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Serum γ-glutamyltransferase and development of impaired fasting glucose or type 2 diabetes in middle-aged Japanese men

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Abstract. Nakanishi N, Nishina K, Li W, Sato M, Suzuki K, Tatara K (Osaka University Graduate School of Medicine, Osaka; and Japan Labor and Welfare Association, Tokyo; Japan). Serum γ-glutamyltransferase and development of impaired fasting glucose or type 2 diabetes in middle-aged Japanese men. *J Intern Med* 2003; **254:** 287–295.

Objective. To investigate the association between serum γ-glutamyltransferase (GGT) and risk for development of diabetes.
Design. Longitudinal study (followed from 1994 to 2001).
Setting. A work site in Japan.
Subjects. A total of 2918 Japanese male office workers aged 35–59 years who did not have impaired fasting glucose (IFG) (a fasting plasma glucose concentration of 6.1–6.9 mmol L^{-1}), type 2 diabetes (a fasting plasma glucose concentration of $\geqslant$7.0 mmol L^{-1} or receipt of hypoglycaemic medication), medication for hypertension or hepatitis, alanine aminotransferase concentrations higher than three times the upper limit of the reference range or a history of cardiovascular disease at study entry.
Main outcome measure. Incidence of IFG or type 2 diabetes over a 7-year period.
Results. With adjustment for potential risk factors for diabetes, the relative risk for IFG compared with serum GGT <16 U L^{-1} was 1.23 (95% CI, 0.79–1.90), 1.50 (CI, 0.97–2.32) and 1.70 (CI, 1.07–2.71) with serum GGT of 16–24, 25–43 and $\geqslant$44 U L^{-1}, respectively (P for trend = 0.014). The respective relative risks for type 2 diabetes compared with serum GGT <16 U L^{-1} were 2.54 (CI, 1.29–5.01), 2.64 (CI, 1.33–5.23) and 3.44 (CI, 1.69–6.70) (P for trend = 0.002). From stratified analyses by body mass index (BMI) and alcohol intake, a stronger linear association between serum GGT and development of IFG or type 2 diabetes was found in men with a BMI $\geqslant$23.2 kg m^{-2} in both those who drank <46 and $\geqslant$46 g day^{-1} of ethanol.
Conclusions. The risk for development of IFG or type 2 diabetes increased in a dose-dependent manner as serum GGT increased in middle-aged Japanese men. The increased relative risk for IFG or type 2 diabetes associated with serum GGT was more pronounced in obese men.

Keywords: impaired fasting glucose, incidence, serum Japanese men, serum γ-glutamyltransferase, type 2 diabetes.

Introduction

Serum γ-glutamyltransferase (GGT) has been widely used as an index of liver dysfunction and a biological marker of alcohol intake [1–3]. In addition to its diagnostic uses, serum GGT has substantial epidemiological significance. Prospective studies have shown a significant relationship between elevated serum GGT and subsequent mortality and morbidity [2, 4–8] and between serum GGT and development of specific conditions, including myocardial infarction and stroke [5, 8–10]. Apart from alcohol, major effects of obesity on serum GGT have been found, and there is increasing evidence linking raised serum GGT concentrations with other cardiovascular risk factors, including hypertension, dyslipidaemia and

physical inactivity [5, 11–13]. Excess deposition of fat in the liver, usually termed nonalcoholic fatty liver disease, has strong associations with elevated serum GGT, obesity, insulin resistance and hyperinsulinaemia [5, 14–17]. These interrelations between serum GGT, obesity, other cardiovascular risk factors and plasma insulin raise the possibility that elevated GGT concentrations predict the development of type 2 diabetes.

As for the association between serum GGT and risk for diabetes, Perry *et al.* [18] have recently demonstrated that a raised serum GGT is an independent risk factor for the development of doctor-diagnosed type 2 diabetes. To further address this issue, using data from serial annual health examinations at the workplace, we carried out a longitudinal population study to examine prospectively the association of serum GGT with development of impaired fasting glucose (IFG) or type 2 diabetes (as diagnosed with the new revised criteria of American Diabetes Association in 1997 [19] for epidemiological studies) in normoglycaemic middle-aged Japanese men. Possible associations between other liver enzymes [aspartate aminotransferase (AST), alanine aminotransferase (ALT), and alkaline phosphatase] and development of IFG or type 2 diabetes have also been examined.

Methods

Study cohort

Our study is an ongoing cohort investigation, designed to clarify risk factors for major diseases, including hypertension, dyslipidaemia, and diabetes amongst Japanese men who were office workers at one of the biggest building contractors in Japan. The Industrial Safety and Health Law in Japan requires the employer to conduct annual health examinations of all employees, and employees are required by law to participate. A signed self-administered questionnaire is part of this examination, and the employee data, which are anonymous, are available for research with the approval of the employer. To evaluate the association of serum GGT with development of IFG or type 2 diabetes, a survey of the incidence of IFG or type 2 diabetes was carried out between 1994 and 2001. All Japanese male office workers aged 35–59 years in May 1994 were invited to take part in a survey ($n = 3694$); the participation rate was 99.6% ($n = 3681$).

Of 3681 potential participants, 700 (19.0%) were excluded: 175 (4.8%) had IFG, 282 (7.7%) had type 2 diabetes, 253 (6.9%) were taking antihypertensive medication, 34 (0.9%) were under treatment for hepatitis, 23 (0.6%) showed ALT concentrations higher than three times the upper limit of the reference range and 32 (0.9%) had a past history of either coronary heart disease or stroke. Thus, the baseline population consisted of 2981 men. We also excluded 63 men who did not participate in consecutive annual health examinations during follow-up. The final study population for analysis therefore consisted of 2918 men. Men in whom IFG or type 2 diabetes was found during repeated surveys during May 2001 were classified as having IFG or type 2 diabetes. To determine the incidence of type 2 diabetes, incidental cases of IFG were followed and were considered type 2 diabetes if they reached that end-point. Altogether, 39 participants who started taking medication for diabetes during the observation period were considered to have type 2 diabetes. Owing to the age range of the study population, all cases of IFG or type 2 diabetes were diagnosed after 35 years of age.

Study design

Fasting plasma glucose concentrations were measured at annual health examinations in May from 1994 to 2001. The participants were asked to fast for at least 8 h and to avoid smoking and heavy physical activity for more than 2 h before the examinations. Blood samples were drawn from an antecubital vein. Glucose was measured using hexokinase-glucose dehydrogenase method (Shino-Test, Tokyo, Japan) with Olympus AU-5000 in 1994 and Olympus AU-5200 in 1995 to 2001 (Olympus Japan, Tokyo, Japan) at FALCO Biosystems Tokyo (Tokyo, Japan). Quality control of the laboratory was assessed internally, and the coefficients of variation between and within assays for plasma glucose were no more than 3% from 1994 to 2001. Normal fasting glucose, IFG and type 2 diabetes were defined using the criteria of American Diabetes Association [19]. Normal fasting glucose was defined as fasting plasma glucose concentration of <6.1 mmol L^{-1}. IFG was defined as fasting plasma glucose concentration of 6.1–6.9 mmol L^{-1}. Type 2

diabetes was defined as fasting plasma glucose concentration of $\geqslant 7.0$ mmol L^{-1} or receipt of hypoglycaemic medications, because an oral glucose tolerance test was not performed for every subject.

Annual health examinations at study entry included medical history, physical examination, anthropometric measurements, biochemical measurements and a questionnaire on health-related behaviours, such as alcohol consumption, smoking and physical activity. Medical history and history of use of prescription drugs were assessed by the examining doctors. Family history of diabetes was defined as a mother, father, sister or brother with diagnosed diabetes. The body mass index (BMI) was used as a measure of overall obesity and was calculated as body weight/height^2 (kg m^{-2}). After a 5-min rest in a quiet room, systolic and diastolic blood pressures were measured in right arm by using a standard mercury sphygmomanometer. Serum total cholesterol and triglycerides were determined by the Olympus AU-5000 using enzymatic method with commercial reagents kits (Wako, Osaka, Japan) and high-density lipoprotein (HDL) cholesterol was assayed using the same enzymatic method after precipitation by polyethylene glycol. Enzyme activities for GGT, AST, ALT, and alkaline phosphatase were measured by the Olympus AU-5000 using commercial reagent kits (International Reagents, Kobe, Japan) based on the principles recommended by the Japan Society of Clinical Chemistry [20]. White blood cell (WBC) counts were determined using a Sysmex E-4000 autoanalyzer (Toa Medical Electronics, Tokyo, Japan).

For health-related behaviours, the questions about alcohol intake included items about frequency of alcohol consumption per week, type of alcoholic beverage and usual amount consumed daily in units of 'go' (a traditional Japanese unit of measurement, by volume, corresponding to 23 g of ethanol). Weekly alcohol intake was calculated and then converted to daily alcohol consumption by using standard Japanese tables. One go is 180 mL of sake and corresponds to one bottle (663 mL) of beer, two single shots (75 mL) of whiskey, or two glasses (180 mL) of wine. Subjects were classified as non-drinkers or current drinkers who averaged 0.1–22.9, 23.0–45.9, 46.0–68.9 or $\geqslant 69.0$ g day^{-1} of ethanol. The questionnaires were also used to ask about smoking habits (never, past or current smoker); past or current smokers were questioned about the number of cigarettes smoked per day and the duration of smoking in years. Participants were inquired about the type and weekly frequency of leisure-time physical activity. Physical exercise was defined as participation in any physical activity such as jogging, bicycling, swimming or tennis, that was performed long enough to sweat.

Statistical analyses

The chi-squared test and one-way analysis of variance were used to analyse statistical differences amongst characteristics of the study participants at enrolment according to serum GGT. Categories of serum GGT were defined by the following quartiles: <16, 16–24, 25–43 and $\geqslant 44$ U L^{-1}. For each participant, person-years of follow-up were calculated from the date of enrolment to the date of the development of IFG or type 2 diabetes or the date of follow-up, whichever occurred first. The follow-up rate was 92.6% of the total potential person-years of follow-up. Cox's proportional hazards models were used to evaluate the association between serum GGT and development of IFG or type 2 diabetes. The data were adjusted first for age alone and then for the following multiple covariates: age, family history of diabetes, BMI, alcohol consumption and physical activity (model 1). The data were also additionally adjusted for systolic blood pressure, concentrations of total cholesterol, triglycerides, fasting plasma glucose, and WBC count (model 2). Potential confounding factors were treated as categorical variables: age, BMI, systolic blood pressure, total cholesterol concentration, triglyceride concentration, fasting plasma glucose concentration and WBC count [graded from 1 to 5 (first to fifth quintiles)]; family history of diabetes (no or yes); alcohol consumption [graded as 1 (none) or as quartile 1 (grade of 2) to quartile 4 (grade of 5) for drinkers]; cigarette smoking [graded as 1 (none) or as quartile 1 (grade of 2) to quartile 4 (grade of 5) for current smokers]; and regular physical exercise [graded from 1 to 3 (hardly ever, once a week, or twice or more a week)]. The linear trends in risks were evaluated by entering indicators for each category of exposure.

Data were analysed by using the SPSS/PC statistical package (SPSS, Chicago, IL, USA). All reported P values are two-tailed and those less than 0.05 were considered to be statistically significant.

Results

The baseline characteristics of the study sample according to serum GGT are shown in Table 1. Tests for differences in baseline characteristics across four groups of serum GGT were significant except for family history of diabetes, regular physical activity and HDL cholesterol. Age, BMI, current drinking, current cigarette smoking, systolic and diastolic blood pressures, fasting plasma glucose, total cholesterol, triglycerides, WBC count, AST, ALT and alkaline phosphatase showed a linear trend to GGT.

Altogether 274 and 166 men developed IFG and type 2 diabetes during the 18 084 and 18 387 person-years of follow-up, respectively (Table 2). With adjustment for potential risk factors for diabetes – age, family history of diabetes, BMI, alcohol consumption, cigarette smoking, and regular physical exercise (model 1) – the relative risk for IFG and type 2 diabetes increased as serum GGT increased. The multivariate-adjusted relative risk for IFG compared with serum GGT <16 U L^{-1} was 1.37 (95% CI, 0.89–2.11), 1.80 (CI, 1.17–2.76), and 2.36 (CI, 1.53–3.63) with serum GGT of 16–24, 25–43 and ⩾44 U L^{-1}, respectively (P for trend <0.001). The respective relative risks for type 2 diabetes compared with serum GGT <16 U L^{-1} were 2.76 (CI, 1.41–5.43), 3.26 (CI, 1.66–6.43) and 4.92 (CI, 2.49–9.70) (P for trend <0.001). Although additional adjustment for systolic blood pressure, total cholesterol, triglycerides, fasting plasma glucose and WBC count (model 2) lowered the magnitude of these associations, a similar, independent and graded association was observed between serum GGT and risk for IFG and type 2 diabetes (P for trend = 0.014 and 0.002, respectively).

To assess the effect of obesity and alcohol consumption on the association between serum GGT and risk for diabetes, we studied the relation between serum GGT and development of IFG or type 2 diabetes according to BMI and drinking status (Table 3). IFG and type 2 diabetes were combined in this analysis, because we assumed a similar mechanism for the development of IFG and type 2 diabetes. Amongst those with a BMI <23.2 kg m^{-2}, with full adjustment for potential risk factors (model 2), no significant associations between serum GGT and risk for IFG or type 2 diabetes were observed in both those who drank <46 and ⩾46 g day^{-1} of ethanol. However, amongst those with a BMI ⩾23.2 kg m^{-2}, the multivariate-adjusted relative risk for IFG or type 2 diabetes increased as serum GGT increased in both those who drank <46 and ⩾46 g day^{-1} of ethanol. The multivariate-adjusted relative risk for IFG or type 2 diabetes compared with serum GGT

Table 1 Baseline characteristics of 2918 Japanese male office workers, according to serum γ-glutamyltransferase

Characteristics	Serum γ-glutamyltransferase, U L^{-1}				
	<16 (n = 646)	16–24 (n = 775)	25–43 (n = 750)	≥44 (n = 747)	P value
Age, years	45.8 ± 6.4	46.5 ± 6.3	46.5 ± 6.0	47.1 ± 5.5	<0.001
Family history of diabetes, %	7.3	7.5	8.4	8.4	0.782
BMI, kg m^{-2}	22.1 ± 2.2	23.1 ± 2.4	23.7 ± 2.6	24.1 ± 2.7	<0.001
Current drinkers, %	73.4	84.0	90.3	94.8	<0.001
Current smokers, %	44.3	48.1	52.0	56.9	<0.001
Regular physical activity at least once a week, %	51.5	50.5	54.5	53.5	0.375
Systolic blood pressure, mmHg	121.4 ± 13.6	125.8 ± 13.4	126.9 ± 14.0	131.7 ± 14.3	<0.001
Diastolic blood pressure, mmHg	73.3 ± 10.1	76.5 ± 10.2	77.6 ± 10.5	81.2 ± 10.6	<0.001
Total cholesterol, mmol L^{-1}	4.69 ± 0.70	4.96 ± 0.75	5.07 ± 0.82	5.23 ± 0.86	<0.001
HDL cholesterol, mmol L^{-1}	1.42 ± 0.30	1.40 ± 0.32	1.38 ± 0.35	1.41 ± 0.35	0.111
Triglycerides, mmol L^{-1}	0.98 ± 0.52	1.25 ± 0.76	1.54 ± 1.07	1.97 ± 1.39	<0.001
Fasting plasma glucose, mmol L^{-1}	4.93 ± 0.34	4.98 ± 0.34	4.99 ± 0.35	5.05 ± 0.37	<0.001
White blood cell count, 10^9 cells per litre	6.14 ± 1.70	6.35 ± 1.60	6.60 ± 1.74	6.63 ± 1.69	<0.001
Aspartate aminotransferase, U L^{-1}	18.0 ± 4.7	20.2 ± 5.3	22.7 ± 6.7	28.5 ± 13.3	<0.001
Alanine aminotransferase, U L^{-1}	15.6 ± 6.6	19.9 ± 9.3	25.5 ± 13.0	33.0 ± 17.3	<0.001
Alkaline phosphatase, U L^{-1}	162.7 ± 30.9	164.3 ± 31.7	167.1 ± 30.7	174.1 ± 37.0	<0.001

Data are mean ± SD unless indicated otherwise.

Table 2 The risk for incidence of impaired fasting glucose and type 2 diabetes during 7 years of follow-up, according to serum γ-glutamyltransferase

	Serum γ-glutamyltransferase, U L^{-1}				
	<16 (n = 646)	16–24 (n = 775)	25–43 (n = 750)	≥44 (n = 747)	P value*
Impaired fasting glucose					
Cases, n	33	60	78	103	
Total person-years	4112	4864	4591	4517	
Rate per 1000 person-years	8.0	12.3	17.0	22.8	
Age-adjusted relative risk (95% CI)	1.0 (referent)	1.51 (0.99–2.31)	2.08 (1.38–3.13)	2.75 (1.86–4.07)	<0.001
Multivariate-adjusted relative risk (95% CI)[a]	1.0 (referent)	1.37 (0.89–2.11)	1.80 (1.17–2.76)	2.36 (1.53–3.63)	<0.001
Multivariate-adjusted relative risk (95% CI)[b]	1.0 (referent)	1.23 (0.79–1.90)	1.50 (0.97–2.32)	1.70 (1.074–2.71)	0.014
Type 2 diabetes					
Cases, n	11	40	47	68	
Total person-years	4188	4897	4682	4620	
Rate per 1000 person-years	2.6	8.2	10.0	14.7	
Age-adjusted relative risk (95% CI)	1.0 (referent)	3.06 (1.57–5.96)	3.75 (1.95–7.23)	5.43 (2.87–10.27)	<0.001
Multivariate-adjusted relative risk (95% CI)[a]	1.0 (referent)	2.76 (1.41–5.43)	3.26 (1.66–6.43)	4.92 (2.49–9.70)	<0.001
Multivariate-adjusted relative risk (95% CI)[b]	1.0 (referent)	2.54 (1.29–5.01)	2.64 (1.33–5.23)	3.44 (1.69–6.70)	0.002

*For linear trend.
[a]Adjusted for age, family history of diabetes, BMI, alcohol intake, cigarette smoking and regular physical activity at study entry.
[b]Additionally adjusted for systolic blood pressure, total cholesterol concentration, triglyceride concentration, plasma fasting glucose concentration and white blood cell count at study entry.

<16 U L^{-1} was 1.57 (CI, 0.76–3.23), 2.12 (CI, 1.05–4.30) and 2.86 (CI, 1.36–6.03) amongst those who drank <46.0 g day^{-1} of ethanol (P for trend = 0.003) and 1.54 (CI, 0.34–6.95), 2.31 (CI, 0.54–9.92) and 3.04 (CI, 0.71–12.94) amongst those who drank ⩾46 g day^{-1} of ethanol (P for trend = 0.017) with serum GGT of 16–24, 25–43, and ⩾44 U L^{-1}, respectively.

As for the association between serum GGT and other liver enzymes, the age-adjusted relative risk for IFG or type 2 diabetes compared with the first quartile was 1.06 (CI, 0.75–1.51), 0.97 (CI, 0.69–1.35) and 1.46 (1.06–2.02) for AST (P for trend = 0.018); 1.00 (CI, 0.71–1.42), 1.15 (CI, 0.82–1.62) and 1.83 (CI, 1.32–2.52) for ALT (P for trend <0.001); and 1.28 (CI, 0.93–1.76), 1.51 (1.12–2.03) and 1.45 (CI, 1.07–1.96) for alkaline phosphatase (P for trend = 0.010) with the second, third and fourth quartiles, respectively. These increased risks were attenuated after full adjustment for potential risk factors (model 2) (top versus bottom fourth relative risk: 1.08 (CI, 0.77–1.52) for AST, 1.19 (CI, 0.84–1.69) for ALT and 1.28 (CI, 0.93–1.75) for alkaline phosphatase.

Discussion

We found a linear relation between serum GGT and risk for development of IFG or type 2 diabetes even after adjustment for potential confounders for diabetes. In contrast, significant associations between serum AST, ALT and alkaline phosphatase, and risk for development of IFG or type 2 diabetes were not observed in the multivariate analyses. From stratified analyses by BMI and alcohol intake, a stronger linear association between serum GGT and development of IFG or type 2 diabetes was found in men with a BMI ⩾23.2 kg m^{-2} in both those who drank <46 and ⩾46 g day^{-1} of ethanol. In this cohort, a U-shaped association was found between alcohol intake and development of IFG or type 2 diabetes over a 7-year period, with the lowest incidence at an intake of 23.0–45.9 g day^{-1} of ethanol [21]. The risk for development of IFG or type 2 diabetes was similar in heavy drinkers who drank 46 g day^{-1} of ethanol and nondrinkers. Hence, the association between serum GGT and risk of IFG or type 2 diabetes cannot be attributed to residual confounding due to alcohol intake.

Table 3 The risk of impaired fasting glucose or type 2 diabetes during 7 years of follow-up by body mass index, alcohol drinking and serum γ-glutamyltransferase

Characteristic	Serum γ-glutamyltransferase, U L^{-1}				
	<16	16–24	25–43	≥44	*P* value*
Body mass index < median (23.2 kg m^{-2})					
Alcohol consumption <46 g day^{-1} of ethanol					
Participants, *n*	364	247	149	85	
Cases, *n*	25	29	19	8	
Total person-years	2278	1530	900	512	
Rate per 1000 person-years	11.0	19.0	21.1	15.6	
Age-adjusted relative risk (95% CI)	1.0 (referent)	1.70 (1.00–2.91)	1.89 (1.04–3.44)	1.38 (0.62–3.07)	0.104
Multivariate-adjusted relative risk (95% CI)†	1.0 (referent)	1.47 (0.84–2.58)	1.70 (0.90–3.22)	1.22 (0.53–2.80)	0.302
Alcohol consumption ≥46 g day^{-1} of ethanol					
Participants, *n*	81	168	172	193	
Cases, *n*	5	17	20	26	
Total person-years	522	1048	1058	1167	
Rate per 1000 person-years	9.6	16.2	18.9	22.3	
Age-adjusted relative risk (95% CI)	1.0 (referent)	1.55 (0.57–4.22)	1.85 (0.69–4.95)	2.07 (0.79–5.42)	0.116
Multivariate-adjusted relative risk (95% CI)†	1.0 (referent)	1.16 (0.42–3.24)	1.39 (0.51–3.81)	0.99 (0.35–2.79)	0.808
Body mass index ≥ median (23.2 kg m^{-2})					
Alcohol consumption <46 g day^{-1} of ethanol					
Participants, *n*	169	232	229	152	
Cases, *n*	11	26	40	32	
Total person-years	1074	1371	1343	868	
Rate per 1000 person-years	10.2	19.0	29.8	36.9	
Age-adjusted relative risk (95% CI)	1.0 (referent)	1.82 (0.90–3.68)	2.83 (1.45–5.51)	3.57 (1.80–7.09)	<0.001
Multivariate-adjusted relative risk (95% CI)†	1.0 (referent)	1.57 (0.76–3.23)	2.12 (1.05–4.30)	2.86 (1.36–6.03)	0.003
Alcohol consumption ≥46 g day^{-1} of ethanol					
Participants, *n*	32	128	200	317	
Cases, *n*	2	12	25	65	
Total person-years	200	806	1168	1831	
Rate per 1000 person-years	10.0	14.9	21.4	35.5	
Age-adjusted relative risk (95% CI)	1.0 (referent)	1.47 (0.33–6.55)	2.10 (0.50–8.86)	3.42 (0.84–13.99)	0.001
Multivariate-adjusted relative risk (95% CI)†	1.0 (referent)	1.54 (0.34–6.95)	2.31 (0.54–9.92)	3.04 (0.72–12.94)	0.017

*For linear trend.

†Adjusted for age, family history of diabetes, BMI, alcohol intake, cigarette smoking, regular physical activity, systolic blood pressure, total cholesterol concentration, triglyceride concentration, plasma fasting glucose concentration and white blood cell count at study entry.

Our results are consistent with those of Perry *et al.* [18] and indicate that serum GGT is associated with an increased risk for IFG or type 2 diabetes and that this association is more pronounced in obese men. Although the mechanism of how serum GGT increases the risk for diabetes remains to be elucidated, the findings that elevated serum GGT is associated with a higher risk of diabetes and that obesity may modify or strengthen these associations are biologically plausible. One explanation for our findings is that raised serum GGT reflects fatty acid changes in the liver or hepatic steatosis, and that this in turn reflects pathophysiological changes predating the development of type 2 diabetes. In fact, serum GGT is closely linked with body fat [5, 11–13, 16]. Obesity is associated with insulin resistance and hyperinsulinaemia [22], which are related to increased GGT and hepatic steatosis [14–16]. Steatosis appears when insulin secretion is sufficient to block free fatty acid oxidation, but not sufficient to block free fatty acid mobilization from adipose tissue [23]. Free fatty acids are the major substrate for hepatic triglyceride synthesis, explaining the strong association between triglycerides and

fasting insulin, and markedly cytotoxic [24], which may lead to alterations in hepatic cell membrane function and to increased release of GTT [16, 25–27]. Furthermore, the hepatic synthesis of triglycerides increases in obesity as a consequence of hyperinsulinaemia in combination with increased concentrations of substrate-free fatty acids and glucose [28]. As serum GGT is strongly associated with concentrations of triglycerides in this study, the stronger association between serum GGT and development of IFG or type 2 diabetes in obese individuals may be a reflection of the increased prevalence of fatty liver in more obese individuals.

A second pathophysiological mechanism is also possible. Fatty acid change is a characteristic response of the liver to the proinflammatory cytokine tumour necrosis factor-α [29, 30]. Fatty acid change and concomitant raised serum GGT may reflect inflammation, which may impair insulin signalling both in the liver and other organ systems. Furthermore, WBCs, one major component of the inflammatory process, are increased by cytokines, especially interleukin-6 [31]. In this study, as serum GGT is highly associated with WBC count and some features of low-graded inflammation, especially high triglycerides, are identical to the components of the insulin resistance syndrome [22], elevated GGT could be the expression of subclinical inflammation or an insulin-resistant state, which would represent the underlying mechanism [32, 33]. It is therefore suggested that measurements of other inflammatory markers including C-reactive protein by a validated high-sensitivity assay be added in an attempt to substantiate this hypothesis.

Our study had several limitations. First, serum GGT during follow-up was not included in the analysis. In this study, serum GGT at study entry was highly associated with that at the date of diagnosis of IFG or type 2 diabetes or at the end of follow-up (Spearman's rank correlation coefficient: 0.755, $P < 0.001$). This indicates that those who had higher serum GGT at study entry tended to do so during follow-up. The observed associations between serum GGT at baseline and the increased risk for IFG or type 2 diabetes may reflect the effects of serum GGT over an observation period.

Secondly, bias in case-finding could have occurred. Specifically, men with high serum GGT are more likely to visit a doctor for reasons other than diabetes, thus diabetes could have been found by chance. However, because all incidental cases were found by periodic annual screening in our study, such bias is unlikely to have occurred. Furthermore, participants in our normoglycaemic cohort, particularly those in the older age groups, might not be typical of the general population. Those whose plasma glucose concentration was already above borderline values or who reported taking drugs for hypertension or having a past history of cardiovascular disease during the initial examination were excluded. Because hypertension is a recognized risk factor for diabetes [34, 35], exclusion of hypertensive persons would bias the study towards a particularly healthy study population at a low risk for diabetes. The selection of men with rigorously normal fasting plasma glucose concentration at study entry could have had an effect on the observations.

Finally, we could not include several confounding variables in this study, such as waist-to-hip ratio and fasting insulin concentration. The central pattern of distribution, with its increased waist-to-hip ratio, is associated with more insulin resistance than is the peripheral pattern of distribution [36, 37]. Individuals with the central pattern are more likely to have glucose intolerance and hyperinsulinaemia resulting from insulin resistance [38, 39]. Therefore, visceral adiposity (waist-to-hip ratio) and fasting insulin concentration should be included in future studies.

Despite these potential limitations, our findings, which were obtained from a cohort of middle-aged Japanese men, support the conclusion that an elevated, albeit normal, serum GGT is associated with a higher risk for development of IFG or type 2 diabetes.

Conflict of interest

No conflict of interest was declared.

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