Participants and Methods

Study design and population

The study population consisted of Japanese men who worked for a metal products factory in Toyama prefecture, Japan that employed approximately 4400 male and 2600 female workers. The Industrial Safety and Health Law in Japan requires all employers to conduct annual health examinations on all employees. The details of this study population have been reported previously [20]. Of all the employees who underwent an annual health examination in 1996 (>90%), 4011 male employees aged 20–54 years were enrolled as potential participants and were followed-up for 6 years from 1996 to 2002 by annual health examinations. Female employees were not included, due to the small number of newly developed cases of hyperuricemia during the follow-up period in female workers. Employees aged 55 years or older were also excluded, as they were required to retire by the end of the follow-up period. The present cohort study was approved by the Institutional Review Committee of Kanazawa Medical University for Ethical Issues.

Of the 4011 potential participants, 701 were excluded due to one or more of the following criteria: baseline hyperuricemia, defined as a serum uric acid >416.4 µmol/l (or 7.0 mg/dl) based on the Japanese guidelines for the management of hyperuricemia and gout [21] and/or taking medication for hyperuricemia (n=529); baseline renal dysfunction, defined as a serum creatinine >114.9 µmol/l (or 1.3 mg/dl) set as the upper normal limit for men in our laboratory (n=9); missing information at the time of the baseline survey (n=44); or failure to obtain information in the follow-up survey (n=119). The remaining 3310 normouricemic participants were included in the final analyses as the eligible study population.

Baseline examination

Data collected at study entry were age, medical history, alcohol drinking, smoking and exercise habits, anthropometric indices, blood pressure, serum liver enzymes including GGT, uric acid, creatinine, total cholesterol, and blood glucose. Blood samples were obtained by cubital venipuncture and then transported to a single laboratory (BML, Inc., Toyama, Japan) for analysis. Serum GGT levels were measured by the L-gamma-glutamyl-*p*-nitroanilide method using an automatic analyzer (H-7450; Hitachi, Ltd., Tokyo, Japan), and AST and ALT were determined by an ultraviolet method. Uric acid was measured enzymatically, total cholesterol by an enzymatic method, and creatinine by the Jaffe colorimetric method. Glycated hemoglobin (HbA1c) levels were measured by high-performance liquid chromatography using an automated analyzer (Hi-AUTOA1C HA8121; Kyoto Daiichi Kagaku Co., Ltd, Kyoto, Japan). Quality control of the HbA1c measurements was performed using the standard certified by the Japan Diabetes Society (JDS), with the values of HbA1c being converted to values of both the National Glycohemoglobin Standardization Program (NGSP) and the International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) for Système International (SI) units. The former conversion was performed using the formula provided by the JDS; NGSP-HbA1c (%)=JDS-HbA1c (%) + 0.4 [22], and the latter conversion using the IFCC-NGSP master equation; IFCC-HbA1c (mmol/mol)=10.93×NGSP-HbA1c (%) − 23.50 [23,24]. Measurements of anthropometric indices and blood pressure were carried out by trained staff, with body mass index (BMI) calculated as weight in kilograms divided by the square of height in meters.

Data were collected using a self-administered questionnaire at study entry and included age, medical history, alcohol drinking, smoking, and exercise habits. Alcohol drinking habits were evaluated by determining whether a participant was a nondrinker or a drinker. The drinkers were then asked to estimate the frequency of their alcohol intake during a typical week or month. The quantity of alcoholic beverages consumed during each typical period was also reported, using units of major alcoholic beverages in Japan, with one unit (approximately 23 g of alcohol for each beverage) defined as 2 drinks. The average weekly alcohol intake (drinks/week) was calculated for each participant using data on the frequency and quantity of alcohol intake. Smoking habits were classified as never, former, or currently smoking. Exercise habits were evaluated in terms of frequency and intensity of exercise (no, light, moderate, or hard exercise).

Follow-up survey

To identify incident cases of hyperuricemia during the follow-up period, the participants underwent annual health examinations until the end of 2002. The procedures for the measurement of serum uric acid and obtaining the medical histories were the same throughout the follow-up period. Hyperuricemia was defined as serum uric acid>416.4 µmol/l [21] and/or taking medication for hyperuricemia. This was the same definition used for exclusion at baseline.

Statistical analysis

The study participants were grouped according to their GGT level at baseline: GGT≤19, 20–39, 40–59, 60–79, or ≥80 U/l. The risk of incident hyperuricemia was compared in these 5 groups. Hazard ratios and their corresponding 95% confidence intervals (CIs) for incident hyperuricemia were calculated for each of the GGT groups, with the GGT≤19 group acting as the reference. A Cox proportional hazards regression model was carried out that incorporated the following variables as covariates: age (years as a continuous variable), serum uric acid at baseline (µmol/l as a continuous variable), BMI (kg/m^2 as a continuous variable), alcohol intake (nondrinking, 0.1–9.9, 10.0–19.9, 20.0–29.9, 30.0–39.9, or ≥40.0 drinks/week, using 5 dummy variables with nondrinkers as the reference), smoking habits (current, former, or never smoked, using 2 dummy variables with never smoked as the reference), exercise habits (hard, moderate, light, or no exercise, using 3 dummy variables with no exercise as the reference), serum creatinine (µmol/l as a continuous variable), systolic blood pressure (mm Hg as a continuous variable), medication for hypertension (yes or no), serum total cholesterol (mmol/l as a continuous variable), medication for hypercholesterolemia (yes or no), NGSP-HbA1c (% as a continuous variable), medication for diabetes (yes or no) and either lnAST or lnALT. The trend of the relationship between GGT and the risk of incident hyperuricemia was also examined in a multivariate Cox model using the continuous variable, lnGGT, instead of the GGT category. The hazard ratio associated with an increase of one standard deviation in lnGGT (i.e., one geometric standard deviation) was then calculated. Natural logarithmic transformations of GGT, AST, and ALT were used to normalize the distribution due to their skewed distributions. The risk of incident hyperuricemia was compared in participants grouped according to baseline AST, as well as in participants grouped according to baseline ALT; ≤19, 20–39, 40–59, 60–79, or ≥80 U/l. Hazard ratios were calculated using a similar multivariate Cox model that incorporated lnGGT instead of lnAST or lnALT.

In order to avoid the potential confounding effect of alcohol drinking, obesity, metabolic disorders, and elevated levels of serum aminotransferases on incident hyperuricemia, similar analyses were then conducted after all the study participants had been stratified according to the presence or absence of alcohol drinking, obesity (BMI≥25.0 kg/m^2 [25]), metabolic disorders [any combination of hypertension (systolic blood pressure≥140 mm Hg, diastolic blood pressure≥90 mm Hg [26] and/or taking medication for hypertension], hypercholesterolemia [serum total cholesterol≥6.21 mmol/l (or 240 mg/dl) [27] and/or taking medication for hypercholesterolemia) and/or diabetes (NGSP-HbA1c≥6.5% [28] and/or taking medication for diabetes)], and clinically high serum aminotransferases (AST≥40 U/l and/or ALT≥40 U/l [1]). The significance of the interaction was tested using an interaction term for the categorical variables in the multivariate Cox model.

The statistical analyses were performed using the Statistical Package for the Social Sciences Version 12.0J for Windows (SPSS Japan Inc., Tokyo, Japan). All probability values were 2-tailed and the significance level was set at $p<0.05$.

Results

Characteristics of the study population

At baseline, the mean age±standard deviation of the 3310 study participants was 38.5±9.6 years, mean serum uric acid was 327.0±54.1 μmol/l, and median values (interquartile range) of GGT, AST, and ALT were 19 (13–29) U/l, 21 (18–24) U/l, and 19 (15–27) U/l, respectively (see ○ **Table 1** for other baseline characteristics).

The baseline characteristics of the 3310 study participants grouped according to GGT are summarized in ○ **Table 1**. The prevalence of participants with a GGT of ≤19, 20–39, 40–59, 60–79, or ≥80 U/l was 53.4%, 32.0%, 7.6%, 3.1%, and 4.0%, respectively. Mean age was marginally but significantly higher in participants with higher GGT levels than those with lower GGT levels. Similarly, the mean values of serum uric acid, BMI, systolic and diastolic blood pressure, serum total cholesterol, and HbA1c and the median values of alcohol intake, AST and ALT, and the rate of current smoking and taking medication for hypertension and hypercholesterolemia were significantly higher in participants with higher GGT levels than those with lower GGT levels.

GGT and the risk of incident hyperuricemia

The study included 16673 person-years of follow-up, with the mean±standard deviation follow-up period being 5.0±1.7 years. During follow-up, 529 incident cases of hyperuricemia were identified. The crude incidence rate of hyperuricemia in the study population was 31.7 per 1000 person-years.

The incidence rate of hyperuricemia rose with increasing GGT levels (○ **Table 2**). The positive relationship between GGT and the risk of incident hyperuricemia remained in a dose-dependent manner after adjustment for major confounders including age, baseline serum uric acid, BMI, alcohol intake, and use of antihypertensive medication. This positive relationship was attenuated only marginally even after further adjustment for either AST or ALT. The relationship between GGT and the risk of incident hyperuricemia was significant after further adjustment for AST and ALT (p-value for trend = 0.006 and 0.02, respectively), with the multivariate-adjusted hazard ratio associated with an increase of one standard deviation in lnGGT (1.86 as one geometric standard deviation of GGT) being 1.17 (95% CI: 1.05–1.30) and 1.14 (95% CI: 1.02–1.28), respectively. The relationship between the serum aminotransferases (AST and ALT) and the risk of incident hyperuricemia showed a similar positive, dose-dependent association without taking GGT into account in the analyses. However, the corresponding relationship was no longer present after further adjustment for GGT.

As shown in ○ **Table 3**, the nature of the relationship between GGT and the risk of incident hyperuricemia was consistent across strata of alcohol drinking habits, obesity status, metabolic status, and serum aminotransferase levels. The trend was either significant or borderline significant in several subgroups of these factors, with the multivariate-adjusted hazard ratio associated with an increase of one standard deviation in lnGGT being similar to the overall hazard ratio. The corresponding hazard ratio for their counterpart subgroups was also broadly similar, although it was not statistically significant. There was no significant interaction between increased GGT and alcohol drinking, obesity, metabolic disorders, or high serum aminotransferases for the risk of incident hyperuricemia (p-value for interaction = 0.57 for alcohol drinking, 0.26 for obesity, 0.98 for metabolic disorders, and 0.95 for high serum aminotransferases).

Discussion

The present cohort study demonstrated that there was a positive, dose-response relationship between GGT and the risk of incident hyperuricemia in normouricemic Japanese men, even within the normal range of GGT (≤60 U/l in our laboratory). Regardless of the presence or absence of alcohol drinking, obesity, metabolic disorders, and high serum aminotransferases, it is likely that a similar positive relationship is observed. This indicates the positive relationship we observed may be independent of alcohol intake, obesity status, metabolic status, and serum aminotransferase levels, which are major determinants of GGT [9, 10, 29]. On the other hand, there appeared to be no relationship between serum aminotransferases and the risk of incident hyperuricemia within the normal-to-mildly high range of AST and AST when the effect of GGT on the development of hyperuricemia was taken into account.

Our study confirms the findings of relevant cross-sectional studies, which observed an independent positive relationship between GGT and serum uric acid [9–13], and importantly provides evidence on causality for this relationship. This causality is supported by the CARDIA study in the United States [5], which reported that future serum uric acid appeared to increase with increasing GGT levels. However, the analyses in this earlier longitudinal study did not take into account either baseline uric acid levels, alcohol intake, obesity status, or serum aminotransferase levels. To the best of our knowledge, our data are the first to indicate that increased GGT levels predict subsequent development of hyperuricemia independently of alcohol intake, obesity status, and serum aminotransferase levels.

Although our study could not elucidate the underlying mechanism for the positive relationship between GGT levels and the risk of incident hyperuricemia, the following mechanisms are possible. Recently, GGT has become regarded as a marker of oxidative stress [10, 30–32], whereas uric acid acts as an antioxidant [33–35]. Taken together, these properties may provide a rationale for the positive relationship between GGT levels and the risk of incident hyperuricemia, and suggest that oxidative

Table 1 Baseline characteristics of the 3310 normouricemic male study participants in Toyama, Japan (1996).

	Overall	Serum gamma-glutamyltransferase (U/l)					p-Values for heterogeneity[a]
		≤19	20–39	40–59	60–79	≥80	
Participants	3310	1766	1058	252	101	133	
Age (years)	38.5±9.6	36.0±9.9	40.6±8.4	42.0±7.9	45.3±6.9	43.6±7.6	<0.001
Serum uric acid (μmol/l)	327.0±54.1	321.1±52.7	331.8±54.8	336.3±52.8	335.3±58.9	342.9±56.9	<0.001
Height (cm)	169.1±6.1	169.7±6.2	168.8±6.0	168.2±6.1	166.9±6.1	166.7±5.5	<0.001
Weight (kg)	65.3±8.9	63.7±8.3	66.8±8.9	68.6±10.0	68.3±8.5	67.4±10.4	<0.001
Body mass index (kg/m^2)	22.8±2.8	22.1±2.5	23.4±2.7	24.2±3.1	24.5±2.6	24.2±3.2	<0.001
Alcohol intake (drinks/wk)[b]	6.0 (0–14.0)	3.0 (0–10.0)	10.0 (2.0–20.0)	14.5 (6.0–28.0)	14.0 (8.2–28.0)	24.0 (12.0–31.0)	<0.001
Cigarette smoking habits							<0.001
Never smoked	31.4	35.3	28.8	20.2	20.8	27.8	
Former smoker	9.9	9.7	10.8	11.1	7.9	4.5	
Currently smoking	58.8	55.0	60.4	68.7	71.3	67.7	
Exercise habits							0.03
None	63.7	61.8	64.8	66.3	73.3	68.4	
Light exercise	18.4	18.0	19.3	19.0	19.8	15.0	
Moderate exercise	12.5	14.2	10.8	11.5	5.0	11.3	
Hard exercise	5.4	6.1	5.1	3.2	2.0	5.3	
Serum creatinine (μmol/l)	85.2±10.1	86.1±9.8	84.7±10.4	84.3±10.9	82.7±10.2	81.2±9.6	<0.001
Systolic blood pressure (mm Hg)	122.7±13.4	121.0±12.6	123.5±13.5	125.4±13.3	128.4±15.7	129.7±15.4	<0.001
Diastolic blood pressure (mm Hg)	74.2±10.4	71.9±9.8	75.7±10.2	78.1±10.1	80.2±10.9	80.5±11.0	<0.001
Medication for hypertension	2.5	1.2	3.1	3.2	8.9	7.5	<0.001
Serum total cholesterol (mmol/l)	5.07±0.87	4.87±0.81	5.24±0.84	5.44±0.83	5.35±0.94	5.46±1.20	<0.001
Medication for hypercholesterolemia	0.8	0.6	0.9	0.4	2.0	3.0	0.02
Glycated hemoglobin							
NGSP-HbA1c (%)	5.42±0.53	5.35±0.49	5.47±0.53	5.55±0.54	5.50±0.72	5.48±0.64	<0.001
IFCC-HbA1c (mmol/mol)	35.7±5.8	35.0±5.4	36.3±5.8	37.2±5.9	36.7±7.9	36.4±7.0	<0.001
Medication for diabetes	0.5	0.3	0.5	0.8	3.0	0	0.002
Serum gamma-glutamyltransferase (U/l)	19 (13–29)	14 (12–16)	26 (22–31)	47 (43–52)	66 (63–73)	111 (94–153)	<0.001
Serum aspartate aminotransferase (U/l)	21 (18–24)	19 (16–22)	22 (19–26)	25 (22–30)	27 (23–34)	34 (28–46)	<0.001
Serum alanine aminotransferase (U/l)	19 (15–27)	16 (13–20)	23 (18–30)	28 (23–37)	29 (23–41)	40 (28–57)	<0.001

Data are presented for the total study group and also grouped according to serum gamma-glutamyltransferase level. Values are expressed as mean±standard deviation, median (interquartile range), or the % of participants in that category

[a] One-way analysis of variance, Kruskal-Wallis test, or a chi-square test was used to compare each risk factor in the 5 groups of serum gamma-glutamyltransferase level

[b] One drink of an alcoholic beverage is defined as containing 11.5 g of ethanol

Table 2 Hazard ratios for the incidence of hyperuricemia, grouped according to each serum liver enzyme level in 3310 normouricemic male participants over 6 years of follow-up (1996–2002).

	Serum gamma-glutamyltransferase/aspartate aminotransferase/alanine aminotransferase (U/l)					1 SD in ln (each enzyme)	p-Values for trend[a]
	≤19	20–39	40–59	60–79	≥80		
Serum gamma-glutamyltransferase and hyperuricemia						(1.86 as 1 GSD of GGT)	
Participants	1766	1058	252	101	133		
Person-years of follow-up	9180	5198	1240	464	591		
Cases of hyperuricemia[b]	187	210	59	28	45		
Crude incidence rate[c]	20.4	40.4	47.6	60.3	76.1		
HR (95% CI), model 1[d]	1.00 (reference)	1.56 (1.27–1.92)	1.70 (1.25–2.29)	2.22 (1.47–3.36)	2.31 (1.64–3.24)	1.28 (1.18–1.38)	<0.001
HR (95% CI), model 2[d]	1.00 (reference)	1.34 (1.07–1.66)	1.31 (0.94–1.82)	1.59 (1.02–2.47)	1.62 (1.11–2.35)	1.15 (1.05–1.26)	0.004
HR (95% CI), model 3[d]	1.00 (reference)	1.34 (1.08–1.68)	1.33 (0.94–1.86)	1.61 (1.02–2.55)	1.67 (1.09–2.57)	1.17 (1.05–1.30)	0.006
HR (95% CI), model 4[d]	1.00 (reference)	1.32 (1.05–1.67)	1.28 (0.90–1.83)	1.56 (0.98–2.47)	1.57 (1.02–2.41)	1.14 (1.02–1.28)	0.02
Serum aspartate aminotransferase and hyperuricemia						(1.36 as 1 GSD of AST)	
Participants	1342	1843	97	14	14		
Person-years of follow-up	6865	9228	443	74	63		
Cases of hyperuricemia[b]	169	323	30	4	3		
Crude incidence rate[c]	24.6	35.0	67.7	54.1	47.6		
HR (95% CI), model 1[d]	1.00 (reference)	1.06 (0.87–1.28)	1.54 (1.03–2.29)	1.79 (0.66–4.82)	2.42 (0.77–7.58)	1.13 (1.04–1.23)	0.003
HR (95% CI), model 2[d]	1.00 (reference)	0.91 (0.74–1.11)	1.13 (0.74–1.71)	1.26 (0.45–3.53)	2.20 (0.70–6.98)	1.05 (0.96–1.14)	0.34
HR (95% CI), model 5[d]	1.00 (reference)	0.85 (0.69–1.05)	0.86 (0.54–1.36)	0.94 (0.33–2.69)	1.69 (0.52–5.44)	0.97 (0.88–1.08)	0.60
Serum alanine aminotransferase and hyperuricemia						(1.63 as 1 GSD of ALT)	
Participants	1672	1349	198	56	35		
Person-years of follow-up	8609	6752	906	253	153		
Cases of hyperuricemia[b]	205	236	62	14	12		
Crude incidence rate[c]	23.8	35.0	68.4	55.3	78.4		
HR (95% CI), model 1[d]	1.00 (reference)	1.21 (1.00–1.46)	1.61 (1.21–2.14)	1.51 (0.88–2.60)	1.96 (1.10–3.51)	1.17 (1.08–1.26)	<0.001
HR (95% CI), model 2[d]	1.00 (reference)	1.09 (0.89–1.33)	1.26 (0.92–1.74)	1.27 (0.72–2.24)	2.04 (1.09–3.81)	1.09 (0.99–1.20)	0.09
HR (95% CI), model 5[d]	1.00 (reference)	1.00 (0.81–1.25)	1.06 (0.74–1.52)	1.03 (0.56–1.88)	1.58 (0.80–3.10)	1.01 (0.90–1.13)	0.86

HR: hazard ratio; CI: confidence interval; SD: standard deviation; GGT: gamma-glutamyltransferase; AST: aspartate aminotransferase; ALT: alanine aminotransferase; GSD: geometric standard deviation

[a] The trend for the relationship between each serum liver enzyme (GGT, AST, and ALT) and the risk of incident hyperuricemia were tested in a Cox model using continuous variables for lnGGT, lnAST, and lnALT, respectively

[b] Hyperuricemia is defined as a serum uric acid >416.4 µmol/l and/or taking medication for hyperuricemia

[c] The crude incident rate is expressed in per 1000 person-years

[d] Five different Cox proportional hazards regression models were used: model 1 was adjusted for age and serum uric acid at baseline; model 2 was adjusted for the same covariates used in model 1 in addition to body mass index, alcohol intake, smoking and exercise habits, serum creatinine, systolic blood pressure, serum total cholesterol, NGSP-HbA1c, and medications for hypertension, hypercholesterolemia, and diabetes; and models 3, 4 and 5 were adjusted for the same covariates used in model 2 in addition to lnAST, lnALT, and lnGGT, respectively

Table 3 Hazard ratios for the incidence of hyperuricemia, grouped according to serum gamma-glutamyltransferase level in 3 310 normouricemic male participants over 6 years of follow-up (1996–2002), stratified according to alcohol drinking habits, obesity status, metabolic status, and serum aminotransferase levels.

	Serum gamma-glutamyltransferase (U/l)					1 SD in lnGGT (1.86 as 1 GSD)	p-Values for trend[a]
	≤19	20–39	40–59	60–79	≥80		
Nondrinkers							
Participants	608	193	38	9	7		
Person-years of follow-up	3208	973	185	38	31		
Cases of hyperuricemia[b]	51	34	8	3	3		
Crude incidence rate[c]	15.9	34.9	43.2	78.9	96.8		
HR (95 % CI), model 1[d]	1.00 (reference)	1.42 (0.90–2.25)	1.66 (0.78–3.53)	1.85 (0.56–6.10)	4.17 (1.29–13.43)	1.35 (1.10–1.66)	0.005
HR (95 % CI), model 2[d,e]	1.00 (reference)	1.19 (0.73–1.95)	1.53 (0.66–3.54)	2.07 (0.45–9.56)	3.79 (1.12–12.83)	1.28 (0.99–1.65)	0.06
HR (95 % CI), model 3[d,e]	1.00 (reference)	1.24 (0.74–2.09)	1.64 (0.67–4.02)	2.43 (0.46–12.98)	4.60 (1.07–19.78)	1.32 (0.97–1.81)	0.08
HR (95 % CI), model 4[d,e]	1.00 (reference)	1.16 (0.67–2.01)	1.47 (0.60–3.63)	1.92 (0.37–10.09)	3.47 (0.82–14.73)	1.25 (0.90–1.73)	0.18
Drinkers							
Participants	1158	865	214	92	126		
Person-years of follow-up	5972	4225	1055	426	560		
Cases of hyperuricemia[b]	136	176	51	25	42		
Crude incidence rate[c]	22.8	41.7	48.3	58.7	75.0		
HR (95 % CI), model 1[d]	1.00 (reference)	1.55 (1.23–1.96)	1.65 (1.19–2.30)	2.14 (1.36–3.35)	2.14 (1.48–3.08)	1.25 (1.14–1.36)	<0.001
HR (95 % CI), model 2[d]	1.00 (reference)	1.36 (1.06–1.74)	1.31 (0.92–1.88)	1.58 (0.99–2.54)	1.54 (1.03–2.29)	1.13 (1.02–1.25)	0.02
HR (95 % CI), model 3[d]	1.00 (reference)	1.36 (1.06–1.75)	1.32 (0.92–1.91)	1.60 (0.99–2.60)	1.57 (0.99–2.49)	1.14 (1.02–1.29)	0.03
HR (95 % CI), model 4[d]	1.00 (reference)	1.35 (1.05–1.75)	1.31 (0.89–1.92)	1.58 (0.96–2.58)	1.53 (0.97–2.42)	1.13 (1.00–1.27)	0.05
Nonobese individuals							
Participants	1548	763	154	57	83		
Person-years of follow-up	8104	3765	776	261	375		
Cases of hyperuricemia[b]	153	146	32	14	29		
Crude incidence rate[c]	18.9	38.8	41.2	53.6	77.3		
HR (95 % CI), model 1[d]	1.00 (reference)	1.63 (1.28–2.06)	1.64 (1.11–2.42)	2.22 (1.26–3.91)	2.23 (1.47–3.38)	1.27 (1.15–1.39)	<0.001
HR (95 % CI), model 2[d]	1.00 (reference)	1.40 (1.09–1.80)	1.38 (0.91–2.09)	1.63 (0.90–2.95)	1.68 (1.07–2.65)	1.17 (1.04–1.30)	0.007
HR (95 % CI), model 3[d]	1.00 (reference)	1.38 (1.07–1.78)	1.34 (0.88–2.04)	1.57 (0.86–2.88)	1.54 (0.91–2.60)	1.15 (1.01–1.31)	0.03
HR (95 % CI), model 4[d]	1.00 (reference)	1.33 (1.02–1.74)	1.28 (0.83–1.98)	1.51 (0.82–2.78)	1.48 (0.88–2.47)	1.12 (0.98–1.28)	0.09
Obese individuals[b]							
Participants	218	295	98	44	50		
Person-years of follow-up	1076	1433	464	203	216		
Cases of hyperuricemia[b]	34	64	27	14	16		
Crude incidence rate[c]	31.6	44.7	58.2	69.0	74.1		
HR (95 % CI), model 1[d]	1.00 (reference)	1.15 (0.75–1.77)	1.37 (0.82–2.31)	1.80 (0.94–3.45)	1.94 (1.05–3.58)	1.23 (1.06–1.44)	0.008
HR (95 % CI), model 2[d]	1.00 (reference)	1.02 (0.64–1.61)	1.01 (0.57–1.81)	1.51 (0.73–3.15)	1.42 (0.70–2.89)	1.12 (0.93–1.34)	0.25
HR (95 % CI), model 3[d]	1.00 (reference)	1.10 (0.68–1.77)	1.21 (0.65–2.26)	1.90 (0.86–4.20)	1.96 (0.87–4.41)	1.23 (0.98–1.53)	0.07
HR (95 % CI), model 4[d]	1.00 (reference)	1.15 (0.70–1.90)	1.26 (0.65–2.43)	1.88 (0.84–4.20)	1.93 (0.83–4.47)	1.23 (0.98–1.53)	0.07

Table 3 Continued.

	Serum gamma-glutamyltransferase (U/l)					1 SD in lnGGT (1.86 as 1 GSD)	p-Values for trend[a]
	≤19	20–39	40–59	60–79	≥80		
Individuals without any metabolic disorder							
Participants	1458	727	156	56	71		
Person-years of follow-up	7578	3645	761	272	330		
Cases of hyperuricemia[b]	148	127	34	16	23		
Crude incidence rate[c]	19.5	34.8	44.7	58.8	69.7		
HR (95% CI), model 1[d]	1.00 (reference)	1.45 (1.13–1.86)	1.62 (1.10–2.38)	2.19 (1.28–3.76)	2.17 (1.37–3.45)	1.28 (1.15–1.42)	<0.001
HR (95% CI), model 2[d,f]	1.00 (reference)	1.24 (0.95–1.61)	1.17 (0.76–1.78)	1.57 (0.88–2.79)	1.54 (0.94–2.51)	1.14 (1.01–1.29)	0.04
HR (95% CI), model 3[d,f]	1.00 (reference)	1.25 (0.95–1.63)	1.18 (0.77–1.83)	1.60 (0.89–2.89)	1.59 (0.92–2.75)	1.15 (1.00–1.32)	0.05
HR (95% CI), model 4[d,f]	1.00 (reference)	1.22 (0.92–1.62)	1.14 (0.73–1.79)	1.53 (0.84–2.79)	1.49 (0.86–2.56)	1.13 (0.98–1.31)	0.10
Individuals with metabolic disorders (any combination of hypertension, hypercholesterolemia and/or diabetes[b])							
Participants	308	331	96	45	62		
Person-years of follow-up	1602	1553	479	192	261		
Cases of hyperuricemia[b]	39	83	25	12	22		
Crude incidence rate[c]	24.3	53.4	52.2	62.5	84.3		
HR (95% CI), model 1[d]	1.00 (reference)	1.73 (1.17–2.55)	1.75 (1.05–2.91)	2.21 (1.14–4.30)	2.40 (1.40–4.10)	1.24 (1.10–1.41)	0.001
HR (95% CI), model 2[d]	1.00 (reference)	1.50 (0.99–2.26)	1.60 (0.92–2.79)	1.74 (0.85–3.57)	1.66 (0.90–3.09)	1.14 (0.98–1.33)	0.08
HR (95% CI), model 3[d]	1.00 (reference)	1.50 (0.99–2.28)	1.62 (0.90–2.89)	1.76 (0.83–3.72)	1.70 (0.81–3.56)	1.16 (0.97–1.40)	0.10
HR (95% CI), model 4[d]	1.00 (reference)	1.50 (0.97–2.32)	1.61 (0.86–2.98)	1.75 (0.82–3.74)	1.67 (0.80–3.50)	1.14 (0.95–1.36)	0.17
Individuals with normal serum aminotransferases							
Participants	1734	938	190	71	51		
Person-years of follow-up	9025	4639	949	337	230		
Cases of hyperuricemia[b]	182	180	39	17	13		
Crude incidence rate[c]	20.2	38.8	41.1	50.4	56.5		
HR (95% CI), model 1[d]	1.00 (reference)	1.51 (1.22–1.88)	1.59 (1.11–2.27)	1.89 (1.13–3.16)	1.91 (1.07–3.41)	1.27 (1.13–1.41)	<0.001
HR (95% CI), model 2[d]	1.00 (reference)	1.29 (1.03–1.63)	1.23 (0.84–1.81)	1.36 (0.79–2.34)	1.46 (0.80–2.65)	1.13 (0.99–1.28)	0.06
HR (95% CI), model 3[d]	1.00 (reference)	1.35 (1.06–1.71)	1.34 (0.91–1.98)	1.51 (0.87–2.63)	1.75 (0.94–3.27)	1.18 (1.04–1.35)	0.01
HR (95% CI), model 4[d]	1.00 (reference)	1.34 (1.05–1.71)	1.30 (0.87–1.95)	1.43 (0.82–2.50)	1.55 (0.84–2.88)	1.16 (1.01–1.33)	0.04
Individuals with clinically high serum aminotransferases[b]							
Participants	32	120	62	30	82		
Person-years of follow-up	155	559	291	127	361		
Cases of hyperuricemia[b]	5	30	20	11	32		
Crude incidence rate[c]	32.3	53.7	68.7	86.6	88.6		
HR (95% CI), model 1[d]	1.00 (reference)	1.66 (0.63–4.36)	1.91 (0.70–5.20)	3.14 (1.02–9.65)	2.81 (1.04–7.58)	1.28 (1.08–1.51)	0.005
HR (95% CI), model 2[d]	1.00 (reference)	1.67 (0.61–4.57)	1.37 (0.45–4.18)	2.39 (0.67–8.47)	1.67 (0.54–5.20)	1.11 (0.89–1.40)	0.36
HR (95% CI), model 3[d]	1.00 (reference)	1.93 (0.69–5.39)	1.46 (0.48–4.47)	2.49 (0.70–8.84)	1.53 (0.49–4.79)	1.07 (0.85–1.35)	0.57
HR (95% CI), model 4[d]	1.00 (reference)	1.52 (0.54–4.23)	1.23 (0.39–3.84)	2.05 (0.56–7.56)	1.40 (0.43–4.61)	1.07 (0.85–1.36)	0.56

HR: hazard ratio; CI: confidence interval; SD: standard deviation; GGT: gamma-glutamyltransferase; GSD: geometric standard deviation

[a] The trend for the relationship between GGT and the risk of incident hyperuricemia were tested in a Cox model using a continuous variable for lnGGT; [b] Hyperuricemia is defined as a serum uric acid >416.4 µmol/l and/or taking medication for hyperuricemia; obesity as a body mass index ≥25.0 kg/m^2; hypertension as a systolic blood pressure ≥140 mmHg, diastolic blood pressure ≥90 mmHg and/or taking medication for hypertension; hypercholesterolemia as a serum total cholesterol ≥6.21 mmol/l and/or taking medication for hypercholesterolemia; diabetes as a NGSP-HbA1c ≥6.5% and/or taking medication for diabetes; and clinically high serum aminotransferases a serum aspartate aminotransferase ≥40 U/l, and/or serum alanine aminotransferase ≥40 U/l; [c] Crude incident rate is expressed in per 1000 person-years; [d] Four different Cox proportional hazards regression models were used: model 1 was adjusted for age and serum uric acid at baseline; model 2 was adjusted for the same covariates used in model 1 in addition to body mass index, alcohol intake, smoking and exercise habits, serum creatinine, systolic blood pressure, serum total cholesterol, NGSP-HbA1c, and medications for hypertension, hypercholesterolemia, and diabetes; and models 3 and 4 were adjusted for the same covariates used in model 2 in addition to ln(serum aspartate aminotransferase) and ln(serum alanine aminotransferase), respectively; [e] This Cox model did not incorporate alcohol intake; [f] This Cox model did not incorporate medications for hypertension, hypercholesterolemia, or diabetes

stress plays a key role in this relationship. Another possible mechanism concerning insulin resistance may also partially underlie the positive relationship observed between GGT and hyperuricemia, although this may be less applicable in nonobese than obese individuals. There is evidence that GGT is inversely correlated with insulin sensitivity [36,37], and especially associated with intraabdominal visceral fat accumulation and hepatic steatosis [38,39]. Compensatory hyperinsulinemia to insulin resistance may cause hyperuricemia as a result of decreased urinary excretion of uric acid [40]. In addition, insulin resistance may disturb the glycolysis pathway and induce diversion of glycolytic intermediates towards uric acid [41]. However, our results indicated the positive relationship between GGT and hyperuricemia was independent of diabetic status, although it is important to note this is a different condition than insulin resistance. The corresponding relationship also appeared to be independent of ALT levels, another marker of hepatic steatosis found frequently in obese individuals [1,42].

Several limitations should be acknowledged in the present study. First, we could not consider dietary patterns due to no information being collected. As some foods (i.e., meat and coffee) influence both GGT and uric acid levels [29,43–45], it is possible that intake of these foods may have had a potential confounding effect on the positive relationship we observed. However, there is evidence that these foods are likely to have a smaller influence on both GGT and uric acid levels than alcohol from the point of view of a common lifestyle [29,43–46]. Second, with the exception of antihypertensive medication, we did not collect information on other factors that may have increased serum uric acid levels, such as other medications and diseases [15]. Third, we assumed that the baseline characteristics including GGT, alcohol drinking habits, BMI, and serum aminotransferases remained unchanged throughout the follow-up period. Fourth, we defined metabolic disorders without considering serum triglycerides and high-density lipoprotein cholesterol due to the limited availability of data on these factors. Fifth, as our study participants consisted solely of male workers in one factory, it is necessary to use caution when generalizing our results. Finally, although 16.6% (n=551) of the eligible participants dropped out in mid-study with a median follow-up of 4 years, they were included in the final analyses. Analyses after excluding these dropout participants provided very similar results (data not shown). Furthermore, the participants that dropped out during the follow-up had similar baseline characteristics including GGT and baseline serum uric acid levels to those who completed follow-up, except for a slightly higher mean age (3.2 years higher). We therefore assumed that these withdrawals from the study would not have had a significant influence on our results.

In conclusion, this study in normouricemic Japanese men showed that GGT may predict the development of hyperuricemia independent of other relevant factors such as alcohol intake, obesity status, metabolic status, and serum aminotransferase levels. Further investigations are warranted to elucidate the underlying mechanism responsible for the positive relationship between GGT and the risk of incident hyperuricemia.

Acknowledgements

The present study was supported by a Grant-in-Aid from the Ministry of Health, Labour and Welfare, Health and Labour Sciences Research Grants, Japan (H17-Kenkou-007, H18-Junkankitou [Seishuu]-Ippan-012, H19-Junkankitou [Seishuu]-Ippan-021, H20-Junkankitou [Seishuu]-Ippan-013, H22-Junkankitou [Seishuu]-Ippan-005, H23-Junkankitou [Seishuu]-Ippan-005) and by the Japan Arteriosclerosis Prevention Fund.

Affiliations

[1] Department of Epidemiology and Public Health, Kanazawa Medical University, Uchinada, Japan
[2] Department of Health Science, Shiga University of Medical Science, Otsu, Japan
[3] Department of Social and Environmental Medicine, Kanazawa Medical University, Uchinada, Japan
[4] School of Health Sciences, College of Medical, Pharmaceutical and Health Sciences, Kanazawa University, Kanazawa, Japan
[5] Department of Human Science and Fundamental Nursing, Toyama University School of Nursing, Toyama, Japan
[6] Department of Occupational and Environmental Medicine, Graduate School of Medicine, Chiba University, Chiba, Japan

References

1 *Pratt DS, Kaplan MM.* Evaluation of abnormal liver-enzyme results in asymptomatic patients. N Engl J Med 2000; 342: 1266–1271
2 *Hanigan MH, Frierson HF Jr.* Immunohistochemical detection of gamma-glutamyl transpeptidase in normal human tissue. J Histochem Cytochem 1996; 44: 1101–1108
3 *Miura K, Nakagawa H, Nakamura H, Tabata M, Nagase H, Yoshida M, Kawano S.* Serum gamma-glutamyl transferase level in predicting hypertension among male drinkers. J Hum Hypertens 1994; 8: 445–449
4 *Lee DH, Ha MH, Kim JR, Gross M, Jacobs DR Jr.* Gamma-glutamyltransferase, alcohol, and blood pressure. A four year follow-up study. Ann Epidemiol 2002; 12: 90–96
5 *Lee DH, Jacobs DR Jr, Gross M, Kiefe CI, Roseman J, Lewis CE, Steffes M.* Gamma-glutamyltransferase is a predictor of incident diabetes and hypertension: the Coronary Artery Risk Development in Young Adults (CARDIA) Study. Clin Chem 2003; 49: 1358–1366
6 *Lee DH, Ha MH, Kim JH, Christiani DC, Gross MD, Steffes M, Blomhoff R, Jacobs DR Jr.* Gamma-glutamyltransferase and diabetes – a 4 year follow-up study. Diabetologia 2003; 46: 359–364
7 *André P, Balkau B, Vol S, Charles MA, Eschwège E; DESIR Study Group.* Gamma-glutamyltransferase activity and development of the metabolic syndrome (International Diabetes Federation Definition) in middle-aged men and women: Data from the Epidemiological Study on the Insulin Resistance Syndrome (DESIR) cohort. Diabetes Care 2007; 30: 2355–2361
8 *Nakanishi N, Suzuki K, Tatara K.* Serum gamma-glutamyltransferase and risk of metabolic syndrome and type 2 diabetes in middle-aged Japanese men. Diabetes Care 2004; 27: 1427–1432
9 *Yokoyama H, Moriya S, Homma Y, Ogawa T.* Association between gamma-glutamyl transpeptidase activity and status of disorders constituting insulin resistance syndrome. Alcohol Clin Exp Res 2003; 27 (Suppl): 22S–25S
10 *Bo S, Gambino R, Durazzo M, Guidi S, Tiozzo E, Ghione F, Gentile L, Cassader M, Pagano GF.* Associations between gamma-glutamyl transferase, metabolic abnormalities and inflammation in healthy subjects from a population-based cohort: a possible implication for oxidative stress. World J Gastroenterol 2005; 11: 7109–7117
11 *Lim JS, Kim YJ, Chun BY, Yang JH, Lee DH, Kam S.* The association between serum GGT level within normal range and risk factors of cardiovascular diseases. J Prev Med Public Health 2005; 38: 101–106 (in Korean, abstract and tables in English)
12 *Onat A, Uyarel H, Hergenç G, Karabulut A, Albayrak S, Sari I, Yazici M, Keleş I.* Serum uric acid is a determinant of metabolic syndrome in a population-based study. Am J Hypertens 2006; 19: 1055–1062
13 *Afzali A, Weiss NS, Boyko EJ, Ioannou GN.* Association between serum uric acid level and chronic liver disease in the United States. Hepatology 2010; 52: 578–589
14 *Campion EW, Glynn RJ, DeLabry LO.* Asymptomatic hyperuricemia. Risks and consequences in the Normative Aging Study. Am J Med 1987; 82: 421–426
15 *Richette P, Bardin T.* Gout. Lancet 2010; 375: 318–328
16 *Hall AP, Barry PE, Dawber TR, McNamara PM.* Epidemiology of gout and hyperuricemia. A long-term population study. Am J Med 1967; 42: 27–37

17 *Yü T, Gutman AB.* Uric acid nephrolithiasis in gout. Predisposing factors. Ann Intern Med 1967; 67: 1133–1148
18 *Iseki K, Ikemiya Y, Inoue T, Iseki C, Kinjo K, Takishita S.* Significance of hyperuricemia as a risk factor for developing ESRD in a screened cohort. Am J Kidney Dis 2004; 44: 642–650
19 *Weiner DE, Tighiouart H, Elsayed EF, Griffith JL, Salem DN, Levey AS.* Uric acid and incident kidney disease in the community. J Am Soc Nephrol 2008; 19: 1204–1211
20 *Sakurai M, Miura K, Takamura T, Ishizaki M, Morikawa Y, Nakamura K, Yoshita K, Kido T, Naruse Y, Kaneko S, Nakagawa H.* J-shaped relationship between waist circumference and subsequent risk for Type 2 diabetes: an 8-year follow-up of relatively lean Japanese individuals. Diabet Med 2009; 26: 753–759
21 *Japanese Society of Gout and Nucleic Acid Metabolism.* Guideline for management of hyperuricemia and gout. 2nd ed. Osaka: Medical Review Co., Ltd, 2010 (in Japanese)
22 *The Committee of the Japan Diabetes Society on the diagnostic criteria of diabetes mellitus.* Report of the Committee on the classification and diagnostic criteria of diabetes mellitus. Diabetol Int 2010; 1: 2–20
23 *Hoelzel W, Weykamp C, Jeppsson JO, Miedema K, Barr JR, Goodall I, Hoshino T, John WG, Kobold U, Little R, Mosca A, Mauri P, Paroni R, Susanto F, Takei I, Thienpont L, Umemoto M, Wiedmeyer HM; IFCC Working Group on HbA1c Standardization.* IFCC reference system for measurement of hemoglobin A1c in human blood and the national standardization schemes in the United States, Japan, and Sweden: a method-comparison study. Clin Chem 2004; 50: 166–174
24 *Geistanger A, Arends S, Berding C, Hoshino T, Jeppsson JO, Little R, Siebelder C, Weykamp C; IFCC Working Group on Standardization of Hemoglobin A1c.* Statistical methods for monitoring the relationship between the IFCC reference measurement procedure for hemoglobin A1c and the designated comparison methods in the United States, Japan, and Sweden. Clin Chem 2008; 54: 1379–1385
25 *The Examination Committee of Criteria for 'Obesity Disease' in Japan, Japan Society for the study of Obesity.* New criteria for 'obesity disease' in Japan. Circ J 2002; 66: 987–992
26 *Mancia G, De Backer G, Dominiczak A, Cifkova R, Fagard R, Germano G, Grassi G, Heagerty AM, Kjeldsen SE, Laurent S, Narkiewicz K, Ruilope L, Rynkiewicz A, Schmieder RE, Boudier HA, Zanchetti A, Vahanian A, Camm J, De Caterina R, Dean V, Dickstein K, Filippatos G, Funck-Brentano C, Hellemans I, Kristensen SD, McGregor K, Sechtem U, Silber S, Tendera M, Widimsky P, Zamorano JL, Erdine S, Kiowski W, Agabiti-Rosei E, Ambrosioni E, Lindholm LH, Viigimaa M, Adamopoulos S, Agabiti-Rosei E, Ambrosioni E, Bertomeu V, Clement D, Erdine S, Farsang C, Gaita D, Lip G, Mallion JM, Manolis AJ, Nilsson PM, O'Brien E, Ponikowski P, Redon J, Ruschitzka F, Tamargo J, van Zwieten P, Waeber B, Williams B; Management of Arterial Hypertension of the European Society of Hypertension; European Society of Cardiology.* 2007 Guidelines for the Management of Arterial Hypertension: The Task Force for the Management of Arterial Hypertension of the European Society of Hypertension (ESH) and of the European Society of Cardiology (ESC). J Hypertens 2007; 25: 1105–1187 Errata: J Hypertens 2007; 25: 1749
27 *National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III).* Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) final report. Circulation 2002; 106: 3143–3421
28 *International Expert Committee.* International Expert Committee report on the role of the A1C assay in the diagnosis of diabetes. Diabetes Care 2009; 32: 1327–1334
29 *Nakanishi N, Nakamura K, Nakajima K, Suzuki K, Tatara K.* Coffee consumption and decreased serum gamma-glutamyltransferase: a study of middle-aged Japanese men. Eur J Epidemiol 2000; 16: 419–423
30 *Kugelman A, Choy HA, Liu R, Shi MM, Gozal E, Forman HJ.* gamma-Glutamyl transpeptidase is increased by oxidative stress in rat alveolar L2 epithelial cells. Am J Respir Cell Mol Biol 1994; 11: 586–592
31 *Lim JS, Yang JH, Chun BY, Kam S, Jacobs DR Jr, Lee DH.* Is serum gamma-glutamyltransferase inversely associated with serum antioxidants as a marker of oxidative stress? Free Radic Biol Med 2004; 37: 1018–1023
32 *Yamada J, Tomiyama H, Yambe M, Koji Y, Motobe K, Shiina K, Yamamoto Y, Yamashina A.* Elevated serum levels of alanine aminotransferase and gamma glutamyltransferase are markers of inflammation and oxidative stress independent of the metabolic syndrome. Atherosclerosis 2006; 189: 198–205
33 *Ames BN, Cathcart R, Schwiers E, Hochstein P.* Uric acid provides an antioxidant defense in humans against oxidant- and radical-caused aging and cancer: a hypothesis. Proc Natl Acad Sci USA 1981; 78: 6858–6862
34 *Nieto FJ, Iribarren C, Gross MD, Comstock GW, Cutler RG.* Uric acid and serum antioxidant capacity: a reaction to atherosclerosis? Atherosclerosis 2000; 148: 131–139
35 *Muraoka S, Miura T.* Inhibition by uric acid of free radicals that damage biological molecules. Pharmacol Toxicol 2003; 93: 284–289
36 *Wallace TM, Utzschneider KM, Tong J, Carr DB, Zraika S, Bankson DD, Knopp RH, Kahn SE.* Relationship of liver enzymes to insulin sensitivity and intra-abdominal fat. Diabetes Care 2007; 30: 2673–2678
37 *Thamer C, Tschritter O, Haap M, Shirkavand F, Machann J, Fritsche A, Schick F, Häring H, Stumvoll M.* Elevated serum GGT concentrations predict reduced insulin sensitivity and increased intrahepatic lipids. Horm Metab Res 2005; 37: 246–251
38 *Wagenknecht LE, Langefeld CD, Scherzinger AL, Norris JM, Haffner SM, Saad MF, Bergman RN.* Insulin sensitivity, insulin secretion, and abdominal fat: the Insulin Resistance Atherosclerosis Study (IRAS) Family Study. Diabetes 2003; 52: 2490–2496
39 *Marchesini G, Brizi M, Morselli-Labate AM, Bianchi G, Bugianesi E, McCullough AJ, Forlani G, Melchionda N.* Association of nonalcoholic fatty liver disease with insulin resistance. Am J Med 1999; 107: 450–455
40 *Facchini F, Chen YD, Hollenbeck CB, Reaven GM.* Relationship between resistance to insulin-mediated glucose uptake, urinary uric acid clearance, and plasma uric acid concentration. JAMA 1991; 266: 3008–3011
41 *Leyva F, Wingrove CS, Godsland IF, Stevenson JC.* The glycolytic pathway to coronary heart disease: a hypothesis. Metabolism 1998; 47: 657–662
42 *Saito Y, Yagyu K, Hattori Y, Ohno K, Okamoto N, Takahashi A, Ohtsuka S, Ohtsuka T, Maeda K, Okamoto K, Kato T.* A study on fatty liver in health examination participants. Nippon Eiseigaku Zasshi 1989; 44: 953–961 (in Japanese)
43 *Lee DH, Steffen LM, Jacobs DR Jr.* Association between serum gamma-glutamyltransferase and dietary factors: the Coronary Artery Risk Development in Young Adults (CARDIA) Study. Am J Clin Nutr 2004; 79: 600–605
44 *Choi HK, Liu S, Curhan G.* Intake of purine-rich foods, protein, and dairy products and relationship to serum levels of uric acid: the Third National Health and Nutrition Examination Survey. Arthritis Rheum 2005; 52: 283–289
45 *Choi HK, Curhan G.* Coffee, tea, and caffeine consumption and serum uric acid level: the third national health and nutrition examination survey. Arthritis Rheum 2007; 57: 816–821
46 *Choi HK, Curhan G.* Beer, liquor, and wine consumption and serum uric acid level: the Third National Health and Nutrition Examination Survey. Arthritis Rheum 2004; 51: 1023–1029

Reaction Time as a Predictor of Mortality: The Radiation Effects Research Foundation Adult Health Study

MICHIKO YAMADA, MD, MASAKI SHIMIZU, MD, FUMIYOSHI KASAGI, PHD, AND HIDEO SASAKI, MD

Objective: We investigated the association between reaction time (RT) and mortality in middle-aged and older atomic bomb survivors and their unexposed controls over a period of 30 years. **Methods:** During 1970–72, 4912 participants of the Adult Health Study cohort in Hiroshima, Japan, underwent biologic tests including RT. Mortality was followed to the end of 2003. **Results:** In a multivariate-adjusted model, the hazard ratio (HR) for 1-standard deviation increments of RT was 1.08 (95% confidence interval [CI] = 1.03–1.13) for men, 1.22 (95% CI = 1.16–1.28) for women, and 1.13 (95% CI = 1.09–1.16) for all. When the analysis was performed by sex, age, and follow-up period, a consistent increase of mortality with increments of RT was observed. The HR for mortality for the highest RT quintile was higher than that of the lowest quintile in all sex-age groups. A significant positive association between mortality risk and RT was observed even after 20 years of follow-up ($p = .03$ in men, $p < .001$ in women). RT and radiation dose were risk factors for mortality independent of conventional risk factors such as smoking, high blood pressure, and diabetes mellitus. Interaction between RT and radiation dose had no significant effect on mortality in men. Although increased radiation dose reduced the HR for mortality per RT increment in women, RT and radiation dose were still significant predictors of mortality. **Conclusions:** RT is a consistently strong predictor of mortality. Although mortality risk increased with radiation dose, radiation did not accelerate the relationship between RT and mortality.
Key words: reaction time, mortality, longitudinal study, atomic bomb survivors.

RT = reaction time; **AHS** = Adult Health Study; **RERF** = Radiation Effects Research Foundation; **DM** = diabetes mellitus; **DS02** = the Radiation Effects Research Foundation 2002 Dosimetry System; **HR** = hazard ratio; **CI** = confidence interval.

Reaction time (RT), an index of physiologic and cognitive function, indicates processing speed and involves many factors such as visual cognition, sensory-motor speed, motor control/motor speed, and attention (1,2). The UK Health and Lifestyle Survey, which includes the largest population-based sample of RT data, reported a precise, sex-dependent pattern of RT increasing with age, but the study was only cross sectional (1). The Baltimore Longitudinal Study of Aging showed, over a period of 8 years, that both simple and choice RTs are strongly associated with age (2). These findings support the notion that RT can be a useful marker of aging.

Because RT is a measure of processing speed, which is one domain of cognitive function, the relationship between RT and mortality is of interest in terms of the broader association between cognition and mortality (3). Although it is unclear whether the association between cognition and mortality is pervasive or specific to particular functions, processing speed is a strong predictor of mortality (4–6). Measurements of processing speed other than RT include the Digit Symbol Substitution Test and inspection time (3).

To date, few findings on an association between mortality and RT have been reported in population-based samples (7–9). Although significant age and sex differences in RT have been reported (1,2), there are no reports on age-sex–specific relationships between RT and mortality because of the small number of available study participants. The Scottish Twenty-07 Study (7) involved participants approximately 56 years old, and the Baltimore Longitudinal Study of Aging (8) included only men. The UK Health and Lifestyle Survey, which is the only study that covers both sexes of all adult ages, reported that mortality increased with RT after adjusting for age, sex, socioeconomic status, health behaviors, and health status (9). When the analysis was split into age groups and follow-up periods, however, results were not consistent. A significant association was noted in the 20- to 39-year-old group and the 60-plus-year-old group, but not in the 40- to 59-year-old group (9). In the Baltimore Longitudinal Study of Aging, a significant increase in mortality risk was detected before 10 years of follow-up but not after (8).

The Adult Health Study (AHS) is a population-based longitudinal clinical study of atomic bomb survivors and unexposed controls. Comprising one of the largest cohort studies of middle-aged and elderly persons, it constitutes an ongoing investigation that was first begun in 1958 by the Atomic Bomb Casualty Commission and was continued by its successor, the Radiation Effects Research Foundation (RERF) (10). In the AHS, a battery of noninvasive physiologic tests designed to indicate physiologic age was used in 1970 to 1972 in a cross-sectional survey to test the hypothesis that ionizing radiation hastens aging (11). The tests included handgrip strength, auditory acuity, vibration perception, skin elasticity, and RT. At that time, no association between radiation and specific physiologic functions was found (12). Here, using AHS data collected for 30 years, we investigated the association between RT and mortality and whether radiation affected that association.

PARTICIPANTS AND METHODS

The study participants were drawn from 6129 members of the AHS cohort who underwent a battery of noninvasive age-related physiologic function tests that included RT between July 1970 and June 1972 in Hiroshima. Details of the examinations performed are described elsewhere (10). The RERF institutional review boards (the Research Protocol Review Committee and the

From the Department of Clinical Studies (M.Y., M.S., H.S.), Radiation Effects Research Foundation, Hiroshima and Nagasaki, Japan; Tokai Hokuriku Regional Bureau of Health and Welfare (M.S.), Nagoya, Japan; Institute of Radiation Epidemiology (F.K.), Radiation Effects Association, Tokyo, Japan; and The Health Management and Promotion Center (H.S.), Hiroshima Atomic Bomb Casualty Council, Hiroshima, Japan.

Address correspondence and reprint requests to Michiko Yamada, MD, Department of Clinical Studies, Radiation Effects Research Foundation, 5-2 Hijiyama Park, Minami-ku, Hiroshima 732-0815, Japan. E-mail: yamada@rerf.or.jp

Received for publication January 16, 2012; revision received October 22, 2012.
DOI: 10.1097/PSY.0b013e3182822b4a

0033-3174/13/7502–0154

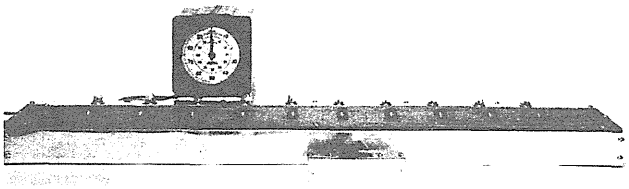

Figure 1. Light extinction instrument used to measure reaction time.

Human Investigation Committee) approved this study. All participants provided informed consent.

Reaction Time

RT was measured with a light extinction testing instrument. (Fig. 1) The instrument consists of a row of 10 neon pilot lights mounted on a rectangular board. The lights are lit in a fixed order, and the participant must extinguish each as it becomes illuminated by pressing the switch beneath the board. RT was defined as the length of time between the extinguishing of the 1st light and the extinguishing of the 10th lamp and was measured by a standard electric timer accurate to 0.01 second. Before measurement, participants practiced with a separately mounted switch. Analysis was restricted to 4912 participants 35 to 74 years old (1695 men and 3217 women) who had no physical or cognitive impairment and who had undergone a clinical examination.

Radiation Dose Estimates

We used individual estimates of weighted kerma dose, in which survivor location and shielding by house and terrain were taken into account based on the RERF 2002 dosimetry system (DS02) (13). We calculated weighted kerma doses as weighted sums of the γ-ray and neutron components, giving the neutron component a weight of 10 and truncating the DS02 estimates to 4 Gy in consideration of the imprecision for proximal survivors (14). Table 1 shows the distribution of the participants by DS02 category; 12.3% of the participants had doses of 1 Gy or greater, and 45.9% were unexposed (dose <0.001 Gy).

Clinical Examination

The clinical examination included a medical history, a general physical examination, determination of height and body weight and serum cholesterol concentration, and a self-administrated questionnaire. Body mass index was calculated as body weight (in kilograms) divided by the square of the height (in meters). Blood pressure was measured by using a sphygmomanometer on the right arm with the participant seated. Information on tobacco smoking and alcohol intake was obtained in 1965 to 1968 with a self-administrated questionnaire. Smoking habit and alcohol intake categories were "never," "former," and "current." Diabetes mellitus (DM) prevalence was determined by medical history and the use of medication.

Mortality Follow-Up

Mortality was followed for the entire study sample from the time of the physiologic tests and clinical examinations in 1970 to 1972 to the end of 2003. Deaths were routinely identified through Japan's *Koseki* (obligatory household registry) system, and the degree of ascertainment was essentially complete. All deaths not caused by external causes were included in our analysis.

Statistical Analysis

We used multiple regression analysis to examine the association between RT and sex, age, sex-age interaction, and radiation dose. We used the Cox proportional hazard model to calculate the hazard ratio (HR) of mortality associated with RT under various models including sex, age, radiation dose, and their interaction term with RT (model 1: no adjustment; model 2: including radiation dose; model 3: including radiation dose and interaction

TABLE 1. Baseline Characteristics of the Study Participants

Variable	Total	Men	Women
No. participants	4912	1695	3217
Age at baseline, y	54.4 (10.9)	55.5 (11.1)	53.8 (10.7)
Systolic blood pressure, mm Hg	127.4 (23.6)	131.0 (23.6)	125.5 (23.4)
Diastolic blood pressure, mm Hg	79.8 (26.2)	81.9 (26.1)	78.7 (26.2)
Total cholesterol, mg/dl	192.3 (37.0)	183.7 (34.3)	200.7 (37.6)
Body mass index, kg/m^2	22.3 (3.5)	21.6 (3.1)	22.6 (3.6)
% Current smokers	32.7	68.4	13.9
% Current alcohol drinkers	34.3	70.0	15.7
% With diabetes mellitus	8.7	14.3	5.7
Reaction time, s			
M (SD)	9.26 (2.72)	9.10 (2.76)	9.34 (2.69)
Median	8.6	8.5	8.7
1st, 3rd quartile	7.6, 10.2	7.4, 10.0	7.6, 10.3
Minimum, maximum	5.0, 50.7	5.0, 45.6	5.3, 50.7
No. participants with <0.001 Gy	2253	772	1481
Dose among exposed participants, Gy	0.83 (1.13)	0.92 (1.20)	0.81 (1.10)

Continuous variables are shown as M (SD). M = mean; SD = standard deviation.

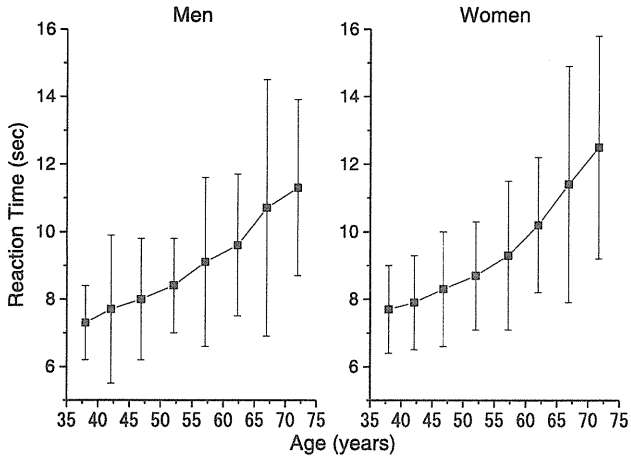

Figure 2. Mean and SD of reaction time for each sex by age. SD = standard deviation.

between RT and radiation dose; model 4: including age, sex, radiation dose, and interaction between RT and radiation dose; model 5: including age, and sex; model 6: including age, sex, and interaction between RT and sex; model 7: including age, sex, and interaction between RT and age).

We also estimated the HR of mortality associated with RT, after adjusting for potential confounding factors singly and multivariately. The factors considered in the model that were obtained at baseline (i.e., at the time of RT measurements) were age, body mass index, systolic blood pressure, total cholesterol, smoking and drinking habits, DM, and radiation dose, all of which are known risk factors for cardiovascular disease or cancer. The significance of interaction between RT and other covariate was assessed by testing their product.

In the sex-age-specific analyses, we used 3 age categories (35–54, 55–64, and 65–74 years at baseline). In the sex-age–specific RT quintiles analysis, we estimated HR with the lowest quintile as the reference. In the aforementioned analyses, we excluded deaths that occurred in the 2 years that followed the baseline examination so as to separate measurements that were predictive of future events from those caused by latent disease. We calculated secular trends of multivariate-adjusted HR for 1-standard deviation (SD) increments of RT of all deaths by dividing up the follow-up period based on the time since baseline examination (within 5 years, after 5 years, after 10 years, after 15 years, and after 20 years). We used SAS software for all analyses (SAS version 9.1; SAS Institute, Cary, NC, USA).

RESULTS

Table 1 shows the baseline characteristics of the study participants. RT was positively associated with age in both sexes (Fig. 2), and the association was stronger in women than in men (p = .04). No radiation effect on RT was evident in either sex (p = .11 in men, p = .09 in women). Figure 3 shows the Kaplan-Meier survival curve by sex.

After the exclusion of deaths (81 men and 112 women) that occurred during the 2 years after baseline and due to external causes, 1171 male and 1593 female deaths remained for analysis. RT was a consistent and significant mortality risk in various models including age, sex, radiation dose, and interaction between RT and each variable (Table 2). The HRs of mortality associated with RT were similar when potential confounding factors were adjusted singly and multivariately (Tables 3 and 4). The multivariate-adjusted model revealed that mortality trend had a significant, positive association with age, systolic blood pressure, total cholesterol, smoking, DM, radiation dose, and RT (Table 4). In the sex-specific analysis, the HRs of mortality for 1-SD increments of RT and 1-Gy increments of radiation dose were significant (Table 4), and the interaction effect on mortality between RT and radiation dose was not significant in men (p = .97) and significant and negative in women (estimate, −0.032; standard error of the estimate = 0.01174). Although the HR for 1-SD increments of RT was reduced approximately 10% among women exposed to 1 Gy of radiation versus those unexposed, RT and radiation dose were still significant predictors of mortality (p < .001 for RT and radiation dose). The multivariate-adjusted HR for all deaths for each 1-SD increment of RT was significantly higher in all sex-age groups, except that of men aged 65 to 74 years at baseline (Table 5). When RT categories were divided into quintiles for each age and sex group (Fig. 4), the HRs for mortality were significantly higher for the highest quintile than for the lowest quintile (the reference group). The HR for the highest quintile in men was 1.63 (95% confidence interval [CI] = 1.13–2.38) for the 35- to 54-year-old group, 1.41 (95% CI = 1.03–1.93) for the 55- to 64-year-old group, and 2.00 (95% CI = 1.42–2.81) for the 65- to 74-year-old group; in women, it was 1.39 (95% CI = 1.02–1.92) for the 35- to 54-year-old group, 1.51 (95% CI = 1.13–2.03) for the 55- to 64-year-old group, and 1.67 (95% CI = 1.28–2.18) for 65- to 74-year-old group.

The significantly increased HR for mortality for 1-SD increment of RT was observed during the entire follow-up period. It was highest in both men (HR = 1.07, 95% CI = 1.01–1.12) and women (HR = 1.23, 95% CI = 1.10–1.34) during the initial 5 years, but even after 20 years, a significant positive association between mortality risk and RT was observed in both men (HR = 1.06, 95% CI = 1.00–1.12, p = .03) and women (HR = 1.17, 95% CI = 1.10–1.24, p < .001).

DISCUSSION

In this study, we demonstrated a consistent and significant association between RT and mortality in middle-aged and elderly

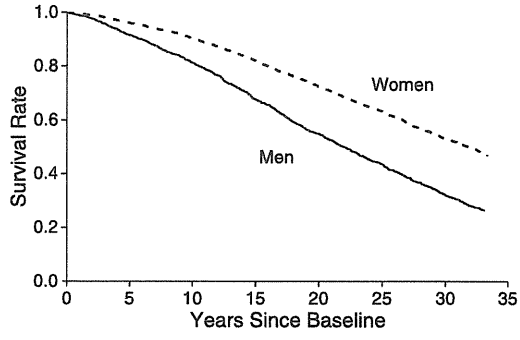

Figure 3. Kaplan-Meier survival curve by sex.

TABLE 2. Hazard Ratio (HR) of Mortality Associated With Reaction Time (RT) Based on Various Models

Model	HR (95% CI)[a]						
	Reaction Time (1 SD)	Dose (1 Gy)	Interaction of RT and Dose	Age (10 y)	Sex (Women/Men)	Interaction of RT and Sex	Interaction of RT and Age (10 y)
1	1.37 (1.35–1.39)***						
2	1.38 (1.35–1.40)***	1.13 (1.08–1.17)***					
3	1.36 (1.33–1.38)***	0.75 (0.66–0.86)***	1.12 (1.08–1.16)***				
4	1.15 (1.11–1.18)***	1.26 (1.08–1.47)**	0.98 (0.94–1.02)	2.73 (2.61–2.86)***	0.52 (0.48–0.56)***		
5	1.14 (1.11–1.17)***			2.71 (2.59–2.84)***	0.51 (0.48–0.55)***		
6	1.08 (1.04–1.13)***			2.67 (2.55–2.80)***	0.32 (0.25–0.40)***	1.14 (1.07–1.20)***	
7	1.41 (1.05–1.87)*			3.01 (2.58–3.52)***	0.51 (0.48–0.55)***		0.97 (0.93–1.01)

[a] Cox proportional hazard model.
* $p < .05$.
** $p < .01$.
*** $p < .001$.
CI = confidence interval; SD = standard deviation.

persons, even when we split the participants into sex-age groups and follow-up period groups by sex. In a previous analysis of this cohort, we found grip strength and biologic score (the first principal component score of five physiologic functions) to be consistent predictors of all causes of mortality for 30 years (15,16), a finding similar to the association between RT and mortality found in this study. This result is consistent with a report indicating that certain domains of physiologic function predict mortality in the elderly (17). Intercorrelations between physiologic functions (11,18) in middle-aged and elderly populations may support a similar association between various physiologic functions and mortality.

We found no previous reports of a significant association between RT and mortality across different age groups or of an association that persisted for more than 10 years of follow-up, but studies conducted for men and women including broad age ranges and a long follow-up period are few (8,9).

RT was measured somewhat differently in this study from previous studies, where the procedures were simpler and the latent time measured was shorter. In this study, the entire length of time between the extinguishing of the 1st and the 10th lamps was measured, and the mean RT value was approximately 9 seconds. The Health and Lifestyle Survey (8) and the Baltimore Longitudinal Study of Aging (9) used different visual and auditory stimuli. Both used the mean or median time in milliseconds between each stimulus to the time of response, and almost all of the individual RTs fell within a period of 1 second. The movement from one switch to another switch was required in the Light Extinction Test used in this study, but in the previously mentioned two studies, the participants activated the switches without hand movement. The strong relationship between RT and mortality in the present study may be explained by the greater proportion of time allowed in the Light Extinction Test for hand movement. Indeed, control over and execution of the movement itself contribute more to the association between RT and mortality than RT test decision time (8). In addition, we confirmed the significant association between RT and mortality among restricted unexposed participants seen in other cohort studies (8,9) (data not shown).

TABLE 3. Hazard Ratio (HR) of Mortality Per SD Increment of Reaction Time in Age-, Sex-, and Other Variable–Adjusted Models[a]

Added Variable for Adjustment	HR	95% CI
None	1.14	(1.11–1.17)***
SBP (for 10 mm Hg)	1.13	(1.10–1.16)***
Total cholesterol (10 mg/dl)	1.14	(1.11–1.17)***
Body mass index (1 kg/m^2)	1.14	(1.11–1.17)***
Smoking (current versus nonsmoker)	1.14	(1.11–1.17)***
Drinking (current versus nondrinker)	1.14	(1.11–1.18)***
Diabetes mellitus (yes versus no)	1.15	(1.11–1.18)***
Radiation (1 Gy)	1.14	(1.11–1.17)***

[a] Cox proportional hazard model.
*** $p < .001$.
SD = standard deviation; CI = confidence interval; SBP, systolic blood pressure.

TABLE 4. Multivariate-Adjusted Hazard Ratio (HR) of Mortality

Variable	HR[a] (95% CI)		
	Total	Men	Women
Age (10 y)	2.49 (2.37–2.62)***	2.48 (2.27–2.72)***	2.54 (2.37–2.72)***
SBP (for 10 mm Hg)	1.11 (1.09–1.13)***	1.12 (1.10–1.16)***	1.09 (1.07–1.11)***
Total cholesterol (10 mg/dl)	0.97 (0.96–0.98)***	0.97 (0.95–0.99)*	0.98 (0.96–1.00)*
Body mass index (1 kg/m^2)	0.99 (0.97–1.00)*	0.99 (0.97–1.01)	0.99 (0.98–1.01)
Smoking (current versus nonsmoker)	1.77 (1.61–1.99)***	1.33 (1.10–1.63)**	1.60 (1.40–1.83)***
Drinking (current versus nondrinker)	1.09 (0.97–1.19)	1.05 (0.91–1.20)	0.89 (0.77–1.02)
Diabetes mellitus (yes versus no)	1.63 (1.45–1.83)***	1.40 (1.19–1.63)***	1.82 (1.51–2.18)***
Radiation dose (1 Gy)	1.17 (1.11–1.22)***	1.16 (1.08–1.24)***	1.18 (1.11–1.25)***
Reaction time (1 SD[b])	1.13 (1.09–1.16)***	1.08 (1.03–1.13)***	1.22 (1.16–1.28)***

[a] Cox proportional hazard model.
[b] Sex specific.
* $p < .05$.
** $p < .01$.
*** $p < .001$.
CI = confidence interval; SBP = systolic blood pressure; SD = standard deviation.

RT is a measure of processing speed, which is one representative domain of cognitive function (3,19). The association between RT and mortality reported here supports an association between cognition and mortality (5). Various measurements including RT, the Digit Symbol Substitution Test, and inspection time are used to assess processing speed (3). There are studies showing that the Digit Symbol Substitution Test is a strong, independent predictor of mortality and that processing speed is a specific marker of central nervous system aging (4,20–22), but all study follow-up periods were less than 10 years. A strong correlation between physiologic function and cognitive function was observed in a cross-sectional study (23), and cognitive function was shown to predict future physiologic function in a longitudinal study (24). Because the physiologic function of grip strength was a predictor of all mortalities in this cohort (15,16) and in others (25,26), the persistent effect of baseline RT on mortality for 30 years might be caused by cross-sectional and longitudinal interrelationship between cognitive and physiologic functions. After adjustment for age and sex, there was little attenuation of the HR associated with RT when potential confounding factors at baseline were added.

In this study, the only one to examine RT as a predictor of mortality among atomic bomb survivors, we found no association between radiation dose and RT at baseline, which is in agreement with a previous RERF report (12). The effect of ionizing radiation on aging was a subject of research in the late 1940s to the 1960s (27,28), and more recent research has revived concern (29). Biologic mechanisms associated with aging, such as oxidative stress and DNA aberration, have been shown to be associated with radiation, as well (29). Recent epidemiological studies of atomic bomb survivors showed an increase not only in neoplastic diseases but also in nonneoplastic diseases including those of the circulatory and respiratory system (30). Similar degenerative changes relevant to aging and radiation exposure in the hematological system (31) and T-cell function (32) were found among AHS participants. However, other age-related markers including postmortem morphological changes such as neurofibril tangle and senile plaques do not show radiation effects (33). As of now, whether ionizing radiation exposure accelerates aging is controversial, especially for relatively low-dose exposures compared with those delivered during radiation therapy (29). The HR for radiation dose found in this study is compatible with a pre-

TABLE 5. Multivariate-Adjusted Hazard Ratio (HR) of Mortality Per SD Increment of Reaction Time by Age at Baseline

Age at Baseline, y	Total		Men		Women	
	Deaths/No. at Risk	HR (95% CI)	Deaths/No. at Risk	HR (95% CI)	Deaths/No. at Risk	HR (95% CI)
35-54	773/2493	1.18 (1.11–1.26)***	341/749	1.15 (1.04–1.26)**	432/1744	1.23 (1.13–1.33)***
55-64	972/1264	1.17 (1.10–1.24)***	429/491	1.18 (1.07–1.29)***	543/773	1.17 (1.07–1.27)***
65-74	1019/1076	1.13 (1.06–1.18)***	401/413	1.06 (0.97–1.14)	618/663	1.28 (1.17–1.38)***

HR for sex-age–specific 1-SD increment of reaction time was estimated by using the Cox proportional hazard model, after adjusting for age, body mass index, systolic blood pressure, total cholesterol, smoking and drinking habits, diabetes mellitus, and radiation dose. SD = standard deviation; CI = confidence interval.
** $p < .01$.
*** $p < .001$.

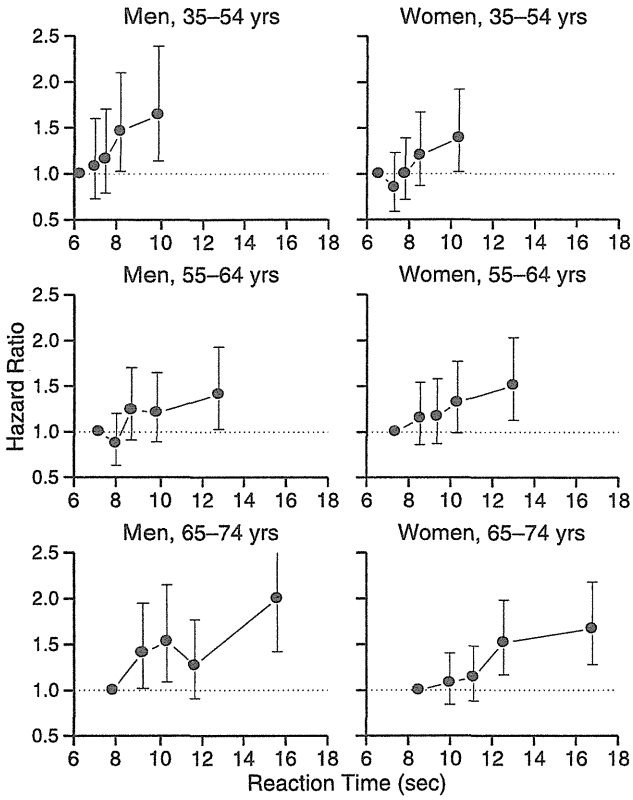

Figure 4. Multivariate-adjusted HR and 95% CI for each reaction time category divided into quintiles for each age and sex group, with the lowest quintile group used as the reference point. HR = hazard ratio; CI = confidence interval.

vious report on mortality in atomic bomb survivors (30). Also, although the biologic implications of the attenuated relationship between RT and mortality among exposed women are unknown, at least radiation did not accelerate that relationship.

A major limitation of this study is that certain factors that might affect the observed relationship between RT and mortality, such as education, physical activity, and occupation, could not be adjusted for the analysis because of the number of participants for whom there was inadequate information. Another limitation is that we used total mortality, not cause-specific mortality. Nevertheless, RT was shown to be a strong and consistent predictor of mortality in a large sample of atomic bomb survivors and unexposed controls that covered a wide age range over a period of 30 years. Radiation did not accelerate the relationship between RT and mortality.

Source of Funding and Conflicts of Interest: The RERF, Hiroshima and Nagasaki, Japan, is a private, nonprofit foundation funded by the Japanese Ministry of Health, Labour and Welfare and the US Department of Energy, the latter in part through Department of Energy Award DE-HS0000031 to the National Academy of Sciences. This publication was supported by RERF Research Protocols RP Nos. 2-75 and 4-86 and, in part, by a Grant-in-Aid from the Ministry of Health, Labour and Welfare, Health and Labor Sciences Research Grants, Japan (H20-Junkankitou [Seishuu]: Ippan-013 and H23-Junkankitou [Seishuu]: Ippan-005). The views of the authors do not necessarily reflect those of the two governments. Professional editing was provided by SciWrite Biomedical Writing and Editing Services. The authors report no conflicts of interest.

REFERENCES

1. Der G, Deary IJ. Age and sex differences in reaction time in adulthood: results from the United Kingdom Health and Lifestyle Survey. Psychol Aging 2006;21:62–73.
2. Fozard JL, Vercryssen M, Reynolds SL, Hancock PA, Quilter RE. Age differences and changes in reaction time: the Baltimore Longitudinal Study of Aging. J Gerontol 1994;49:P179–89.
3. Deary IJ, Johnson W, Starr JM. Are processing speed tasks biomarkers of cognitive aging? Psychol Aging 2010;25:219–28.
4. Pavlik VN, de Moraes SA, Szklo M, Knopman DS, Mosley TH Jr, Hyman DJ. Relation between cognitive function and mortality in middle-aged adults: the atherosclerosis risk in communities study. Am J Epidemiol 2003;157:327–34.
5. Sabia S, Gueguen A, Marmot MG, Shipley MJ, Ankri J, Singh-Manoux A. Does cognition predict mortality in midlife? Results from the Whitehall II cohort study. Neurobiol Aging 2010;31:688–95.
6. Smits CH, Deeg DJ, Kriegsman DM, Schmand B. Cognitive functioning and health as determinants of mortality in an older population. Am J Epidemiol 1999;150:978–86.
7. Deary IJ, Der G. Reaction time explains IQ's association with death. Psychol Sci 2005;16:64–9.
8. Metter EJ, Schrager M, Ferrucci L, Talbot LA. Evaluation of movement speed and reaction time as predictors of all-cause mortality in men. J Gerontol A Biol Sci Med Sci 2005;60:840–6.
9. Shipley BA, Der G, Taylor MD, Deary IJ. Cognition and all-cause mortality across the entire adult age range: health and lifestyle survey. Psychosom Med 2006;68:17–24.
10. Yamada M, Wong FL, Fujiwara S, Akahoshi M, Suzuki G. Noncancer disease incidence in atomic bomb survivors, 1958–1998. Radiat Res 2004; 161:622–32.
11. Hollingsworth JW, Hashizume A, Jablon S. Correlation between tests of aging in Hiroshima subjects, an attempt to define "physiologic" age. Yale J Biol Med 1965;38:11–36.
12. Belsky JL, Moriyama I, Fujita S, Kawamoto S. Aging Studies in Atomic Bomb Survivors. Hiroshima: Radiation Effects Research Foundation; 1978.
13. Cullings HM, Fujita S, Funamoto S, Grant EJ, Kerr GD, Preston DL. Dose estimation for atomic bomb survivor studies: its evolution and present status. Radiat Res 2006;166:219–54.
14. Pierce DA, Stram DO, Vaeth M. Allowing for random errors in radiation dose estimates for the atomic bomb survivor data. Radiat Res 1990;123: 275–84.
15. Sasaki H, Kasagi F, Yamada M, Fujita S. Grip strength predicts cause-specific mortality in middle-aged and elderly persons. Am J Med 2007; 120:337–42.
16. Kasagi F, Yamada M, Sasaki H, Fujita S. Biologic score and mortality based on a 30-year mortality follow-up: Radiation Effects Research Foundation Adult Health Study. J Gerontol A Biol Sci Med Sci 2009;64:865–70.
17. Lyyra TM, Leskinen E, Heikkinen E. A cohort study found good respiratory, sensory and motor functions decreased mortality risk in older people. J Clin Epidemiol 2005;58:509–16.
18. Anstey KJ, Dear K, Christensen H, Jorm AF. Biomarkers, health, lifestyle, and demographic variables as correlates of reaction time performance in early, middle, and late adulthood. Q J Exp Psychol A 2005;58:5–21.
19. Duff K, Beglinger LJ, Moser DJ, Paulsen JS. Predicting cognitive change within domains. Clin Neuropsychol 2010;24:779–92.
20. Fried LP, Kronmal RA, Newman AB, Bild DE, Mittelmark MB, Polak JF, Robbins JA, Gardin JM. Risk factors for 5-year mortality in older adults: the Cardiovascular Health Study. JAMA 1998;279:585–92.
21. Rosano C, Newman AB, Katz R, Hirsch CH, Kuller LH. Association between lower digit symbol substitution test score and slower gait and greater risk of mortality and of developing incident disability in well-functioning older adults. J Am Geriatr Soc 2008;56:1618–25.
22. Swan GE, Carmelli D, LaRue A. Performance on the digit symbol substitution test and 5-year mortality in the Western Collaborative Group Study. Am J Epidemiol 1995;141:32–40.
23. Anstey KJ, Luszcz MA, Sanchez L. A reevaluation of the common factor theory of shared variance among age, sensory function, and cognitive function in older adults. J Gerontol B Psychol Sci Soc Sci 2001;56:P3–11.

24. Elovainio M, Kivimaki M, Ferrie JE, Gimeno D, De Vogli R, Virtanen M, Vahtera J, Brunner EJ, Marmot MG, Singh-Manoux A. Physical and cognitive function in midlife: reciprocal effects? A 5-year follow-up of the Whitehall II study. J Epidemiol Community Health 2009;63:468–73.
25. Al Snih S, Markides KS, Ray L, Ostir GV, Goodwin JS. Handgrip strength and mortality in older Mexican Americans. J Am Geriatr Soc 2002;50:1250–6.
26. Rantanen T. Muscle strength, disability and mortality. Scand J Med Sci Sports 2003;13:3–8.
27. Mewissen DJ, Comar CL, Trum BF, Rust JH. A formula for chronic radiation dosage versus shortening of life span: application to a large mammal. Radiat Res 1957;6:450–9.
28. Upton AC, Kimball AW, Furth J, Christenberry KW, Benedict WH. Some delayed effects of atom-bomb radiations in mice. Cancer Res 1960; 20(pt 2):1–60.
29. Richardson RB. Ionizing radiation and aging: rejuvenating an old idea. Aging (Albany NY) 2009;1:887–902.
30. Ozasa K, Shimizu Y, Suyama A, Kasagi F, Soda M, Grant EJ, Sakata R, Sugiyama H, Kodama K. Studies of the mortality of atomic bomb survivors, Report 14, 1950–2003: an overview of cancer and noncancer diseases. Radiat Res 2012;177:229–43.
31. Wong FL, Yamada M, Tominaga T, Fujiwara S, Suzuki G. Effects of radiation on the longitudinal trends of hemoglobin levels in the Japanese atomic bomb survivors. Radiat Res 2005;164:820–7.
32. Kusunoki Y, Yamaoka M, Kubo Y, Hayashi T, Kasagi F, Douple EB, Nakachi K. T-cell immunosenescence and inflammatory response in atomic bomb survivors. Radiat Res 2010;174:870–6.
33. Sasaki H, Kodama K, Yamada M. A review of forty-five years study of Hiroshima and Nagasaki atomic bomb survivors. Aging. J Radiat Res (Tokyo) 1991;32:310–26.

Advance Publication by-J-STAGE

Circulation Journal
Official Journal of the Japanese Circulation Society
http://www.j-circ.or.jp

Comparison of Predictability of Future Cardiovascular Events Between Chronic Kidney Disease (CKD) Stage Based on CKD Epidemiology Collaboration Equation and That Based on Modification of Diet in Renal Disease Equation in the Japanese General Population

– Iwate KENCO Study –

Masaki Ohsawa, MD; Kozo Tanno, MD; Kazuyoshi Itai, PhD; Tanvir Chowdhury Turin, PhD; Tomonori Okamura, MD; Akira Ogawa, MD; Kuniaki Ogasawara, MD; Tomoaki Fujioka, MD; Toshiyuki Onoda, MD; Yuki Yoshida, MD; Shin-ichi Omama, MD; Yasuhiro Ishibashi, MD; Motoyuki Nakamura, MD; Shinji Makita, MD; Fumitaka Tanaka, MD; Toru Kuribayashi, PhD; Tomiko Koyama, PhD; Kiyomi Sakata, MD; Akira Okayama, MD

Background: Whether estimated glomerular filtration rate (eGFR) calculated using the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) Study equation ($eGFR_{CKDEPI}$) improves risk prediction compared to that calculated using the Modification of Diet in Renal Disease (MDRD) study equation ($eGFR_{MDRD}$) has not been examined in a prospective study in Japanese people.

Methods and Results: Participants (n=24,560) were divided into 4 stages (1, ≥90; 2, 60–89 (reference); 3a, 45–59; 3b+ <45 $ml \cdot min^{-1} \cdot 1.73 m^{-2}$) according to $eGFR_{CKDEPI}$ or $eGFR_{MDRD}$. Endpoints were all-cause death, myocardial infarction (MI) and stroke. Area under the receiver operating characteristic curves (95% confidence intervals) for predicting all-cause death, MI and stroke by $eGFR_{CKDEPI}$ vs. $eGFR_{MDRD}$ were 0.680 (0.662–0.697) vs. 0.582 (0.562–0.602); 0.718 (0.665–0.771) vs. 0.642 (0.581–0.703); and 0.656 (0.636–0.676) vs. 0.576 (0.553–0.599), respectively. Multivariate-adjusted Cox regression and Poisson regression analysis results were similar for adjusted incidence rates and adjusted hazard ratios in each corresponding stage between the 2 models and no differences were found in model assessment parameters. Net reclassification improvement (NRI) for predicting all-cause death, MI and stroke were estimated to be 6.7% (P<0.001), −1.89% (P=0.029) and −0.20% (P=0.421), respectively.

Conclusions: Better discrimination was achieved using $eGFR_{CKDEPI}$ than $eGFR_{MDRD}$ on univariate analysis. NRI analysis indicated that the use of $eGFR_{CKDEPI}$ instead of $eGFR_{MDRD}$ offered a significant improvement in reclassification of death risk.

Key Words: Chronic kidney disease; CKD-EPI equation; Estimated glomerular filtration rate; MDRD equation; Model assessment

Received August 2, 2012; revised manuscript received December 14, 2012; accepted January 15, 2013; released online February 21, 2013 Time for primary review: 39 days

Department of Hygiene and Preventive Medicine, Iwate Medical University, Iwate (M.O., K.T., T. Onoda, K.S.); Department of Neurosurgery (A. Ogawa, K.O., Y.Y., S.O.), Department of Urology (T.F.), Department of Internal Medicine (Y.I., M.N., S.M., F.T.), Iwate Medical University, Morioka; Department of Health and Physical Education, Faculty of Education, Iwate University, Morioka (T. Kuribayashi); Iwate Health Service Association, Morioka (T. Koyama); The First Institute of Health Service, Japan Anti-Tuberculosis Association, Tokyo (K.I., A. Okayama); Department of Preventive Medicine and Public Health, Keio University, Tokyo (T. Okamura), Japan; and Department of Medicine, University of Calgary, Calgary, Alberta (T.C.T.), Canada

Mailing address: Masaki Ohsawa, MD, Department of Hygiene and Preventive Medicine, Iwate Medical University, 2-1-1 Nishitokuta, Yahaba-cho, Iwate 020-3694, Japan. E-mail: masakio@iwate-med.ac.jp

ISSN-1346-9843 doi:10.1253/circj.CJ-12-0982

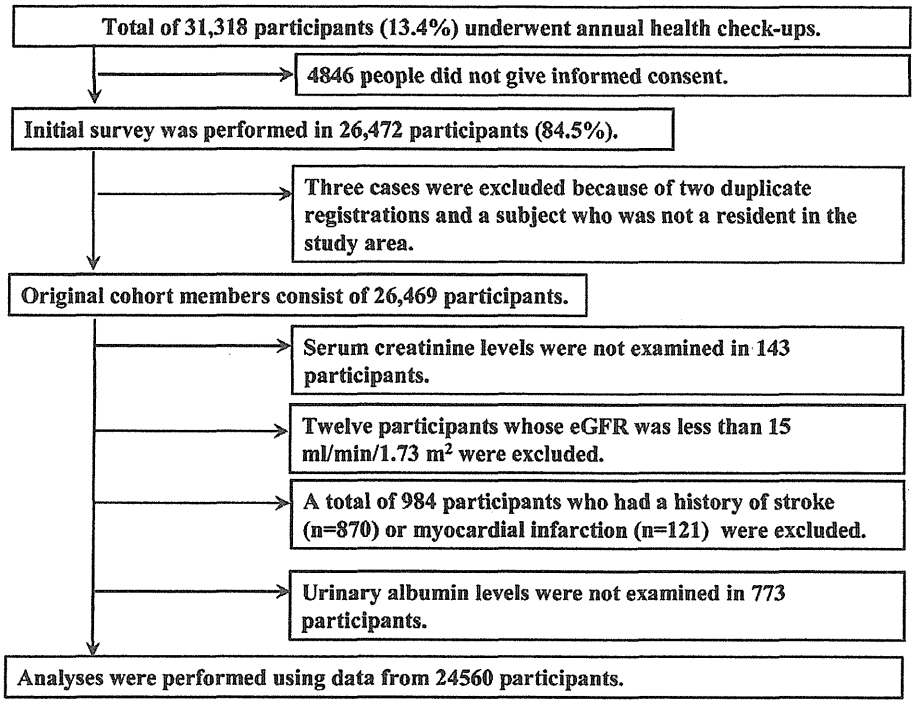

Figure 1. Flow chart of the procedure used to select participants for the Iwate KENCO study. The original cohort consisted of 26,469 participants. We excluded a total of 1,909 participants who lacked data for serum creatinine (n=143), 984 participants with a history of myocardial infarction or stroke (n=984), and 785 participants who lacked data used for analysis. eGFR, estimated glomerular filtration rate.

Chronic kidney disease (CKD) contributes not only to the risk for development of end-stage renal disease but also to the risk for cardiovascular morbidity and mortality.[1] CKD also increases risks for cardiovascular morbidity and mortality in Japanese people.[2–6] The National Kidney Foundation and the American Heart Association have proposed using CKD in cardiovascular risk stratification and treatment guidelines.[1,7] Defining and staging of kidney disease requires combining information on kidney damage, usually detected using albuminuria, and decreased renal function, usually based on glomerular filtration rate (GFR).[7]

The Modification of Diet in Renal Disease (MDRD) Study equation is the most widely used equation.[8] Recently, the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) developed a new equation.[9] The CKD-EPI Study equation was shown to be more accurate than the MDRD Study equation for the calculation of estimated GFR (eGFR).[9] Both the MDRD equation modified by a Japanese coefficient[10] and CKD-EPI equation modified by a Japanese coefficient[11] have recently been developed. The CKD-EPI equation modified by a Japanese coefficient was also shown to be more accurate than the MDRD equation modified by a Japanese coefficient using inulin clearance as a gold standard.[11]

The development of a more accurate equation for eGFR requires reclassification of CKD stage using the new equation instead of the old equation and confirmation of the concordance of CKD stage between the 2 models. Reclassification of CKD stage and correlations between the 2 equations were examined for US subjects.[9,12] Horio et al also examined proportions in each CKD stage separately for the 2 equations.[11]

The development of a more accurate equation for eGFR also requires reassessment of the predictability of CKD in prospective longitudinal studies using the new equation for eGFR. Comparisons of risk predictabilities have been widely used in the cardiovascular field, and new statistical methods have also been developed.[13–20] Recently, Matsushita et al used a new statistical method for comparing risk predictabilities between the 2 types of eGFR.[12] They showed that improved reclassification of CKD stage was observed using $eGFR_{CKDEPI}$ instead of $eGFR_{MDRD}$, with statistical significance in all 4 endpoints (death, myocardial infarction [MI], stroke and end-stage renal disease).

They noted, however, that considerable racial difference existed in correlations between the 2 types of eGFR and in risk predictabilities. They also noted that similar analyses should be performed for ethnicities other than white and African-American people.[12] It is necessary to compare correlations between the 2 types of eGFR and compare risk predictability for CKD stage between the 2 CKD stage models based on $eGFR_{CKDEPI}$ and $eGFR_{MDRD}$ in other ethnicities including Japanese people. Therefore, we compared correlations between the 2 types of eGFR and compared the risk predictability of models based on $eGFR_{CKDEPI}$ and on $eGFR_{MDRD}$ using traditional and newly developed techniques in statistics to examine model accuracy in univariate- and multivariate-adjusted analyses based on a prospective study in Japanese people.

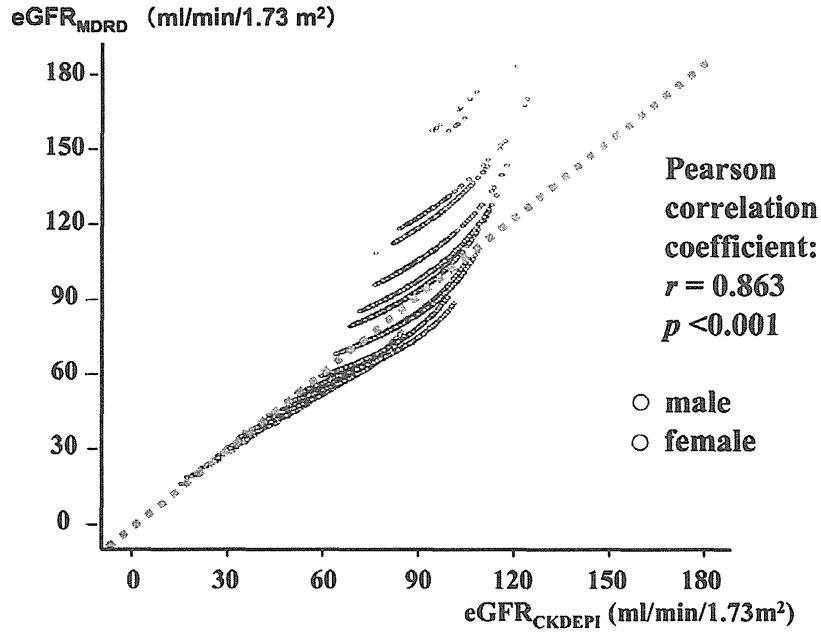

Figure 2. Relationship between eGFRCKDEPI and eGFRMDRD. Points along the diagonal dotted line, accordance between eGFRCKDEPI and eGFRMDRD; points below the diagonal dotted line, underestimation by eGFRMDRD; points over the diagonal dotted line, overestimation by eGFRMDRD. eGFRCKDEPI, estimated glomerular filtration rate (eGFR) calculated using the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) Study equation; eGFRMDRD, eGFR calculated using the Modification of Diet in Renal Disease (MDRD) study equation.

Methods

Subjects

The subjects were participants of the Iwate-Kenpoku cohort (Iwate-KENCO) study. The Iwate KENCO Study was designed to determine the effects of traditional risk factors and new biomarkers on cardiovascular morbidity and mortality in the Japanese general population. The study area is a typical rural area of Japan (**Figure S1**) with a low move-out/move-in population and a high proportion of elderly people. The methodology of the Iwate-KENCO study has been described elsewhere.[21,22] The initial surveys were carried out from 2002 to 2004. The original cohort study members consisted of 26,469 participants. We excluded participants who lacked data for serum creatinine (n=143), participants with a history of MI or stroke (n=984), and participants who lacked data for at least 1 factor that was used for analysis as an explanatory variable (n=785). Finally, we analyzed data from 24,560 participants (**Figure 1**). The study was approved by the Medical Ethics Committee of Iwate Medical University and conducted in accordance with the guidelines of the Declaration of Helsinki.

Measurements

The initial examinations consisted of a questionnaire, measurements of blood pressure and anthropometric data, and blood tests. Serum creatinine level was measured using an enzymatic assay on an automated analyzer (HITACHI 7700). The methods for measuring serum lipid profile, plasma glucose level, and plasma glycosylated hemoglobin (HbA_{1c}) have been previously described.[21] In this analysis, HbA_{1c} level (National Glycohemoglobin Standardization Program [NGSP] equivalent value) was modified by adding 0.4% to the estimated value (the Japan Diabetes Society [JDS] value) according to the Guidelines of the JDS.[23] Urine albumin was assessed quantitatively using an immunonephelometric method (N-antiserum albumin, Dade Behring) and urine creatinine was measured quantitatively on enzymatic colorimetric test.[24] The urine albumin-creatinine ratio (UACR) was used because the accuracy of the ratio in comparison to 24-h urine sample has been demonstrated in previous studies.[25,26] The data-gathering methodology has been previously described.[21]

Classification and Definition

eGFR was calculated using both MDRD and CKD-EPI equations modified by a Japanese coefficient (eGFRCKDEPI and eGFRMDRD) as shown in **Table S1**.[10,11] Participants were divided into 4 categories (1, $\geq 90\,ml \cdot min^{-1} \cdot 1.73\,m^{-2}$; 2, 60–89 $ml \cdot min^{-1} \cdot 1.73\,m^{-2}$; 3a, 45–59 $ml \cdot min^{-1} \cdot 1.73\,m^{-2}$; 3b+, $<45\,ml \cdot min^{-1} \cdot 1.73\,m^{-2}$) according to both GFR based on eGFRCKDEPI and on eGFRMDRD. Albuminuria was defined as the presence of microalbuminuria or macroalbuminuria. Microalbuminuria was defined as a UACR of 30–299 mg/g and macroalbuminuria was defined as a UACR ≥300 mg/g.

Hypertension was defined as systolic blood pressure (SBP) ≥140 mmHg, diastolic blood pressure ≥90 mmHg, use of antihypertensive agents or a combination of these. Diabetes was defined as plasma glucose ≥200 mg/dl, plasma HbA_{1c} (NSGP equivalent value) ≥6.5%, use of anti-diabetes agents or a combination of these. Dyslipidemia was defined as serum total cholesterol (TC) ≥220 mg/dl, serum high-density-lipoprotein cholesterol (HDLC) <40 mg/dl, use of anti-hyperlipidemia agents or a combination of these. Regular alcohol drinking was defined as drinking ≥5 days/week. Exercise habit was defined as doing exercise for at least 60 min 8 days per month.

Outcome Measures

In this cohort study, the endpoints were all-cause death, incident MI and incident stroke. To ascertain subjects' vital status, follow-up surveys were performed in 2006 and 2009. The investigators visited each municipality and reviewed the Basic Resident Register sheets in each local government to confirm the dates of death and move-out of participants. Persons who were known to be alive at the end of follow-up and those who had moved away from the study area were treated as censored

Table 1. Subject Characteristics vs. CKD Stage

CKD stage (ml·min^{-1}·1.73 m^{-2})	eGFR ≥90	60≤eGFR<90	45≤eGFR<60	eGFR <45
CKD-EPI equation (no. subjects)	2,504	20,607	1,259	190
Age (years)	43.0±9.0	63.9±9.3	73.0±7.6	75.2±6.8
BMI (kg/m^2)	23.3±3.6	24.1±3.2	24.3±3.3	24.6±3.4
SBP (mmHg)	116±17.7	128±19.9	133±19.8	134±21.5
TC (mg/dl)	189±33.7	202±32.6	202±33.2	200±38.6
HDLC (mg/dl)	61.4±14.5	59.6±14.9	55.6±14.4	53.3±14.4
HbA_{1c} (mg/dl)	5.33±0.77	5.53±0.66	5.59±0.65	5.64±0.68
SCr (mg/dl)	0.6±0.1	0.7±0.1	1.0±0.1	1.4±0.3
UACR† (mg/g)	10.7 (6.6–19.1)	14.9 (8.5–29.3)	18.8 (9.5–47.2)	56.2 (20.0–317)
Comorbid conditions and habits (%)				
Microalbuminuria	332 (13.3)	4,668 (22.7)	374 (29.7)	77 (40.5)
Macroalbuminuria	21 (0.8)	351 (1.7)	66 (5.2)	49 (25.8)
Hypertension	352 (14.1)	8,653 (42.0)	752 (59.7)	123 (64.7)
Diabetes mellitus	76 (3.0)	1,070 (5.2)	87 (6.9)	19 (10.0)
Dyslipidemia	540 (21.6)	7,600 (36.9)	534 (42.4)	92 (48.4)
Current smoker	577 (23.0)	2,395 (11.6)	120 (9.5)	16 (8.4)
Past smoker	230 (9.2)	2,295 (11.1)	230 (18.3)	34 (17.9)
Regular drinker	579 (23.1)	3748 (18.2)	184 (14.6)	25 (13.2)
MDRD equation (no. subjects)	4,449	17,128	2,711	272
Age (years)	56.2±12.2	62.7±10.9	68.7±8.5	73.3±8.2
BMI (kg/m^2)	23.8±3.5	24.0±3.2	24.4±3.3	24.6±3.4
SBP (mmHg)	124±20.1	127±20.0	131±19.6	134±20.9
TC (mg/dl)	197±33.5	201±32.8	204±32.9	201±38.3
HDLC (mg/dl)	60.7±14.8	59.7±14.9	57.1±14.7	54.2±13.8
HbA_{1c} (mg/dl)	5.54±0.90	5.50±0.61	5.54±0.58	5.62±0.71
SCr (mg/dl)	0.5±0.1	0.7±0.1	0.9±0.1	1.3±0.3
UACR (mg/g)†	15.3 (8.9–30.8)	14.0 (8.1–27.4)	15.4 (8.2–35.5)	38.7 (14.8–180)
Comorbid conditions and habits (%)				
Microalbuminuria	1,076 (24.2)	3,594 (21.0)	677 (25.0)	104 (38.2)
Macroalbuminuria	66 (1.5)	263 (1.5)	103 (3.8)	55 (20.2)
Hypertension	1,452 (32.6)	6,805 (39.7)	1,451 (53.5)	172 (63.2)
Diabetes mellitus	273 (6.1)	815 (4.8)	137 (5.1)	27 (9.9)
Dyslipidemia	1,396 (31.4)	6,090 (35.6)	1,151 (42.5)	129 (47.4)
Current smoker	743 (16.7)	2,110 (12.3)	237 (8.7)	18 (6.6)
Past smoker	402 (9.0)	1,952 (11.4)	398 (14.7)	37 (13.6)
Regular drinker	944 (21.2)	3,175 (18.5)	387 (14.3)	30 (11.0)

Data given as mean±SD, n (%) or †median (interquartile range).
AMI, acute myocardial infarction; BMI, body mass index; CKD, chronic kidney disease; CKD-EPI, Chronic Kidney Disease Epidemiology Collaboration; eGFR, estimated glomerular filtration rate; HbA_{1c}, glycosylated hemoglobin; HDLC, high-density lipoprotein cholesterol; MDRD, Modification of Diet in Renal Disease; SBP, systolic blood pressure; SCr, serum creatinine; TC, total cholesterol; UACR, urinary albumin to creatinine ratio.

cases.

Stroke events were identified by assessing the Iwate Prefecture Stroke Registration program, which included the entire area where the subjects lived. Details of this registry have been described previously.[22,27] The medical records of all medical facilities within the survey area were verified every year to ensure complete capture of all data from 2006 to 2009 by the physicians and trained research nurses. Incidents of acute MI (AMI) were identified by accessing data from Northern Iwate Heart Disease Registry Consortium, which has been collecting data since 2002. The registration of AMI was based on the criteria of the MONICA study. Details of this registry have been described previously.[22,28] To verify the accuracy of the data, physicians and trained research nurses also checked the medical records of the referral hospitals.

Statistical Analysis

The relationship between $eGFR_{CKDEPI}$ and $eGFR_{MDRD}$ was illustrated using a scatter diagram. Pearson correlation coefficient (r) between $eGFR_{CKDEPI}$ and $eGFR_{MDRD}$ was calculated. Concordance between each corresponding CKD stage according to $eGFR_{CKDEPI}$ and $eGFR_{MDRD}$, was examined on a cross-table for the 4 CKD categories of $eGFR_{CKDEPI}$ and $eGFR_{MDRD}$. Baseline characteristics are listed according to CKD stage based on both $eGFR_{CKDEPI}$ and $eGFR_{MDRD}$ and according to endpoint category. Risk factor-related variables are expressed as mean±SD, proportions (expressed as percentages) or median (interquartile range). Proportion of each CKD stage was compared between GFR_{CKDEPI} and $eGFR_{MDRD}$ by chi-square test.

We defined follow-up time as the period from the initial survey to the first outcome or end of observation. Individuals

Table 2. Baseline Subject Characteristics vs. Endpoint

	All subjects	No events survivor	Died	Incident AMI	Incident stroke
Subjects (n)	24,560	22,246	851	78	605
Age (years)	62.3±11.4	61.3±11.3	71.3±9.0	70.3±7.6	69.9±8.1
Male	8,368 (34.1)	7,334 (33.0)	320 (37.6)	55 (70.5)	96 (36.0)
BMI (kg/m^2)	24.0±3.3	24.0±3.3	23.5±3.5	24.3±3.2	24.3±3.5
SBP (mmHg)	127±20.1	126±20.0	133±20.5	134±19.8	138±20.6
TC (mg/dl)	201±33.0	201±33.0	192±33.9	206±32.6	198±32.7
HDLC (mg/dl)	59.5±14.9	59.7±14.8	56.4±15.4	51.3±14.2	57.5±14.9
HbA$_{1c}$ (%)	5.52±0.67	5.50±0.65	5.63±0.91	5.77±0.72	5.67±0.83
SCr (mg/dl)	0.7±0.2	0.7±0.2	0.8±0.2	0.8±0.2	0.7±0.2
eGFR$_{CKDEPI}$ (ml·min^{-1}·1.73m^{-2})	77.6±10.6	78.4±10.3	71.0±12.0	69.3±10.8	72.2±10.3
eGFR$_{MDRD}$ (ml·min^{-1}·1.73m^{-2})	75.9±15.4	76.5±15.2	71.8±16.8	68.3±14.1	72.1±15.1
UACR (mg/g)†	14.5 (8.2–29.1)	14.0 (8.0–27.4)	19.5 (10.4–49.6)	20.4 (9.3–51.9)	22.8 (12.6–51.8)
Comorbid conditions and habits					
Microalbuminuria	5,453 (22.6)	4,664 (21.3)	273 (34.0)	24 (32.4)	218 (37.8)
Macroalbuminuria	487 (2.0)	373 (1.7)	47 (5.5)	4 (5.1)	29 (4.8)
Hypertension	9,881 (40.2)	8,529 (38.3)	447 (52.5)	51 (65.4)	412 (68.1)
Diabetes mellitus	1,252 (5.1)	1,028 (4.6)	88 (10.3)	8 (10.3)	60 (9.9)
Dyslipidemia	8,770 (35.7)	7,945 (35.7)	274 (32.2)	35 (44.9)	215 (35.5)
Current smoker	3,110 (12.7)	2,761 (12.4)	184 (21.6)	19 (24.4)	108 (17.9)
Past smoker	2,789 (11.4)	2,451 (11.0)	176 (20.7)	16 (20.5)	97 (16.0)
Regular drinker	4,537 (18.5)	4,085 (18.4)	217 (25.5)	15 (19.2)	155 (25.6)
CKD stage based on eGFR$_{CKDEPI}$ (ml·min^{-1}·1.73m^{-2})					
GFR ≥90	2,508 (10.2)	2,470 (11.1)	26 (3.1)	1 (1.3)	11 (1.8)
60≤GFR<90	20,612 (83.9)	18,716 (84.1)	695 (81.7)	62 (79.5)	521 (86.1)
45≤GFR<60	1,259 (5.1)	951 (4.3)	96 (11.3)	14 (17.9)	61 (10.1)
GFR <45	190 (0.8)	118 (0.5)	34 (4.0)	1 (1.3)	12 (2.0)
CKD stage based on eGFR$_{MDRD}$ (ml·min^{-1}·1.73m^{-2})					
GFR ≥90	4,453 (18.1)	4,209 (18.9)	112 (13.2)	5 (6.4)	69 (11.4)
60≤GFR<90	17,133 (69.7)	15,582 (70.0)	568 (66.7)	48 (61.5)	428 (70.7)
45≤GFR<60	2,711 (11.0)	2,280 (10.2)	132 (15.5)	24 (30.8)	94 (15.5)
GFR <45	272 (1.1)	184 (0.8)	39 (4.6)	1 (1.3)	14 (2.3)

Data given as mean±SD, n (%) or †median (interquartile range).
Abbreviations as in Table 1.

who were free of outcomes by 5-year follow-up were subjected to censoring. Receiver operating characteristic (ROC) curves were drawn and the areas under the curves (AUROC) were calculated for each equation to compare the discrimination abilities of the 2 models. Crude, sex- and age-adjusted, and multivariate-adjusted mortality and incidence rates of AMI and stroke (/1,000 person-years) were determined in the 4 groups according to eGFR (CKD stages 1, 2, 3a and 3b+) on Poisson regression analysis. Multivariate-adjusted mortality and incidence rates were estimated after adjusting for age, sex, SBP, body mass index, TC, HDLC, HbA$_{1c}$, existence of albuminuria, smoking habit, regular drinking habit and exercise habit.

Relative risks for all-cause death, incident AMI, and incident stroke were estimated in each category and compared with the reference group (60≤eGFR<90 ml·min^{-1}·1.73 m^{-2}) in a Cox regression model for the same explanatory variables as those used in Poisson regression analysis separately for the 2 models of eGFR$_{CKDEPI}$ and eGFR$_{MDRD}$ CKD stage. The performances of the multivariate models were quantified using Harrell's concordance statistics (Harrell's C).[14] The Akaike information criterion (AIC)[29] and Bayesian information criterion (BIC)[30] were also estimated for these models. We also quantified the degree of correct reclassification by estimating net reclassification improvement (NRI) using cross-categories of eGFR for both equations.[20,31]

All P-values were 2-tailed, and P<0.05 was considered to be statistically significant. Statistical analysis was performed using PASW version 18.0 (IBM Japan, Tokyo, Japan) and STATA version STATA/SE 11 (STATA, College Station, TX, USA).

Results

Figure 2 shows a scatter graph of eGFR$_{CKDEPI}$ and eGFR$_{MDRD}$. The overall correlation between eGFR$_{CKDEPI}$ and eGFR$_{MDRD}$ was relatively good (r=0.863, P<0.001.). Using eGFR$_{CKDEPI}$ as the gold standard, eGFR$_{MDRD}$ mildly to moderately underestimated GFR in persons with eGFR$_{CKDEPI}$ 45–90 ml·min^{-1}·1.73 m^{-2}. Because most of the present participants belonged to this category, eGFR$_{MDRD}$ was likely to underestimate GFR in the total subjects. In contrast, eGFR$_{MDRD}$ moderately to greatly overestimated GFR in persons with eGFR$_{CKDEPI}$ 90–120 ml·min^{-1}·1.73 m^{-2}, especially in female participants. **Table S2** is a cross-table of the 4 CKD categories of eGFR$_{CKDEPI}$ and eGFR$_{MDRD}$. Underestimation of eGFR$_{MDRD}$ was observed in 30%