP <80 mm Hg).[13] Thus, our findings are concordant with those of the APCSC and J-LIT studies. However, it should be emphasized that our study cohort consisted exclusively of a general Asian population in contrast with the APCSC. In addition, hypercholesterolemia patients in the J-LIT study were treated with low-dose simvastatin.

The HRs for ischemic stroke did not differ between the normal and high LDL-C groups at the same BP levels. However, most cohort studies in Japan report that total cholesterol and LDL-C are not risk factors for total stroke[28] despite their

Table 1. Baseline characteristics of women with respect to blood pressure and serum low-density lipoprotein categories in men

	Normal LDL-C			High LDL-C			
	Optimal BP	Prehypertension	Hypertension	Optimal BP	Prehypertension	Hypertension	*P* value
No.	543	593	491	232	269	271	
Age, y	49.5 (13.2)	54.8 (13.2)**	61.3 (11.4)**	52.4 (12.5)*	56.2 (11.5)**	60.7 (10.8)**	<0.001
Systolic blood pressure, mm Hg	107.6 (7.5)	126.1 (7.3)**	150.8 (17.6)**	108.3 (7.4)*	126.3 (7.7)**	149.3 (16)**	<0.001
Diastolic blood pressure, mm Hg	67.9 (7)	67.9 (7)**	90.1 (10.6)**	69.3 (6.3)	79.5 (6.8)**	90.1 (10.3)**	<0.001
LDL-C, mg/dl	108.7 (20.5)	109.2 (20.7)*	108.6 (21.6)**	159.9 (18.5)**	159.8 (19.4)**	161.5 (22.5)**	<0.001
Fasting blood glucose, mg/dl	96.5 (13)	100.6 (17.8)*	102.6 (19.4)**	98.0 (20.7)**	102.9 (22.7)**	104.2 (21.2)**	<0.001
Body mass index, kg/m^2	21.8 (2.7)	22.7 (2.7)**	23.1 (3)**	22.6 (2.5)*	23.3 (2.6)**	23.8 (2.7)**	<0.001
Diabetes, %	3.5	5.4	7.9	3.4	4.8	10	0.004
Medication for hypertension, %	0	0	49.7	0	0	34.3	
Medication for hypercholesterolemia, %	0	0	0	2.6	5.6	8.5	
Medication for diabetes, %	0.9	2	2	1.3	1.9	5.2	0.001
Smoking, %							<0.001
Current smoker	60.6	49.7	43.6	32.8	44.2	41.7	
Ex-smoker	22.7	29.3	36.9	67.8	32.7	40.6	
Never smoker	16.8	20.9	19.6	43.3	23	17.7	
Alcohol consumption, %							<0.001
Current drinker	74.2	78.8	79.6	65.1	72.9	73.1	
Ex-drinker	2.2	3.9	3.1	3.9	4.8	6.6	
Never drinker	23.6	17.4	17.3	31	22.3	20.3	

Age, systolic blood pressure, diastolic blood pressure, low-density lipoprotein cholesterol (LDL-C), fasting blood glucose, and body mass index were analyzed by analysis of variance. The percentages of diabetes, medication for hypertension, medication for hypercholesterolemia, medication for diabetes, smoking, and alcohol consumption were analyzed by the χ^2 test. Data are expressed as mean (SD) or percentages. Optimal BP: systolic blood pressure <120 mm Hg and diastolic blood pressure <80 mm Hg. Prehypertension: systolic blood pressure 120–139 mm Hg or diastolic blood pressure 80–89 mm Hg. Hypertension: systolic blood pressure ≥140 mm Hg, diastolic blood pressure ≥90 mm Hg, or the use of antihypertensive medication. Normal LDL-C: fasting LDL-C <140 mg/dl. High LDL-C: fasting LDL-C ≥140 mg/dl or the use of medication for hypercholesterolemia. Diabetes: fasting plasma glucose ≥126 mg/dl or the use of antidiabetic medication.

Abbreviations: BP, blood pressure; LDL-C, low-density lipoprotein cholesterol.

*P < 0.05, **P < 0.001: Bonferroni test (with normal LDL-C and optimal BP as the reference).

Table 2. Baseline characteristics of women with respect to blood pressure and serum LDL-C categories in women

	Normal LDL-C						High LDL-C						
	Optimal BP		Prehypertension		Hypertension		Optimal BP		Prehypertension		Hypertension		*P* value
No.	837		427		311		364		403		410		
Age, y	44.9	(11.3)	53.2	(12.4)**	61.9	(10.4)**	53.3	(10.8)**	57.9	(10.1)**	62.5	(8.9)**	<0.001
Systolic blood pressure, mm Hg	104.8	(8.1)	126.3	(7.1)**	151.8	(16.9)**	106.9	(7.5)	127.5	(6.8)**	151.4	(16.6)**	<0.001
Diastolic blood pressure, mm Hg	66	(6.6)	77.4	(6.8)**	86.3	(11.2)**	67	(6.8)	77.7	(7)**	86.8	(11.3)**	<0.001
LDL-C, mg/dl	108.9	(19.4)	114.2	(18.2)	115.4	(18.1)	163.3	(23.6)**	167.2	(26.6)**	168	(26.3)**	<0.001
Fasting blood glucose, mg/dl	91.3	(8.3)	96.2	(14.7)**	100.1	(20.5)**	93.5	(11.4)	98.6	(22.7)**	101.9	(18.3)**	<0.001
Body mass index, kg/m^2	20.9	(2.65)	22.2	(3.03)**	23.2	(3.81)**	21.7	(2.82)*	22.9	(3.08)**	23.8	(3.43)**	<0.001
Diabetes, %	1.2		2.3		6.1		1.4		4.5		6.3		<0.001
Medication for hypertension, %	0		0		37.6		0		0		39.3		
Medication for hypercholesterolemia, %	0		0		0		3.6		5.2		7.6		
Medication for diabetes, %	0.6		0.9		2.3		0.5		1.5		2		0.09
Smoking, %													<0.001
Current smoker	16.4		9.8		8.7		13.7		10.2		7.6		
Ex-smoker	3.8		3.5		2.3		2.7		3.2		5.6		
Never smoker	79.8		86.7		89.1		83.5		86.6		86.8		
Alcohol consumption, %													<0.001
Current drinker	41.1		33		29.3		26.1		29.3		27.8		
Ex-drinker	2		1.6		1.9		1.6		1.7		1.7		
Never drinker	56.9		65.3		68.8		72.3		69		70.5		

Age, systolic blood pressure, diastolic blood pressure, low-density lipoprotein cholesterol (LDL-C), fasting blood glucose, and body mass index were analyzed by analysis of variance. The percentages of diabetes, medication for hypertension, medication for hypercholesterolemia, medication for diabetes, smoking, and alcohol consumption were analyzed by the χ^2 test. Data are expressed as mean (SD) or percentages. Optimal BP: systolic blood pressure <120 mm Hg and diastolic blood pressure <80 mm Hg. Prehypertension: systolic blood pressure 120–139 mm Hg or diastolic blood pressure 80–89 mm Hg. Hypertension: systolic blood pressure ≥140 mm Hg, diastolic blood pressure ≥90 mm Hg, or the use of antihypertensive medication. Normal LDL-C: fasting LDL-C <140 mg/dl. High LDL-C: fasting LDL-C ≥140 mg/dl or the use of medication for hypercholesterolemia. Diabetes: fasting plasma glucose ≥126 mg/dl or the use of antidiabetic medication. When mean body mass index was examined in women, there were 836 women with normal blood pressure and normal LDL-C.

Abbreviation: BP, blood pressure.

*$P < 0.05$, **$P < 0.001$: Bonferroni test (with normal LDL-C and optimal BP as the reference).

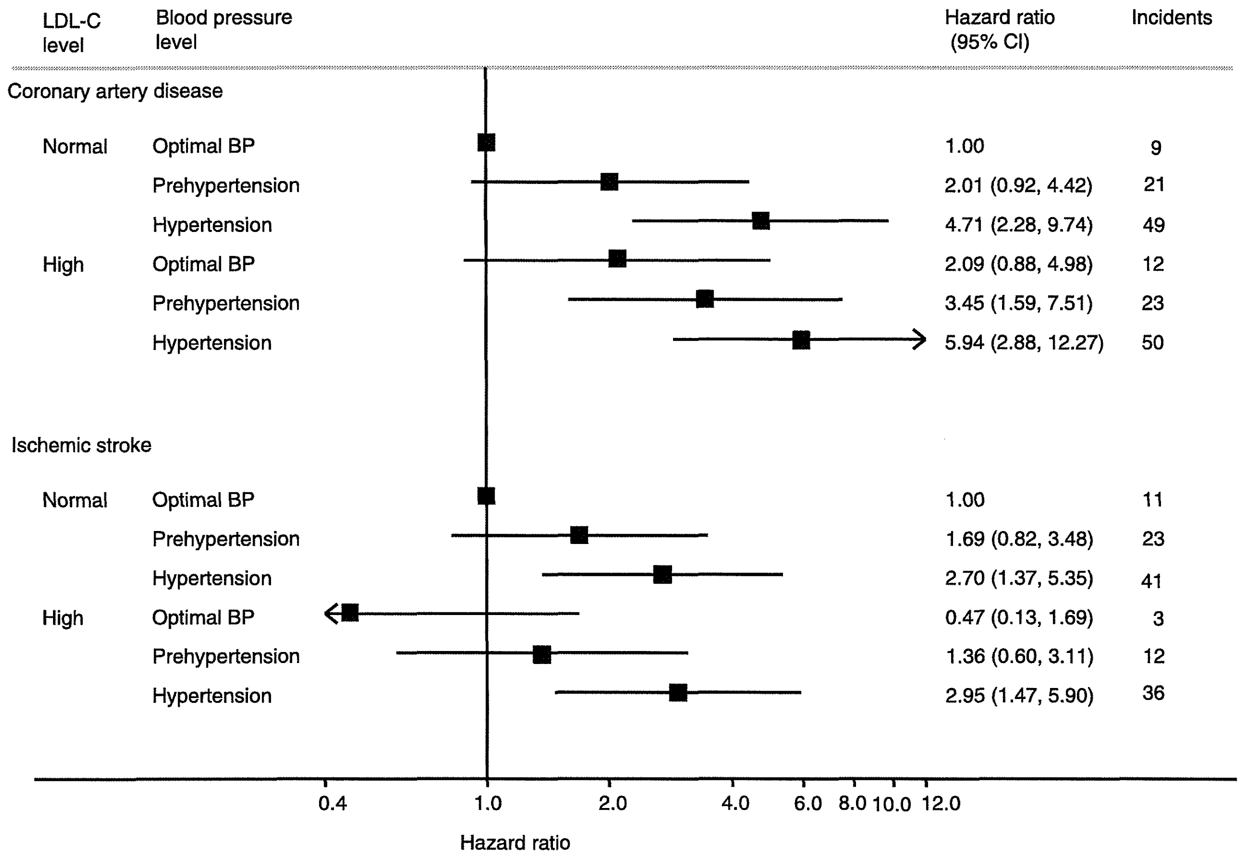

Figure 1. Hazard ratios (HRs) for coronary artery disease and stroke by blood pressure (BP) group with respect to low-density lipoprotein cholesterol (LDL-C; mg/dl) categories adjusted for age, high-density lipoprotein cholesterol, diabetes, smoking status, and alcohol consumption. ■ indicate HR estimates: ▬ indicate 95% confidence intervals (CIs) by Cox proportional hazard model stratified by sex. The reference group had optimal BP (systolic BP <120 mm Hg and diastolic BP <80 mm Hg) and normal LDL-C levels (LDL-C <140 mg/dl). $P = 0.48$ for CAD; $P = 0.39$ for ischemic stroke.

weak association with ischemic stroke.[29] Given these contradictory results between clinical trials and cohort studies, the effects of statins on the prevention of stroke should be interpreted cautiously. Several studies report cholesterol-lowering statins are beneficial for the prevention of stroke in hypertensive patients.[30,31] The post hoc analysis of the Management of Elevated Cholesterol in the Primary Prevention Group of Adult Japanese study revealed that pravastatin effectively reduced the incidence of cardiovascular disease, particularly ischemic stroke, in individuals with both hypertension and mildly elevated cholesterol.[30] Meanwhile, the Anglo–Scandinavian Cardiac Outcomes Trial–Lipid Lowering Arm showed that atorvastatin significantly reduces CAD, stroke, and cardiovascular disease even in hypertensive patients without dyslipidemia.[31] Although there have been numerous clinical trials for statin therapy, the primary endpoint of these studies was CAD, with ischemic stroke as the secondary endpoint. Furthermore, statins have pleiotropic effects that help prevent cardiovascular disease, including anti-inflammatory effects, improved vascular endothelial function, and plaque stabilization. These factors may explain the significant discrepancy between cohort studies and clinical trials with respect to the efficacy of statins in stroke prevention. Therefore, further investigation is required to clarify the role of statin therapy in stroke prevention. However, BP management should be the first priority in ischemic stroke prevention irrespective of LDL-C level.

Our study used a stratified Cox model that included 3 BP and 2 LDL-C categories, as well as their interaction terms and confounders. This combined model is statistically appropriate for the investigation of interactions between risk factors and disease outcomes. Furthermore, LDL-C and hypertension, which were the main targets of this study, and the abovementioned confounding factors encompass all major risk factors in the Framingham risk score,[22] the European SCORE chart,[21] and the Japanese Atherosclerosis Society risk chart[19] for predicting future coronary events.

We found that the HRs for CAD in high LDL-C group were higher than those in normal LDL-C at the same BP levels in the Japanese population. However, the apparent effect modification of LDL-C was not detected in the relation between BP and CAD. There are 3 possibilities to explain these results; no interaction, low statistical power, and bias. Our results suggested that BP and LDL-C were mutually independent risk factors and no interaction exist between them. This result was not as similar to other previous studies, and explanations were needed to claim the independence. A second possibility is that lack of statistical power induces the results; a very small number of events was assigned in each category. The third possibility is that the single assessment of BP and LDL-C at the baseline survey and the fact that we did not evaluate the longitudinal trend for each risk factor may have underestimated the relationship between these conditions and CAD because of regression dilution bias.[32] All of these misclassification diluted the HRs and made the effect modification obscure. We are not quite sure which is the correct answer for this issue, but we believe our description of the BP, LDL-C, and CAD relationship among an Asian population gives important insight to future epidemiological studies.

In conclusion, to the best of our knowledge, this study is the first epidemiological study to examine the combined association between BP and LDL-C on the incidences of cardiovascular disease subtypes in an Asian population only. The results show that risk for CAD due to hypertension or prehypertension in the high LDL-C group was higher than those in the normal LDL-C group, although there were no statistical interaction between BP and LDL-C. Accordingly, BP and LDL-C should be managed for the early prevention of CAD in Japanese and other Asian individuals with hypertension, prehypertension, and high LDL-C levels. Furthermore, large-scale epidemiological studies should carefully assess the association between BP and LDL-C with the incidence of cardiovascular disease subtypes among Asian populations.

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DISCLOSURE

The authors declared no conflict of interest.

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Original Article

Serum Apolipoprotein B-48 Concentration Is Associated with a Reduced Estimated Glomerular Filtration Rate and Increased Proteinuria

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Aim: Apolipoprotein B-48 (apoB-48) is a constituent of chylomicrons and their remnants (chylomicron remnants). A high concentration of serum apoB-48 is suspected to be a major risk factor for the development of atherosclerotic cardiovascular disease. Proteinuria and a reduced estimated glomerular filtration rate (eGFR) are independent risk factors for cardiovascular events and renal dysfunction. In the present study, we examined whether the serum apoB-48 concentration is associated with renal dysfunction.

Methods: A total of 264 patients was enrolled and classified into four groups according to the eGFR and level of proteinuria: a high eGFR (>60 mL/min/1.73 m^2) without proteinuria (≥1+ by urine dipstick) (n=50); a high eGFR with proteinuria (n=75); a low eGFR (>60 mL/min/1.73 m^2) without proteinuria (n=74); and a low eGFR with proteinuria (n=65). Biochemical markers of lipid metabolism, including the fasting serum apoB-48 concentration, were compared between the four groups.

Results: The serum log-apoB-48 and log-apoB-48/TG levels were significantly higher in the patients with a high eGFR with proteinuria, low eGFR with proteinuria and low eGFR without proteinuria than in those with a high eGFR without proteinuria, with the most significant differences for these parameters. The eGFR was found to be significantly correlated with the log-apoB-48 and log-apoB-48/TG levels, whereas urinary protein was found to be significantly correlated with the log-apoB-48 level only. A multiple regression analysis indicated that the log-apoB-48/TG level was a significant determinant of a reduced eGFR.

Conclusions: Both a low eGFR (<60) and proteinuria (≥1+) are independent determinants of a high apoB-48 concentration. Taken together, the present results suggest that an increased serum apoB-48 concentration contributes to an increased risk of cardiovascular events.

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Key words: Apolipoprotein B-48, Chylomicron remnants, Chronic kidney disease, eGFR, Proteinuria

Introduction

Proteinuria is an independent risk factor for the progression of chronic kidney disease (CKD) and the development of cardiovascular disease (CVD)[1, 2]. Patients with CKD are at a substantially increased risk of developing atherosclerosis, and approximately 40% of CKD patients have CVD even before they reach end-stage renal failure requiring hemodialysis[3]. The cardiovascular mortality is 30 times higher among hemodialysis patients than individuals from the general population[4]. In an effort to standardize the stage

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of CKD, a system was recently developed based on the calculation of the estimated glomerular filtration rate (eGFR) using the serum creatinine level and age[5]. Recent data have demonstrated the impact of albuminuria, as well as eGFR, on the incidence of CVD, and it is evident that both measurements together confer an enhanced risk of CVD[6]. Recently, the prevalence of CKD has been increasing, approximately 13.3 million adults with CKD living in Japan[7]. Therefore, CKD is a major health problem worldwide, and its prevention is an urgent issue.

Dyslipidemia associated with CKD is characterized by an increased level of serum triglycerides (TG) and a decreased level of high-density lipoprotein (HDL) cholesterol (HDL-C)[8]. A high level of low-density lipoprotein (LDL) cholesterol (LDL-C) has been established as a strong risk factor for CVD; however, it is not consistently observed among patients with CKD[9]. It has also been reported that individuals with CKD have elevated levels of TG-rich lipoproteins (TRL), including chylomicrons (CM), very-low-density lipoproteins (VLDL) and their remnant lipoproteins[5]. Furthermore, many basic studies have suggested that remnant lipoproteins are associated with an increased risk of cardiovascular disease[10]. The accumulation of CM remnant (CM-R) particles, the hydrolyzed products of CMs produced by lipoprotein lipase (LPL), are related to foam cell formation among macrophages and may promote the development of atherosclerosis in the arterial wall[11, 12]. It is presumed that measurements of the serum apoB-48 concentration can be used to evaluate the synthesis and metabolism of CM and CM-R, as each contains one apolipoprotein B-48 (apoB-48) molecule per particle[13]. We previously developed a novel enzyme-linked immunosorbent assay (ELISA)[14, 15] and chemiluminescent enzyme immunoassay (CLEIA)[16] for conveniently determining the serum apoB-48 concentration. In our previous study, we reported that the serum apoB-48 concentrations are substantially elevated in patients with CKD of stage 4 (eGFR: 15-29 mL/min/1.73 m^2) and 5 (eGFR: <15 mL/min/1.73 m^2)[17]. Recently, Japanese guidelines for the diagnosis and treatment of CKD[18] were released that indicate that both an increased GFR and the existence of proteinuria may exacerbate the pathogenesis of CKD. However, few studies conducted to date have investigated the association between the fasting serum apoB-48 concentration and proteinuria and/or eGFR.

In the current study, we examined whether the serum apoB-48 concentration is associated with renal dysfunction in patients with a low eGFR and/or proteinuria.

Materials and Methods

Subjects

The study subjects included 264 patients who attended the outpatient clinic of Osaka University Hospital (Osaka, Japan) or who were admitted to the hospital. Subjects taking lipid-lowering medications and those with heart, liver or thyroid disease were excluded. All subjects provided their informed consent, and the research protocol was approved by the Ethics Committee of Osaka University Hospital.

Measurements of the Renal Function, Serum Lipids and Apolipoproteins

In each subject, a sample of blood was drawn from the cubital vein after an overnight fast (over 12 hours), and the serum was collected following brief centrifugation. The basal performance of the recently developed CLEIA for apoB-48 measurement kit (Fujirebio Inc., Tokyo, Japan) used in this study has been previously reported[16], and the assay was carried out on the Lumipulse *f* fully automated immunoassay analyzer (Fujirebio, Inc.). The serum total cholesterol (TC), TG and HDL-C levels were determined according to enzymatic methods, while the serum LDL-C levels were determined using the direct method (Sekisui Medical Co., Ltd., Tokyo, Japan). The non-HDL-cholesterol levels were calculated by subtracting the value of HDL-C from that of serum TC. The serum creatinine levels were measured using a photometric assay (Wako, Osaka, Japan). The urine test results were classified as (−), (±), (+), (2+) or (3+).

The eGFR values were subsequently estimated using the new equation proposed by the Japanese Society of Nephrology[19]: eGFR (mL/min/1.73 m^2) = $194 \times \text{S-Cr}^{-1.094} \times \text{Age}^{-0.287}$ (for female patients, the total value was multiplied by 0.739). An eGFR of < 60 mL/min/1.73 m^2 is generally considered to be indicative of CKD; therefore, subjects with an eGFR of <60 mL/min/1.73 m^2 (eGFR <60) were defined as having a low eGFR. As for proteinuria, (−) and (±) urine test results were defined as normal; all other results [(≥1+);(+), (2+) or (3+)] were defined as indicating proteinuria. All samples were treated in accordance with the Helsinki Declaration.

Statistical Analysis

Before the analysis, skewed variables (TG, apoB-48 and apoB-48/TG ratio) were logarithmically transformed to improve data normalization. The data are expressed as the mean ± SD for continuous variables and percentages for categorical variables. The significance of differences in the systolic BP, diastolic BP,

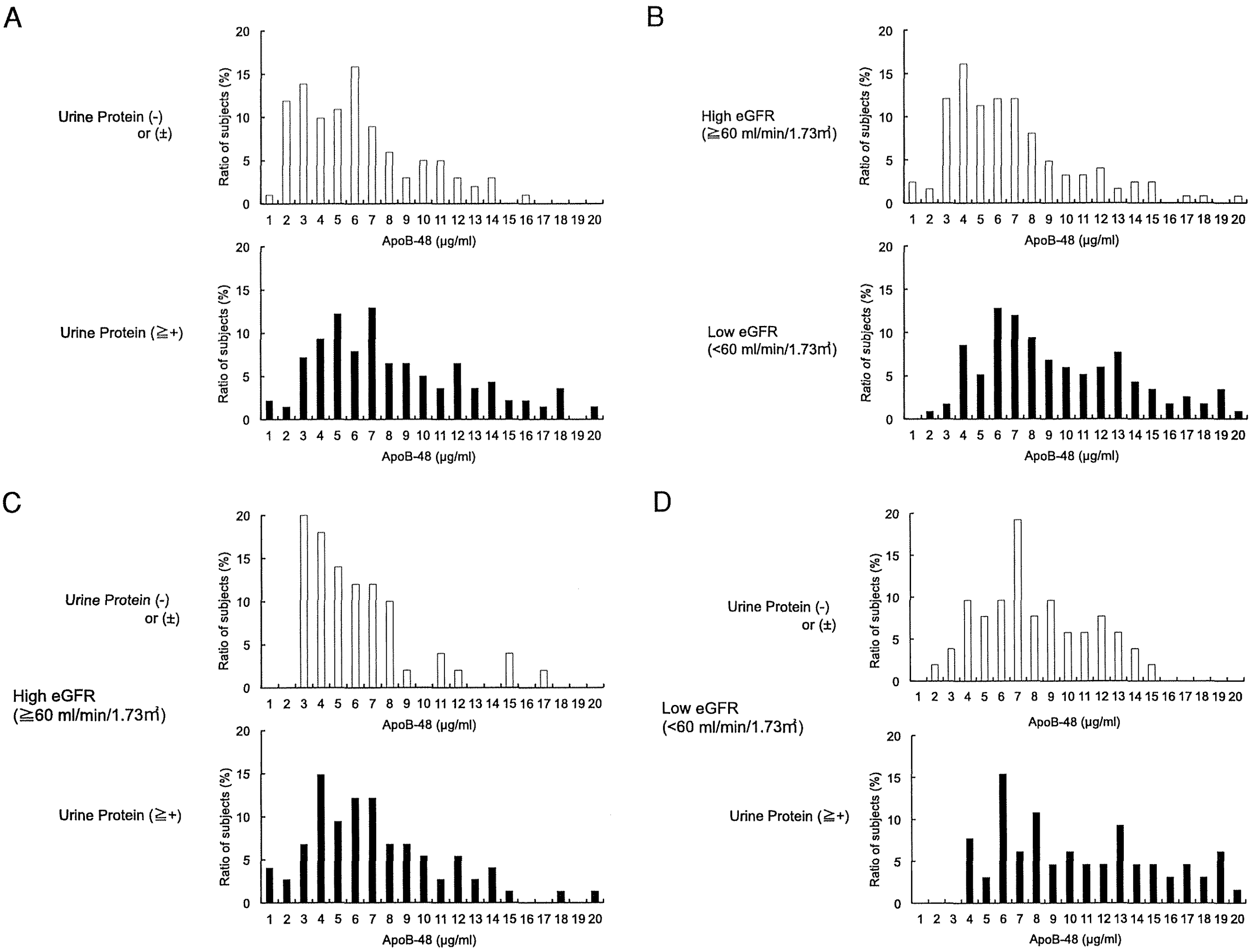

Fig. 1. Distribution of the fasting serum apoB-48 concentrations in the subjects with and without urinary protein, the subjects with a low and high eGFR and the subjects with both eGFR and proteinuria.

Serum apoB-48 concentration = 1 represents a concentration between 0.0 and 1.0 μg/mL.
A) Fasting serum apoB-48 concentrations in the subjects without urinary protein (open squares, $n = 124$) and with urinary protein (closed squares, $n = 140$).
B) Fasting serum apoB-48 concentrations in the subjects with a high eGFR (open squares, $n = 125$) and a low eGFR (closed squares, $n = 139$).
C) Fasting serum apoB-48 concentrations in the subjects with a combination of a high eGFR without urinary protein (open squares, $n = 50$) and a high eGFR with urinary protein (closed squares, $n = 75$).
D) Fasting serum apoB-48 concentrations in the subjects with a combination of a low eGFR without urinary protein (open squares, $n = 74$) and a high eGFR with urinary protein (closed squares, $n = 65$).

UA, TC, TG, HDL-C, LDL-C, non-HDL-C, log-apoB-48 and log-apoB-48/TG values between the four groups was tested using an analysis of variance (ANOVA) with the least significant difference (LSD) test as a post hoc test and the Bonferroni correction for multiple comparisons. The significance of differences with respect to the prevalence of diabetes and drug treatment were tested using the χ^2 test. Correlations between the kidney function parameters (eGFR and urinary protein) and various other parameters were analyzed using Spearman's rank correlation coefficient, and a stepwise multiple logistic regression analysis was performed to determine independent predictors of CKD. The statistical analyses were conducted using the JMP8 software program (SAS Institute, Cary, NC). Statistical significance was established at a p value of < 0.05.