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Oxidized LDL, Gamma-glutamyltransferase (GGT) and adverse outcomes in older adults

Belinda Spoto, DrBiol[1], **Francesco Mattace-Raso, MD PhD**[2], **Eric J. Sijbrands, MD PhD**[2], **Graziella D'Arrigo, DrBiostat PhD**[1], **Giovanni Tripepi, DrBiostat PhD**[1], **Stefano Volpato, MD**[3], **Stefania Bandinelli, MD**[4], **Luigi Ferrucci, MD PhD**[5], and **Carmine Zoccali, MD**[1]

[1]National Council of Research and Institute of Clinical Physiology CNR-IFC, Reggio Calabria Unit, Italy [2]Department of Internal Medicine, Erasmus University Medical Center, Rotterdam, The Netherlands [3]Department of Science and Medicine, University of Ferrara, Ferrara, Italy [4]Geriatric Unit, Azienda Sanitaria di Firenze, Florence, Italy [5]Longitudinal Studies Section, National Institute on Aging, Baltimore, Maryland, USA

Abstract

Background/Objectives—Gamma-glutamyltransferase (GGT) is a biomarker of liver disease and oxidative stress which was associated with all-cause and cardiovascular (CV) mortality in the general population and in patients with high risk conditions. This study aims at assessing whether oxLDL modifies the relationship between GGT, all-cause and CV mortality in elderly individuals from the general population.

Design—Observational longitudinal study.

Setting—Population-based cohort of older individuals (>65 years) free of liver disease.

Address all correspondence and offprint requests to: Prof. Carmine Zoccali. National Council of Research and Institute of Clinical Physiology (CNR-IFC), Ospedali Riuniti, 89124 Reggio Calabria, ITALY c/o EUROLINE, Via Vallone Petrara 55-57, 89100 Reggio Calabria, ITALY. carmine.zoccali@tin.it, **FAX**: 0039-0965-26879, **Phone**: 0039-0965-397010, or Dr Belinda Spoto. National Council of Research and Institute of Clinical Physiology (CNR-IFC), Ospedali Riuniti, 89124 Reggio Calabria, ITALY c/o EUROLINE, Via Vallone Petrara 55-57, 89100 Reggio Calabria, ITALY. belinda.spoto@tin.it, **FAX**: 0039-0965-26879, **Phone**: 0039-0965-397028.

CONFLICT OF INTEREST

All the authors declare no conflict of interest.

AUTHOR CONTRIBUTIONS

Belinda Spoto: conceived the study and wrote the paper with Carmine Zoccali, and performed statistical data analysis with Graziella D'Arrigo and Giovanni Tripepi. Francesco Mattace-Raso: critically revised the manuscript and provided significant intellectual contribution.

Eric Sijbrands: critically revised the manuscript and provided significant intellectual contribution.

Graziella D'Arrigo: performed statistical analysis and interpreted the results

Giovanni Tripepi: performed statistical analysis and interpreted the results

Stefano Volpato: critically revised the manuscript, provided significant intellectual contribution and was co-responsible of data handling/collection.

Stefania Bandinelli: critically revised the manuscript, provided significant intellectual contribution and was co-responsible of data handling/collection.

Luigi Ferrucci: supervised data collection and management, critically revised the manuscript and provided significant intellectual contribution.

Carmine Zoccali: conceived the study and wrote the paper with Belinda Spoto. Critically revised the manuscript and provided significant intellectual contribution.

Participants—One thousand and thirty-eight individuals from the Invecchiare in Chianti (InCHIANTI) study.

Measurements—serum GGT level, oxidized low-density lipoprotein (oxLDL), CV comorbidities, all-cause and CV mortality.

Results—The median age of the study population (n=1038) was 74 years (inter-quartile range: 69–79), 152 individuals (15%) had past CV events. During a median follow-up of 9 years, 401 individuals died, 168 of them (42%) for CV causes. In adjusted analyses, GGT predicted all-cause mortality (HR for 20U/L increase in serum GGT: 1.11, 95% CI: 1.02–1.21, P=0.02) and CV mortality HR: 1.17, 95%CI: 1.03–1.33; P=0.02). Furthermore, in an analysis for interaction circulating oxLDL amplified the effect of GGT on all-cause mortality (P=0.003).

Conclusions—Circulating oxLDL amplifies the effect of GGT on mortality in the elderly. The mechanism for this association remains unknown and requires further research, including studying the potential role of GGT in oxidative stress. These results are consistent with the hypothesis of a causal role of GGT in the CV morbidity and mortality in older individuals and indicate that oxLDL plays a crucial role in the interpretation of the link between GGT and the risk of adverse clinical events in this population.

Keywords

GGT; mortality; CV events; oxidized LDL; elderly

INTRODUCTION

Aging is characterized by a progressive decline of anatomic integrity and function across multiple tissues and organs. A number of mechanisms have been proposed to drive the aging process, including accumulation of damaged macromolecules due to oxidative stress (1).

Gamma-glutamyltransferase (GGT), a multifaceted biomarker impinging upon oxidative stress (2,3), emerged as a risk factor for all-cause and cardiovascular (CV) mortality in population-based studies independent of liver disease and alcohol intake (4, 5). GGT has been detected within atherosclerotic plaques of cerebral and coronary arteries where it co-localizes with oxidized low-density lipoprotein (oxLDL) (6, 7). In theory, such a co-localization may be key to the interpretation of oxidative stress damage in the arterial system.

In the sole study in an elderly cohort testing the relationship between GGT and mortality and CV events, this biomarker was a direct predictor of adverse clinical outcomes (8) which contrasts with age-stratified analyses in community-based studies where GGT predicted mortality in the young and middle age strata but not in the elderly (9–12). Herein, we tested the relationship between GGT and all-cause and CV mortality in a population-based cohort of elderly individuals (n=1038) from the Invecchiare in Chianti study, which enrolled a random sample of people older than 65 years living in the Chianti area in Tuscany and followed up them for a median time of 9 years. In light of the pathophysiological relationship between GGT and oxLDL alluded to before (6), a pre-specified goal of the

present study was testing whether oxLDL modifies the relationship between GGT, CV and all-cause mortality.

METHODS

Study population

The elderly InCHIANTI (13) population included 1155 participants aged between 65 and 102 years randomly selected from residents in the Chianti geographic area. The baseline data collection started in September 1998 and lasted until March 2000; thereafter, participants were fully re-evaluated every 3 years and the fourth follow-up is still ongoing.

The present study was performed on 1038 participants out of the 1155 original cohort because we excluded individuals with missing serum GGT (n=112), documented liver disease (n=4) or because of extremely high serum level of GGT (n=1) identified as a statistically significant outlier (GGT= 565 U/L) by Grubbs' test ($P<0.001$).

Alcohol consumption was assessed by self-reported daily intake of wine, beer and spirits. The content of ethyl alcohol was calculated as follows: 5 g ethyl alcohol in 100 mL of beer, 13 g in 100 mL of wine and 50 g in 100 ml of spirits. In agreement with WHO Guidelines, 40 g/day ethyl alcohol for males and 20 g/day for females were taken as a cut-off for identifying heavy drinkers.

Follow-up and incident study outcomes

The primary outcomes were all-cause and CV mortality. After the enrolment, individuals were monitored for a median of 9 years (ranging from 0.15 – 10.5 years). CV deaths were classified following ICD9 diagnosis codes from 410 to 438.

Laboratory measurements

Serum GGT was measured through an enzymatic colorimetric assay using a Roche analyzer (Roche Diagnostics, GmbH, Mannheim, Germany). The minimum detectable threshold was 3 U/L and the measure range was 3–1200 U/L. The intra-assay coefficient of variation (CV) was 1.5% and the inter-assay CV was 1.4%. The normal values considered for GGT were: 10–50 U/L in men and 10–38 U/L in women. Oxidized LDL was measured using an enzyme-linked immunoassay (ELISA) kit (Mercodia AB, Uppsale, Sweden). The intra-assay and the inter-assay CV was 6% and 5%, respectively. High sensitivity C-reactive protein (CRP) was measured by a colorimetric competitive immunoassay that uses purified protein and polyclonal anti-CRP antibodies. The minimum detectable threshold was 0.03 mg/L and the inter-assay CV was 5%. Homocysteine were measured by fluorimetric polarized immunoassay method (IMX, Abbott Laboratories). The minimum detectable threshold was 0.5 μmol/L and the inter-assay CV was 4.3%.

Statistical analysis

Cross-sectional data were analyzed by univariate and multiple linear, logistic and Cox regression analyses. Multiple models included GGT as well as traditional risk factors (age, gender, smoking, diabetes, LDL cholesterol and blood pressure), factors peculiar to liver

disease (transaminases, alkaline phosphatase, alcohol consumption), BMI, hemoglobin, oxidized LDL, C-reactive protein (CRP), homocysteine and creatinine clearance. To account for over-fitting (i.e. when the number of covariates overcame 1 variable every 10 study outcomes) a shrinkage correction was applied to Cox and logistic regression models (14). A backward elimination strategy was applied to logistic regression analysis. Interaction analysis was performed by the standard linear combination method. The proportionality assumption was tested by analyzing the Schoenfeld residuals and no violation was found. The functional form of key-covariates (including interaction term) was investigated by the analysis of Martingale residuals and the use of both risk factor (GGT) and effect modifier (oxLDL) as continuous variables resulted to be the best functional form for capturing the risk of all-cause and CV mortality explained by this biomarker. To assess whether early deaths could affect the study results a sensitivity analysis excluding patients who died within the first year from the enrolment was carried out. Furthermore, to minimize the potential distortion of heavy drinking (> 40 g/day ethyl alcohol for males and 20 g/day for females) a sensitivity analysis excluding heavy drinkers was performed. Data were expressed as odds ratio (OR), hazard ratios (HR), 95% confidence intervals (CI) and P values, as appropriate. All analyses were performed by standard statistical packages (SPSS for Windows Version 9.0.1, 11 Mar-1999, Chicago, Illinois, USA; STATA/SE 9.0 StataCorp LP, TX, USA).

RESULTS

The main demographic and clinical characteristics of the study population are summarized in Table 1. The study cohort included 1038 subjects (43% males) with a median age of 74 years (inter-quartile range: 69–79). Biochemical parameters, including liver enzymes, were in the normal range and the mean value and SD of oxLDL was 42±13 U/L.

Clinical and functional correlates of GGT

Serum levels of GGT had a left skewed distribution with a median value of 19 U/L (inter-quartile range: 14 to 28 U/L). In a multiple linear regression model, including all univariate correlates of GGT (Supplementary Table S1), the independent correlates of this biomarker were ALT, male gender, C reactive protein, alcohol consumption, alkaline phosphatase, hemoglobin, CV comorbidities, oxLDL and homocysteine whereas AST, creatinine clearance, BMI, smoking, age, systolic BP, LDL cholesterol, diabetes and lipid lowering agents were not (P ranging from 0.28 to 0.99) (Supplementary Table S1)

GGT and past CV events

At baseline, 207 past CV events occurred in 152 individuals. In detail, myocardial infarction in 25 cases, coronary surgery/angioplasty in 16 cases, stroke in 36 cases, angina in 34 cases, peripheral vascular disease in 96 cases. In logistic regression analysis, GGT adjusted for age and sex was associated (P=0.03) with past CV events (OR:1.14, 95% CI: 1.01–1.27). In a parsimonious backward logistic regression model adjusting for age, sex, CRP, alcohol consumption, ALT and diabetes the OR of a 20 U/L increase in GGT for the risk of past CV events was 1.23 (95%CI:1.06–1.43; P=0.01). Furthermore, the GGT-CV link was confirmed in a multiple logistic regression model [(OR:1.24, CI 95%:1.07–1.44; P=0.005); (shrinkage

corrected OR:1.19, CI 95%:1.02–1.39; P=0.02)] adjusting for the full list of traditional and non-traditional risk factors considered in this study (Supplementary Table S2).

GGT, all-cause and CV mortality

During the follow-up period (median 9 years, range 0.15– 10.5 years), 401 individuals died, 168 of them (42%) for cardiovascular causes. In an age and sex adjusted Cox regression model, 20U/L increase in serum GGT signaled a parallel 10% increase in the risk of all-cause mortality (HR:1.10, 95% CI:1.03–1.18, P=0.007). In a multiple Cox regression analysis adjusting for the same set of variables applied in the multiple logistic regression model (Supplementary Table S3), GGT was confirmed as an independent risk factor of mortality [HR (20 U/L increase):1.11, 95% CI:1.02–1.21, P=0.02]. Similarly, GGT predicted CV mortality both in age and sex adjusted model (HR:1.12, 95%CI:1.01–1.24; P=0.04) and in an analysis [(HR:1.17, 95%CI:1.03–1.34; P=0.02); shrinkage corrected HR: 1.17, 95%CI: 1.02–1.33; P=0.02)] adjusting for the same set of variables applied in the multiple logistic regression model. In sensitivity analyses (N=937) excluding patients who died for malignancies or within 1 year from the enrolment, the HR of GGT for all-cause [HR (20 U/L increase):1.13, 95%CI:1.02–1.25, P=0.02] and CV death (shrinkage corrected HR: 1.17, 95%CI:1.03–1.34, P=0.02) remained the same.

GGT and clinical outcomes: effect modification by oxidized low-density lipoprotein (LDL)

Because oxLDL and GGT reflect reactive oxygen species burden, we hypothesized that coexistence of high oxLDL and GGT may amplify the risk for all-cause and CV mortality in the elderly population of the InCHIANTI study. Oxidized LDL per sé failed to show a meaningful link with all-cause and CV death (HR:0.99 for both outcomes). However, oxLDL amplified the effect of GGT on all-cause mortality both in age and sex adjusted Cox models (P for the effect modification=0.001) and adjusted analyses (P for interaction=0.003) (Table 2). As shown in Figure 1, the risk excess for all-cause mortality portended by a fixed increase in GGT (20 U/L) was progressively higher across increasing values of oxLDL. A sensitivity analysis excluding individuals who were heavy drinkers (n=194), confirmed the oxLDL-GGT interaction for all-cause mortality (P<0.001). Of note, this interaction was specific because no similar effect modification existed for AST, ALT, CRP, smoking, alcohol consumption and other traditional or non-traditional risk factors (P ranging from 0.16 to 0.97).

An interaction analysis carried out to test the effect modification by oxLDL on the GGT-CV mortality link (Supplementary Table S4) showed a similar trend in age and sex adjusted Cox model (P for interaction=0.02) but this interaction just failed to achieve the formal statistical significance in both a fully adjusted analysis (P for interaction=0.08) and a sensitivity analysis (P=0.18) excluding heavy drinkers.

DISCUSSION

In this cohort study conducted in older persons living in the Chianti area of Italy, serum GGT levels associated with history of CV disease and predicted the risk for all-cause and CV death independently of other risk factors, including liver disease and alcohol

consumption. Furthermore, in a pre-specified interaction analysis circulating oxLDL amplified the effect of GGT on mortality.

Meta-analyses of studies in the general population and in high risk conditions (4, 5) coherently showed that GGT predicts an excess risk for death and fatal CV events. Importantly, the excess risk by GGT for these outcomes is largely independent of liver disease and alcohol consumption, i.e. the two major environmental factors responsible for raised GGT in human diseases. Of note the strength of the association between GGT and all-cause mortality was second only to that by age and CRP and the same outcome, further emphasising the relevance of non-traditional risk factors in the elderly (15). The vast majority of these studies were based on cohorts of young and middle-aged adults (4, 5). Until now just one community study specifically focused on an elderly population (the Rancho Bernardo study) (8). In this elderly population, GGT emerged as an independent predictor of all-cause and CV death. This observation contrasts with age-stratified analyses in the Minnesota Heart Survey where GGT was unrelated to CV death in people older than 70 years (9). Similarly, GGT failed to predict CV mortality in men older than 55 years in the British Regional Heart study cohort (12). Remarkably, an age-dependent attenuation of the health risk signalled by GGT was registered not only for the independent risk of CV death (16) but also for the incident risk of cancer (17). The age-dependent attenuation of the risk by GGT on major clinical outcomes suggests that the duration of exposure to this risk factor is critical to explain its link with adverse outcomes. In other words, shorter life expectancy in elderly people and competing risks by other diseases may prevent capturing any underlying link between GGT and mortality or CV disease in the elderly.

The InCHIANTI study is based on a cohort of prevalently healthy elderly people. Life expectancy in Tuscany (85 years for women and 80 years for men) is among the longest worldwide and the InCHIANTI study has a quite long follow up with a median time of observation of 9 years. In this prevalently healthy cohort in subjects free of liver disease at baseline, GGT emerged as coherent predictor of death and fatal CV events independently of traditional (Framingham) and non-traditional risk factors including alcohol intake as well as CRP (18) and homocysteine (19), two established predictors of adverse outcomes in the elderly. High GGT is considered as a major biomarker of non-alcoholic fatty liver disease (NAFLD) (20) which is seen as a manifestation of metabolic syndrome (21). However, both in our study and in the Rancho Bernardo study(8) the link between GGT and mortality was largely independent of BMI and other variables underlying the metabolic syndrome suggesting that the independent risk of GGT for adverse clinical outcomes may underlie mechanism other than the metabolic syndrome.

GGT plays a crucial role in oxidative processes favouring the cellular supply of glutathione (GSH), the major thiol antioxidant in human body (2). GGT is ubiquitously expressed to the cell-surface where it promotes the extracellular catabolism of GSH, allowing for precursor amino acids to be internalized and reused for intracellular GSH synthesis in a continuous “GSH cycling” across the plasma membrane (22). Accordingly, GGT associates directly with F2-isoprostanes, an established marker of oxidative stress (23) and inversely with serum antioxidants (24, 25). On the other hand, experimental data exists indicating that GGT

may per sé trigger the production of ROS via a sulphur di-aminoacid (cysteinyl-glycine) generated from GSH hydrolysis (22, 26).

Because GGT in atherosclerotic plaques co-localizes with oxLDL, this co-localization may be critical in the pathogenesis of atherosclerosis (6), a possibility supported by the observation that circulating GGT is bound to LDL (25). In a pre-specified interaction analysis we found an effect modification by oxLDL for the risk of death predicted by GGT levels. We observed a similar interaction for fatal CV events but, perhaps due to the relatively limited number of events, this effect just failed to achieve statistical significance (P=0.07). Such an interaction suggests that GGT levels may underlie a mechanism which amplifies the toxic effects of oxLDL on the vascular system or vice versa.

In conclusion, in this cohort study in a population of relatively healthy elderly people, GGT is directly associated with the incident risk of all-cause and CV death independently of a large set of potential confounders and circulating oxLDL amplifies the effect of GGT on all-cause but not CV mortality in older adults. This study supports the contention that GGT may have a role in oxidative stress-mediated adverse health outcomes in the elderly.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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REFERENCES

1. Kregel KC, Zhang HJ. An integrated view of oxidative stress in aging: basic mechanisms, functional effects, and pathological considerations. Am J Physiol Regul Integr Comp Physiol. 2007; 292(1):R18–R36. [PubMed: 16917020]
2. Whitfield JB. Gamma glutamyl transferase. Crit Rev Clin Lab Sci. 2001; 38(4):263–355. [PubMed: 11563810]
3. Huseby NE. Multiple forms of serum gamma-glutamyltransferase. Association of the enzyme with lipoproteins. Clin Chim Acta. 1982; 124(1):103–112. [PubMed: 6127175]
4. Kunutsor SK, Apekey TA, Seddoh D, et al. Liver enzymes and risk of all-cause mortality in general populations: a systematic review and meta-analysis. International Journal of Epidemiology. 2014; 43(1):187–201. [PubMed: 24585856]
5. Long Y, Zeng F, Shi J, et al. Gamma-glutamyltransferase predicts increased risk of mortality: a systematic review and meta-analysis of prospective observational studies. Free Radic Res. 2014; 48(6):716–728. [PubMed: 24684379]

6. Paolicchi A, Minotti G, Tonarelli P, et al. Gamma-glutamyl transpeptidase-dependent iron reduction and LDL oxidation--a potential mechanism in atherosclerosis. J Investig Med. 1999; 47(3):151–160.
7. Paolicchi A, Emdin M, Ghliozeni E, et al. Images in cardiovascular medicine. Human atherosclerotic plaques contain gamma-glutamyl transpeptidase enzyme activity. Circulation. 2004; 109(11):1440. [PubMed: 15037540]
8. Loomba R, Doycheva I, Bettencourt R, et al. Serum γ-Glutamyltranspeptidase Predicts All-cause, Cardiovascular and Liver Mortality in Older Adults. Journal of Clinical and Experimental Hepatology. 2013; 3(1):4–11. [PubMed: 23997501]
9. Lee DH, Buijsse B, Steffen L, et al. Association between serum gamma-glutamyltransferase and cardiovascular mortality varies by age: the Minnesota Heart Survey. Eur J Cardiovasc Prev Rehabil. 2009; 16(1):16–20. [PubMed: 18753951]
10. Fraser A, Thinggaard M, Christensen K, et al. Alanine aminotransferase, gamma-glutamyltransferase (GGT) and all-cause mortality: results from a population-based Danish twins study alanine aminotransferase, GGT and mortality in elderly twins. Liver International: Official Journal of the International Association for the Study of the Liver. 2009; 29(10):1494–1489. [PubMed: 19686309]
11. Strasak AM, Kelleher CC, Klenk J, et al. Longitudinal change in serum gamma-glutamyltransferase and cardiovascular disease mortality: a prospective population-based study in 76,113 Austrian adults. Arteriosclerosis, Thrombosis, and Vascular Biology. 2008; 28(10):1857–1865.
12. Wannamethee G, Ebrahim S, Shaper AG. Gamma-glutamyltransferase: determinants and association with mortality from ischemic heart disease and all causes. Am J Epidemiol. 1995; 142(7):699–708. [PubMed: 7572939]
13. Ferrucci L, Bandinelli S, Benvenuti E, et al. Subsystems contributing to the decline in ability to walk: bridging the gap between epidemiology and geriatric practice in the InCHIANTI study. J Am Geriatr Soc. 2000; 48(12):1618–1625. [PubMed: 11129752]
14. Steyerberg, E. Clinical prediction models: Modern estimation methods. NY: Springer-Verlag New York; 2009. p. 232-234.
15. Mukamal KJ, Kronmal RA, Tracy RP, et al. Traditional and novel risk factors in older adults: cardiovascular risk assessment late in life. Am J Geriatr Cardiol. 2004; 13(2):69–80. [PubMed: 15010653]
16. Ruttmann E, Brant LJ, Concin H, et al. Gamma-glutamyltransferase as a risk factor for cardiovascular disease mortality: an epidemiological investigation in a cohort of 163,944 Austrian adults. Circulation. 2005; 112(14):2130–2137. [PubMed: 16186419]
17. Strasak AM, Rapp K, Brant LJ, et al. Association of gamma-glutamyltransferase and risk of cancer incidence in men: a prospective study. Cancer Research. 2008; 68(10):3970–3977. [PubMed: 18483283]
18. Strandberg TE, Pitkala KH, Tilvis RS. Predictors of mortality in home-dwelling patients with cardiovascular disease aged 75 and older. J Am Geriatr Soc. 2009; 57(2):279–284. [PubMed: 19207143]
19. de Ruijter W, Westendorp RG, Assendelft WJ, den Elzen WP, de Craen AJ, le Cessie S, Gussekloo J. Use of Framingham risk score and new biomarkers to predict cardiovascular mortality in older people: population based observational cohort study. BMJ. 2009; 338:a3083. [PubMed: 19131384]
20. Ghouri N, Preiss D, Sattar N. Liver enzymes, nonalcoholic fatty liver disease, and incident cardiovascular disease: a narrative review and clinical perspective of prospective data. Hepatology. 2010; 52(3):1156–1161. [PubMed: 20658466]
21. Vanni E, Bugianesi E, Kotronen A, et al. From the metabolic syndrome to NAFLD or vice versa? Dig Liver Dis. 2010; 42(5):320–330. [PubMed: 20207596]
22. Paolicchi A, Dominici S, Pieri L, et al. Glutathione catabolism as a signaling mechanism. Biochem Pharmacol. 2002; 64(5–6):1027–1035. [PubMed: 12213602]
23. Lee DH, Jacobs DR Jr, Gross M, et al. Gamma-glutamyltransferase is a predictor of incident diabetes and hypertension: the Coronary Artery Risk Development in Young Adults (CARDIA) Study. Clinical Chemistry. 2003; 49(8):1358–1366. [PubMed: 12881453]

24. Lee DH, Gross MD, Jacobs DR Jr. Cardiovascular Risk Development in Young Adults S. Association of serum carotenoids and tocopherols with gamma-glutamyltransferase: the Cardiovascular Risk Development in Young Adults (CARDIA) Study. Clinical Chemistry. 2004; 50(3):582–588. [PubMed: 14726472]

25. Lim JS, Yang JH, Chun BY, et al. Is serum gamma-glutamyltransferase inversely associated with serum antioxidants as a marker of oxidative stress? Free Radical Biology & Medicine. 2004; 37(7):1018–1023. [PubMed: 15336318]

26. Drozdz R, Parmentier C, Hachad H, et al. gamma-Glutamyltransferase dependent generation of reactive oxygen species from a glutathione/transferrin system. Free Radical Biology & Medicine. 1998; 25(7):786–792. [PubMed: 9823544]

27. Paolicchi A, Emdin M, Passino C, et al. Beta-lipoprotein- and LDL-associated serum gamma-glutamyltransferase in patients with coronary atherosclerosis. Atherosclerosis. 2006; 186(1):80–85. [PubMed: 16112119]

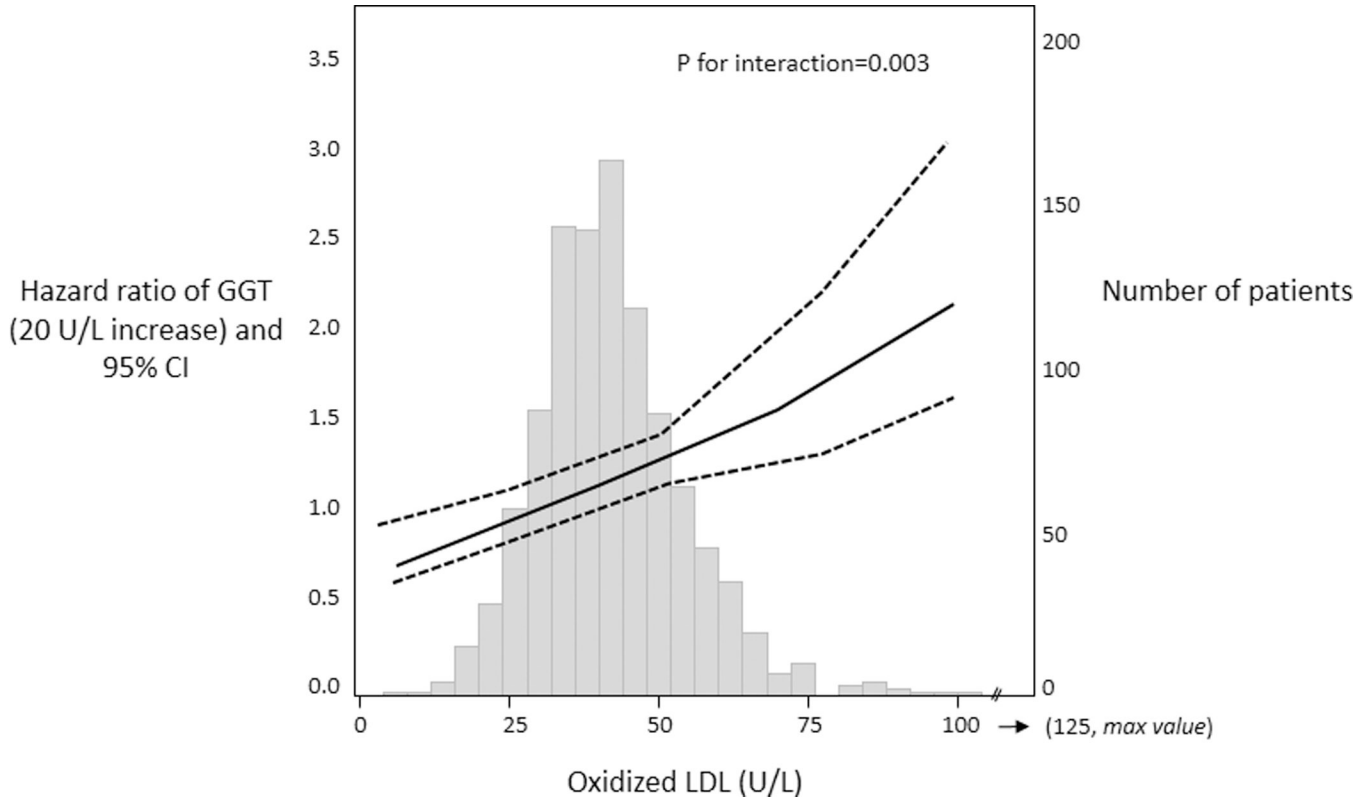

Figure 1.

Effect modification of oxLDL levels on the GGT-mortality link (adjusted for age, gender, smoking, BMI, LDL cholesterol, C reactive protein, systolic BP, alkaline phosphatase, hemoglobin, alcohol consumption, AST, ALT, homocysteine, diabetes, creatinine clearance and past CV events). The hazard ratio for all-cause mortality portended by a fixed increase (20U/L) in serum GGT is reported on the left scale. The continuous line represents the shape of the hazard ratios throughout oxLDL levels and the dotted lines the corresponding 95% CI. In the background the distribution of oxLDL is plotted and the number of patients corresponding to each column of the histogram is reported on the right scale.

Table 1

Main Demographic, Somatometric and Clinical Characteristics of the Study Population

	N=1038
Age (years)	74 (69–79)
Males (%)	454 (43)
Diabetes (%)	114 (11)
Smoking (%)	141 (14)
CV comorbidities (%)	152 (15)
Alcohol consumption (g/day)	8.8 (0–29.9)
BMI (kg/m^2)	27 (15–30)
Systolic BP (mmHg)	150±20
Diastolic BP (mmHg)	84±9
Lipid-lowering agents	45 (4)
Glucose (mg/dL)	89 (81–100)
LDL cholesterol (mg/dL)	136±34
Haemoglobin (g/dL)	13.7 ±1.4
C reactive protein (µg/mL)	2.8 (1.3–5.9)
Alkaline phosphatase (U/L)	203 (168–246)
Homocysteine (µmol/L)	14.5 (12.2–17.8)
GGT (U/L)	19 (14–28)
AST (U/L)	20 (17–23)
ALT (U/L)	17 (13–22)
oxLDL (U/L)	42±13
Creatinine clearance (ml/min/1.73m^2)	65±19

Past CV events were defined as the presence of at least one of the following documented comorbidities at enrolment: myocardial infarction, angina, peripheral vascular disease, stroke or coronary surgery/angioplasty.

Data are expressed as mean± SD, median and inter-quartile range or as percent frequency as appropriate.

Abbreviations: BMI=body mass index; BP=blood pressure; LDL=low-density lipoproteins; GGT= gamma-glutamyltransferase; AST= aspartate aminotransferase; ALT=alanine aminotransferase; oxLDL= oxidized low-density lipoproteins.

Table 2

Multiple Cox regression model of the oxLDL-GGT interaction for all-cause mortality

	Units of increase	All-cause mortality	
		Hazard ratio (CI 95%)	**P value**
GGT oxLDL GGT × oxLDL (*interaction term*)	20 U/L 1 U/L 20 U^2/L^2	**See** Figure 1	**P for interaction=0.003**
Age	1 year	1.13 (1.11–1.15)	0.001
Male gender		1.26 (0.98–1.63)	0.07
Current smokers	yes/no	1.91 (1.41–2.59)	0.001
BMI	1 Kg/m^2	1.01 (0.98–1.04)	0.59
LDL cholesterol	1 mg/dl	0.99 (0.98–0.99)	0.007
C reactive protein	1 μg/mL	1.01 (1.01–1.02)	0.006
Systolic blood pressure	1 mmHg	1.00 (0.99–1.01)	0.26
Alkaline phosphatase	1 U/L	1.00 (1.00–1.01)	0.005
Hemoglobin	1 g/dL	1.04 (0.96–1.12)	0.34
Alcohol consumption	1 g/day	0.99 (0.98–0.99)	0.04
AST	1 U/L	1.01 (0.99–1.04)	0.21
ALT	1 U/L	0.98 (0.96–0.99)	0.02
Diabetes	yes/no	1.17 (0.85–1.62)	0.33
Creatinine clearance	1 ml/min/1.73m^2	1.00 (0.99–1.01)	0.70
Past CV events	yes/no	1.43 (1.11–1.85)	0.006
Homocysteine	μmol/L	1.02 (1.01–1.03)	0.002

Abbreviations: GGT= gamma-glutamyltransferase; oxLDL=oxidized low-density lipoproteins; BMI=body mass index; LDL=low-density lipoproteins; AST= aspartate aminotransferase; ALT=alanine aminotransferase