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DIABETES MELLITUS

Serum gamma-glutamyl transferase level and diabetes mellitus among US adults

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Abstract Serum gamma-glutamyl transferase (GGT), a marker of oxidative stress, has been shown to be associated with diabetes mellitus in some population-based studies, but not all. Also, it is not clear if there is a continuous dose-response relationship in this association, or if this association is evident only beyond a particular threshold level of GGT. We examined the association between serum GGT and diabetes mellitus in a representative sample of US adults aged ≥20 years, in a cross-sectional study involving 7,976 National Health and Nutrition Examination Survey 1999–2002 participants. Diabetes mellitus was defined as a fasting glucose ≥ 126mg/dl, nonfasting glucose ≥200 mg/dl, or use of oral hypoglycemic medication or insulin ($n = 805$). Higher serum GGT levels were positively associated with diabetes mellitus, independent of, alcohol consumption, body mass index, hypertension and other confounders. Multivariable odds ratio (95% confidence interval) comparing quartile 4 of GGT (>33 U/L) to quartile 1 (<15U/L) was 2.33 (1.59–3.41), P-trend < 0.0001. This association persisted in separate analysis among men and women. In nonparametric models, the positive association between serum GGT and diabetes appeared to be present across the full range of GGT, without any threshold effect. Higher serum GGT levels are positively associated with diabetes mellitus.

Keywords Gamma-glutamyl transferase · Diabetes mellitus · Gender · NHANES

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Introduction

Serum gamma-glutamyl transferase (GGT), an enzyme responsible for extracellular catabolism of glutathione and a marker of oxidative stress [1] has been shown to be associated with cardiovascular disease [2], peripheral arterial disease [3], and hypertension [4]. Several prospective [5–10] and cross-sectional studies [11–13] have reported a positive association between serum GGT and diabetes mellitus. In this context, we studied the association between serum GGT and diabetes mellitus in a nationally representative sample of US adults participating in the National Health and Nutrition Examination Survey (NHANES) (1999–2002). We also employed nonparametric analytical techniques to graphically examine the dose-response nature of the association between serum GGT levels and diabetes mellitus.

Methods

Study population

The NHANES 1999–2002 collected data on a nationwide, probability sample of the civilian non institutionalized US population. Standardized questionnaires were administered in the home, followed by a detailed physical examination at a mobile examination center. NHANES 1999–2002 uses a

stratified, multistage, clustered sample with oversampling of Mexican Americans and non-Hispanic blacks to ensure adequate sample size for analysis. Detailed descriptions of the complex survey design, interviewing procedures, physical and laboratory examinations conducted have been published before and are available online [14].

Of the 10,291 adults ≥20 years of age who participated in the interview and examination component of NHANES (1999–2002), participants without history of cardiovascular disease (coronary heart disease, myocardial infarction, angina, stroke or heart failure) and with data on serum GGT and other important covariates were included for this analysis ($n = 7{,}976$). Diabetes was defined using American Diabetes Association criteria as follows: a serum glucose ≥126 mg/dl after fasting for a minimum of 8 h, a serum glucose ≥200 mg/dl for those who fasted <8 h before their NHANES visit, or self-reported current use of oral hypoglycemic medication or insulin [15].

Questionnaires were used to collect information on age, gender, smoking status, alcohol intake, educational attainment, self-reported history of diabetes, hypertension, and intake of oral hypoglycemic drug or insulin administration or antihypertensive medication. Food frequency questionnaire was used to collect information on coffee intake over the past month. Dietary intake of fruit, red meat and energy consumption was assessed from a single 24 h recall. Information on anthropometric, physical and laboratory examination components were obtained during the medical examination center (MEC) examination. Patients were considered hypertensive if they reported current blood pressure-reducing medication use and/or had systolic blood pressures ≥140 mm Hg and/or diastolic blood pressures ≥90 mm Hg. Body Mass Index (BMI) was calculated as weight in kilograms divided by the square of height in meters. Plasma glucose was measured at the University of Missouri Diabetes Diagnostic Laboratory using a modified hexokinase enzymatic method. Glycosylated hemoglobin was also measured at the University of Missouri using a boronate affinity high-performance liquid chromatography system. Serum GGT concentration was assayed with a Hitachi 737 Analyzer (Boehringer–Mannheim Diagnostics, Indianapolis, IN, USA) at White Sands Research Center, Alamogordo, New Mexico (USA); coefficients of variation for GGT ranged from 1.9 to 3.5% [16].

Because of its skewed distribution, serum GGT was log transformed (base 2) when analyzed as a continuous variable. We analyzed serum GGT levels as quartiles: <15 U/L, 15–21 U/L, 22–33 U/L, and >33 U/L. As initial analysis based on gender-specific GGT quartiles render similar results as GGT quartiles for the whole cohort and there is no effect modification by gender, GGT cutoffs for the whole cohort were used to simplify the presented results. The odds ratio (OR) and (95% confidence interval [CI]) of diabetes mellitus were calculated across the quartiles of serum GGT level, with the lowest quartile of GGT (quartile 1) as the reference, in a multivariable logistic regression model adjusted for education categories (<high school, high school, >high school), smoking (never, former, current), alcohol intake (grams/day), waist circumference (cms), hypertension (absent, present), and serum total cholesterol (mg/dl). Trends in the OR of diabetes across increasing serum GGT category were examined modeling GGT categories as an ordinal variable. To specifically examine the effect of coffee intake on our results, we repeated the main analysis after additionally adjusting for coffee intake (cups/day) in the multivariable model. Sample weights that account for the unequal probabilities of selection, oversampling, and nonresponse were applied for all analyses using SUDAAN (version 8.0; Research Triangle Institute, Research Triangle Park, NC) and SAS (version 9.2.; SAS institute, Cary, NC) softwares; SEs were estimated using the Taylor series linearization method. To examine the dose-response relationship of the observed association between GGT level and diabetes mellitus without linearity assumptions, we used flexible nonparametric logistic regression employing the generalized additive modeling approach (R system for statistical computing, available from Comprehensive R Archive Network [http://www.CRAN.R-project.org]) to calculate odds of diabetes mellitus [17, 18], adjusting for all covariates in the multivariable model; the predicted odds of diabetes mellitus were then plotted against increasing GGT levels (both on the log scale). All statistical analyses were performed using SAS version 9.1.

Results

Among 7,976 adults ≥20 years of age included in the current analysis, 805 participants had diabetes mellitus. Table 1 shows selected baseline characteristics by quartiles of serum GGT. Participants with higher levels of GGT were more likely to be: women, older, smokers (ever and current) and hypertensives; more likely to have below high school education, consumed more alcohol per day, greater waist circumference, and higher BMI, glycosylated hemoglobin levels and total cholesterol than those with lower levels of serum GGT. The prevalence of diabetes mellitus increased with increasing quartiles of serum GGT.

In Table 2, we present the odds ratios of diabetes by quartiles of GGT for the whole cohort and stratified by gender. In the whole cohort, increasing quartiles of GGT levels were positively associated with diabetes mellitus in the multivariable-adjusted model; corresponding models for assessing trend across GGT quartiles were also

Table 1 Characteristics of the study population by categories of serum gamma-glutamyltransferase (GGT) levels

Characteristics	Serum GGT quartiles				*P*-trend[†]
	Quartile 1 (<15 U/L)	Quartile 2 (15–21 U/L)	Quartile 3 (21–33 U/L)	Quartile 4 (>33 U/L)	
Number at risk	2,209	1,829	1,990	1,948	
Women (%)	76.5 ± 0.9	55.3 ± 1.1	41.0 ± 1.1	34.2 ± 1.0	<0.0001
Age (years)	44.40 ± 19.74	50.34 ± 19.11	50.98 ± 17.82	50.34 ± 16.08	<0.0001
Education categories (%)[a]					
Below high school	27.4 ± 0.9	32.0 ± 1.0	34.4 ± 1.0	38.1 ± 1.0	<0.0001
High school	22.8 ± 0.8	21.6 ± 0.9	23.2 ± 0.9	24.2 ± 0.9	0.2836
Above high school	49.4 ± 1.0	46.1 ± 1.1	42.2 ± 1.1	37.3 ± 1.0	<0.0001
Smoking (%)[a]					
Ever smoker	38.2 ± 1.0	46.1 ± 1.1	50.6 ± 1.1	58.2 ± 1.1	<0.0001
Current smoker	16.0 ± 0.7	18.8 ± 0.9	22.4 ± 0.9	28.8 ± 1.0	<0.0001
Alcohol intake (grams/day)	4.63 ± 19.26	7.83 ± 30.54	10.08 ± 31.49	17.01 ± 45.08	<0.0001
Body mass index (kg/m^2)	26.56 ± 5.67	27.90 ± 6.15	29.15 ± 5.98	29.53 ± 6.13	<0.0001
Waist circumference (cm)	91.02 ± 13.88	95.49 ± 14.81	99.82 ± 14.39	101.47 ± 14.88	<0.0001
Total cholesterol (mg/dl)	196.84 ± 42.77	197.78 ± 38.85	202.23 ± 41.46	208.31 ± 41.87	<0.0001
Hypertension (%)[a]	24.5 ± 0.9	33.1 ± 1.1	41.9 ± 1.1	43.6 ± 1.1	<0.0001
Glycosylated hemoglobin (%)	5.26 ± 0.71	5.50 ± 0.91	5.71 ± 1.16	5.82 ± 1.35	<0.0001
Diabetes mellitus (%)	4.3 ± 0.4	8.5 ± 0.6	12.3 ± 0.7	15.7 ± 0.8	<0.0001

[a] Data presented are row percentages or mean values and corresponding standard error (SE)

[†] *P*-trend estimated from linear regression or logistic regression models, as appropriate, with GGT categories as an ordinal variable

Table 2 Association between serum gamma-glutamyltransferase (GGT) level and diabetes mellitus

	Whole cohort (n = 7,976)		Men (n = 3,788)		Women (n = 4,188)	
	No. at risk (cases)	Multivariable OR (95% CI)[a]	No. at risk (cases)	Multivariable OR (95% CI)[a]	No. at risk (cases)	Multivariable OR (95% CI)[a]
Quartile 1 (<15 U/L)	2,209 (96)	1 (referent)	517 (32)	1 (referent)	1,692 (64)	1 (referent)
Quartile 2 (15–21 U/L)	1,829 (157)	0.99 (0.67, 1.46)	817 (78)	1.19 (0.65, 2.18)	1,012 (79)	0.92 (0.58, 1.44)
Quartile 3 (21–33 U/L)	1,990 (246)	1.60 (1.02, 2.53)	1,174 (134)	1.90 (0.99, 3.64)	816 (112)	1.40 (0.83, 2.38)
Quartile 4 (>33 U/L)	1,948 (306)	2.33 (1.59, 3.41)	1,280 (178)	2.79 (1.45, 5.39)	668 (128)	2.17 (1.39, 3.39)
P-trend		<0.0001		0.0005		<0.0001
Log-transformed serum GGT (U/L)	7,976 (805)	1.47 (1.31, 1.65)	3,788 (422)	1.51 (1.27, 1.81)	4,188 (383)	1.44 (1.21, 1.70)

[a] Adjusted for age (years), sex (men, women), education categories (<high school, high school, >high school), smoking (never, former, current), alcohol intake (grams/day), waist circumference (cms), hypertension (absent, present), and serum total cholesterol (mg/dl)

statistically significant. When serum GGT was analyzed as a continuous variable, the positive association with diabetes persisted. In gender-specific analysis, the positive association between serum GGT and diabetes was consistently present both among men (OR = 2.79) and women (OR = 2.17), comparing quartile 4 with quartile 1 of serum GGT.

We then employed nonparametric models to examine if the observed positive association between higher quartiles of GGT and diabetes mellitus was present across the full range of GGT levels available in the study (Fig. 1). This analysis reveals a continuous positive association between GGT and diabetes mellitus with increasing GGT levels without a threshold effect.

In a supplementary analysis, to examine if the observed association between serum GGT and diabetes is explained by inflammation, we additionally adjusted for CRP (mg/dl) in the multivariable model and the results were essentially similar. Compared to GGT quartile 1 (referent), the OR (95% CI) of diabetes was 0.99 (0.67–1.46) in quartile 2,

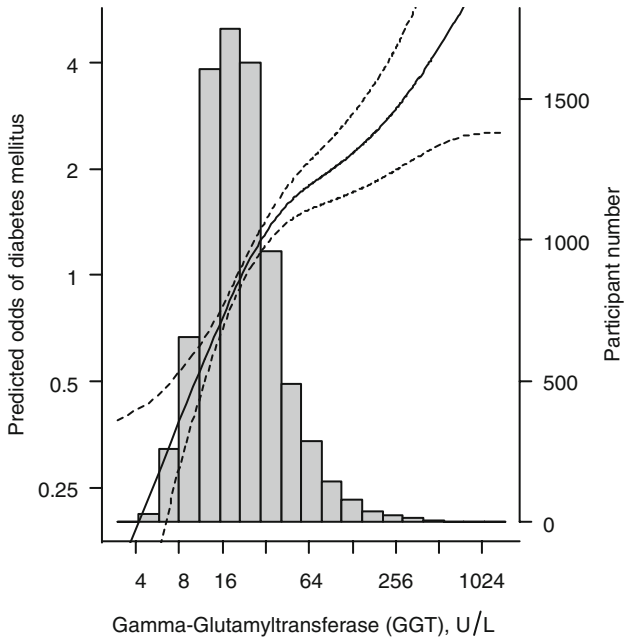

Fig. 1 Multivariable-adjusted odds of diabetes mellitus according to serum gamma-glutamyltransferase (GGT) level. *Solid thick line* represents the predicted odds of diabetes from nonparametric logistic regression; *dashed lines*, 95% confidence limits for the nonparametric logistic regression estimates. The nonparametric logistic regression was adjusted for age (years), sex (men, women), education categories (<high school, high school, >high school), smoking (never, former, current), alcohol intake (grams/day), waist circumference (cms), hypertension (absent, present), and serum total cholesterol (mg/dl). *X* axis: Serum gamma-glutamyltransferase (GGT) level (U/L) plotted in log scale. *Y1* axis: Predicted odds of diabetes mellitus plotted in log scale. *Y2* axis: Participant number for each serum gamma-glutamyltransferase (GGT) level

1.60 (1.00–2.53) in quartile 3 and 2.31 (1.57–3.39) in quartile 4 (P-trend < 0.0001). Similarly, additionally adjusting for coffee intake (cups/day) did not materially alter the results. Compared to GGT quartile 1 (referent), the OR (95% CI) of diabetes was 1.01 (0.68–1.50) in quartile 2, 1.55 (1.00–2.40) in quartile 3 and 2.23 (1.52–3.27) in quartile 4 (P-trend < 0.0001). In a third supplementary analysis, we additionally adjusted for intake of fruit (servings/day), red meat (servings/day), and total energy intake (kilocalories/day) in the multivariable model; the results were essentially similar. Compared to GGT quartile 1 (referent), the OR (95% CI) of diabetes was 1.01 (0.69–1.48) in quartile 2, 1.56 (1.01–2.41) in quartile 3 and 2.27 (1.52–3.39) in quartile 4 (P-trend $= 0.0032$). Finally, when we replaced waist circumference with BMI in the multivariable model, the results were not materially different. Compared to GGT quartile 1 (referent), the OR (95% CI) of diabetes was 1.03 (0.67–1.58) in quartile 2, 1.62 (1.03–2.55) in quartile 3 and 2.33 (1.60–3.39) in quartile 4 (P-trend $= 0.0011$).

Discussion

In a nationally representative sample of US adults ≥20 years, we found that serum GGT level was positively associated with diabetes mellitus in a dose-dependent manner, independent of age, sex, education, smoking, alcohol intake, waist circumference, hypertension and serum cholesterol level. In a subsequent analysis employing nonparametric models, the observed positive association between serum GGT level and diabetes mellitus was present continuously across the full range of GGT.

Serum GGT is also a marker of alcohol consumption [19]. In our study, the association between serum GGT and diabetes mellitus was present even after adjusting for grams of daily alcohol intake, suggesting an effect independent of alcohol use. The association appeared to be independent of CRP levels, a marker of inflammation [20]. Also our results are consistent with most [1, 5–10, 12] but not all [21, 22] previous studies that examined the association between serum GGT and diabetes mellitus. Plausible mechanisms that support the association of serum GGT with diabetes mellitus include the role of GGT in oxidative stress [1], insulin resistance [10], and hepatic inflammation which impairs insulin signaling in liver and other organs [21].

The main study limitation is the cross-sectional nature of NHANES which limits making causal inferences in the association between serum GGT and diabetes. In addition, GGT data was based on a single measurement, limiting the precision of the elevated GGT estimates. A reliability study using NHANES data III showed intra-individual variability of 13.8% for serum GGT measurements [23].

In conclusion, serum GGT levels were found to be positively associated with diabetes mellitus in a nationally representative sample of US adults. These results suggest an independent role for oxidative stress in diabetes mellitus. Also, studies have shown associations between serum GGT and hypertension, high lipid levels and the metabolic syndrome. If these associations are confirmed in future prospective studies in diverse populations, higher levels of this relatively inexpensive marker could serve as a risk stratification tool for cardiovascular disease and diabetes mellitus.

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Ethical Approval: This study followed the recommendations of Declaration of Helsinki. Subjects signed a consent form, and approval was obtained from the Human Subjects Committee in the U.S. Department of Health and Human Service.

References

1. Lee DH, Blomhoff R, Jacobs DR Jr. Is serum gamma glutamyltransferase a marker of oxidative stress? Free Radic Res. 2004;38(6):535–9. doi:10.1080/10715760410001694026.
2. Lee DH, Silventoinen K, Hu G, Jacobs DR Jr, Jousilahti P, Sundvall J, et al. Serum gamma-glutamyltransferase predicts non-fatal myocardial infarction and fatal coronary heart disease among 28,838 middle-aged men and women. Eur Heart J. 2006;27(18):2170–6. doi:10.1093/eurheartj/ehl086.
3. Shankar A, Li J, Klein BE, Javier NF, Klein R. Serum gamma-glutamyltransferase level and peripheral arterial disease. Atherosclerosis. 2008;199(1):102–9. doi:10.1016/j.atherosclerosis.2007.10.009.
4. Shankar A, Li J. Association between serum gamma-glutamyltransferase level and prehypertension among US adults. Circ J. 2007;71(10):1567–72. doi:10.1253/circj.71.1567.
5. Nakanishi N, Nishina K, Li W, Sato M, Suzuki K, Tatara K. Serum gamma-glutamyltransferase and development of impaired fasting glucose or type 2 diabetes in middle-aged Japanese men. J Intern Med. 2003;254(3):287–95. doi:10.1046/j.1365-2796.2003.01198.x.
6. Lee DH, Ha MH, Kim JH, Christiani DC, Gross MD, Steffes M, et al. Gamma-glutamyltransferase and diabetes-a 4 year follow-up study. Diabetologia. 2003;46(3):359–64.
7. Meisinger C, Lowel H, Heier M, Schneider A, Thorand B. Serum gamma-glutamyltransferase and risk of type 2 diabetes mellitus in men and women from the general population. J Intern Med. 2005;258(6):527–35. doi:10.1111/j.1365-2796.2005.01572.x.
8. Lee DH, Silventoinen K, Jacobs DR Jr, Jousilahti P, Tuomileto J. Gamma-Glutamyltransferase, obesity, and the risk of type 2 diabetes: observational cohort study among 20,158 middle-aged men and women. J Clin Endocrinol Metab. 2004;89(11):5410–4. doi:10.1210/jc.2004-0505.
9. Perry IJ, Wannamethee SG, Shaper AG. Prospective study of serum gamma-glutamyltransferase and risk of NIDDM. Diabetes Care. 1998;21(5):732–7. doi:10.2337/diacare.21.5.732.
10. Nilssen O, Forde OH. Seven-year longitudinal population study of change in gamma-glutamyltransferase: the Tromso Study. Am J Epidemiol. 1994;139(8):787–92.
11. Kim DJ, Noh JH, Cho NH, Lee BW, Choi YH, Jung JH, et al. Serum gamma-glutamyltransferase within its normal concentration range is related to the presence of diabetes and cardiovascular risk factors. Diabet Med. 2005;22(9):1134–40. doi:10.1111/j.1464-5491.2005.01581.x.
12. Lee DH, Jacobs DR Jr, Gross M, Kiefe CI, Roseman J, Lewis CE, et al. Gamma-glutamyltransferase is a predictor of incident diabetes and hypertension: the Coronary Artery Risk Development in Young Adults (CARDIA) Study. Clin Chem. 2003;49(8):1358–66. doi:10.1373/49.8.1358.
13. Lim JS, Lee DH, Park JY, Jin SH, Jacobs DR Jr. A strong interaction between serum gamma-glutamyltransferase and obesity on the risk of prevalent type 2 diabetes: results from the Third National Health and Nutrition Examination Survey. Clin Chem. 2007;53(6):1092–8. doi:10.1373/clinchem.2006.079814.
14. National Center for Health Statistics NHANES. 2006. 1999–2002 Survey Questionnaires, Examination component and Laboratory componenets. Available at: www cdc gov/nchs/data/nhanes/guidelines1 pdf. Accessed 17 Jan 2007.
15. American Diabetes Association. Standards of medical care in diabetes–2007. Diabetes Care. 2007;30(Suppl 1):S4–41.
16. National Center for Health Statistics. 2006. Lab methods NHANES 1999–2002. Available at: www cdc gov/NCHS/data/nhanes/nhanes-01-02/140 b_met_uric_acid pdf. Accessed 21 Feb 2007.
17. Figueiras A, Cadarso-Suarez C. Application of nonparametric models for calculating odds ratios and their confidence intervals for continuous exposures. Am J Epidemiol. 2001;154(3):264–75. doi:10.1093/aje/154.3.264.
18. Hastie T, Tibshirani R. Generalized additive models, vol. 1. London: Chapman and Hall; 1990.
19. Teschke R, Brand A, Strohmeyer G. Induction of hepatic microsomal gamma-glutamyltransferase activity following chronic alcohol consumption. Biochem Biophys Res Commun. 1977;75(3):718–24. doi:10.1016/0006-291X(77)91531-5.
20. Ridker PM, Morrow DA. C-reactive protein, inflammation, and coronary risk. Cardiol Clin. 2003;21(3):315–25. doi:10.1016/S0733-8651(03)00079-1.
21. Vozarova B, Stefan N, Lindsay RS, Saremi A, Pratley RE, Bogardus C, et al. High alanine aminotransferase is associated with decreased hepatic insulin sensitivity and predicts the development of type 2 diabetes. Diabetes. 2002;51(6):1889–95. doi:10.2337/diabetes.51.6.1889.
22. Ohlson LO, Larsson B, Bjorntorp P, Eriksson H, Svardsudd K, Welin L, et al. Risk factors for type 2 (non-insulin-dependent) diabetes mellitus. Thirteen and one-half years of follow-up of the participants in a study of Swedish men born in 1913. Diabetologia. 1988;31(11):798–805. doi:10.1007/BF00277480.
23. Lazo M, Selvin E, Clark JM. Brief communication: clinical implications of short-term variability in liver function test results. Ann Intern Med. 2008;148(5):348–52.