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ORIGINAL ARTICLE

The *GCKR* rs780094 polymorphism is associated with susceptibility of type 2 diabetes, reduced fasting plasma glucose levels, increased triglycerides levels and lower HOMA-IR in Japanese population

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It was recently reported that *GCKR* rs780094 was associated with fasting plasma glucose (FPG) and triglyceride (TG) levels in various ethnic populations (A allele for low FPG and high TG). An association between *GCKR* rs780094 and type 2 diabetes mellitus (T2DM) (A allele for low risk) has also been reported. We examined the association between *GCKR* rs780094 and T2DM in Japanese subjects by analyzing 488 cases and 398 controls. A meta-analysis was performed involving two previous association studies. We also analyzed the association between the single-nucleotide polymorphism and clinical parameters in the general Japanese population (n=1854). In the case–control study, the A allele of *GCKR* rs780094 was associated with a reduced risk of T2DM (odds ratio=0.711 (95% confidence interval=0.589–0.859), $P=4.2\times10^{-4}$). A meta-analysis confirmed the association of *GCKR* rs780094 with T2DM susceptibility. In the general Japanese population, subjects with the A/A genotype had lower levels of FPG, fasting plasma insulin and homeostasis model assessment of insulin resistance than those with the G/G genotype. Conversely, subjects with the A/A genotype had higher levels of TG than those with the G/G genotype. We replicated *GCKR* rs780094 as a marker of T2DM susceptibility in Japanese subjects. This suggests that *GCKR* rs780094 is a common variant for T2DM susceptibility in various ethnic groups.
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Keywords: case–control study; *GCKR*; meta-analysis; single-nucleotide polymorphism; type 2 diabetes

INTRODUCTION

Glucokinase (GCK), a glucose sensor, has a pivotal role in maintaining blood glucose homeostasis in the liver and in pancreatic β-cells. GCK activity is regulated by the glucokinase regulatory protein (GCKR), which binds to GCK in the nucleus and inhibits its activity in the presence of fructose-6-phosphate.[1,2] In the presence of fructose-1-phosphate, GCKR permits GCK to exert its activity in the cytoplasm. *GCKR*-deficient mice have reduced *GCK* expression and show impaired glycemic control,[3] suggesting that the GCK pool in the nucleus maintained by GCKR is critical for GCK function in glucose metabolism. In contrast, the adenoviral-mediated hepatic overexpression of *GCKR* in mice was reported to improve fasting plasma glucose (FPG) levels and insulin sensitivity, and increased plasma triglyceride (TG) levels.[4]

A single-nucleotide polymorphism (SNP) rs780094 is located in an intron of the glucokinase regulatory protein gene (*GCKR*). Associations between this A allele and either higher fasting serum TG levels, lower FPG levels, less insulin resistance or a lower risk of type 2 diabetes mellitus (T2DM) were identified in genome-wide association studies and have been confirmed in the European population.[5,6] A meta-analysis confirmed the comprehensive association of this SNP with the opposite effects on fasting TG levels and FPG by analyzing Caucasians, African Americans, Hispanics, Chinese, Malays and Asian Indians and the association of this SNP with a decreased risk of T2DM in European populations.[7] Recently, similar findings were also replicated in the case of Han Chinese.[8] In a Japanese study, a significant association of the A allele of *GCKR* rs780094 with lower

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plasma TG levels and a marginal association with T2DM were reported.[9]

Rare mutations in the human glucokinase (*GCK*) gene have been reported to be a cause of maturity-onset diabetes of the young.[10] *GCK* rs179988, which is located in the promoter region of *GCK*, was reported to be associated with FPG[11] and T2DM. An interaction effect of *GCKR* rs780094 and *GCK* rs179988 on metabolic traits has also been reported.[5,12]

In this study, we examined the association between either *GCKR* rs780094 or *GCK* rs179988 and the risk of T2DM in Japanese subjects, and carried out a meta-analysis by combining the collected data with those from two previous studies. We also analyzed the association between these SNPs and some clinical parameters in the general Japanese population.

MATERIALS AND METHODS

Case–control subjects

The clinical characteristics of subjects are summarized in Table 1. All T2DM subjects were recruited from the Ehime University Hospital and the Ehime Prefectural Hospital in Japan. Diabetes mellitus was diagnosed according to the 1998 ADA criteria.[13] Nondiabetic control subjects were selected based on the absence of a personal and familial history of diabetes in their first-degree relatives, as well as either normal glucose tolerance based on a 75 g oral glucose tolerance test, or HbA1c levels under 5.6% with FPG levels under 110 mg per 100 ml. Detail of the selection criteria have been reported in previous studies.[14,15]

The sample size for the present association study allowed us to detect a susceptibility polymorphism, which had a high minor allele frequency (MAF) and a high relative risk with an appropriate statistical power. We assume that S and N are susceptibility and normal alleles, respectively. When the genotype relative risk is assumed to be 1.50 for SN, and 2.25 for SS, the population frequency of S is 45.9% as the G allele of *GCKR* rs780094, and the prevalence of diabetes is 6.9% based on IDF e-Atlas (http://www.eatlas.idf.org/About_e_Atlas/), the penetrance for genotypes of SS, SN and NN could be calculated as 0.117, 0.078 and 0.052, respectively. Under this condition, a significant difference in the allele frequency between 488 cases and 398 controls can be detected with a power $>99\%$. For a significant association to be detected with a power $>80\%$ in this study, the genotype relative risk of a susceptible allele should be >1.29 for *GCKR* rs780094 with MAF=0.459.[16,17] It should be noted that these relative risks are higher than those currently reported by genome-wide association studies on T2DM, namely ~ 1.2. When the relative risk is 1.2, the power for MAF=0.459 is 53.5%.

General population

General population subjects are community-dwelling subjects in the Ehime prefecture, a rural area located in western Japan with a population of $\sim 11\,000$ inhabitants.[14] The subjects were recruited during a community-based annual medical checkup processes. The sample population was composed of 2895 middle-aged to elderly residents. In this general population, 1854 subjects who were undergoing no hyperglycemic or hyperlipidemic treatment were enrolled in the analysis. The baseline clinical characteristics of subjects were obtained from personal health records evaluated during their medical checkup. Additional characteristics, such as medication and a history of cardiovascular disease, were obtained by interviews using a questionnaire. A homeostasis model assessment of insulin resistance (HOMA-IR; (fasting immunoreactive insulin ($\mu U\,ml^{-1}$)$\times$fasting glucose (mg per 100 ml))/405) was used as an index of insulin resistance. A homeostasis model assessment for β-cells (HOMA-β; (360$\times$fasting immunoreactive insulin ($\mu U\,ml^{-1}$)/(fasting glucose (mg per 100 ml)–63)) was used as an index of insulin secretion. Plasma low-density lipoprotein (LDL) cholesterol values were estimated using the following formula: total cholesterol–HDL cholesterol–triglyceride/5, where HDL cholesterol stands for high-density lipoprotein cholesterol. With regard to CRP levels, 1766 out of 1854 subjects were measured because of sample availability and, with regard to HOMA-β, 1849 out of 1854 subjects were used for the calculation based on inappropriate levels of fasting blood glucose.

Ethical issue

This study was approved by the ethics committee of the Ehime University Graduate School of Medicine, and informed consent was obtained from all subjects.

Genotyping

Genomic DNA was extracted from peripheral blood, using a QIAamp DNA blood kit (Qiagen GmbH, Hilden, Germany). The *GCKR* rs780094 and *GCK* rs1799884 were analyzed by a TaqMan probe assay (Applied Biosystems, Foster City, CA, USA) using commercially available primers and probes purchased from the Assay-on-Demand system (C_2862873_10 and C_8304645_10, respectively). An ABI PRISM 7900HT sequence detector (Applied Biosystems) was used to measure the fluorescence levels of PCR products.

The genotype call rate was 98.6 and 97.2% for *GCKR* rs780094 and *GCK* rs179988, respectively, in the case–control study, and 99.1 and 99.5% for *GCKR* rs780094 and *GCK* rs179988, respectively, in the general population study. The *P*-value of deviation from Hardy–Weinberg equilibrium was 0.527, 0.498 and 0.261 in cases, controls and the general populations, respectively, for *GCKR* rs780094, and 0.207, 0.572 and 0.704 in cases, controls and the general populations, respectively, for *GCK* rs179988.

Statistical analysis

Values are expressed as the mean ± s.d. Differences in allele frequency between T2DM and control subjects were assessed by χ^2-test. The pooled odds ratio (OR) for allele frequency was estimated by the fixed effects model (Mantel–Haenszel method). Mean values among genotypes were assessed by an analysis of variance. The age-, gender- and body mass index-adjusted *P*-values of each genotype were analyzed from multiple regression equations, which include the *GCKR* genotype (G/G=0, G/A=1, A/A=2) in an additive model, or the *GCKR* genotype (G/A or A/A vs GG) as two dummy variables (G/G (0, 0), G/A (1, 0), A/A (0, 1)). To assess isolated effects of the *GCKR* rs780094 genotype on clinical parameters, multiple regression analyses involving FPG, insulin and TG, each

Table 1 Clinical characteristics of case–control subjects

	Type 2 diabetes patients n=*506*	*Control subjects* n=*402*	P-*value*
Sex (male/female)	280/226	214/188	0.528
Age (years)	60 ± 11	59 ± 9	0.038
Age at onset (years)	49 ± 12	—	—
Duration of diabetes (years)	11 ± 9	—	—
Body mass index ($kg\,m^{-2}$)	24 ± 4	23 ± 3	0.032
Fasting blood glucose (mg per 100 ml)	166 ± 58	92 ± 8	<0.001
HbA1C (%)	8.0 ± 1.9	4.9 ± 0.3	<0.001

Values are mean ± s.d.

as a dependent variable, and the *GCKR* rs780094 genotype, age, gender (male=1, female=2), body mass index, FPG and TG as independent variables were performed. When the effect on FPG, insulin or TG was analyzed, the corresponding factor was not included as an independent variable. In these analyses, the genotypes for *GCKR* rs780094, A/A, G/A and G/G, were denoted by two dummy variables (c1, c2)=(0, 0), (1, 0) and (0, 1), respectively. Statistical analyses were performed using a commercially available statistical software package (JMP 7.0, SAS, Chary, NC, USA). Null hypotheses were rejected at a level of significance of $P<0.05$.

RESULTS

The A allele of *GCKR* rs780094 was associated with the reduced risk of T2DM

We first examined the association between *GCKR* rs780094 and T2DM in 488 cases and 398 controls who have been successfully genotyped (Table 1 and 2). The overall frequency of the A allele was 54.1%, which is higher than that in the European population and similar to that in the Han Chinese and Japanese population reported previously.[8,9,18] The A allele of *GCKR* rs780094 was associated with a reduced risk of T2DM (OR=0.711 (95% confidence interval=0.589–0.859), $P=4.2\times10^{-4}$) as previously reported in a Danish study.[5] To further clarify T2DM susceptibility, a meta-analysis combining previous reports, from which we were able to use genotype frequency data, was performed. In a meta-analysis combining a previous Japanese study (Table 2),[9] the A allele of *GCKR* rs780094 was found to be associated with a reduced risk of T2DM (OR=0.858 (95% confidence interval=0.788–0.934), $P=4.0\times10^{-4}$). Furthermore, a meta-analysis combining the Japanese and Danish studies also showed this association (OR=0.898 (95% confidence interval=0.854–0.945), $P=3.2\times10^{-5}$).[5,9] No significant association between *GCK* rs179988 and T2DM was found (Table 2).

The A/A genotype of *GCKR* rs780094 is associated with higher fasting plasma TG, lower FPG, lower fasting plasma insulin and lower HOMA-IR.

We next examined the association between these SNPs and some clinical parameters in the general Japanese population ($n=1854$) (Table 3). A multiple regression analysis adjusted for age, gender and body mass index revealed that subjects with the A/A genotype of *GCKR* rs780094 had higher levels of TG ($P=0.028$), total cholesterol ($P=0.011$) and LDL cholesterol ($P=0.04$) compared with those with the G/G genotype. In contrast, subjects with the A/A genotype of *GCKR* rs780094 had lower levels of FPG ($P=0.008$), fasting insulin ($P=0.008$) and HOMA-IR ($P=0.002$) compared with those with the G/G genotype (Table 3). In an additive model adjusted for age, gender and body mass index, the A allele was also found to be associated with lower FPG ($P=0.004$), lower insulin ($P=0.0127$), lower HOMA-IR ($P=0.004$), higher TG ($P=0.0176$), higher total cholesterol ($P=0.002$) and higher LDL cholesterol ($P=0.016$) (Table 3). Although it may be too conservative to perform multiple testing, we performed the adjustment of multiple testing in two ways, Bonferonni's correction and Holm's correction. The associations with FPG and HOMA-IR remained significant in both Bonferonni's ($P=0.043$ and 0.042, respectively (raw P-value$\times11$)) and Holm's corrections ($P=0.035$ (raw P-value$\times9$) and 0.037 (raw P-value $\times10$), respectively) and, those with insulin and TG are marginally significant in Holm's correction ($P=0.09$ (raw P-value$\times8$) and 0.10 (raw P-value $\times6$), respectively) in an additive model. No association was found between *GCKR* rs780094 and HOMA-β. Compared with the G/G genotype, the associations of the A/A genotype of *GCKR* rs780094 with lower FPG ($\beta=-0.067$, $P=0.026$), lower fasting insulin ($\beta=-0.075$, $P=0.003$) and higher fasting TG ($\beta=0.099$, $P=0.001$) levels remained significant, after adjustment for age, gender, and also FPG, insulin and TG when not included in a dependent variable, in a multiple regression analysis (Table 4). No association was observed between *GCK* rs179988 and metabolic traits; namely, FPG, IRI, HOMA-IR, HOMA-β and TG (Supplementary Table 1).

DISCUSSION

In this study, we confirmed that the A allele of *GCKR* rs780094 is associated with a reduced susceptibility to T2DM in Japanese subjects. A meta-analysis that included combining two previous reports further reinforced and confirmed this association. Horikawa *et al.*[9] reported that the association between this SNP and T2DM was marginal, based on an analysis of data for 1921 subjects with T2DM and 1622 controls. The meta-analysis using combining data in the present report revealed a significant association of *GCKR* rs780094 with T2DM in Japanese subjects. The association was further confirmed by combining two previous reports in which data for Danes and Japanese were analyzed. It was recently reported that the A allele of *GCKR* rs780094 is associated with a reduced risk of T2DM in Han

Table 2 Association of GCKR and GCK polymorphism with type 2 diabetes

	Genotype (n)						*Allele frequency*				*Allelic model*	
	T2DM			*Control*			*T2DM*		*Control*		*OR (95% CI)*	P-*value*
GCKR rs780094	*GG*	*GA*	*AA*	*GG*	*GA*	*AA*	*G*	*A*	*G*	*A*		
Current study	124	237	127	64	198	133	0.497	0.503	0.413	0.587	0.711 (0.589–0.859)	4.2×10^{-4}
Horikawa *et al.*[9]	421	903	534	312	782	492	0.470	0.530	0.443	0.557	0.899 (0.818–0.989)	0.029
Sparso *et al.*[5]	1755	1681	442	2066	2234	591	0.669	0.331	0.651	0.349	0.921 (0.865–0.981)	0.010
Pooled OR												
Japanese	—	—	—	—	—	—	—	—	—	—	0.858 (0.788–0.934)	4.0×10^{-4}
All study	—	—	—	—	—	—	—	—	—	—	0.898 (0.854–0.945)	3.2×10^{-5}
GCK rs1799884	TT	TC	CC	TT	TC	CC	T	C	T	C	—	—
Current study	19	134	341	9	112	280	0.174	0.826	0.162	0.838	1.090 (0.849–1.399)	0.500

Abbreviations: CI, confidence interval; GCK, glucokinase; GCKR, glucokinase regulatory protein; OR, odds ratio; T2DM, type 2 diabetes mellitus.
Differences in allele frequency between T2DM and control subjects were assessed by the χ^2-test. Type 2 diabetes is defined by fasting blood glucose levels $\geqslant$126 mg per 100 ml, or an occasional blood glucose level $\geqslant$200 mg per 100 ml and/or the current use of oral antidiabetic agents. Nondiabetic control subjects in the case–control sample were selected based on an absence of personal and familial history of diabetes among their first-degree relatives, as well as either normal glucose tolerance, confirmed by a 75 g oral glucose tolerance test, or HbA1c levels $<$5.6 with fasting plasma glucose levels $<$110 mg per 100 ml.

Table 3 Association between GCKR rs780094 polymorphism and metabolic parameters in general population

		Mean ± s.d.			P-*value*			
	Total	*GCKR rs780094 genotype*			*ANOVA*	*Regression analysis*		
							Genotype (vs GG)	
		GG	*GA*	*AA*		*Additive*		
Number of subjects	*1854*	*337*	*879*	*638*			*GA*	*AA*
Sex (male/female)	825/1029	155/182	405/474	265/373	—	—	—	—
Age (years)	62 ± 13	60 ± 13	62 ± 13	62 ± 13	—	—	—	—
BMI (kg m^{-2})	23 ± 3	24 ± 3	23 ± 3	23 ± 3	—	—	—	—
Glucose (mg per 100 ml)	97 ± 18	98 ± 20	97 ± 20	95 ± 14	0.006	0.004	0.361	0.008
Insulin (μU ml^{-1})	6.5 ± 4.7	7.2 ± 5.7	6.4 ± 4.2	6.2 ± 4.7	0.007	0.013	0.029	0.008
HOMA-IR	1.6 ± 1.4	1.8 ± 1.7	1.6 ± 1.2	1.5 ± 1.3	0.003	0.004	0.020	0.002
HOMA-β	74 ± 49	78 ± 52	74 ± 47	73 ± 50	0.261	0.838	0.132	0.133
Triglyceride (mg per 100 ml)	111 ± 70	107 ± 69	109 ± 63	114 ± 78	0.214	0.017	0.442	0.028
Total cholesterol (mg per 100 ml)	203 ± 34	201 ± 34	200 ± 35	207 ± 33	2.0×10^{-4}	0.002	0.708	0.011
LDL cholesterol (mg per 100 ml)	119 ± 32	117 ± 31	117 ± 32	122 ± 32	0.010	0.016	0.944	0.040
HDL cholesterol (mg per 100 ml)	62 ± 16	63 ± 16	61 ± 15	63 ± 16	0.163	0.879	0.094	0.578
hsCRP (mg per 100 ml)	0.10 ± 0.17	0.11 ± 0.20	0.10 ± 0.17	0.09 ± 0.14	0.278	0.140	0.214	0.119
HMW adiponectin (μg ml^{-1})	6.1 ± 4.3	5.8 ± 4.1	6.1 ± 4.5	6.2 ± 4.2	0.389	0.662	0.743	0.775
Resistin (mg per 100 ml)	12 ± 7	11 ± 8	11 ± 7	12 ± 7	0.913	0.860	0.998	0.881

Abbreviations: ANOVA, analysis of variance; BMI, body mass index; GCKR, glucokinase regulatory protein; HDL, high-density lipoprotein; HOMA-β, homeostasis model assessment for β-cells; HOMA-IR, homeostasis model assessment of insulin resistance; LDL, low-density lipoprotein.
Values are expressed as mean ± s.d. Mean values among genotypes were assessed by ANOVA. The age-, gender- and BMI-adjusted *P*-values of each genotype were analyzed by multiple regression analysis, which include the *GCKR* genotype (G/G=0, G/A=1, A/A=2) in additive models, or the *GCKR* genotype(G/A or AA vs GG) as two dummy variables (G/G (0, 0), G/A (1, 0), A/A (0, 1)). None of the subjects were taking antihyperlipidemic and antihyperglycemic agents. HOMA-IR was calculated as fasting blood glucose×insulin/405. HOMA-β was calculated as (360×fasting immunoreactive insulin (μU ml^{-1})/(fasting blood glucose (mg per 100 ml)−63)). LDL cholesterol was calculated as total cholesterol–HDL cholesterol–triglyceride/5.

Table 4 Multiple regression analysis for blood glucose, insulin and triglyceride

	Glucose		*Insulin*		*Triglyceride*	
	β	P-*value*	β	P-*value*	β	P-*value*
Age (years)	0.133	<0.001	−0.081	<0.001	0.034	0.114
Sex (male)	0.193	<0.001	−0.266	<0.001	0.192	<0.001
BMI (kg m^{-2})	0.062	0.016	0.418	<0.001	0.091	<0.001
Glucose (mg per 100 ml)			0.170	<0.001	0.031	0.176
Triglyceride (log-normalized)	0.032	0.176	0.229	<0.001	—	—
Insulin (log-normalized)	0.244	<0.001	—	—	0.315	<0.001
GCKR rs780094 genotype						
GG	Reference		Reference		Reference	
GA	−0.021	0.485	−0.039	0.124	0.051	0.086
AA	−0.067	0.026	−0.075	0.003	0.099	0.001

Abbreviations: BMI, body mass index; *β*, unstandardized regression coefficient.
Multiple regression analysis involving either glucose, insulin or triglyceride as a dependent variable was performed as described in Materials and methods. When each of glucose, TG and insulin was involved as dependent variables, each of these was not involved as independent variables. Otherwise, age (years), gender (male=1, female=2), BMI, glucose, triglyceride, insulin or *GCKR* rs780094 genotype (G/A or A/A vs G/G) were involved as independent variables.

Chinese.[8] These findings suggest that *GCKR* rs780094 is one of the common variants that lead to susceptiblity to T2DM in various ethnic populations.

In this study, the allele frequency of the *GCKR* rs780094 A allele in Japanese was 54%, which is higher than that for Europeans (35%),[6] but similar to that for Han Chinese (55%)[8] and in previous reports for Japanese.[9] On the basis of the findings reported herein, it can be concluded that analyzing a large number of subjects can result in the identification of susceptible loci to T2DM and that this can be confirmed for various ethnic groups, even though the minor allele frequency of the SNP is different among them.

We also found that *GCKR* rs780094 had an opposite effect on fasting TG levels and FPG levels by analyzing a group of general Japanese general subjects comprised of 1854 individuals. The A/A genotype of *GCKR* rs780094 was associated with increased TG and reduced FPG. These findings are in agreement with reports dealing with the European and Han Chinese populations.[6,8,18] Most recently, the A allele of *GCKR* rs780094 has been reported to be associated with reduced FPG and increased TG in the Japanese population as well as in the Sri Lankan population.[19] This study confirmed the effect of *GCKR* rs780094 on FPG and TG in the Japanese population. A possible mechanism for the opposite effect on plasma glucose and TG has been proposed; namely, increased glucose utilization and glycolytic flux in the liver could cause the downregulation of PEPCK and glucose-6-phosphatase, and the upregulation of GCK and fatty acid synthase, changes in which could result in increased TG production and a decreased hepatic glucose output. Although the associations between the A allele of *GCKR* rs780094 and higher levels of fasting TG were replicated in earlier studies, the associations between this SNP and either lower levels of FPG, lower levels of fasting insulin or reduced HOMA-IR were not consistently detected. It is also possible that *GCKR* rs780094 may exert a direct effect on TG levels, which does not involve glucose metabolism resulting in an upregulation of lipogenesis in the liver. In fact, no association between *GCK* polymorphism and TG levels have been reported.[12]

Subjects who carried the A/A genotype of *GCKR* rs780094 were more insulin sensitive, as evidenced by HOMA-IR, and this appears to contribute to the protective effect of the A allele of *GCKR* rs780094 on T2DM. This finding is consistent with some previous reports for

Europeans,[5,18] but not with a recent report with regard to Han Chinese.[8] The report in which the Han Chinese were analyzed found an association of the A allele of *GCKR* rs780094 with HOMA-β but not with HOMA-IR. The report concluded that the protective effect of the A allele of *GCKR* rs780094 against T2DM appeared to be mediated through improved insulin secretion from pancreatic β-cells. This difference in the effect of *GCKR* rs780094 on insulin secretion and insulin resistance between Japanese and Han Chinese suggests that GCKR may function differently in the liver and pancreatic β-cells by interacting with genetic and environmental factors.

We also detected an association of the A/A genotype of *GCKR* rs780094 with higher LDL cholesterol and total cholesterol levels as reported by Sparso *et al*.[5] We failed to observe an association between this SNP and hsCRP, as previously reported.[18] Increased TG levels and elevated LDL cholesterol levels are thought to be risk factors for atherosclerosis. In this study, the A/A genotype of *GCKR* rs780094 was associated with both higher TG and higher LDL cholesterol levels, but not with hsCRP, which is a marker of atherosclerosis and inflammation. Lower glucose levels and lower insulin resistance (lower HOMA-IR), which are associated with the A/A genotype of *GCKR* rs780094, may contribute to protection against atherosclerosis.

Rare mutations of *GCK* are known to be associated with maturity-onset diabetes of the young, and a common variant, *GCK* rs1799884 (*GCK*-30A), has been shown to be associated with an increased risk of T2DM, hyperglycemia or impaired insulin secretion in Europeans and Han Chinese.[8,11,20,21] In this study, an association of *GCK* rs1799884 with either the risk of T2DM, glucose levels or insulin release was not detected, as assessed by HOMA-β. The lack of this association in Japanese may be due to genetic factors rather than *GCK* rs1799884, which have stronger effects on insulin secretion in the Japanese. SNPs in *KCNQ1*, a gene expressed in pancreatic islets, have recently been reported to be associated with T2DM in Japanese.[22]

In summary, we replicated a significant association of the A allele of *GCKR* rs780094 with a reduced risk of T2DM, as well as its opposite effect on increased fasting TG levels and decreased FPG in Japanese subjects. These findings suggest that *GCKR* rs780094 is a common variant for T2DM susceptibility in various ethnic groups. The mechanisms underlying the association between this SNP and T2DM, FPG or TG are currently unclear and further studies will be required to clarify these points.

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