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Gamma Glutamyl Transferase and Metabolic Syndrome, Cardiovascular Disease, and Mortality Risk The Framingham Heart Study

Douglas S. Lee, Jane C. Evans, Sander J. Robins, Peter W. Wilson, Irene Albano, Caroline S. Fox, Thomas J. Wang, Emelia J. Benjamin, Ralph B. D'Agostino, Ramachandran S. Vasan

Objective—To determine whether serum γ -glutamyl transferase (GGT) predicts cardiovascular disease (CVD) morbidity and mortality, accounting for temporal changes in known CVD risk factors and C-reactive protein (CRP).

Methods and Results—In 3451 Framingham Study participants (mean age 44 years, 52% women) we examined the relations of GGT with CVD risk factors, and prospectively determined the risk of new-onset metabolic syndrome, incident CVD, and death. GGT was positively associated with body mass index, blood pressure, LDL cholesterol, triglycerides, and blood glucose in cross-sectional analysis ($P < 0.005$). On follow-up (mean 19 years), 968 participants developed metabolic syndrome, 535 developed incident CVD, and 362 died. The risk of metabolic syndrome increased with higher GGT (multivariable-adjusted hazard ratio [HR] per SD increment log-GGT, 1.26 [95%CI: 1.18 to 1.35]). Adjusting for established CVD risk factors (as time-dependent covariates updated quadriennially) and baseline CRP, a 1-SD increase in log-GGT conferred a 13% increase in CVD risk ($P = 0.007$) and 26% increased risk of death ($P < 0.001$). Individuals in the highest GGT quartile experienced a 67% increase in CVD incidence (multivariable-adjusted HR 1.67, 95%CI: 1.25 to 2.22).

Conclusion—An increase in serum GGT predicts onset of metabolic syndrome, incident CVD, and death suggesting that GGT is a marker of metabolic and cardiovascular risk. (*Arterioscler Thromb Vasc Biol.* 2007;27:127-133.)

Key Words: biomarkers ■ gamma glutamyl transferase ■ risk factor ■ cardiovascular disease ■ metabolic syndrome ■ mortality

Gamma;-glutamyl transferase (GGT) has been regarded as a biomarker of hepatobiliary disease and alcohol consumption/abuse.¹ However, GGT is elaborated by extra-hepatic tissues including the kidney, epididymis, fibroblasts, lymphocytes, and lung.²⁻⁴ Accumulating experimental evidence suggests an important role for GGT in extracellular catabolism of glutathione, the principal thiol antioxidant in humans. GGT enhances the availability of cysteine to promote intracellular glutathione (GSH) resynthesis, thereby counteracting oxidant stress.^{2,5,6} GGT adsorbs onto circulating low-density lipoprotein cholesterol (LDL) and can catalyze its oxidation.⁷ It is expressed in the atheromatous core of coronary plaques, where it colocalizes with oxidized LDL and foam cells.⁸ GGT may also be proinflammatory, because it mediates interconversion of the glutathione-containing inflammatory mediator leukotriene C4 into leukotriene D4.⁹

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Parallel evidence from epidemiological studies suggest that higher serum GGT is associated with development of cardiovascular disease (CVD) risk factors, including diabetes, hypertension, dyslipidemia,¹⁰⁻¹³ and the metabolic syndrome.¹⁰ GGT levels correlate positively with novel cardiovascular risk factors such as C-reactive protein (CRP), fibrinogen, F2-isoprostanes,¹⁴ and inversely with antioxidant levels.¹⁵ Prior studies associated increased GGT with mortality attributable to ischemic heart and cerebrovascular disease,¹⁶⁻¹⁸ but have not addressed whether serum GGT reflects greater burden of CVD risk factors^{12,13,19} or whether GGT has incremental prognostic utility beyond these risk factors.^{20,21} Although prior studies have had unique strengths, they did not adjust for established cardiovascular risk factors or CRP^{16,22-24} and had limited end point selection.²⁴

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We examined the cross-sectional clinical correlates of serum GGT and evaluated, longitudinally, whether higher levels predicted future CVD events and mortality in the Framingham Heart Study. We hypothesized that increasing serum GGT would be associated with elevated risk of new-onset metabolic syndrome, incident CVD, and all-cause mortality after accounting for established and novel cardiovascular risk factors. We postulated that GGT would predict CVD risk even after adjusting for vascular risk factors as time-dependent variables during follow-up.

Methods

Study Participants

The Framingham Offspring Study began in 1971 with the enrollment of 5124 offspring of the original cohort participants (and their spouses).²⁵ The second examination cycle (1978–1982), was attended by 3853 offspring participants (1864 men, 1989 women). Of these, 402 were excluded for the following reasons: missing GGT data ($n=234$, 6%), prevalent CVD ($n=151$, 4%), and missing covariate data ($n=17$, 0.4%). Prior CVD was defined as presence of coronary heart disease (myocardial infarction, coronary insufficiency, angina pectoris), peripheral vascular disease (intermittent claudication), cerebrovascular disease (stroke or transient ischemic attack), or heart failure.²⁶ At each quadrennial Heart Study examination, participants underwent standardized measurements of blood pressure (BP), anthropometry, medical history, physical examination, and laboratory assessment of cardiovascular risk factors. All participants provided written informed consent and the study protocol was approved by the Institutional Review Board of the Boston Medical Center.

Measurements and Definitions

Systolic and diastolic BP were the average of two physician-obtained measurements performed after participants had rested at least 5 minutes in a sitting position, using a mercury sphygmomanometer. Hypertension was defined as a systolic BP ≥ 140 mm Hg or a diastolic BP ≥ 90 mm Hg, or the use of antihypertensive medication. Current smoking was self-reported and was defined as having smoked cigarettes regularly within the prior year. Alcohol consumption was defined based on self-reported average weekly intake. Serum triglycerides, total and HDL cholesterol, and blood glucose were measured after an overnight fast. Diabetes was defined by fasting blood glucose ≥ 126 mg/dL or the use of oral hypoglycemic agents or insulin.

Participants underwent phlebotomy after an overnight fast (between 10 to 12 hours), typically between 7.30 AM and 9 AM. Blood was immediately centrifuged, and plasma and serum specimens were stored at -20°C until assayed. Uniform measurement of GGT activity in serum was performed using spectrophotometry by detecting the liberation of *p*-nitroaniline at 405 nm, resulting from the reaction of γ -glutamyl-*p*-nitroanilide + glycylglycine (Quest Diagnostics [MedPath]).²⁷ High-sensitivity C-reactive protein (CRP) was measured with a Dade Behring BN100 nephelometer from specimens also obtained at the second offspring examination cycle. The average intra-assay coefficient of variation for CRP was 3.8%.

Cross-Sectional Correlates of GGT

We evaluated the association of baseline serum GGT with CVD risk factors and clinical covariates including age, sex, systolic and diastolic BP, hypertension, LDL and HDL cholesterol, serum triglycerides, fasting blood glucose, diabetes, body mass index (BMI), smoking status, and alcohol consumption. We compared serum GGT levels according to presence of metabolic syndrome at baseline, using modified National Cholesterol Education Program (NCEP) criteria, which required at least three of: (1) elevated triglycerides, ≥ 150 mg/dL; (2) HDL cholesterol < 40 mg/dL [men] or < 50 mg/dL [women]; (3) BP ≥ 130 mm Hg systolic, ≥ 85 mm Hg diastolic, or on antihypertensive therapy; (4) fasting blood glucose ≥ 100 mg/dL;

and (5) BMI ≥ 30 kg/m².²⁸ We substituted BMI for increased waist circumference because measurements of waist were not obtained at baseline examination.

Prospective Follow-Up for Incident Events

Participants were followed prospectively for development of metabolic syndrome, incident CVD (fatal or non-fatal coronary heart disease, peripheral vascular disease, cerebrovascular disease, or heart failure), and death over a maximum follow-up duration of 20 years. All CVD events and deaths were systematically reviewed by a three-investigator panel after evaluating all available office and hospitalization records, laboratory test results, death certificates, and autopsy reports.

Statistical Analysis

Cross-Sectional Correlates of GGT

The distribution of GGT values was right-skewed, therefore a natural log-transformation was applied. To account for an upward shift in log-GGT in men relative to women, we standardized the distribution (mean=0, SD=1) within each sex. The distributions of serum triglyceride and alcohol consumption were skewed, and were also log-transformed. Cross-sectional correlates of GGT were identified using sex-pooled multiple linear regression analysis. Each potential correlate was examined separately in age/sex-adjusted models. Variables that were statistically significant at $\alpha=0.05$ in these models were evaluated in multivariable analysis with forward stepwise selection; covariates significant at $\alpha=0.15$ were retained.

Longitudinal Analysis of GGT and Clinical Events

We used Cox proportional hazards regression to examine the association of baseline GGT with: (1) metabolic syndrome, (2) incident CVD, and (3) all-cause mortality, over 20 year follow-up. We constructed Cox models for pooled sexes because formal tests of interaction (sex \times log-GGT) were not statistically significant for any outcome. Initially, we determined the risk associated with a one-standard deviation increment in standardized log-transformed GGT. Cutpoints for sex-specific quartiles were defined based on the GGT distribution of all participants at baseline (prior to exclusions). We compared the risk of events in quartiles 2 to 4 relative to the lowest quartile, and also tested for linear trend across quartiles.

For new-onset metabolic syndrome, the primary analysis examined events over the entire study duration (20 years), after excluding participants with metabolic syndrome at baseline. We also examined the risk of metabolic syndrome according to GGT during short-term follow-up (8-years). New-onset metabolic syndrome was defined by presence of the modified NCEP diagnostic criteria at any subsequent quadriennial examination.²⁹ Because ascertainment of metabolic syndrome required attendance at Heart Study examinations (whereas CVD or death are ascertained irrespective of Heart Study visits), we terminated follow-up at the last examination date if >2 consecutive examination cycles were unattended. Cox models were adjusted initially for factors unrelated to the metabolic syndrome definition: age, sex, alcohol consumption, and log-CRP. In secondary analysis, we evaluated whether GGT predicted new-onset metabolic syndrome after additional adjustment for BMI, fasting blood glucose, systolic and diastolic BP, serum triglycerides, and smoking.

For analyses relating GGT to risk of incident CVD and death, we constructed age/sex-adjusted cumulative incidence curves to illustrate risk across GGT quartiles. Cox models estimating risk of incident CVD and mortality were adjusted for age, sex, BMI, diabetes, systolic BP, antihypertensive treatment, total/HDL cholesterol ratio, current smoking, and alcohol consumption at baseline. Additionally, we adjusted for serum creatinine concentration and education level (postsecondary versus non) as an indicator of socioeconomic status in CVD models. Furthermore, we adjusted for aspartate and alanine aminotransferases (AST, ALT), because reports have linked these enzymes to CVD and metabolic syndrome.^{30,31} Additionally, we adjusted for: (1) baseline CRP; (2) baseline CRP and all other covariates modeled as time-dependent variables (updated at each subsequent quadriennial Framingham

TABLE 1. Baseline Participant Characteristics by Sex-Specific GGT Quartile

Sex-Specific Serum GGT Level (units/liter)	Total Sample	Q1 Men	Q2 Men	Q3 Men	Q4 Men
		1–11 Women 1–6	12–16 Women 7–9	17–24 Women 10–13	25–99 Women 14–88
Age, years (SD) [†]	44 (10)	42 (10)	42 (10)	45 (10)	46 (9)
Women, n (%)	1790 (52)	356 (44)	546 (57)	421 (53)	467 (53)
Body mass index (kg/m ²) [†]					
Mean, SD	25.6 (4.3)	24.6 (3.5)	25.0 (4.0)	25.7 (4.4)	27.0 (5.0)
<25, n (%)	1721 (50)	484 (60)	520 (54)	381 (48)	336 (38)
25–29, n (%)	1250 (36)	257 (32)	339 (35)	313 (39)	341 (38)
≥30, n (%)	480 (14)	63 (8)	100 (11)	107 (13)	210 (24)
Alcohol consumption, n (%) ^{*†}					
None	811 (24)	213 (27)	235 (24)	197 (25)	166 (19)
≤14/wk (M), ≤7/wk (F)	1738 (50)	476 (59)	506 (53)	405 (50)	351 (39)
>14/wk (M), >7/wk (F)	902 (26)	115 (14)	218 (23)	199 (25)	370 (42)
Systolic BP, mm Hg (SD) [†]	122 (16)	118 (14)	119 (15)	123 (17)	127 (17)
Diastolic BP, mm Hg (SD) [†]	78 (10)	75 (8)	76 (10)	79 (10)	81 (9)
Hypertension, n (%) [†]	602 (17)	78 (10)	124 (13)	165 (21)	235 (26)
Treated hypertension, n (%) [†]	327 (9)	31 (4)	60 (6)	71 (9)	165 (19)
Lipid levels (SD)					
Total cholesterol, mg/dL [†]	203 (38)	191 (36)	198 (36)	207 (39)	216 (38)
HDL cholesterol, mg/dL	49 (13)	49 (13)	49 (13)	48 (14)	48 (14)
Total/HDL ratio [†]	4.5 (1.6)	4.2 (1.3)	4.3 (1.4)	4.7 (1.7)	4.9 (1.7)
LDL cholesterol, mg/dL [†]	130 (35)	122 (33)	127 (34)	134 (37)	137 (35)
Triglycerides (serum), mg/dL ^{*†}	105 (80)	81 (51)	91 (60)	108 (73)	139 (109)
Aspartate aminotransferase (AST), IU/L (SD)	21.1 (11.7)	18.1 (7.8)	19.1 (9.6)	20.8 (11.2)	26.3 (15.0)
Alanine aminotransferase (ALT), IU/L (SD)	25.6 (17.7)	19.4 (8.7)	21.7 (13.3)	25.5 (13.4)	35.7 (25.3)
Serum creatinine concentration, mg/dL (SD)	1.16 (0.24)	1.15 (0.23)	1.15 (0.23)	1.18 (0.24)	1.16 (0.24)
Fasting blood glucose, mg/dL (SD) [†]	98 (19)	96 (16)	96 (14)	99 (17)	103 (25)
Diabetes, n (%) [†]	121 (4)	18 (2)	20 (2)	30 (4)	53 (6)
Current smoker, n (%) [†]	1254 (36)	252 (31)	349 (36)	287 (36)	366 (41)
CRP, mg/L, median (IQR) ^{*†}	1.02 (2.06)	0.65 (1.24)	0.82 (1.67)	1.11 (2.21)	1.79 (3.02)

IQR=Interquartile Range; * tests for trend across quartiles performed using log-transformed values; [†]P for quartile trend <0.001

examination attended). We examined the discrimination of models that included clinical covariates and log-GGT with and without log-CRP to determine the incremental value of the latter after accounting for GGT, using the c-statistic. In Cox models, we confirmed that the assumption of proportionality of hazards was met.³² Statistical analyses were performed using SAS version 8.2 (Cary, NC) and a two-sided probability value ≤0.05 was considered statistically significant.

Results

Cross-Sectional Correlates of GGT

Participants in higher GGT quartiles were older, had higher BMI, and were more likely to have hypertension, and elevated lipids, fasting blood glucose, and CRP (Table 1; $P<0.001$ for quartile trend). In the highest quartile, 81.4% of men and 86.9% of women had GGT values within the normal reference range (eg, men ≤50 U/L, women ≤40 U/L). Cross-sectionally, presence of the metabolic syndrome was associated with higher GGT in men (24.9 ± 15.3 versus 18.9 ± 14.7 U/L; $P<0.001$) and women (19.8 ± 15.0 versus 11.4 ± 9.2 U/L; $P<0.001$). In stepwise multiple regression models (see the supplemental materials, avail-

able online at <http://atvb.ahajournals.org>), log-GGT was positively associated with age ($P=0.009$), male sex, smoking, BMI, LDL cholesterol, serum triglycerides, alcohol consumption, diastolic BP, hypertension treatment ($P\leq 0.001$ for all), and fasting blood glucose ($P=0.004$). The above factors explained 33% of the interindividual variability in GGT; sex, serum triglycerides, and alcohol consumption were principal correlates explaining a large degree of variation. There was weak positive correlation of log-GGT with log-CRP (Pearson's $r=0.27$, $P<0.001$), which was of consistent magnitude in men ($r=0.26$) and women ($r=0.27$).

Serum GGT and Incidence of the Metabolic Syndrome

On follow-up, 419 participants (16%, 192 women) developed metabolic syndrome at 8 years, and 968 individuals (37%, 479 women) developed metabolic syndrome over a 20-year period. In multivariable Cox models adjusted for age, sex, alcohol consumption, and CRP, higher GGT was associated with greater risk of developing the metabolic syndrome with a 134% (8-year) to 76% (20-year) increased risk in the top

TABLE 2. Hazards Ratios for Metabolic Syndrome Onset According to GGT Levels

	Log-GGT, 1-SD Increment	Quartile 1	Quartile 2	Quartile 3	Quartile 4	Quartile Trend
Onset of Metabolic Syndrome within 8 years						
Age/sex-adjusted	1.39* (1.26–1.52)	Referent	1.40 [†] (1.04–1.87)	1.76* (1.30–2.39)	2.26* (1.69–3.01)	1.30* (1.19–1.42)
Adjusted for age, sex, and alcohol	1.45* (1.32–1.60)	Referent	1.46 [†] (1.08–1.96)	1.83* (1.35–2.48)	2.54* (1.89–3.41)	1.35* (1.23–1.48)
Additional adjustment for CRP	1.38* (1.25–1.53)	Referent	1.51 [‡] (1.11–2.06)	1.64 [‡] (1.19–2.27)	2.34* (1.72–3.19)	1.30* (1.18–1.43)
Onset of Metabolic Syndrome within 20 years						
Age/sex-adjusted	1.29* (1.21–1.38)	Referent	1.21 [†] (1.01–1.44)	1.49* (1.23–1.80)	1.85* (1.54–2.22)	1.23* (1.16–1.30)
Adjusted for age, sex, and alcohol	1.33* (1.24–1.42)	Referent	1.23 [†] (1.03–1.48)	1.53* (1.26–1.85)	1.99* (1.65–2.40)	1.26* (1.19–1.33)
Additional adjustment for CRP	1.26* (1.18–1.35)	Referent	1.23 [†] (1.02–1.49)	1.36 [‡] (1.11–1.66)	1.76* (1.45–2.13)	1.20* (1.12–1.27)

Value of 1-SD log-GGT=0.6
[†] $P < 0.05$, [‡] $P \leq 0.01$, * $P \leq 0.001$

quartile relative to the lowest (Table 2). In models evaluating log-GGT, a 1-SD increment in log-CRP was associated with a 1.38-fold (95%CI; 1.25 to 1.53, $P < 0.001$) and 1.26-fold (95%CI; 1.18 to 1.35, $P < 0.001$) risk of metabolic syndrome at 8 and 20 years, respectively. The association of GGT with new-onset metabolic syndrome remained robust in models adjusted for serum AST and ALT (data not shown).

Adjusting for age, sex, BMI, fasting glucose, systolic BP, diastolic BP, log-triglycerides, alcohol consumption, smoking status, and log-CRP, the association of GGT with metabolic syndrome remained significant. The hazards ratios (HR) per increment in GGT quartile were 1.14 (95%CI; 1.04 to 1.26, $P < 0.01$) and 1.09 (95%CI; 1.02 to 1.16, $P < 0.01$) in Cox models with 8-year and 20-year follow-up, respectively.

Serum GGT and CVD and Mortality Risk

A total of 65 900 person-years of observation was available in 3451 participants for incident CVD and death. On follow-up (mean 19.1±3.0 years), 535 participants (15.5%; 173 women) experienced incident CVD, and 362 individuals died (10.5%; 131 women). Age/sex-adjusted cumulative incidence of CVD and death (Figures 1 and 2) displayed an increasing gradient of risk across GGT quartiles (log-rank $P < 0.001$ for both outcomes).

In multivariable models adjusting for established risk factors and CRP, log-GGT was positively related to CVD incidence and a graded increase in CVD risk was observed across GGT quartiles (Table 3). The association of GGT with CVD was maintained in models incorporating CVD risk factors as time-varying covariates (Table 3). A 1-SD increment in log-CRP was associated with a 1.20-fold (95%CI; 1.08 to 1.33, $P < 0.001$) risk of incident CVD. After additionally adjusting for serum creatinine concentration and education level, a 1-SD increment in GGT was still associated with a 15% increase in CVD (HR 1.15, 95%CI; 1.05 to 1.27, $P = 0.004$). Those in the highest GGT quartile had a 1.66-fold risk (95%CI; 1.22 to 2.26, $P = 0.001$), and a significant trend was present across quartiles ($P < 0.001$).

In multivariable analyses of mortality, the risk increased across GGT quartiles, remaining robust even after adjustment for log-CRP, and risk factors modeled as time-varying covariates (Table 4). Accounting for log-CRP and all other risk factors as time-varying covariates, a 1-SD increment in log-GGT was associated with a 26% increased risk of death. A 1-SD increment in log-CRP was associated with a 1.31-fold (95%CI; 1.16 to 1.47, $P < 0.001$) risk in the latter models. The associations of GGT with incident CVD and death were maintained after adjustment for serum AST and ALT (data not shown).

Adjusting for clinical covariates (eg, age, sex, BMI, diabetes, systolic BP, total/HDL cholesterol ratio, current smoking, alcohol consumption) and log-GGT, the c-statistic for CVD risk was 0.785 (95%CI; 0.766 to 0.804). When log-CRP was added, the c-statistic increased minimally to 0.786 (95%CI; 0.767 to 0.805). Similarly, the model for mortality

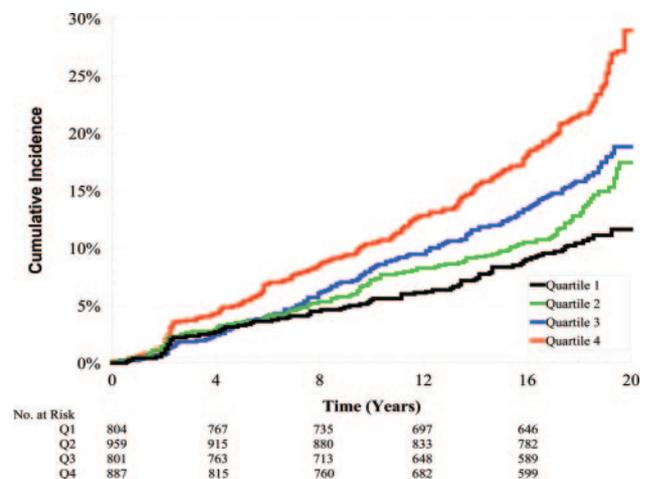


Figure 1. Age/sex-adjusted cumulative incidence of CVD by GGT quartile. Numbers at risk are not the same in each quartile because cut points were determined on all participants with available GGT data (before exclusions).

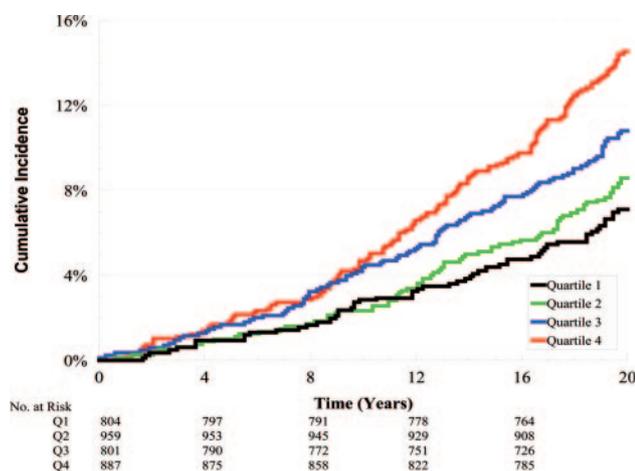


Figure 2. Age/sex-adjusted cumulative incidence of mortality by GGT quartile. Numbers at risk are not the same in each quartile because cut points were determined on all participants with available GGT data (before exclusions).

including clinical covariates and log-GGT had a c-statistic of 0.799 (95%CI; 0.778 to 0.821), and addition of log-CRP increased it minimally to 0.802 (95%CI; 0.780 to 0.823). There was no significant interaction between GGT and CRP for CVD or mortality prediction.

Discussion

Principal Findings

The principal findings of our investigations are three-fold. First, serum GGT levels were related cross-sectionally to CVD risk

factors, notably increased age, male sex, dyslipidemia, BMI, glycemia, BP, and smoking. Second, higher serum GGT was associated prospectively with increased incidence of the metabolic syndrome, above and beyond conventional risk factors including CRP. Third, serum GGT was positively associated with incident CVD and death, after accounting for CRP and hepatic enzymes. Because GGT was associated with the metabolic syndrome prospectively, we adjusted for established CVD risk factors as time-varying covariates, and the association of GGT with CVD and mortality remained, suggesting that GGT risk occurs by mechanisms other than promotion/development of known risk factors. Overall, our data suggest that serum GGT predicts development of the CVD risk factor cluster that constitutes the metabolic syndrome, CVD events, and mortality.

Comparison With Prior Research

Prior studies suggested that higher GGT levels predicted all-cause mortality in patients with myocardial infarction or coronary artery disease,^{20,21} and in middle-aged individuals free of preexisting coronary disease.^{16,24} Prior studies were limited by use of death certificate diagnoses of coronary heart disease, and none addressed whether GGT predicted vascular risk via promotion of established risk factors.¹⁶ Our observations relating GGT to fatal and nonfatal incident CVD events in a community-based sample complement prior studies reporting that higher GGT is associated with cardiovascular death.³³ We expand on prior work by demonstrating that GGT is associated with incident CVD even after accounting for baseline CRP, and risk factors modeled as time-varying covariates.

TABLE 3. CVD Event Rates and Hazards Ratios According to GGT Levels

	Log-GGT, 1-SD Increment	Quartile 1	Quartile 2	Quartile 3	Quartile 4	Quartile Trend
% With Incident CVD						
Events	NA	10.5%	12.1%	16.7%	23.8%	NA
Age/sex-adjusted rates/1000 person-years	NA	6.8	8.6	10.6	14.6	NA
Hazard Ratios (95% CI)						
Models Adjusting for Conventional Risk Factors at Baseline[§]						
Age/sex-adjusted	1.28* (1.18–1.38)	Referent	1.22 (0.92–1.62)	1.53 [‡] (1.16–2.01)	2.11* (1.63–2.74)	1.29* (1.19–1.40)
Multivariable-adjusted	1.14 [‡] (1.04–1.24)	Referent	1.16 (0.87–1.54)	1.30 (0.98–1.72)	1.61* (1.22–2.11)	1.17* (1.08–1.28)
Models Adjusting for Conventional Risk Factors and CRP at Baseline[§]						
Age/sex- and CRP-adjusted	1.20* (1.10–1.31)	Referent	1.28 (0.96–1.72)	1.53 [‡] (1.15–2.04)	1.88* (1.43–2.48)	1.23* (1.13–1.33)
Adjusted for multiple variables+CRP	1.11 [†] (1.02–1.22)	Referent	1.23 (0.92–1.65)	1.35 [†] (1.01–1.81)	1.61 [‡] (1.20–2.14)	1.16 [‡] (1.06–1.27)
Models Adjusting for Conventional Risk Factors as Time-Varying Covariates and CRP at Baseline[§]						
Multivariable-adjusted	1.16* (1.07–1.26)	Referent	1.19 (0.89–1.58)	1.38 [†] (1.05–1.82)	1.69* (1.29–2.21)	1.19* (1.10–1.29)
Adjusted for multiple variables+CRP	1.13 [‡] (1.03–1.24)	Referent	1.26 (0.94–1.68)	1.40 [†] (1.05–1.88)	1.67* (1.25–2.22)	1.18* (1.08–1.28)

Value of 1-SD log-GGT=0.6; NA= not applicable; [†]P<0.05, [‡]P≤0.01, *P≤0.001

[§]Adjusted for age, sex, BMI, diabetes, systolic BP, total/HDL cholesterol ratio, current smoking, and alcohol consumption

TABLE 4. Mortality Rates and Hazards Ratios According to GGT Levels

	Log-GGT, 1-SD Increment	Quartile 1	Quartile 2	Quartile 3	Quartile 4	Quartile Trend
% Mortality						
Events	NA	6.3%	7.4%	12.2%	16.1%	NA
Age/sex-adjusted rates/1000 person-years	NA	3.9	4.6	6.1	8.1	NA
Hazard Ratios (95% CI)						
Models Adjusting for Conventional Risk Factors at Baseline [§]						
Age/sex-adjusted	1.32* (1.20–1.46)	Referent	1.25 (0.87–1.79)	1.70 [‡] (1.21–2.39)	2.21* (1.60–3.05)	1.31* (1.19–1.45)
Multivariable-adjusted	1.25* (1.13–1.38)	Referent	1.21 (0.84–1.74)	1.62 [‡] (1.14–2.29)	1.94* (1.38–2.73)	1.26* (1.13–1.39)
Models Adjusting for Conventional Risk Factors and CRP at Baseline [§]						
Age/sex- and CRP-adjusted	1.27* (1.15–1.40)	Referent	1.20 (0.83–1.74)	1.65 [‡] (1.17–2.34)	1.94* (1.39–2.72)	1.26* (1.14–1.39)
Adjusted for multiple variables+CRP	1.23* (1.10–1.37)	Referent	1.17 (0.81–1.71)	1.61 [‡] (1.13–2.29)	1.83* (1.29–2.60)	1.23* (1.11–1.37)
Models Adjusting for Conventional Risk Factors as Time-Varying Covariates and CRP at Baseline [§]						
Multivariable-adjusted	1.30* (1.17–1.44)	Referent	1.25 (0.87–1.79)	1.73 [‡] (1.23–2.44)	2.16* (1.54–3.02)	1.30* (1.18–1.44)
Adjusted for multiple variables+CRP	1.26* (1.13–1.40)	Referent	1.21 (0.83–1.75)	1.67 [‡] (1.18–2.37)	1.95* (1.38–2.76)	1.26* (1.13–1.40)

Value of 1-SD log-GGT=0.6; NA= not applicable; [†] $P<0.05$, [‡] $P\leq 0.01$, * $P\leq 0.001$

[§]Adjusted for age, sex, BMI, diabetes, systolic BP, total/HDL cholesterol ratio, current smoking, and alcohol consumption

Although GGT was weakly correlated with CRP in our sample and in prior studies,³⁴ CRP did not abrogate the predictive value of GGT for clinical events. First, adjustment for CRP did not attenuate the association of GGT with CVD or mortality. Second, there was minimal additional effect on model discrimination when CRP was added to a model comprised of clinical covariates and GGT. Finally, there was no statistical interaction between GGT and CRP. Our findings suggest that GGT, a routinely-available metabolic marker and indicator of oxidative stress, is a significant predictor of CVD and mortality events independent of CRP. Our findings suggest that GGT will be an important component of future biomarker and multimarker approaches to cardiovascular risk evaluation.

Potential Mechanisms of GGT Effect

Mechanisms that explain the contribution of GGT to CVD and mortality have not been fully elucidated. GGT is associated with hepatic steatosis³⁵ and insulin resistance,^{22,23} and is a predictor of incident hypertension³⁶ and diabetes.^{13,37} Although we observed that the relations of GGT to cardiovascular events and death remained robust after accounting for fasting glucose and components of the metabolic syndrome, it is conceivable that such adjustment incompletely accounts for hepatic insulin resistance and/or steatosis.³⁸ The activity of ectoenzymatic GGT may also modulate the redox status of protein thiols at the cell surface, leading to production of reactive oxygen species and membrane-permeable hydrogen peroxide.³⁹ As noted previously, GGT contributes to oxidative stress pathways in several organ systems, localizes to atheromatous plaques containing oxidized LDL, and is proinflammatory, further implicating this protein in atherogenesis.^{34,40,41}

Strengths and Limitations

The strengths of our investigation are its prospective design, consistent definition and validation of CVD events, complete longitudinal ascertainment of deaths, accounting for risk factors as time-varying covariates, and adjustment for CRP. The biological plausibility that GGT mediates vascular risk is reflected by the strength of the associations, temporal relations between baseline GGT and future vascular risk, and consistency of the results across several analyses. Several limitations of our approach merit comment. Establishing that GGT is a “risk factor” for CVD would require additional mechanistic studies that further assess systemic oxidative stress, and evaluate hepatic steatosis and insulin resistance. We did not obtain repeated GGT or CRP measurements but used baseline values, which is a potential limitation because changes could occur over time.²³ Also, we did not extend this study to other emerging biomarkers of vascular risk. Nonetheless, GGT assays are widely available analytes which are routinely measurable in clinical laboratories. Lastly, the overwhelming majority of our sample was white, limiting the generalizability to other ethnicities.

Conclusions

In our community-based sample, higher GGT levels predicted CVD, mortality, and development of the metabolic syndrome. The association of GGT with adverse cardiovascular outcomes and death was robust after adjustment for traditional cardiac risk factors and CRP. Our study suggests that further investigation of GGT will provide insights into the pathogenesis of CVD and better define the clinical utility of this marker.

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Disclosures

None.

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Online Table I. Multiple stepwise regression of GGT on clinical covariates

Variable	β Estimate	ΔR^2	p-value
Triglyceride levels*	0.2318	0.1555	<0.001
Male	0.3019	0.0847	<0.001
Alcohol consumption†	0.1061	0.0401	<0.001
Diastolic blood pressure	0.0057	0.0195	<0.001
BMI	0.0150	0.0082	<0.001
Hypertension treatment	0.1658	0.0060	<0.001
Age	0.0027	0.0036	0.009
Current smoking	0.0354	0.0021	<0.001
LDL cholesterol	0.0010	0.0018	<0.001
Fasting glucose	0.0015	0.0016	0.004

* Log-transformed triglyceride levels

† Log-transformed weekly alcohol consumption