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ORIGINAL ARTICLE

A cross-sectional study on association of calcium intake with blood pressure in Japanese population

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To investigate the association of calcium intake independently of other nutrients already known as predictors of hypertension, a cross-sectional study was carried out on the same population in Japan as used for the INTERSALT study. Dietary calcium intake was estimated from a 1-day 24-h recall. Sodium and potassium intakes were evaluated by 24-h urinary excretion. Data from 476 subjects aged 20–59 years, 230 men and 246 women, were analysed. The mean dietary calcium intake ranged from 557 to 608 mg/day among men, and from 528 to 639 mg/day among women. Among men, the pooled estimate of the regression coefficients of blood pressure (mm Hg) per 100 mg increase of calcium intake, adjusted for age and body mass index (BMI), were −0.42 mm Hg for systolic blood pressure (SBP) and −0.35 mm Hg for diastolic blood pressure (DBP), but there was no statistical significance. Among women, the pooled estimates of regression coefficients adjusted for age and BMI were −0.92 mm Hg for SBP and −0.83 mm Hg for DBP with statistical significance. After adjustment for age, BMI, alcohol intake and urinary excretion of sodium and potassium, the pooled estimate of calcium intake was −0.66 mm Hg for DBP with statistical significance and − 0.70 mm Hg for SBP. A significant negative association of calcium intake with blood pressure was observed among the subjects in Osaka. Our study suggests that increased calcium intake may provide a benefit of lowering blood pressure independently of other minerals such as sodium and potassium.
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Introduction

There have been many epidemiological studies concerned with the effect of calcium intake on blood pressure.[1,2] A meta-analysis of published results found a small protective effect of calcium intake on blood pressure.[3,4] However, the results of the different studies were inconsistent.[5–16] Some methodological problems were considered to be responsible for this heterogeneity. For example, differences in the method of sampling, dietary data collection and assessing calcium intake are possible causes.[1] In addition, high correlation between calcium and other nutrients has led to problems with multicolinearity, which must be considered. We should also consider the interaction with other well-known predictors for hypertension, such as sodium, potassium, obesity and alcohol intake.[17–19]

The aim of this report is to investigate the association of calcium intake with blood pressure, independently of other minerals already known as predictors of hypertension. It has been designed as a reinvestigation of the Japanese population which participated in the INTERSALT study,[17] who showed a high sodium intake and low potassium intake as risk factors for high blood pressure. Among the 52 centres of the INTERSALT study, three Japanese centres were in a group of high sodium intake and low potassium intake.[20] One of the main aims of this reinvestigation was to investigate the trends of blood pressure and intake of these electrolytes on which we have already reported.[21] Another aim was to investigate the factors related to blood pressure by adding 24-h recall for estimation of dietary intake, because some minerals like calcium are estimated better by dietary data than urinary excretion measurement.

We examined the association of calcium intake

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with blood pressure in men and women separately, by each district (Osaka, Toyama, Tochigi), independent of body mass index (BMI), alcohol intake, and urinary excretion of sodium and potassium.

Materials and methods

This is a cross-sectional study to investigate the effects of electrolytes intake on blood pressure based on the Japanese INTERSALT study. The methodology for subject selection, blood pressure measurements and urine collection was the same as for the INTERSALT study.[22] This methodology has been described previously.[21] The same three centres serving three districts in Japan were used as the basis for the new study. Detailed characteristics of these districts, Osaka, Tochigi and Toyama, were described in a previous study.[21] To summarise, 200 people with complete measurements were recruited from each centre from each of eight sex–age strata. Within each stratum, a total of 25 people were selected at random, with one extra person selected in case one subject failed to make their appointment or to complete the 24-h urine collection procedure. The response rates in Osaka, Tochigi and Toyama were 30%, 28%, and 56%, respectively.

Blood pressure measurements were performed after a 30-min period during which smoking, eating and vigorous exercise were prohibited. Immediately prior to the measurements the subject was asked to empty their bladder and assume a sitting position and rest for 5 min. Then, using a random-zero sphygmomanometer the blood pressure was measured twice on the right arm and the mean value adopted. Diastolic blood pressure (DBP) was recorded as Korotkoff's phase 5.

Calcium intake was estimated from a 1-day 24-h recall which was done by dietician's interviews based on 24-h dietary records by participants and calculated using the fourth revision of the standard Japanese food table. Urine collection was performed by the total urine collection method. Sodium and potassium intakes were evaluated from 24-h urinary excretion. Calcium intake was evaluated from 24-h dietary recall. The 24-h collection was started immediately after voiding the spot urine. To ensure that the end of the 24-h urine collection was appropriately completed, the participants were asked to return to the clinic shortly before the 24 hours were over, and the final urine specimen was then collected. The amount of alcohol consumed during the 1 week prior to the investigation was calculated by having the subjects recall. Height and weight were measured with the subject wearing only socks and light clothes. Questionnaires were used to ascertain the completeness of the 24-h urine collection.

The sex and age distributions of the subjects are shown in Table 1. In the present analysis, 476 subjects aged 20–59 years for whom urine collections, dietary recall and blood pressure measurements were complete were selected. The subjects who were under treatment for hypertension were eliminated. In Tochigi centre the participation rate was the lowest because urbanisation in this area has caused a decrease in a viable source of subject recruitment.

Statistical methods

Age adjusted means of blood pressure and mineral intake were calculated by analysis of covariance. Adjustment was also made for age, BMI, alcohol intake and other nutrients by using multiple regression analysis. To adjust for drinking habits, alcohol intakes were stratified into three groups (0, 1–299 ml, and ⩾300 ml absolute alcohol/week) with two 0, 1 variables, that is, no alcohol intake *vs* 1–299 ml/week (0,1) and no alcohol intake *vs* ⩾300 ml/week (0,1).

Results

Table 2 shows age-adjusted mean of variables and prevalence of alcohol intake by sex in each centre. Among men, there were statistically significant differences between centres in 24-h urinary sodium excretion. Dietary calcium intake was the highest in Osaka, followed by Tochigi and Toyama (with means of 608, 589 and 557 mg/day, respectively). Toyama was characterised by its higher urinary sodium excretion compared with others. Among women, there were statistically significant differences between centres in DBP, urinary sodium and potassium excretion and urinary sodium/potassium ratio, and dietary calcium intake. Dietary calcium intake was the highest in Tochigi, followed by Osaka and Toyama (with means of 639, 597 and

Table 1 Age and sex distribution of participants in each centre

Age class	*Men*				*Women*				*Total*
	Toyama	*Osaka*	*Tochigi*	*Total*	*Toyama*	*Osaka*	*Tochigi*	*Total*	
20–29	25	25	2	52	25	21	7	53	105
30–39	25	24	12	61	25	25	16	66	127
40–49	25	24	12	61	24	23	21	68	129
50–59	24	22	10	56	23	21	15	59	115
Total	99	95	36	230	97	90	59	246	476

Table 2 Age adjusted mean of variables and prevalence of alcohol intake by sex in each centre

Variables	Men							Women						
	Toyama (n = 99)		Osaka (n = 95)		Tochigi (n = 36)		P	Toyama (n = 97)		Osaka (n = 90)		Tochigi (n = 59)		P
	Mean	s.e.	Mean	s.e.	Mean	s.e.		Mean	s.e.	Mean	s.e.	Mean	s.e.	
BMI (kg/m^2)	22.9	(0.3)	23.3	(0.3)	22.4	(0.5)		22.1	(0.3)	22.0	(0.3)	23.0	(0.4)	
Systolic BP (mm Hg)	119.2	(1.3)	116.7	(1.3)	115.9	(2.2)		111.5	(1.5)	113.6	(1.5)	108.3	(1.9)	
Diastolic BP (mm Hg)	73.6	(1.1)	71.2	(1.1)	70.7	(1.8)		69.3	(1.1)	66.7	(1.1)	62.7	(1.4)	**
Urinary electrolytes excretion														
Sodium (mmol/day)	211.8	(6.8)	191.2	(6.9)	176.8	(11.4)	*	179.5	(5.4)	141.4	(5.7)	181.1	(7.1)	***
Potassium (mmol/day)	51.9	(1.5)	51.6	(1.5)	47.6	(2.5)		46.8	(1.5)	42.6	(1.6)	51.6	(2.0)	**
Na/K ratio	4.17	(0.12)	3.79	(0.12)	3.96	(0.2)		3.98	(0.12)	3.56	(0.12)	3.64	(0.15)	*
Dietary intake														
Calcium (mg/day)	557	(29)	608	(29)	589	(48)		528	(27)	597	(28)	639	(35)	*
Alcohol														
1–299 ml/wk (%)	58.2		59.3		46.8				40.0[a]		50.9[a]		39.5[a]	
300 ml/wk and more (%)	20.4		10.2		38.9			—		—		—		

P, analysis variance *$P < 0.05$ **$P < 0.01$ ***$P < 0.001$; *n*, sample size in group. [a]Together with 1–299 ml/wk, and 300 ml/wk and more.

528 mg/day, respectively). Urinary sodium and potassium excretions were lower in Osaka than those of Toyama and Tochigi. DBP was the highest in the subjects in Toyama centre, followed by Osaka and Tochigi.

Table 3 shows the Pearson correlation between variables by sexes. No significant correlations were found between blood pressure and nutrients such as calcium intake, urinary sodium and potassium excretion, and sodium/potassium ratio. Calcium intake correlated with potassium excretion among men, and potassium excretion and sodium/potassium ratio among women. Calcium intake increased with age among women. Analysis by each centre also showed the same results for men and women.

The results of multiple regression analysis of systolic blood pressure (SBP) and DBP with dietary calcium intake are shown in Tables 4 and 5. Among men, the pooled estimates of the regression coefficients adjusted for age and BMI was –0.42 mm Hg for SBP by 100 mg increase of calcium intake and –0.35 mm Hg for DBP, but neither coefficient was statistically significant. The relationship of dietary calcium intake to blood pressure after adjustment for age, BMI, alcohol intake, and urinary excretion of sodium or potassium were also not significant. Among women, the pooled estimates of regression coefficients adjusted for age and BMI were –0.92 mm Hg for SBP by 100 mg increase of calcium intake and –0.83 mm Hg for DBP by 100 mg increase of calcium intake, both of which were significant. After adjustment for age, BMI, alcohol intake and urinary excretion of sodium and potassium, the pooled estimate was –0.66 mm Hg for DBP by 100 mg increase of calcium intake with statistical significance, and –0.70 mm Hg for SBP by 100 mg increase of calcium intake. By each centre, negative significant association of calcium intake with blood pressure was seen in the subjects in Osaka.

Urinary sodium and potassium excretion did not correlate significantly with SBP and DBP except for the negative correlation between sodium and SBP in Toyama among men (Table 5). Among women, there was no significant relationship between urinary excretion of sodium and potassium and blood pressure in pooled estimation and in the subjects in Osaka. In the subjects in Toyama, there was significant negative association of urinary sodium excretion with blood pressure. In Tochigi, there was significant inverse association of urinary potassium excretion with blood pressure.

From our study, we found significant negative association of calcium intake with blood pressure among women, but not for men. The strongest and significant relation between calcium intake and blood pressure was observed in Osaka. These associations were observed independently of other confounders such as BMI, alcohol intake, and urinary sodium and potassium excretions.

Table 3 Pearson correlation between variables

		SBP	DBP	Calcium intake
Men	Age	0.23**	0.33**	0.10
	BMI	0.18**	0.31**	0.05
	Sodium excretion	0.02	0.03	0.09
	Potassium excretion	0.03	0.05	0.19**
	Na/K ratio	0.01	–0.03	–0.06
	Calcium intake	–0.08	–0.04	—
Women	Age	0.44**	0.47**	0.22**
	BMI	0.31**	0.34**	0.00
	Sodium excretion	0.04	0.08	0.10
	Potassium excretion	0.01	0.03	0.35**
	Na/K ratio	0.02	0.01	–0.26**
	Calcium intake	–0.06	–0.06	—

Na/K, sodium/potassium ratio. ** $P < 0.01$.

Table 4 Multiple correlation coefficients of systolic and diastolic blood pressure with dietary calcium intake (100 mg/day) and 24-h urinary sodium and potassium excretion (mmol/day) adjusted for age, BMI, alcohol intake and each other in men

	Adjusted for age, BMI			*Adjusted for age, BMI and others*[a]								
	BP-Calcium			*BP-Calcium*			*BP-Sodium*			*BP-Potassium*		
	Coeff	*s.e.*	*P*	*Coeff*	*s.e.*	*P*	*Coeff*	*s.e.*	*P*	*Coeff*	*s.e.*	*P*
Systolic blood pressure												
All	–0.42	(0.30)		–0.43	(0.31)		–0.04	(0.02)		–0.01	(0.07)	
Toyama	–0.17	(0.44)		–0.09	(0.44)		–0.05	(0.02)	*	0.11	(0.11)	
Osaka	–0.45	(0.56)		–0.47	(0.62)		0.03	(0.03)		–0.06	(0.14)	
Tochigi	–0.84	(0.68)		–0.97	(0.76)		0.00	(0.05)		0.00	(0.17)	
Diastolic blood pressure												
All	–0.35	(0.24)		–0.33	(0.24)		–0.02	(0.01)		–0.01	(0.06)	
Toyama	–0.53	(0.39)		–0.44	(0.38)		–0.04	(0.02)		–0.03	(0.09)	
Osaka	–0.15	(0.41)		–0.05	(0.46)		0.00	(0.02)		–0.09	(0.10)	
Tochigi	–0.13	(0.51)		–0.28	(0.55)		–0.02	(0.04)		0.10	(0.17)	

[a]BP-Calcium: adjusted for age, BMI, alcohol intake, sodium, potassium; BP-Sodium: adjusted for age, BMI, alcohol intake, calcium, potassium; BP-Potassium: adjusted for age, BMI, alcohol intake, calcium, sodium, Coeff, coefficient; s.e. standard error; $^{*}P < 0.05$, $^{**}P < 0.01$.

Table 5 Multiple correlation coefficients of systolic and diastolic blood pressure with dietary calcium intake (100 mg/day) and 24-h urinary sodium and potassium excretion (mmol/day) adjusted for age, BMI, alcohol intake and each other in women

	Adjusted for age BMI			*Adjusted for age, BMI and others*[a]								
	BP-Calcium			*BP-Calcium*			*BP-Sodium*			*BP-Potassium*		
	Coeff	*s.e.*	*P*	*Coeff*	*s.e.*	*P*	*Coeff*	*s.e.*	*P*	*Coeff*	*s.e.*	*P*
Systolic blood pressure												
All	–0.92	(0.34)	**	–0.70	(0.37)		–0.01	(0.02)		–0.10	(0.08)	
Toyama	–0.63	(0.47)		–0.65	(0.48)		–0.05	(0.02)	*	0.03	(0.10)	
Osaka	–1.78	(0.65)	**	–1.12	(0.53)	*	–0.01	(0.05)		0.05	(0.14)	
Tochigi	–0.27	(0.59)		0.61	(0.63)		0.09	(0.04)	*	–0.47	(0.14)	**
Diastolic blood pressure												
All	–0.83	(0.25)	**	–0.66	(0.27)	*	–0.07	(0.01)		–0.07	(0.06)	
Toyama	–0.36	(0.33)		–0.35	(0.33)		–0.04	(0.02)	*	–0.01	(0.07)	
Osaka	–1.12	(0.48)	*	–1.65	(0.71)	*	–0.02	(0.04)		0.08	(0.11)	
Tochigi	–0.77	(0.46)		–0.16	(0.50)		–0.07	(0.03)	*	–0.33	(0.11)	**

[a]BP-Calcium: adjusted for age, BMI, alcohol intake, sodium, potassium; BP-Sodium: adjusted for age, BMI, alcohol intake, calcium, potassium; BP-Potassium: adjusted for age, BMI, alcohol intake, calcium, sodium.
Coeff, coefficient; s.e., standard error; $^{*}P < 0.05$, $^{**}P < 0.01$.

Discussion

The purpose of this study was to investigate the association between dietary calcium intake and blood pressure while taking account of other nutrient intakes recognised as risk factors for hypertension. The role of dietary calcium intake on blood pressure is still being debated. There is heterogeneity of results from observational studies concerned about effect of dietary calcium intake on blood pressure. Pryer *et al*[1] listed several reasons of heterogeneity; the difference in methods of dietary data collection and assessment, considerable variation across the studies in the control for confounding factors and a high degree of correlation between nutrients. In our study, calcium intake correlated with urinary potassium excretion. Some studies considered the sodium and potassium, but they were estimated from dietary assessment method rather than 24-h urinary excretion, which is recognised as a better way of estimating sodium and potassium intake.[23,24]

To overcome these factors of heterogeneity, we used a highly standardised method for data collection and assessment. We used 24-h urinary excretion for estimation of sodium and potassium intake. This study was designed as a reinvestigation of the INTERSALT study, 8 years after it was first carried out. This was a study to investigate trends in blood pressure and urinary sodium and potassium excretion in the Japanese centres which participated. We used the same method as the INTERSALT study for sampling, measurement of blood pressure, and urinary sodium and potassium excretion. We added to this study a 24-h dietary recall for estimation of nutrients. Then we investigated the

relation after full adjustment for age, BMI, alcohol intake, and sodium and potassium intake.

However, we must also note a potential drawback with our data. Because the participation rate of Tochigi centre was low noted above, the subjects in Tochigi centre do not necessarily represent the target population. Because we obtained only one single 24-h dietary data and single 24-h urine collection, there might be intra-individual variation. It has been well documented that high ratios of within/between person variation can attenuate the absolute values of regression and correlation coefficients and can reduce their statistical power.[25–27] Correction for the regression dilution bias was not possible and linear regression between dietary calcium intake and urinary electrolytes and blood pressure might be underestimated.

Instead of the drawback of our study, the present study shows a consistent negative association of dietary calcium intake with both SBP and DBP in men and women in the pooled estimate and in analysis by the three centres. These associations were seen independently of other confounders. One of our interests of this study is whether the inverse association between dietary calcium intake and blood pressure exists in a population with characteristic dietary habit. The notable dietary features of Japan are high intake of salt and low intake of potassium as shown in the INTERSALT study and low calcium intake. In our target population, the mean calcium intake is 528–639 mg/day, which is much lower than in United States and Europe. In addition, as shown in our previous report, sodium consumption remains high while potassium consumption is still low. Already, Iso *et al*[10] reported the inverse association between dietary calcium intake and blood pressure among Japanese people. However, they used dietary estimation for sodium intake and did not consider interaction with potassium intake.

In our population, the associations of calcium intake with blood pressure among women are stronger than those of men. We found a statistical significant association between calcium intake and blood pressure among women, but not among men. The pooled regression slope adjusted for age, BMI and urinary sodium and potassium excretion between dietary calcium and blood pressure among women was −0.70 mm Hg/100 mg per day for SBP and −0.66 mm Hg/100 mg per day for DBP. It is not apparent whether there is a difference in calcium sensitivity between men and women, as described in Geleinjnse and Grobbee.[2] Hamet *et al*[27] reported the protective effect of calcium was most evident at a higher level of sodium intake. However, we did not find such a result. On the contrary, the protective effect of calcium might be more evident at a lower level of sodium intake, because we found the strongest inverse relation between calcium intake and blood pressure in Osaka centre, in which sodium intake was the lowest of the three centres.

Our study suggests that increased calcium intake may provide a benefit of lowering blood pressure independently with other well-known factors. In Japan, for prevention of osteoporosis, increasing the calcium intake, particularly among women, is one of the main issues of public health. This might contribute to the prevention and control of high blood pressure.

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