Serum γ-Glutamyltransferase: Independent Predictor of Risk of Diabetes, Hypertension, Metabolic Syndrome, and Coronary Disease

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Serum γ-glutamyltransferase (GGT) is associated with oxidative stress and hepatic steatosis. The extent to which its value in determining incident cardiometabolic risk (coronary heart disease (CHD), metabolic syndrome (MetS), hypertension and type 2 diabetes) is independent of obesity needs to be further explored in ethnicities. After appropriate exclusions, a cohort of 1,667 adults of a general population (age 52 ±11 years) was evaluated prospectively at 4 year's follow-up using partly Cox proportional hazard regressions. GGT activity was measured kinetically, and values were log-transformed for analyses. MetS was identified by Adult Treatment Panel-III criteria modified for male abdominal obesity. Median (interquartile range) GGT activity was 24.9 (17.0; 35.05) U/l in men, 17.0 (12.3; 24.0) U/l in women. In linear regression analysis, while smoking status was not associated, (male) sex, sex-dependent age, alcohol usage, BMI, fasting triglycerides and C-reactive protein (CRP) were significant independent determinants of circulating GGT. Each 1-s.d. increment in (= 0.53 ln GGT) GGT activity significantly predicted in each sex incident hypertension (hazard ratio (HR) 1.20 (95% confidence interval (CI) 1.10; 1.31)), and similarly MetS, after adjustment for age, alcohol usage, smoking status, BMI and menopause. Strongest independent association existed with diabetes (HR 1.3 (95% CI 1.1; 1.5)) whereas GGT activity tended to marginally predict CHD independent of total bilirubin but not of BMI. Higher serum total bilirubin levels were protective against CHD risk in women. We conclude that elevated serum GGT confers, additively to BMI, risk of hypertension, MetS, and type 2 diabetes but only mediates adiposity against CHD risk.

Obesity (2012) **20,** 842–848. doi:10.1038/oby.2011.136

INTRODUCTION

Elevation in serum γ-glutamyltransferase (GGT) activity, previously ascribed to alcohol intake or liver disease, has been shown to predict morbidity and mortality independent of these (1,2). Modest increases within normal range may be an early marker of cellular oxidative stress (3) and explain the strong associations of serum GGT with many cardiovascular risk factors and disease. Oxidative stress, assessed by circulating prostaglandin F2α levels, is recognized to be related to obesity (4). Indeed, BMI was observed to be a determinant of GGT concentrations in both genders in the Tromsö study (5). Increases in GGT activity have been found to predict hypertension (6,7), as well as incident cases of type 2 diabetes (8–10). GGT activity also predicted all-cause and coronary heart disease (CHD) mortality, independent of alcohol intake or liver disease (1,2). Metabolic syndrome (MetS) also was found to be associated with increased GGT activity in a prospective study of Japanese men (10), in the Framingham Study (2), and cross-sectionally among Turkish adults (11).

Despite the availability of considerable knowledge in the topic, further information is needed in regard to the magnitudes of risk associated between GGT activity and the individual cardiometabolic disorders, the extent of ethnic variation therein and to what extent the observed associations are dependent on excess adiposity.

Serum bilirubin is recognized to act as an antioxidant; and prospective cohort studies have shown that higher serum bilirubin concentrations are associated with decreased risk for CHD (12,13). In dyslipidemic patients with MetS, mean bilirubin levels decreased progressively with the number of MetS components as mean GGT activity increased (14). Whether an interaction exists between the two stated oxidation-related variables with respect to diabetes or CHD is not defined.

Studies examining the relationship of serum GGT levels with each of the risks of hypertension, MetS, diabetes and CHD in the same population-based cohort might provide clues to the preferential oxidative pathways operative in regard to different

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Received 8 November 2010; accepted 11 April 2011; published online 2 June 2011. doi:10.1038/oby.2011.136

cardiometabolic risks; such information might eventually be used in risk assessment or prevention. Turks have a high prevalence of MetS (15) and enhanced low-grade inflammation (16), hence, are suitable to explore such associations. We, therefore, aimed to examine: (i) the determinants of GGT activity, including sex, age, and inflammatory mediators, (ii) to explore prospectively its associations with the development of individual cardiometabolic disorders independent of adiposity measures, and (iii) to determine whether serum total bilirubin concentrations act as antioxidant against type 2 diabetes or CHD in a population sample representative of middle-aged and elderly Turkish adults.

METHODS AND PROCEDURES

This study sample is formed by the cohort of the Turkish Adult Risk Factor Study, a prospective survey on the prevalence of cardiac disease and risk factors in a representative sample of adults in Turkey carried out biennially since 1990 in 59 communities throughout all geographical regions of the country (17). Partial logistic support was provided by the Turkish Ministry of Health. Written informed consent was obtained from all participants. Data were obtained by history of the past years via a questionnaire, physical examination of the cardiovascular system, and recording of a resting electrocardiogram. During the 2003–04 screening serum GGT concentrations were determined in 1,875 participants, aged 33–84 years. Exclusions (11%) were made as follows: 56 individuals with GGT values over 100 U/l to minimize confounding by unrecognized hepatic disease and heavy usage of alcohol, hormone replacement therapy or use of lipid lowering drugs (64 subjects), type 2 diabetes with or without renal impairment (97 persons). No subjects existed with recent myocardial infarction or known hepatic disease. This left 1,667 nondiabetic persons (821 men and 846 women) for analysis in this study. Control was made for menopause in 409 women (48.4%), for 245 persons with alcohol intake up to one to four times weekly (moderate) in regression analyses.

Measurement of risk factors

Blood pressure was measured with an aneroid sphygmomanometer (Erka, Bad Tölz, Germany) in the sitting position on the right arm, and the mean of two recordings 3-min apart was recorded. Weight was measured in light indoor clothes using scales. Waist circumference was measured—with the subject standing and wearing only underwear, at the level midway between the lower rib margin and the iliac crest. BMI was computed as weight divided by height squared (kg/m^2). In regard to cigarette smoking, never smokers, past smokers, and current smokers formed the categories. Anyone who drank alcoholic drinks between once a month and four times per week was considered as moderate user.

Blood samples were collected in a 11-h or longer fasting state in this study except for 15% of individuals. Samples were spun at 1,000 g for 10 min and shipped on cooled gel packs at 2–5 °C to Istanbul to be stored in deep-freeze at −75 °C, until analyzed at the Yıldız Technical University in the same city. Serum GGT activity was assayed by the kinetic method using Glucana as substrate (Thermo Trace, Noble Park, Australia) with a Hitachi 902 autoanalyzer, normal range being reported as £50 U/l in men, £30 U/l in women. PreciNorm U and PreciPath U universal control sera were used as controls. Inter- and intra-assay coefficient of variation for GGT were 1.2/1.1% and 1.5/1.6%, respectively. Serum concentrations of total cholesterol, fasting triglycerides, glucose, high-density lipoprotein cholesterol (HDL-C plus 2nd generation, directly without precipitation) and total bilirubin were determined by using enzymatic kits from Roche Diagnostics (Mannheim, Germany) with a Hitachi 902 autoanalyzer. Low-density lipoprotein cholesterol values were computed according to the Friedewald formula. Uric acid was determined enzymatically by Infinity kit utilizing modified Trinder method. Serum concentrations of complement C3, high-sensitivity C-reactive protein (CRP), apolipoprotein A-I, and B were measured by Behring kits and nephelometry (Behring Diagnostics, Westwood, MA). Concentrations of insulin were determined by the chemiluminescent immunometric method using Roche kits and Elecsys 1010 immunautoanalyzer (Roche Diagnostics).

Diagnosis of CHD was based on the presence of angina pectoris, of a history of myocardial infarction with or without accompanying Minnesota codes of the electrocardiogram (18), or on a history of myocardial revascularization (in one-quarter of patients). Among women, age >45 years was prerequisite for a definitive diagnosis added to isolated typical angina. Electrocardiogram changes of "ischemic type" of greater than minor degree (codes 1.1-2, 4.1-2, 5.1-2, 7.1) were considered as myocardial infarct sequelae or myocardial ischemia, respectively.

Hypertension was defined as a blood pressure ³140 mm Hg and/or ³90 mm Hg, and/or use of antihypertensive medication. Metabolic syndrome was identified when three out of the five criteria of the National Cholesterol Education Program (Adult Treatment Panel-III) (19) were met, modified for prediabetes (fasting glucose ³100 mg/dl (>5.55 mmol/) (20) and further for male abdominal obesity using cutoff of ³95 cm, as assessed in the Turkish Adult Risk Factor study (21). Fasting triglyceride values of the previous participation in the survey were taken into account in few instances of missing data. Diabetes was diagnosed with the criteria of the American Diabetes Association (20), namely by self-report or when plasma fasting glucose was ³7.0 mmol/l or when 2-h postprandial glucose was >11.1 mmol/l.

Data analysis

Descriptive parameters were shown as mean ± s.d. or in %. Due to skewed distribution, values derived from log-transformed (geometric) means were used for GGT, CRP, and insulin. Pearson correlation was used for continuous variables, and Spearman correlation for log-transformed and categorical variables. GGT was dichotomized by the near-median ≥25 U/l in men, and ≥17 U/l in women to evaluate the distribution of baseline characteristics in participants. Quartiles were formed of sex-specific serum total bilirubin, a marker of oxidative stress, using cutoff points of 0.4, 0.585, ≥0.80 mg/dl in men and 0.3, 0.465, ≥0.60 mg/dl in women. Multiple linear regression analyses were performed. Since logarithmic values were used for the dependent variable GGT in linear regression models, the β-coefficient of an independent variable was calculated by log-transforming the obtained values and using the exponents corresponding to the standard deviation. The relative risk for GGT by logistic regression or Cox regression analyses was expressed in terms of 1-s.d. increment which corresponded to ln 0.53. Statistical analyses were performed using SPSS-10 for Windows (Nr. 9026510; SPSS, Chicago, IL). A value of $P < 0.05$ on the two-tail test was considered statistically significant.

RESULTS

In 1,667 nondiabetic subjects at baseline, 101 (6.1%) developed diabetes, and of 1,533 persons free of CHD at baseline, 118 (7.7%) developed CHD over a period of 3.93 (±1.0, range 2–5) years for either outcome. Median (interquartile range) GGT activity at baseline was 24.9 (17.0; 35.05) U/l in 820 men, 17.0 (12.3; 24.0) U/l in 847 women.

Correlates of GGT

High compared with low GGT concentrations significantly distinguished practically all risk factors related to MetS and cardiovascular disease including alcohol usage and CRP, except smoking status, HDL-cholesterol and apo A-I (**Table 1**). Gender was similar in the two groups ($P = 0.28$).

Table 1 Means (s.d.) of baseline characteristics in the study sample stratified by sex-specific low/high GGT values (*n* = 1,667)

	Low GGT (<25/17 U/l)			High GGT (≥25/17 U/l)			
	n	Mean	s.d.	*n*	Mean	s.d.	*P* value
Age, years	857	51.7	11.7	810	52.4	9.8	0.17
GGT,[a] mIU/l	857	13.75	1.42	810	32.3	1.5	<0.001
Total bilirubin, mg/dl	623	0.59	0.37	588	0.56	0.3	0.067
Waist circumference, cm	842	91.5	12	797	97.7	10.6	<0.001
BMI, kg/m^2	842	28.1	5.1	797	30.3	5.0	<0.001
Systolic BP, mm Hg	843	122.9	20.9	802	127.5	20.2	<0.001
Diastolic BP, mm Hg	843	78.6	11.4	801	81.5	10.6	<0.001
Fasting glucose, mmol/l	763	99.3	44.7	733	103	37.7	0.09
Total cholesterol, mmol/l	844	4.84	1.02	806	5.24	1.07	<0.001
Fasting triglycerides, mmol/l	729	1.56	0.87	688	1.99	1.14	<0.001
HDL-cholesterol, mmol/l	844	1.14	0.33	806	1.11	0.31	0.07
Apolipoprotein A-I, g/l	656	1.34	0.29	591	1.37	0.34	0.16
Apolipoprotein B, g/l	648	1.01	0.27	597	1.09	0.30	<0.001
Uric acid, µmol/l	856	296.8	85	809	334.3	88.6	<0.001
CRP[a] mg/l	803	1.72	3.26	757	2.63	2.85	<0.001
Complement C3, g/l	588	1.24	0.27	583	1.38	0.26	<0.001
Fasting insulin,[a] mIU/l	606	6.86	1.99	606	9.43	2.02	<0.001
Fibrinogen, g/l	605	3.14	1.02	562	3.24	1.02	0.085
Alcohol usage, *n*, %	857	34	4.0	810	85	10.5	<0.001
Current smoking, *n*, %	857	260	30.3	810	241	29.8	0.39

Some parameters were not measured at baseline in each participant.
BP, blood pressure; CRP, C-reactive protein; GGT, γ-glutamyltransferase; HDL, high-density lipoprotein.
[a]Geometric mean and s.d. values.

Highest correlations of log GGT were observed with fasting triglycerides, waist circumference, alcohol usage, complement C3 and fasting insulin (*r* ranging from 0.2 to 0.3, Table 2). Table 3 depicts a multiple regression analysis for log GGT in a model comprising five independent variables that explained 20% of the variation in GGT values; it showed male sex (values 1.37-fold those in women), usage of moderate alcohol (1.4-fold than in abstainers) and waist circumference (1.16-fold per 1-s.d. increment) to be major determinants of circulating GGT. While smoking was not independently associated, separate analyses in sexes disclosed age to be inversely associated in men but positively in women. Further linear regression analyses for log GGT adjusted for sex, age, alcohol usage, smoking status, and BMI were used with the following additional individual variables that proved to be significantly and independently associated: elevated fasting triglycerides (β-coefficient 1.29 per 1 s.d.), CRP (β-coefficient 1.13) and low-density lipoprotein cholesterol (β-coefficient 1.11).

Prediction of incident cardiometabolic disorders

In multivariable Cox models for incident CHD and diabetes, adjusted for sex, age, and menopause, we first examined whether log GGT and serum total bilirubin quartiles were each of independent predictive value. While GGT was modestly

Table 2 Pearson/Spearman correlations of GGT concentrations (*n* = 1,667)

	n	*r*	*P* value
Waist circumference, cm	1,643	**0.28**	<0.001
BMI, kg/m^2	1,639	**0.14**	<0.001
Systolic BP, mm Hg	1,645	**0.09**	0.001
Diastolic BP, mm Hg	1,645	**0.12**	<0.001
Total cholesterol, mg/dl	1,650	**0.16**	<0.001
Fasting triglycerides, mmol/l	1,417	**0.27**	<0.001
HDL-cholesterol, mmol/l	1,650	**–0.15**	<0.001
Fasting glucose, mmol/l	1,496	**0.11**	<0.001
Uric acid, µmol/l	1,665	**0.06**	0.019
Total bilirubin, mg/dl	1,211	0.04	0.15
Complement C3, g/l	1,171	**0.21**	<0.001
C-reactive protein, mg/l	1,560	**0.19**	<0.001
Fasting insulin[a] mIU/L	1,212	**0.23**	<0.001
Apolipoprotein A-I, g/l	1,247	*–0.05*	0.06
Apolipoprotein B, g/l	1,245	**0.13**	<0.001
Smoking status	1,667	**0.15**	<0.001
Alcohol usage	1,667	**0.21**	<0.001

Some parameters were not measured at baseline in each participant. Significant values are provided in boldface and borderline significant values are italicized.
BP, blood pressure; GGT, γ-glutamyltransferase; HDL, high-density lipoprotein.
[a]Log-transformed values.

Table 3 Linear regression analyses for GGT

	β-coefficient	s.e.	*P* value	β-coefficient	s.e.	*P* value	β-coefficient	s.e.	*P* value
	Total (*n* = 1,641)			Men (*n* = 807)			Women (*n* = 834)		
Constant	10.0	1.15	<0.001	8.65	1.20	<0.001	4.6	1.17	<0.001
Sex, male	**1.37**	1.03	<0.001						
Alcohol intake, yes/no	**1.40**	1.05	<0.001	**1.36**	1.05	<0.001	**1.48**	1.05	0.046
Waist circumference, 11/13 cm[a]	**1.16**	1.00	<0.001	**1.17**	1.03	<0.001	**1.14**	1.00	<0.001
Age, 11 years	1.00	1.002	0.80	**0.94**	1.03	<0.001	**1.08**	1.002	<0.001
Smokers, current vs. never	1.001	1.016	0.42	1.00	1.02	0.84	1.04	1.016	0.15
Variance r^2	0.20; $P < 0.001$			0.15; $P < 0.001$			0.08; $P < 0.001$		

Log-transformed values. Significant values are provided in boldface.
GGT, γ-glutamyltransferase.
[a]Denotes 1-s.d. increment in men/women.

Table 4 Cox regression for prediction of incident CHD and diabetes by serum GGT, adjusted for sex, age and total bilirubin quartiles

	HR	95% CI	HR	95% CI	HR	95% CI
For CHD	Total (*n* = 103/1,125)[a]		Men (*n* = 45/542)[a]		Women (*n* = 58/583)[a]	
Sex, female	0.94	0.42; 2.12				
Age, 11 years[b]	**1.94**	1.59; 2.36	**2.00**	1.49; 2.66	**2.02**	1.27; 3.18
GGT, 1.7-fold[c]	*1.15*	1.00; 1.33	*1.21*	0.97; 1.51	1.12	0.91; 1.36
Bilirubin quartile 2 (0.4–0.58/0.3–0.46)[d]	0.78	0.47; 1.29	0.85	0.38; 1.88	0.72	0.37; 1.41
Bilirubin quartile 3 (0.59–0.79/0.47–0.59)[d]	0.63	0.36; 1.09	0.96	0.44; 2.10	**0.42**	0.19; 0.92
Bilirubin quartile 4 (>0.8/0.6)[d]	0.64	0.37; 1.11	0.65	0.27; 1.57	0.62	0.31; 1.23
Menopause					0.88	0.33; 2.32
For type 2 diabetes	*n* = 85/1,211[a]		*n* = 53/587[a]		*n* = 32/624[a]	
GGT, 1.7-fold[c]	**1.35**	1.16; 1.58	**1.47**	1.20; 1.80	*1.25*	0.96; 1.64
Bilirubin quartile 2 (0.4–0.58/0.3–0.46)[d]	1.05	0.58; 1.90	0.72	0.33; 1.58	1.82	0.68; 4.87
Bilirubin quartile 3 (0.59–0.79/0.47–0.59)[d]	0.91	0.49; 1.68	1.08	0.54; 2.16	0.49	0.12; 1.97
Bilirubin quartile 4 (>0.8/0.6)[d]	1.13	0.63; 2.05	0.82	0.38; 1.77	1.80	0.66; 4.87
Menopause					0.85	0.23; 3.12

Overall samples vary depending on exclusion of the prevalent cases for each disorder at baseline. Age adjusted in both models. Individuals taking hormone replacement therapy or f lipid lowering drugs (64 subjects) had been excluded. Significant values are provided in boldface.
CHD, coronary heart disease; CI, confidence interval; GGT, γ-glutamyltransferase; HR, hazard ratio.
[a]Number of cases/number at risk. [b]Denotes 1-s.d. increment. [c]Log-transformed values ≥25/17 vs. <25/17 U/l expressed in terms of 1 s.d. =70% higher geometric mean values. [d]Referent <0.4/<0.3 mg/dl.

predictive of CHD and fairly strongly of diabetes (Table 4), high bilirubin quartiles did not prove to be inversely and significantly associated with these diseases. Though GGT was not associated with CHD in females, women with total bilirubin ≥0.47 mg/dl (in the two highest quartiles) were associated with protection against CHD alone: relative risk (RR) 0.52 (95% confidence interval (CI) 0.28; 0.95) compared with the lowest quartile. While in men RR was not significant with 0.81, in the total study sample, the two highest quartiles disclosed an RR of 0.64 (95% CI 0.40; 1.006).

When BMI was substituted for bilirubin to evaluate the degree of mediation of the GGT associations by adiposity (Table 5), the introduced BMI attenuated the associations of GGT for CHD in women, contrasted to a modest attenuation in men. The converse was true in regard to diabetes, namely, the associations of GGT were modestly attenuated in men but became significant and somewhat stronger in women.

GGT and incidence of hypertension and metabolic syndrome

Table 6 demonstrates prediction of incident hypertension by log GGT in a logistic regression model adjusted for sex, age, menopause and BMI, all powerful determinants of elevated blood pressure. GGT contributed to the associations with an RR of 1.20 (95% CI 1.10; 1.31) per 1-s.d. increment.

After exclusion of 40% of participants who had MetS at baseline, 332 among 975 persons developed MetS during the follow-up. Serum GGT significantly predicted incident MetS at a modest RR in each sex, after adjustment for age (and menopause in women). Further adjustment for alcohol usage and

Table 5 Cox regression for prediction of incident CHD and diabetes by serum GGT, adjusted for sex, age, BMI and menopause

	HR	95% CI	HR	95% CI	HR	95% CI
For CHD	Total (*n* = 116/1,334)[a]		Men (*n* = 52/647)[a]		Women (*n* = 64/687)[a]	
Sex, female	1.15	0.77; 1.70				
Age, 11 years[b]	**1.88**	1.56; 2.26	**1.94**	1.48; 2.58	**1.78**	1.17; 2.74
GGT, 1.7-fold[c]	1.08	0.94; 1.23	1.16	0.94; 1.42	1.02	0.84; 1.23
BMI, kg/m^2, 4.5/5.5[b]	**1.21**	1.04; 1.42	1.20	0.98; 1.47	1.23	0.98; 1.55
Menopause					1.13	0.47; 2.76
For type 2 diabetes	*n* = 99/1,439[a]		*n* = 59/700[a]		*n* = 40/739[a]	
Sex, female	0.66	0.43; 1.02				
Age, 11 years[b]	1.12	0.90; 1.38	**1.33**	1.00; 1.76	1.02	0.56; 1.86
GGT, 1.7-fold[c]	**1.28**	1.11; 1.48	**1.30**	1.07; 1.57	**1.35**	1.07; 1.71
BMI, kg/m^2, 4.5/5.5[b]	**1.54**	1.34; 1.76	**1.44**	1.24; 1.68	**1.66**	1.26; 2.18
Menopause					0.68	0.23; 2.04

Overall samples vary depending on exclusion of the prevalent cases for each disorder at baseline. Significant values are provided in boldface.
CHD, coronary heart disease; CI, confidence interval; GGT, γ-glutamyltransferase; HR, hazard ratio.
[a]Number of cases/number at risk. [b]Denotes 1-s.d. increment in men/women. [c]Log-transformed values ≥25/17 vs. <25/17 U/l expressed in terms of 1 s.d. =70% higher geometric mean values.

Table 6 Logistic regression for prediction of incident hypertension (HT) and metabolic syndrome (MetS) by serum GGT, adjusted for sex, age, menopause and BMI

	RR	95% CI	RR	95% CI	RR	95% CI
For HT	Total (*n* = 476/1,422)[a]		Men (*n* = 224/735)[a]		Women (*n* = 252/678)[a]	
Sex, female	**1.40**	1.07; 1.84				
Age, 11 years[b]	**2.02**	1.78; 2.31	**1.82**	1.52; 2.17	**1.80**	1.30; 2.48
GGT, 1.7-fold[c]	**1.20**	1.10; 1.31	**1.17**	1.03; 1.34	**1.15**	1.01; 1.31
BMI, kg/m^2, 4.5/5.5[b]	**1.75**	1.53; 1.99	**1.98**	1.63; 2.42	**1.60**	1.32; 1.92
Menopause					**1.85**	1.03; 3.32
For MetS	*n* = 338/987[a]		*n* = 157/486[a]		*n* = 181/501[a]	
Sex, female	1.59	1.19; 2.12				
Age, 11 years[b]	**1.20**	1.05; 1.37	1.01	0.83; 1.23	**1.56**	1.09; 2.22
GGT, 1.7-fold[c]	**1.31**	1.18; 1.45	**1.14**	1.07; 1.22	**1.20**	1.04; 1.39
Menopause					0.86	0.45; 1.66
	n = 332/975[a]		*n* = 153/479[a]		*n* = 179/496[a]	
Sex, female	0.93	0.67; 1.30				
Age, 11 years[b]	**1.24**	1.07; 1.44	1.09	0.88; 1.37	**1.64**	1.14; 2.38
GGT, 1.7-fold[c]	**1.17**	1.04; 1.30	1.13	0.95; 1.33	1.14	0.98; 1.32
BMI, kg/m^2, 4.5/5.5[b]	**2.57**	2.12; 3.11	**3.89**	2.78; 5.42	**2.08**	1.63; 2.65
Alcohol usage, yes/no	0.89	0.50; 1.58	0.85	0.45; 1.61	0.01	NS
Menopause					0.77	0.38; 1.53

Overall samples vary depending on exclusion of the prevalent cases for each disorder at baseline. Significant values are provided in boldface.
CI, confidence interval; GGT, γ-glutamyltransferase; NS, not significant; RR, relative risk.
[a]Number of cases/number at risk. [b]Denotes 1-s.d. increment in men/women. [c]Log-transformed values ≥25/17 vs. <25/17 U/l expressed in terms of 1 s.d. =70% higher geometric mean values.

BMI led to reduction of significance to an RR 1.17 (95% CI 1.04; 1.30) per 1-s.d. increment.

DISCUSSION

In a middle-aged population-based sample prone to MetS, we found that GGT activity was associated with age inversely in men but positively in women and furthermore with fasting triglycerides, CRP and low-density lipoprotein cholesterol, all independent of BMI and lifestyle habits. High bilirubin quartiles were not inversely associated with incident diabetes and only in women with CHD, independent of circulating GGT. In logistic regression models adjusted for sex, age, menopause

and BMI, GGT contributed to the association with incident hypertension with an RR of 1.20, significantly predicted incident MetS at a similar RR which was in part BMI-mediated. In similarly adjusted Cox regression models, GGT activity was most strongly predictive of diabetes independently in each sex (hazard ratio (HR) 1.30 per 1-s.d. increment). Yet introduction of BMI in the models attenuated the modest predictive ability of GGT for incident CHD in women, contrasted to a minor attenuation in men. Thus GGT activity, likely a modest independent CHD risk mediator in males, is part of the proinflammatory state/oxidative stress in Turkish females, for which other evidences had been reported (16,22).

Elevated general levels and independent covariates

General levels of GGT activity in the study population was substantially higher than those reported for other populations. In four large community-based studies (2,8,23,24) median values ranged between 15–20 U/l in men and 9–11 U/l in women, while current values were higher by 1.4-fold and 1.7-fold, respectively. This reflects, in our opinion, the true difference in GGT levels (rather than variation in measurement methodology) given the high prevalence of MetS and diabetes among Turks, particularly women.

Apart from the well-known GGT determinants of male sex and alcohol intake, fasting triglycerides, CRP and sex-dependent age were the major covariates of GGT activity in the present study. A rise concomitant with age was not unexpected in women, yet the significant moderate independent decline of GGT (by 6%) per 1-s.d. increment in age among men might be ascribed to the concomitant decrease in Turkish men (but not women) of both SHBG and total testosterone with age (25). Serum triglycerides and CRP, recognized mediators of low-grade inflammation and oxidative stress, were documented to be significantly associated with GGT independent of BMI and other covariates. The lack of significant association with high or low GGT activity of serum HDL-C, apo A-I and current smoking is not surprising in this population sample, in view of previously documented dysfunction of HDL and apo A-I and lack of current smoking to confer cardiometabolic risk, especially among Turkish women (16).

Serum bilirubin in prediction of CHD risk

In Cox regression analyses jointly with GGT activity, total bilirubin quartiles were not of independent predictive value for the risk of incident diabetes. Regarding incident CHD risk, quartiles 3 and 4 (values >0.46 mg/dl) proved to confer protection in women, but not in men, and marginally in the whole sample. Turkish women rather than men were demonstrated to be more influenced by low-grade inflammation (16) which might explain the observed inverse association of higher bilirubin with decreased CHD risk being confined to females. Latter findings in women are in essential agreement with those reported (12,13).

Independent influence of serum GGT on metabolic risk but BMI-mediation in CHD risk

The magnitude of the association between GGT activity and outcome has previously been expressed variably, either in quantiles (8,24), per doubling of levels or per log unit increase in GGT (26). In agreement with standard expression and in line with the Framingham study (2), we expressed HRs in terms of 1-s.d. increment in GGT. It is to be noted that such expression makes HRs appear lower than previously reported.

We confirmed findings by Lee and associates (2) regarding incident MetS that GGT predicted this development additively to BMI and other confounders. The magnitude of the HR (1.17) was slightly lower than the 1.26 of the Framingham study (2) in which a longer follow-up was available, but relatively high GGT levels in the lowest quartile in this study sample may also be implicated.

The HR in the independent prediction of hypertension, a component of MetS, was of similar magnitude as that for MetS in the current study suggesting that the contribution of GGT to the latter risk was mainly via the component of hypertension. In contrast, type 2 diabetes was linked to GGT more strongly, namely at an HR of 1.3, roughly paralleling the risk magnitude in women of the KORA study (24). This supports our previously expressed view that enhanced proinflammatory state/oxidative stress has an especially prominent role in diabetes among Turks (16,27).

Results on the prediction of CHD by GGT activity have been divergent. Though most studies have reported significant associations, these were of small magnitude, and heterogeneity existed. A meta-analysis of eight studies with respect to incident CHD (26) yielded a "fully-adjusted" pooled HR for 1-s.d. of GGT that corresponded to 1.10 (1.01; 1.20). Greater heterogeneity was notable among women. Our findings showing no independent predictive value of GGT activity for CHD in women and a marginal value in men are closely in line with results of the pooled meta-analysis. Oxidative processes in atherogenesis presumably merely mediate and are not additive to the adiposity-related factors in women; this seems to be distinct from CRP that has additive features and from GGT with respect to metabolic outcomes of diabetes and hypertension in which it contributes additively to adiposity. In the KORA study, the enhanced development of type 2 diabetes among obese subjects required the interaction of high GGT activity in women but not in men (24) suggesting a sex-dependent independent diabetogenic action.

Elevated serum GGT could be involved in cardiometabolic risk either as a marker of hepatic steatosis (28), with or without hepatic insulin resistance (5), and/or as a mediator of oxidative stress via mediation of extracellular glutathione transport into cells of organ systems (9), or as a mediator of low-grade systemic inflammation (29). The documented predictability of all three metabolic disorders of MetS, hypertension, and diabetes by GGT activity suggests that, as a reflection of oxidative stress, elevated GGT levels are actively involved in the pathogenesis of these disorders.

Strengths and limitations

Availability of a cohort of both genders representative of the general population, exclusion of potentially confounding conditions and, especially, the simultaneous assessment of

the diverse cardiometabolic disorders form the strengths of the present study. A study sample prone to MetS and having high GGT levels, while constituting strength in view of lack of similar samples, may limit somewhat the applicability of conclusions to populations having lower prevalence of MetS. Our use of multiple analyses may be a limitation in interpreting the findings, as may be the relatively limited number of events for diabetes and CHD. Although the identification of CHD included soft endpoints beyond myocardial infarction, this may reflect the natural distribution of CHD, especially in women. The relatively short mean follow-up of 4 years may have precluded the obtaining of higher HRs for GGT activity regarding cardiometabolic risk. Nonetheless, findings were in close agreement with mainstream results in prior studies.

Conclusions

In middle-aged adults with a high prevalence of MetS, serum GGT activity is associated not with smoking status but with male sex, alcohol usage, sex-dependent age, fasting triglycerides, CRP and low-density lipoprotein cholesterol, independent of BMI. High bilirubin quartiles did not contribute to the prediction of type 2 diabetes but higher levels were protective of CHD risk in women, independent of circulating GGT. GGT activity modestly predicted hypertension, MetS, and diabetes in each sex, independent of multiple confounders including BMI, whereby the strongest HR existed regarding diabetes risk. GGT is involved in CHD risk mainly by mediating adiposity. Maintenance of normal triglycerides and CRP, regardless of a coexisting obesity, may be relevant in reducing oxidative stress and risk of diabetes.

ACKNOWLEDGMENTS

We thank the pharmaceutical companies AstraZeneca, Novartis, Pfizer, and SanofiAventis, Istanbul, Turkey, which have supported the Turkish Adult Risk Factor survey over the years. We acknowledge the work of G. Hergenç, PhD, in the laboratory measurements at baseline examination and appreciate the dedicated work in data collection of various coworkers in the survey teams.

DISCLOSURE

The authors declared no conflict of interest.

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